IMPACT OF IRRIGATION WATER ON THE QUALITY OF FRESH PRODUCE

BY

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As the candidate's supervisor, I have approved this dissertation for submission.

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PREFACE

The experimental work described in this dissertation was carried out in the School of Biochemistry, Genetics and Microbiology; University of KwaZulu-Natal (Westville Campus), Durban, South Africa from January 2009 to December 2010, under the supervision of Professor B. Pillay and the co-supervision of Dr. A. O. Olaniran.

These studies represent original work by the author and have not otherwise been submitted in any form for any degree or diploma to any tertiary institution. Where use has been made of the work of others it is duly acknowledged in the text.

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ABSTRACT

The consumption of minimally processed fresh fruit and vegetables has increased over the past years, mostly because of consumers awareness that fresh produce serves as a good source of vitamins, minerals and fibre. Although fresh produce is important for the human diet it may provide an optimal environment for the growth and proliferation of pathogenic microorganisms, from cultivation to processing. Several outbreaks of disease, associated with the consumption of fresh produce, have been reported worldwide. In addition, fresh produce can become contaminated by heavy metals imposing a public health concern. One of the major sources of contamination is irrigation water, as it may contain pathogens and heavy metals from upstream operations. Irrigation water has been previously shown to be associated with the contamination of fresh produce. Therefore the objective of this study was to evaluate the microbial- and heavy metal- content of irrigation water used by local farmers in KwaZulu-Natal (KZN) over a 12-month period, in order to establish a link between the water quality and the safety of fresh produce, and to develop a suitable method to reduce the microbial contamination of fresh produce during both pre- and post-harvest phases. The microbial quality of the water and fresh produce samples was determined using the membrane filtration and standard spread-plate techniques, respectively. The heavy metal content of the water and fresh produce samples were analysed using inductively coupled plasma optical emission spectrophotometry (ICP-OES). Presumptive *Escherichia coli*, *Salmonella* spp., *Shigella* spp. and coliform counts in the water samples were high during the sampling period. Presumptive *E. coli* exceeded the DWAF limit of 2×10^3 cfu/100 ml for *E. coli* in irrigation water, in some instances. High counts of presumptive coliforms, *Shigella* spp. and *Campylobacter* spp. were recorded in the fresh produce, throughout the sampling period. The roots of the plant demonstrated the highest microbial and heavy metal contamination. Leafy vegetables such as spinach and lettuce were more contaminated than the other fresh produce sampled; for example, *Campylobacter* spp. exceeded 4.5×10^5 cfu/g in crisphead lettuce. With regard to the heavy metal content of the irrigation water and the fresh produce, mercury (Hg) exceeded the FAO and WHO limit of 0.001 mg/L, throughout the sampling period, with the highest concentration of 0.057 mg/L obtained from irrigation water. Since the concentrations of Hg in both the irrigation water and fresh produce were the highest during the same period, such as in winter, a clear link can be seen between the irrigation water and fresh produce. The method used during the pre-harvest phase, in order to reduce pathogens from produce, was the effect of *Pseudomonas aeruginosa* on the uptake of pathogens to the fresh produce. Inhibition assays were employed to determine whether *P. aeruginosa* could inhibit the pathogens (*E. coli, Listeria monocytogenes*, *Salmonella* spp. and *Shigella* spp.) tested. Only *L. monocytogenes* was found to be inhibited by *P. aeruginosa*. A greenhouse experiment was employed to prove that *P. aeruginosa* could prevent the uptake of this pathogen, via the roots, into the fresh produce by monitoring the concentration of *L. monocytogenes* in the soil and fresh produce by standard spread-plating. Denaturing gradient gel electrophoresis (DGGE) was also used to monitor the populations of *L. monocytogenes* and *P. aeruginosa* in the soil. Colony counts of *L. monocytogenes* decreased from 6 to 3.5 log cfu/g in the soil during the first 3 weeks of sampling. This decrease was confirmed by DGGE and suggested that this pathogen was inhibited by *P. aeruginosa* in the soil; hence, this pathogen was also not detected in the plant. During the post-harvest phase the effect of different treatment methods on the quality of the final produce was evaluated using tap water, NaCl, chlorine, hydrogen peroxide, blanching and ultra-violet (UV) light. UV light showed the most promise as the quality of this treated produce was better as compared to the other treated produce. A link between irrigation water qualities with that of produce was evident in this study as the highest microbial counts were recorded in summer for both the water and fresh produce samples. The pre-harvest method for the reduction of pathogens from the produce, which was the effect of *P. aeruginosa* on the uptake of pathogens to the produce, was limited as this organism had only inhibited *L. monocytogenes*, of the pathogens tested. Of the post-harvest treatment methods, UV treatment had caused the highest reduction in the microbial load of the fresh produce, with tap water treatment aiding in the survival of these presumptive pathogens. The presence of *P. aeruginosa* and the use of UV light in reducing microbial counts on fresh produce had both shown promise in this study. However, further studies need to be employed in order to optimise these methods before application. In addition, irrigation water should be routinely monitored and properly decontaminated, if necessary, to prevent the transmission of food-borne pathogens to crops. This may curb the problem of food-borne associated disease outbreaks world-wide as irrigation water has been shown, by the current study, as a link to the contaminated state of fresh produce.

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CHAPTER 1: INTRODUCTION AND LITERATURE REVIEW

1.1 INTRODUCTION

It has been estimated that the world's population is approximately 7 billion; however, this estimate is increasing every second, with the amount of people being born superseding the amount dying (Rosenberg, 2011). It has also been predicted that during the next decade, the world's population will be increased by approximately 73 million people every year, thus exerting more pressure on food suppliers. Therefore, meeting the food needs of growing populations will result in increasing incomes for the food industry (Pinstrup-Andersen *et al*., 1999). The food industry in South Africa received 7, 494 billion rand, through supplying the dietary needs of people, during the 4th quarter of 2008 (Statistics South Africa, 2008). The fresh produce industry in South Africa generated approximately R5,273 billion income for the 2006 financial year; with a large amount of farming income being generated by the Free State, KwaZulu-Natal (KZN) and Mpumalanga amounting to R2,043 billion, R1,654 billion and R1,576 billion, respectively (Statistics South Africa, 2006).

The consumption of minimally processed fresh fruit and vegetables has increased over the past years, mostly because consumers now have the knowledge of the benefits of living a healthy lifestyle as fresh produce serve as good sources of vitamins, minerals and fibres (Heaton and Jones, 2008; Yang *et al*., 2009). Scientific research through the last decades has revealed that a diet rich in fruit and vegetables allows for protection against several types of cancer and also lowers the incidence of coronary heart disease (WHO, 2007b). This has led to an increased demand for fresh, ready-to eat fruit and vegetables (Heaton and Jones, 2008; Yang *et al*., 2009). Furthermore, the need to preserve the natural flavour as well as the heat-labile nutrients found in fresh fruit and vegetables has also contributed to this demand for fresh produce (Slifko *et al*., 2000). However, it is important to also consider the risks related to the consumption of fresh minimally processed produce since the final produce do not usually contain any preservatives or anti-microbial agents and seldom undergo high temperature treatments prior to consumption. Therefore produce can provide ideal conditions for the transmission of infectious microorganisms, as well as, harbour high levels of toxic heavy metals (Mudgil *et al*., 2004; Seymour and Appleton, 2001).

Several factors are responsible for the contamination of fresh produce, such as, the type of manure or soil used (Amoah *et al*., 2005), the methods of transport as well as the handling of the produce (Amponsah-Doku *et al*., 2010; Drechsel *et al*., 2000; Sonou, 2001). However, the main source of contamination of fresh minimally processed produce in developing countries has been linked to the quality of irrigation waters used (Amponsah-Doku *et al*., 2010). The quality of the irrigation water used, the methods by which it is used, and also the type of crop grown influences the potential for contamination of the produce (Schneider *et al*., 2006).

1.2 FOOD-BORNE DISEASES AND THE ASSOCIATED MALAISES

Fresh produce are at a greater risk of being contaminated as compared to other food types because these produce are either consumed raw or they undergo minimal processing (Bassett and McClure, 2008). Such produce may retain most of it's microflora after undergoing minimal processing, some of which may be pathogens, thus creating a perturbing safety issue (Francis *et al*., 1999). Due to the present mass production and widespread distribution of food, food-borne disease outbreaks are on the increase, despite the fact that numerous technologies and/or strategies have been used on the issue of food safety (Hall *et al*., 2002; Chan and Chan, 2008). The effect of food-borne disease on humans depends on the health condition of the persons affected. Individuals (identified in literature as YOPI), such as, children, the aged, expectant women, as well as individuals with compromised immune systems, such as in the case of a person infected with human immunodeficiency virus (HIV), are severely affected. Food-borne disease may lead to very grave consequences, including death, for the latter persons (UN, 2007). The estimated financial cost of food-borne illnesses, on the person's affected, with regard to pain and suffering, decreased efficiency and medical costs lies within $10-83 billion per year. The WHO estimated that approximately 2 million children will die each year, in developing countries, as a result of food-borne contamination (WHO, 1996). The financial cost affecting developing countries, such as South Africa, is expected to be much greater. Therefore, food-borne disease outbreaks have become a major global concern (US-FDA, 2004).

Between 1992 and 2006, there were 9891 outbreaks of infectious intestinal diseases that had been reported to the Health Protection Agency (HPA), United Kingdom. Approximately, 23% of these outbreaks were associated with food-borne disease and 82% of this were related to the consumption of prepared salads (Little and Gillespie, 2008). Also, about four million cases of food-borne infectious diseases were reported to occur annually in Australia, there is still a threat of new emerging food-borne pathogens (Australian and New Zealand food authority, 1999). Recently, there were two major outbreaks of *Salmonella* that were linked to the consumption of tomatoes in the U.S.A (CDC, 2007). However, the most frequently encountered food-borne illness in the U.S.A, affecting over 2 million people, is gastrointestinal illness caused by *C. jejuni* (Heaton and Jones, 2008). There have been numerous outbreaks of this illness worldwide (Heaton and Jones, 2008). Ackers *et al*. (1998) reported community outbreaks of *E. coli* O157:H7 infections which had been linked to the consumption of lettuce, with illnesses in 70% of the patients examined. These patients developed bloody diarrhoea and abdominal cramps, due to the consumption of fresh produce infected with this bacterial pathogen (Ackers *et al*., 1998). *Salmonella* infections have also been a major cause of food-borne disease outbreaks world-wide (Smith *et al*., 2007). Therefore, it is evident that food safety, such as in the case of minimally processed fresh produce, during its processing and transport is a global matter, and microbial food-borne pathogens have been shown to be of prime concern (Hall *et al*., 2002).

1.3 MICROBIAL PATHOGENS COMMONLY FOUND ON FRESH PRODUCE

1.3.1 Bacterial pathogens

Fresh produce, as an important source of many nutrients, is used daily in a variety of food preparations, including salads and this provides an ideal environment for the growth and survival of many potential bacterial pathogens, such as *E. coli* (Heaton and Jones, 2008). Prepared salads has therefore served as the main vehicle in the transmission of diseases such as gastrointestinal infection and this has been highlighted by many large outbreaks of disease world-wide, during the last decade (Little and Gillespie, 2008). Fresh produce can be a direct source of food-borne illness because of the fact that these produce are most often eaten raw without any means of controlling or eliminating pathogens before consumption (UN, 2007). The surfaces of fresh produce have been shown to provide optimal conditions for many human pathogens to flourish (Heaton and Jones, 2008). Therefore, the bacterial contamination of fruit and vegetable plant tissues are mainly linked to the surfaces of these produce, whilst the inner tissue of these plants are most often considered sterile (Lund, 1992). However, it has been shown that the application of bacterial pathogens to the surfaces of the fresh produce would consequently result in their internalization over time (De Roever, 1998). Also, this internalization could offer protection to the pathogenic microbes from any post-harvest processing or decontamination step (Bihn and Gravani, 2006). It has been found that outbreaks of infection are increasingly being caused by pathogenic food-borne microorganisms, such as *C. jejuni* (Churruca *et al*., 2007), *E. coli* O157:H7 (Matthews, 2006), *L. monocytogenes* (UN, 2007), *Salmonella* spp. and *Shigella* spp. (Johnston *et al*., 2006).

1.3.1.1 *Campylobacter spp.*

The genus *Campylobacter* has 17 recognized species, some of which are important human and animal pathogens (Korczak *et al*., 2006). Campylobacters are known to be the causes of acute gastroenteritis (WHO, 1996). *C. jejuni* is recognized, worldwide, as a very important cause of food-borne illness and therefore members of this genus are considered as major concerns in the food industry (Churruca *et al*., 2007). The transmission of thermophilic campylobacters often occurs via the oral route. The most important reservoirs of campylobacters, includes birds as well as poultry, but members of this genera are also found in other domestic animals, such as cats, dogs, pigs and cattle (WHO, 1996). Campylobacteriosis, characterized by symptoms, including abdominal pain, fever, queasiness and diarrhoea, is a common disease that is also as a result of infection with certain species of this genus. In about 2-10% of these cases, the disease may eventually result with chronic health problems, which includes reactive arthritis and neurological disorders (WHO, 2007a). There is a range of culture media that are available for the detection of slow growing *Campylobacter* spp. over competitors. Members of this genus typically grow best between 37 °C and 43 °C. These bacteria can be identified based on their colonial morphology, microscopic appearance (Gram stain) and a positive oxidase reaction (Moore *et al*., 2005; Lucey, 2004).

1.3.1.2 *Escherichia coli*

E. coli is a member of the genus Enterobacteriaceae and most strains are inhabitants of the intestinal tract and are always present in faeces and therefore in faecally contaminated water (Francis *et al*., 1999; Barnes *et al*., 2007). The occurrence of *E. coli* in water has always been used as an indicator of potentially hazardous contamination requiring serious attention (WHO, 1996). A serogroup of *E. coli*, namely, O157:H7 has been recognized as the cause of severe intestinal diseases in humans (Williams *et al*., 2007). *E. coli* serogroup O157 causes various diseases, such as mild diarrhoea and haemorrhagic colitis, which is defined by blood-stained diarrhoea that usually occurs with the absence of a fever, but is accompanied by severe abdominal pain. This organism is also the causative agent of "the haemolytic uremic syndrome", which is most common in babies and young children and this disease is characterized by haemolytic anaemia and acute renal failure (WHO, 1996).

Some strains of *E. coli* are enteroinvasive (EIEC) and are able to produce dysentery and are also known to enter into the colonic mucosa, resulting in bloody diarrhoea. Enterotoxigenic *E. coli* (ETEC) may cause infants, children, and adults to acquire a cholera-like syndrome. "ETEC produce either a heat-labile enterotoxin (LT), related to cholera enterotoxin, or a heat-stable enterotoxin (ST); some strains produce both toxins". The potential of ETEC to cause infection is dependant not solely on enterotoxin production but also on their capacity to inhabit the small intestine (WHO, 1996). *E. coli* grows generally within 24 h at 37 °C. The laboratory isolation of ETEC requires its differentiation from other *E. coli* strains. Most *E. coli* O157 isolates do not ferment sorbitol so the incorporation of this substance into media is often used to differentiate these isolates from other *E. coli* strains (Lucey, 2004).

1.3.1.3 *Listeria monocytogenes*

L. monocytogenes is a very persistent, non-spore-forming, facultatively anaerobic pathogen and therefore is able to grow in low O_2 conditions (Maciorowski, 2007; Francis *et al*., 1999). This bacterium is harboured in animal intestines as well as in soil and water and causes human listeriosis, which is a severe illness that is often fatal. The effect of listeriosis includes, muscle aches, fever and serious gastrointestinal symptoms. The infection may reach the nervous system, in which case, the symptoms such as, "headaches, stiff neck, confusion, loss of balance, or convulsions" may also occur. During the past 2 decades, many outbreaks of human listeriosis have been associated with contaminated coleslaw which was prepared using raw cabbage in Canada (UN, 2007). Because of the consequences related to the consumption of food contaminated with this bacterium, many countries have enforced a zero tolerance level for the occurrence of *L. monocytogenes* in foodstuffs (UN, 2007; Curtis and Lee, 1995). For the recovery and isolation of *L. monocytogenes* from produce samples, medium such as Agar Listeria Ottaviani and Agosti (ALOA) were developed primarily as a selective and differential medium for *L. monocytogenes* (Ottaviani *et al*., 1997; Jantzen *et al*., 2006).

1.3.1.4 *Salmonella* spp.

Salmonella spp. are Gram negative, rod-shaped bacteria that belong to the family Enterobacteriaceae. The genus comprises five pathogenic strains namely *S. enterica* serovar Typhimurium, *S. enteriditis*, *S. Heidelberg*, *S. saint-paul* and *S. Montevideo* (Barnes *et al*., 2007; Francis *et al*., 1999). These bacteria grow generally within 24 h at 37 °C. The detection of these genera requires a combination of enrichment media and biochemical testing (Lucey, 2004). *Salmonella* spp. are usually transmitted through the consumption of contaminated foodstuffs (WHO, 2007a). These bacteria are frequently reported as causes of food-borne disease outbreaks; and are found within the intestinal tracts of infected humans and animals. They cause salmonellosis, which is a condition resulting in "diarrhoea, abdominal cramps and fever within 8 to 72 hours after ingestion of the contaminated food" (UN, 2007).

1.3.1.5 *Shigella* spp.

Shigella spp. are known as Gram-negative, non-spore-forming and non-motile bacterial rods (WHO, 1996). This organism is another pathogen of the family Enterobacteriaceae (Barnes *et al*., 2007). These bacteria grow generally within 24 h at 37

°C. The isolation of these bacteria requires using selective media, serology and a biochemical profile (Lucey, 2004). Infection by these bacteria is characterized by bloody diarrhoea as a result of invasion of the colonic mucosa. There have been reasons to suggest that this type of infection process is highly species specific. Of the enteric bacterial pathogens, *Shigella* spp. appears to be the most adapted to infect humans. The usual route of infection is through the direct transmission between vulnerable individuals (WHO, 1996). Muller *et al*. (2009) reported an outbreak of *Shigella sonnei* which involved ten cases in Denmark in April and May. The most likely source of this outbreak was the consumption of fresh, raw sugar peas that had been imported from Africa (Muller *et al*., 2009).

1.3.2 Survival mechanisms of bacteria

Many different processes have been studied for their effectiveness in the removal of pathogens from fresh produce; however, these microbial pathogens have been able to survive such removal processes (Chang and Fang, 2007). Bacteria have been shown to survive unfavourable conditions, such as in the case of *S. enteritidis* where low temperatures have been noted to cause a reduction in the generation rate of this organism; but, however, did not inhibit its growth (Rezende *et al*., 2009). Also, the growth and survival of *S. typhimurium* and *E. coli* O157:H7 on fresh produce (lettuce) within a shelf life of 10-12 days had shown the survivability of these food-borne bacterial pathogens (Chang and Fang, 2007). The question, however, is how did these microorganisms survive? Chaveerach *et al*. (2003) reported that *Campylobacter* species may enter into a viable but non-culturable (VBNC) state under acidic conditions. The conditions under which plants develop imposes extrinsic factors which may manipulate the survival as well as the growth of microbes, while intrinsic factors, such as, the nature of the protective cuticle and epithelium, tissue pH, and the occurrence of antimicrobials can dictate which fresh produce are more likely than others to harbour certain types of microbes in injured tissues. The behaviour of some microbial pathogens may be altered by the presence of soil or faecal material on fresh produce surfaces, which may seep into cut tissues and thereby alter the ecological environment, where these pathogens are present. Also, the growth of moulds in these settings could possibly result in an increased pH, thus enhancing the likelihood of growth of pathogenic bacteria (Beuchat, 2002). Outbreaks of disease in humans have commonly been connected with higher pH of fresh produce which suggests that there may be a relationship between the presence of pathogens at the time of consumption and the pH of the fresh produce (Bassett and McClure, 2008). It has also been shown that microbial penetration of fresh produce is enhanced if the temperature of the fresh produce, itself, is higher than that of its environment (Beuchat, 2002). Therefore, there are specific mechanisms that these organisms can use to ensure their growth and survival under unfavourable conditions. Some of which are discussed below:

1.3.2.1 Viable but non-culturable (VBNC) state

Previously, it had been proposed that some culturable bacteria may enter into a "long-term survival state", when they have been subjected to prolonged starvation or some other stress. This means of survival displayed by bacteria is termed the viable but non-culturable (VBNC) state. In this state, bacteria are not detected by culturable testing (Bogosian and Bourneuf, 2001). Liu *et al*. (2009) performed a study which showed the viability and possible health risks of *E. coli* O157:H7 VBNC cells and also that a combination of starvation with either low temperature or osmotic pressure, allowed for the induction of *E. coli* O157:H7 into a VBNC state, however, it was found that starvation alone did not induce this bacterium into a VBNC state. Ziprin *et al*. (2003) demonstrated that *C. jejuni* had entered into the VBNC state upon suspension of the cells in sterile distilled water with cell viability determined with tetrazolium violet. Besnard *et al*. (2002), determined which "environmental and physico-chemical factors" induce the VBNC state in the food-borne pathogen, *L. monocytogenes*. It was found that in the dark, the incubation temperature was the main factor in the formation of VBNC bacteria. However, natural sunlight quickly produced the VBNC state in *L. monocytogenes* cells. The presence of VBNC *L. monocytogenes* cells, as well as other pathogens that can enter into this state, could possibly pose a major problem since they cannot be detected by traditional culturing methods (Besnard *et al*., 2002).

1.3.2.2 Biofilm formation

Another survival mechanism that may be used by bacteria is the formation of biofilms. During growth and maturation of fresh produce as well as during harvesting, transport, processing, and storage after processing, opportunities arise for the establishment of biofilms. These biofilms can provide protection to individual bacterial cells, due to several structural features, which allow for further development of the biofilm (Beuchat, 2002). Bacteria are able to use biofilm formation as a means of survival, as bacteria appear to instigate biofilm formation in response to various environmental conditions, for instance nutrient availability. They maintain their growth in biofilms provided that there is a fresh supply of nutrients, but they begin to detach from the biofilm's surface as soon as the nutrients have been depleted, they then enter in their planktonic form of growth (Harshey, 2003).

Biofilms are, therefore, described as the growth of surface-associated layers of microbial populations that are matrix-embedded (Ponsonnet *et al*., 2008). Biofilms are comprised of hundreds of cells, of which each cell encounters its own microenvironment owing to chemical gradients which are established by metabolism and diffusion (Teal *et al*., 2006). A number of adherent bacteria occur in natural settings as surface-attached biofilms, and are enclosed within a self-produced extracellular matrix that protect these bacteria from hostile environmental settings (Lebeer *et al*., 2007). Biofilms have been shown to have the ability to influence the efficiency of strategies that are used to control food-borne pathogens on fresh produce. Biofilm formation strengthens the adhesion of these pathogens and thus, provide protection against disinfection after the storage of the contaminated produce (Lapidot *et al*., 2006). The growth of *L. monocytogenes*, in a multi-species biofilm with resistance to sodium hypochlorite, has been previously demonstrated by Norwood and Gilmour (2000). The formation of biofilms may encourage conditions that will protect against death or promote the growth of these pathogenic microbes (Beuchat, 2002). The presence of biofilms have been, previously, observed on the surfaces of many leafy fresh produce, including chinese cabbage, spinach, celery, lettuce, endive, basil and parsley (Buck *et al*., 2003; Morris *et al*., 1997).

1.3.3 Viral pathogens

Fresh produce, apart from sustaining the growth and survival of numerous pathogenic bacteria, can also support the survival of human or animal viruses (Seymour and Appleton, 2001; Ward *et al*., 1982). Food may be contaminated by human or animal viruses through primary contamination (due to the virus being present at harvest time) or secondary contamination (introduced during processing, storage and distribution of the produce) (Ward *et al*., 1982). Viral-contaminated fresh produce are increasingly being recognized as the causes of food-borne viral diseases (Croci *et al*., 2008). Viruses, unlike bacteria, are not able to multiply in or on foodstuffs but they sometimes may be present on fresh produce and remain infectious. Viruses may be present on the surfaces of fresh fruit and vegetables as a consequence of faecal contamination. Several groups of viruses contaminate fresh produce but the main food-borne viral pathogens are those that are known to cause infection via the gastrointestinal tract, such as the gastroenteritis viruses (Seymour and Appleton, 2001). The viruses that are most commonly encountered on fresh vegetables such as, cauliflower, lettuce, potato, peas, pepper and tomatoes, include the cucumber mosaic virus, bean yellow mosaic virus and the tobacco mosaic virus (Masuka *et al*., 1998).

Temperature is a key factor that influences the survival of viruses, depending on the type of virus, with low temperatures favouring their survival. There is evidence that "suggests that the adsorption of viruses to particulate matter and sediments confers substantial protection against inactivating influences". pH or salinity does not appear to affect the survival of most non-enveloped viruses. Enteric viruses are even capable of surviving in the gastrointestinal tract and are therefore known to be acid stable. It is therefore more probable that they will be able to survive low pH processes that are inhibitory to bacterial contaminants and thereby remain infectious (Seymour and Appleton, 2001).

1.4 COMMON HEAVY METAL CONTAMINANTS OF FRESH PRODUCE

In addition to pathogenic microorganisms, chemical contaminants are also of concern, with regards to public health safety (Qadir *et al*., 2008). Chemicals have the ability to cause serious heath risks to consumers if they are able to contaminate fresh produce at significant concentrations (above the acceptable daily intake (ADI)). The contamination of such produce may occur by way of either "naturally occurring substances or by synthetic chemicals" which may be added or which are present during production or processing of these produce (UN, 2007).

Micronutrient elements are known to be necessary for plant development and human nutrition; however, some of these elements, such as copper, chromium, molybdenum, nickel, selenium or zinc can be lethal to both animals and humans at higher concentrations. Other trace elements, such as, Arsenic (As), cadmium (Cd), mercury and lead (Pb) may also be present in fresh produce (McLaughlin *et al*., 1999). Heavy metals are extremely dangerous as a result of their "non-biodegradable nature, long biological half-lives and their potential to accumulate in different body parts". Even low concentrations of heavy metals have detrimental effects on humans and animals because there is no effective mechanism for their removal from the body (Arora *et al*., 2008). For example, Cd is a non-essential element, known to cause harmful effects even at very low levels and may be easily taken up from the soil by plants and can accumulate at high levels (Yang *et al*., 2009).

Heavy metals can also accumulate easily in the edible portions of leafy vegetables (Arora *et al*., 2008), however, the absorption as well as the accumulation of these metals in fresh produce may depend on various parameters, such as, temperature, humidity, pH and nutrient availability (Sharma *et al*., 2007). Consuming heavy metal contaminated fresh produce is therefore of serious health concern, as it can use up some vital nutrients in the body causing a decline in immunological defences, intrauterine growth retardation, impaired psycho-social behaviour, disabilities related to malnutrition and a high prevalence of upper gastrointestinal cancer (Sharma *et al*., 2007; McLaughlin *et al*., 1999). It is therefore very important to identify chemical hazards that are applied to fresh produce.

Toxic substances, such as pesticides, are used in pest control to protect developing crops from harmful insects or competitive weeds or to remove potential vectors of disease. Pesticides can be very harmful to both the environment and human beings and can even symbolize a chemical hazard for consumers, when these produce are unintentionally contaminated by such pesticides (UN, 2007). Besides the fact that pesticides may be a chemical hazard, some of these pesticides can also support the growth of some bacterial species (Ng *et al*., 2005). Pesticides reconstituted using sterile water, to their recommended concentrations, have been shown to support the survival and growth of the inoculated species of *Pseudomonas*, *Salmonella* and *E. coli*, while some of the pesticides reconstituted in various sources of irrigation water (bore, dam and river) were able to support the growth of the bacterial species that were present in the different water types. The most predominant bacterial species in these waters prior to and following storage varied as this was dependant on the water source, however, species of *Pseudomonas*, *Acinetobacter*, *Aeromonas* and various coliforms displayed significant growth (Ng *et al*., 2005). Therefore, the quality of the irrigation water that is applied to crops can serve as a source of pre-harvest contamination (De Roever, 1998). With the intention of reducing the concentration of toxic heavy metal contaminants (e.g. As, Cd, Pb) in irrigation water, standards have also been established for these metals in irrigation water (Table 1.1).

1.5 IRRIGATION WATER AS A SOURCE OF FRESH PRODUCE CONTAMINATION

Irrigation water of debatable quality can be a direct cause of contamination of fresh produce (Gast and Holt, 2000). Whenever irrigation water is collected and then used, there is always the likelihood of pathogens getting into this water and thereafter spreading these pathogens to plants (Fischer, 2004). The quality of irrigation water is therefore imperative since the water comes into direct contact with the edible portions of fresh produce (Schneider *et al*., 2006).

Table 1.1: Recommended maximum concentrations of trace elements in irrigation water (Ayers and Westcot, 1985).

Element	Recommended Maximum Concentration (mg/L)	Remarks
Al (aluminium)	5.0	Can cause non-productivity in acid soils (pH < 5.5), but more alkaline soils at pH > 7.0 will precipitate the ion and eliminate any toxicity.
As (arsenic)	0.10	Toxicity to plants varies widely, ranging from 12 mg/L for Sudan grass to less than 0.05 mg/L for rice.
Be (beryllium)	0.10	Toxicity to plants varies widely, ranging from 5 mg/L for kale to 0.5 mg/L for bush beans.
Cd (cadmium)	0.01	Toxic to beans, beets and turnips at concentrations as low as 0.1 mg/L in nutrient solutions. Conservative limits recommended due to its potential for accumulation in plants and soils to concentrations that may be harmful to humans.
Co (cobalt)	0.05	Toxic to tomato plants at 0.1 mg/L in nutrient solution. Tends to be inactivated by neutral and alkaline soils.
Cr (chromium)	0.10	Not generally recognized as an essential growth element. Conservative limits recommended due to lack of knowledge on its toxicity to plants.
Cu (copper)	0.20	Toxic to a number of plants at 0.1 to 1.0 mg/L in nutrient solutions.
Fe (iron)	5.0	Not toxic to plants in aerated soils, but can contribute to soil acidification and loss of availability of essential phosphorus and molybdenum. Overhead sprinkling may result in unsightly deposits on plants, equipment and buildings.
Li (lithium)	2.5	Tolerated by most crops up to 5 mg/L; mobile in soil. Toxic to citrus at low concentrations (<0.075 mg/L). Acts similarly to boron.
Mn (manganese)	0.20	Toxic to a number of crops at a few-tenths to a few mg/L, but usually only in acid soils.
Mo (molybdenum)	0.01	Not toxic to plants at normal concentrations in soil and water. Can be toxic to livestock if forage is grown in soils with high concentrations of available molybdenum.
Ni (nickel)	0.20	Toxic to a number of plants at 0.5 mg/L to 1.0 mg/L; reduced toxicity at neutral or alkaline pH.
Pd (lead)	5.0	Can inhibit plant cell growth at very high concentrations.
Se (selenium)	0.02	Toxic to plants at concentrations as low as 0.025 mg/L and toxic to livestock if forage is grown in soils with relatively high levels of added selenium. An essential element to animals but in very low concentrations.
Sn (tin) Ti (titanium) W (tungsten)	----	Effectively excluded by plants; specific tolerance unknown.
V (vanadium)	0.10	Toxic to many plants at relatively low concentrations.
Zn (zinc)	2.0	Toxic to many plants at widely varying concentrations; reduced toxicity at pH > 6.0 and in fine textured or organic soils.

It has been acknowledged that the use of water with high levels of enteric bacteria and/or viruses results in an increase in the occurrence of pathogen isolations from harvested produce (De Roever, 1998). Islam *et al*. (2004b) investigated the source of vegetable crop contamination by pathogens in the field. *S. enterica* serovar Typhimurium was added to the irrigation water at 10^5 cfu/ml, in order to determine the persistence of salmonellae in soils, that had been irrigated with this artificially contaminated irrigation water. The contamination on leaf lettuce and parsley grown on such treated soil was also investigated in this study. The contaminated irrigation water was applied once on the plants. The results suggested that the contaminated water played an important role in the occurrence of *Salmonella* on the vegetables and survival in soil for an extended period of time (Islam *et al*., 2004b). Majority of studies have indicated that contamination of crops most likely occurs through direct contact between crops and contaminated water (Stuart, 2006). Avery *et al*. (2008) showed the survival of *E. coli* O157:H7 in the following water types: lake, puddle, river, and animal-drinking trough waters. The population of *E. coli* O157:H7 had declined with time in all the water types tested; however, the cells were still present in 45% of the samples after a period of 2 months. Of the water types tested, nutrient concentrations were the highest in the faecally polluted puddle waters and lowest in lake waters. The survival of *E. coli* 0157:H7 was greater in these two contrasting water types, it was hypothesized that the bacteria may be using different survival mechanisms whilst in these two water types, such as the ability of the bacteria to utilize nutrients may sustain populations for longer periods of time in high nutrient faecally contaminated puddle water, while low nutrient conditions (such as lake waters) may bring about metabolic dormancy (Avery *et al*., 2008). Irrigation water is also a major source of heavy metal contamination of fresh produce. Wastewater that had been mixed with industrial effluent was used for irrigation of vegetables growing in the area of Korangi in Karachi (Pakistan); this water was tested for its heavy metal content, as well as plant samples (Spinach). It was shown that both the irrigation waters and the plant samples tested had greater concentrations of many heavy metals (zinc, iron, manganese, cadmium, nickel, lead) then the recommended concentrations (Saif *et al*., 2005). The list of pathogenic viruses which can occur in polluted water and the diseases attributed to them are represented in Table 1.2.

Table 1.2: Viruses pathogenic to humans which can occur in polluted water and diseases attributed to them (WHO, 1996).

Virus family	Members	No. of serotypes	Diseases caused
Picornaviridae	Human polioviruses	3	Paralysis, meningitis, fever
	Human echoviruses	32	Meningitis, respiratory disease, rash, fever, gastroenteritis
	Human coxsackie-viruses A1-22, 24	23	Enteroviral vesicular pharyngitis, respiratory disease, meningitis, enteroviral vesicular stomatitis with exanthema (hand, foot and mouth disease)
	Human coxsackie-viruses B1-6	6	Myocarditis, congenital heart anomalies, rash, fever, meningitis, respiratory disease, epidemic myalgia (pleurodynia)
	Human enteroviruses 68-71	4	Meningitis, encephalitis, respiratory disease, rash, acute enteroviral haemorrhagic conjunctivitis, fever
	Hepatitis A virus	1	Hepatitis A
Reoviridae	Human reoviruses	3	Unknown
	Human rotaviruses	5	Gastroenteritis, diarrhoea
Adenoviridae	Human adenoviruses	41	Respiratory disease, conjunctivitis, gastroenteritis
Parvoviridae	Adeno-associated viruses	4	Latent infection following integration of DNA into the cellular genome
Caliciviridae	Human caliciviruses	5	Gastroenteritis in infants and young children
	Small round structured viruses (including Norwalk virus)	14	Gastroenteritis, acute viral gastroenteropathy (Winter vomiting disease)
Caliciviridae (?)	Hepatitis E virus	?	Hepatitis E
Unknown	Astroviruses	1	Gastroenteritis, neonatal necrotizing enterocolitis
Papovaviridae	Papillomaviruses	2	Plantar warts

The availability of water is often a critical issue and therefore little notice is given to the microbiological quality of water used for irrigation purposes (Gerba and Choi, 2006). Together with the energy costs that the farmers have to bear, the availability of water often leads the farmers to make choices regarding the type of crops to produce, methods of irrigation that are to be used and the source of water to be used for irrigation (Suslow *et al*., 2003). In areas that have a shortage of water, the available sources of water are subjected to contamination by various factors such as sewage discharge from rural communities, cattle feedlot drainage, grazing animals along the water way, storm-water events and also the return of irrigation water (that is excess water that has been applied to crops that returns into the irrigation system) (Gerba and Choi, 2006). An example is the drainage and run-offs from animal pens after it has rained, leading to the contamination of irrigation water sources (De Roever, 1998). Because the irrigation channels used are commonly small, such changes may cause the quick deterioration of the irrigation water quality (Gerba and Choi, 2006). Farmers may not choose which irrigation water to use on the basis of its quality but rather on the availability of water supplies. In addition, a farmer may interchange water sources for irrigation purposes during a season, periodically with the use of available surface water (Suslow *et al*., 2003).

1.5.1 Sources of water used for irrigation

Whenever water is able to come into contact with produce, the quality of the water, itself, may determine the probability of direct pathogen contamination (UN, 2007). Irrigation waters can be of variable quality, ranging from potable to surface water from different sources such as rivers, streams, ponds, lakes, reservoirs, groundwater from wells, rural water, irrigation ditches and open canals (Bihn and Gravani, 2006; Gast and Holt, 2000).

Some human enteric viruses may be found in sources such as, “septic discharges, leaking sewer lines, or infiltration from lakes, rivers, and oxidation ponds” (Gerba and Choi, 2006). Shaban and Malkawi (2007) used molecular techniques such as reverse transcriptase polymerase chain reaction (RT-PCR) to discover the presence of viruses in different water samples. Water samples such as house ground reservoirs and wastewater that were used for irrigation purposes, revealed the existence of Adenoviruses as well as the Enterovirus group (Shaban and Malkawi, 2007). Abedin *et al*. (2002) showed that the long-term usage of heavy metals, such as As, contaminated groundwater, when applied to crops had resulted in high soil arsenic levels in Bangladesh. Roychowdhury *et al*. (2005) further revealed that the concentrations of As in different portions of plants increased with both an increase in ground water and soil As levels. Therefore, it has been postulated that “contaminated irrigation water has the potential to transmit both chemical and biological hazards to fresh produce” (Schneider *et al*., 2006). Various water sources, for example, rivers, creeks and streams can be sources of contamination as they may contain contaminants from upstream operations, and the use of such waterways for irrigation purposes could ultimately lead to the contamination of crops (Suslow *et al*., 2003). The various sources of water used for irrigation purposes are discussed below:

1.5.1.1 Dam water

Certain countries, for example South Africa, that generally have a dry climate depend on dam waters in order to have continuous irrigation water supplies throughout the four seasons of the year (WRC, 2009). Ahmed *et al*. (2004) tested water samples collected from various dams in Pakistan for the presence of bacteria using the heterotrophic plate count and most probable number methods. The study revealed the presence of pathogenic bacteria, such as, *E. coli*, *Salmonella* spp. and *Shigella* spp (Ahmed *et al*., 2004). In the Nkonkobe district, situated in the Eastern Cape Province of South Africa, surface water was collected from different water sources including the Lenge dam, in order to study the presence of enteric pathogens. This water source was found to have tested positive for *Salmonella* spp. (Momba *et al*., 2006). In farm dams, the ionic composition of the water is likely to echo that of the inward flowing waters. The factors that more frequently alter the concentration of the dam water along with the ionic composition of these waters are evapo-concentration as well as the interference of groundwater. However, the final quality of dam water is a consequence of interactions involving the composition of rainwater, different weathering processes in the catchment and groundwater run-offs (Brainwood *et al*., 2004).

1.5.1.2 Groundwater

Groundwater is a very important source of irrigation water (The groundwater foundation, 2009). It has been alleged that groundwater is less prone to contamination as compared to surface water as groundwater loses most of its microbial load and organic compounds following its natural filtration through rock and clay layers of soil (UN, 2007). However, under some circumstances, groundwater may become contaminated either by surface water or persistent chemicals and other substances present within the soil, itself (UN, 2007). For instance, pesticides and fertilizers may find a way into groundwater supplies with time. Road salt, lethal substances from mining sites, as well as second-hand motor oil may also leach into these supplies. Additionally, unprocessed waste from septic tanks along with poisonous chemicals from underground tanks may contaminate groundwater (The groundwater foundation, 2009).

1.5.1.3 River water

Rivers, creeks, and streams can hold pathogenic microorganisms from upstream activities, such as livestock operations and this source of irrigation water could lead to crop contamination (Suslow *et al*., 2003). Soderstrom *et al*. (2005) associated an outbreak of *E. coli* O157:H7, which occurred in Sweden in 2005, to the consumption of lettuce that was irrigated with water from a river which had been contaminated by cattle faeces. Olaniran *et al*. (2009) conducted a study to investigate the microbiological quality of two rivers in Durban, South Africa, using total coliform and faecal coliform populations as indicators. The results indicated that these water sources were of poor microbiological quality and were not suitable for human consumption. Cai *et al*. (1995) showed that river water used for irrigation in China was contaminated with Cd, from the tailings of the tungsten ore dressing plants and wastewater. It was further deduced in this study that the local people had been exposed to this contamination by Cd for not less than 25 years. According to a meal survey, it was estimated that 99.5% of Cd that was taken in orally had come from growing vegetables and rice, locally. In this case the Cd exposure was within a range that could cause adverse renal effects, with long term exposure (Cai *et al*., 1995).

1.5.1.4 Wastewater

Some countries do not possess the capacity to efficiently treat wastewater before its disposal, therefore large volumes of untreated wastewater end up in urban water bodies, which farmers use for irrigation (Keraita *et al*., 2007). Wastewater is mainly used for crop irrigation, because of its availability, low costs associated with its use, removal problems and the shortage of fresh water (Arora *et al*., 2008). The potential for contamination has increased widely over the years as untreated wastewater has been applied to crops. Wastewater used for crop irrigation has been shown to contain a very high concentration of pathogenic microbes (Heaton and Jones, 2008). Sewage-contaminated irrigation water had been previously linked to hepatitis A disease outbreaks which had been linked to the consumption of contaminated lettuce (Seymour and Appleton, 2001). Wachtel *et al*. (2002) described the contamination of cabbage plant roots irrigated with sewage-polluted stream water by *E. coli*; however, the edible portions of the cabbage plant were not affected. In addition, human enteric viruses have the potential to survive in any water source that has been contaminated by human faeces or by sewage (Seymour and Appleton, 2001). In addition, the use of such water for irrigation purposes, significantly contributes to the heavy metal content of soil (Arora *et al*., 2008).

Industrial wastewater regularly contains increased amounts of metals, metalloids, and volatile or semi-volatile components (Qadir *et al*., 2008). A long-term problem with the use of wastewater for irrigation is the possibility of toxic materials, present in these waters, to accumulate in the soil over years of irrigation. It is also possible that these toxic materials (heavy metals) could accumulate in the soil to such a level that it would be taken up by the plant material, which in turn would accumulate these metals at concentrations that are extremely toxic to man upon consumption of the plant material (WHO, 1989). Wastewater, used for irrigation, has been observed to lead to the accumulation of substantial amounts of toxic heavy metals in vegetables such as mint and spinach (Arora *et al*., 2008).

Since processed wastewater has an elevated nutritive value that may perhaps improve the growth of the plant, decrease fertiliser application rates, and increase efficiency of poor fertility soils, it has been suggested that treated wastewater may be used to irrigate tomatoes that are consumed cooked, but not for samples that are eaten raw, with the constant monitoring of the effluent quality from the treatment plant in order to avoid contamination (Al-Lahham *et al*., 2003). However, the degree of contamination is reliant on both the method of irrigation used and the type of produce, itself (De Roever, 1998), as discussed in the following section:

1.5.2 Irrigation methods

Efficient utilization of water for irrigation is of principal importance, in order to sustain agricultural development, therefore, different methods have been introduced in order to improve the utilization of water as well as to conserve it (Narayanamoorthy, 2004). Irrigation water is delivered to the plants by way of using both overhead and surface (flood irrigation, drip irrigation, sprinkler irrigation and sub-irrigation) methods (Bihn and Gravani, 2006; Ilic *et al*., 2009). The choice of the irrigation process intended for use ultimately plays a chief role in the transmission of contaminants from the irrigation water to fresh produce on the field. In addition, irrigation methods that do not apply water directly to the plant may allow for a lower risk of contamination of the produce. The utilization of surface irrigation methods as opposed to overhead irrigation methods have resulted in increased crop yields and a reduction in plant diseases. However, because of the high costs attributed to the use of these methods, farmers still use alternate irrigation methods (Bihn and Gravani, 2006).

1.5.2.1 Basin Irrigation

Using this type of irrigation method, the water is applied swiftly to moderately level plots bordered by levees. The basin is a minute check. The fields that are irrigated using this type of system are divided into level rectangles; however, a particular flow depth must be retained. The entire field is then flooded and the irrigation water is allowed to penetrate the roots of the plants after beating on to the soil surface (Karami, 2006). The disadvantage with this technique is that because of their level surface, it is sometimes difficult to drain surplus irrigation water rapidly from the basins (Maqsood and Cheema, 2005).

1.5.2.2 Border irrigation

Border systems are much like basin irrigation systems with the exception of the presence of a gradient down the perimeter and there may be a slight cross slope. The irrigation water is applied to row crops in ditches flanked by rows made by tillage implements to allow for the irrigation water flow in a single direction (Karami, 2006). This type of irrigation technique is usually suited for largely mechanized farms as the use of this method is intended to create long continuous field lengths which allows for the ease of machine operations. Borders may be about 800 m or greater in length and about 3-30 m wide but this depends on a range of factors. Border irrigation is not suitable for small-scale farms that involve either hand labour or animal-powered methods of cultivation (Brouwer *et al*., 1988).

1.5.2.3 Drip irrigation

The drip irrigation method is considered to be one of the best methods as it allows for the dripping of irrigation water slowly into the soil using a system of tiny plastic pipes, which is fitted with an outlet called a dripper or an emitter (Korkmaz, 2009). Unlike the flood irrigation method drip irrigation allows for the supply of water directly to the roots of crops, thereby reducing the amount of evaporation and losses of water (Narayanamoorthy, 2004). Drip irrigation is more efficient (90%) than sprinkler systems but requires an expensive installation. Drip irrigation is the most suitable irrigation method for the use of water of poor quality as it can decrease the incidence of disease in plants, which is related to high moisture levels. This technique is also reliable for areas where water is scarce (Korkmaz, 2009).

1.5.2.4 Flood irrigation

Flood irrigation involves the movement of water over and across the agricultural land, by simple gravity flow, with the purpose of wetting and infiltrating the soil. This is the most cost effective method, if the landscape is favourable and the farmers can afford a pump. However, the utilization of water is of low efficiency, making this method of irrigation only appropriate when water is not a limiting factor (Qadir *et al*., 2008). Fischer (2004) suggested that less than 10% of the floodwater is actually used up by plants and the other 90% of the water may be returned for reuse. This high volume of water that moves through the flood system dilutes the concentrations of any pathogens that might be present. However, it has been proven that even a single bacterial cell may cause infection, but the risk of the spread of disease, although not eliminated, may be reduced in this way (Fischer, 2004). Solomon *et al*. (2002) showed using a laser scanning confocal microscope and the cells of *E. coli* O157:H7/pGFP (green fluorescent protein), that lettuce grown by flood irrigation with contaminated water may subsequently result in the contamination of the edible portion of the fresh produce. The results from this study also suggested that the edible portions of any plant can become contaminated, through the movement of the pathogen into the root system of the plant, without the direct exposure of the plant to the pathogen (Solomon *et al*., 2002).

1.5.2.5 Sprinkler irrigation

The irrigation water, in this case, is sprayed over the soil surface through nozzles, within a pressure system (Karami, 2006). Sprinkler systems are about 75-85% efficient (Korkmaz, 2009). Keraita *et al*. (2007) found that overhead methods of irrigation, such as watering cans, sprinklers and spray irrigation, exposed lettuce leaves to irrigation water. Overhead irrigation with the use of sprinklers and watering cans are therefore not advised even though they are most inexpensive options, because they expose the edible portions of the plant directly to the contaminated water (Minhas and Samra, 2004).

1.5.3 Factors influencing the level of contamination of the final produce

Information on the ability of fresh produce to act as vehicles of transmission of disease has come to surface over the last two decades. The factors that influence the ability of the pathogen to get onto or into fresh produce includes, the environment, the length of time between pathogen contact and harvest, and post harvest handling practices (Schneider *et al*., 2006). The type of fresh produce, itself also influences the level of contamination of that produce (UN, 2007). The produce that are grown closer to the ground are more prone to infection since they can easily come into contact with contaminants, either through splashed soil or manure during irrigation (Hanning *et al*., 2008). Fresh produce that possess large surface areas, such as leafy vegetables or even those that have coarse surfaces allow pathogens to adhere more effortlessly to their surfaces and hence the fresh produce are at greater risk of being contaminated (UN, 2007). Leafy vegetables also possess a high water holding capacity and these vegetables are at a greater risk of contamination during the period of irrigation (Ilic *et al*., 2009). Also the time lag between contact with water and the harvest, introduces the risk of hazardous contamination which is greater near harvest time (UN, 2007). If lesions are present or if the plant material is injured, this could also influence the microbial growth because of the nutrients or many phytoalexins and in some cases, the presence of antimicrobial compounds in the exudates (Buck *et al*., 2003).

1.6 OTHER POSSIBLE SOURCES OF CONTAMINATION OF FRESH PRODUCE

1.6.1 Pre-harvest

Besides irrigation water being one of the major transporters of contaminants to fresh produce, other possible pre-harvest sources of contaminants include faeces, dust, insects, soil, inefficiently composted manure, wild and domestic animals, and human handling (Beuchat, 2002). Birds can also serve as an important source of contamination because they have the ability to transfer bacteria over large distances (Fenlon, 1985). Allowing domestic animals easy access to orchards may also result in the contamination of fresh produce, mainly those that are gathered after falling to the ground (Goverd *et al*., 1979; De Roever, 1998), even though competition with other soil microbes and unfavourable environmental conditions may cause a reduction in the numbers of pathogens present (Islam *et al*., 2004a).

1.6.2 Post-harvest

The use of contaminated water for post-harvest treatment and handling of the fresh produce such as, in food processing or preparation, which is referred to as post-harvest contamination is also a major source of human infection (Slifko *et al*., 2000). Pathogens that are present on freshly harvested produce may accumulate in water systems, and may result in post-harvest water that has the potential to contaminate other products (UN, 2007). Other sources of post-harvest contamination include “faeces, human handling, harvesting equipment, transport containers, wild and domestic animals, insects, dust, rinse water, ice, transport vehicles and processing equipment" (Beuchat, 2002). As with animals, it should be assumed that a fraction of the human beings in farm settings may harbour one or many enteric pathogens and as a result, this may contribute to the contamination of fresh produce (Goverd *et al*., 1979; De Roever, 1998). When there is a lack of suitable sanitary hand-washing facilities, there is a possibility that this would heighten the transfer of faecal contamination to the surface of fresh produce (De Roever, 1998). Fresh produce that are handled unhygienically may become contaminated with viral and bacterial pathogens (Seymour and Appleton, 2001). This appears to be particularly important for the transmission of viruses such as hepatitis A, in which the growth of the pathogen on the produce is not of importance. Furthermore, if there is a major delay in terms of transportation to the processing facility, there may be sufficient bacterial replication when the temperatures are increased and humid conditions are maintained (De Roever, 1998).

1.7 PROCESSES TO REDUCE MICROBIAL CONTAMINATION OF FRESH PRODUCE

Several guidelines have been set to regulate the level of both chemical and microbial contaminants in irrigation water and food to safeguard human exposure to these contaminants. For example, the limit for the incidence of faecal coliform bacteria in unrestricted irrigation (for vegetable and salad crops consumed raw) is $\leq 10^3$ faecal coliform bacteria/100 ml (Blumenthal *et al*., 2000). Also, the standard for the presence of *E. coli* in foods as depicted by the committee of microbiological specifications for foods (ICMSF) is $<10^5$ *E. coli*/100g (Suslow *et al*., 2003).

Some processing technologies, such as irradiation can be utilized for the destruction of contaminating microbes; however, these technologies are not always readily accepted by the customers (Bassett and McClure, 2008). Whereas, in the case of wastewater, treatment processes that should be followed are primary and secondary treatment followed by tertiary treatment, with the latter consisting of flocculation, sand filtration and finally, disinfection in order to make sure that the water is free from any microbial pathogens. This treated water may then be used for the irrigation of different crops and produce that can be consumed raw by people without any concern of disease outbreaks (Bouwer, 2000). Palese *et al*. (2009), performed a study for the disinfection of wastewater using two disinfecting agents, namely, peracetic acid and chlorine products, and found that, better results were achieved by using peracetic acid with contact times exceeding 60 min and doses of 2.5 mg/L (Palese *et al*., 2009). Wastewater can also be treated by the use of stabilization ponds as it is an efficient and low-cost method for the removal of pathogens (WHO, 1989).

Han *et al*. (2000) suggested that washing with water alone is not sufficient for the removal of bacteria that are strongly attached to wounded surfaces of vegetables, in this case green peppers. Bassett and McClure (2008) recommended the following washing conditions for fruit; the use of potable water that is at a higher temperature than that of the fruit being washed (e.g. 2-3 ºC higher). This should then be followed by, soaking the fruit for 5-10 min, if possible with agitation. The fruit should then be rinsed with potable water. The fruit should then be dried after washing, either by mechanical means or with warm air. Fruit that have a heavy surface soiling/contamination should be double washed (Bassett and McClure, 2008). For fresh produce, many wash methods have been suggested such as the use of chlorine, however, it has been recommended that additional pre-wash steps should be practiced on fresh produce arriving from the farm. This may include a vigorous pre-wash with brushes or sponges in order to remove excess debris from the produce, or a clear water rinse to remove soil and other debris, prior to using the sanitizer solution (Silva, 2008). UV light (UV-C) as been recognized by the US-FDA (2002) as a disinfectant for the surface treatment of food. However, UV-C damages nucleic acids (Farkes, 1997), and some microbes may be able to repair such damage when exposed to visible light (Zagory and Hurst, 1996; Fonseca and Rushing, 2006).

1.8 SCOPE OF THE PRESENT STUDY

The occurrence of major microbial pathogens on fresh produce is, in some cases, generally low. However, since large quantities of fresh produce are either minimally processed or consumed raw, this product becomes a possible public health hazard. Minimally processed fresh produce are exposed to various environments during growth, harvesting and distribution, and it is likely that these environments may, itself, contribute to the microbial load of the final product. The increase in food-borne disease outbreaks, due to the lack of satisfactory control measures is a global concern. Furthermore, this problem is expected to escalate in the near future, as more people consume minimally processed fresh produce, due to its higher nutritional value (UN, 2007). The risk of contamination of fresh produce is dependant on the type of crop cultivated, the irrigation method used, and the time between the last irrigation and harvest (Stine *et al*., 2005). Therefore, the execution of appropriate site-specific irrigation practices is very important in order to avoid produce contamination and at the same time achieve high-quality harvest results (UN, 2007). Consequently, this study focussed on determining the microbial and chemical quality of different irrigation waters used for cultivating fresh produce by local farmers in KZN. The impact of these microbial contaminants and chemical pollutants on the fresh produce quality was also assessed in order to establish the suitability of this fresh produce for human consumption. These findings are expected to generate new information on the quality of irrigation water used on these farms and provide the basis for any intervention strategies for the improvement of irrigation water quality. The effect of *P. aeruginosa* on the uptake of bacterial pathogens during the cultivation of fresh produce was also investigated. Finally, different post-harvest methods were assessed for their effects on microbial and product quality of fresh produce.

1.8.1 Hypotheses tested

It was hypothesised that contaminated irrigation water used by local farmers in KZN impacted negatively on the microbial and chemical safety of fresh produce. It was further hypothesised that pre- and post-harvest strategies needed improvement in order to ensure the safety and quality of fresh produce.

1.8.2 Objectives

The following objectives were established to test the above hypothesis:

1.8.2.1 To determine the microbiological and chemical quality of different irrigation waters used for growing fresh produce by local farmers in KZN.

1.8.2.2 To determine the effect of *P. aeruginosa* on the uptake of pathogens by fresh produce during the pre-harvest phase.

1.8.2.3 To determine the effect of different post-harvest treatment methods on the quality of minimally processed fresh produce.

1.8.3 Experimental design

In order to achieve the stated objectives, this research was divided into the relevant chapters described below:

Chapter Two: This chapter focuses on the microbial and chemical quality of different irrigation waters and parts of the fresh produce plant, over a 12 month period. The levels of contamination of these samples were compared to relevant standards in order to establish their quality.

Chapter Three: This chapter focuses on the effect of *P. aeruginosa* on the uptake of pathogens to the produce. Inhibition assays were used to determine if *P. aeruginosa* could inhibit the bacterial pathogens tested. A greenhouse experiment was employed to confirm this, using both culturing methods and denaturing gradient gel electrophoresis (DGGE).

Chapter Four: This chapter demonstrates the effect of different post-harvest treatment methods on the quality of the final produce. Reducing sugar, total carbohydrate content, chlorophyll content, microbial load, ascorbic acid content as well as pH and sensory evaluations were conducted in order to determine the most effective treatment method.

Chapter Five: This chapter places the entire research in perspective, by providing an outline of the significant findings reported in chapters 2, 3 and 4 of this dissertation. It also reveals the short-comings of this study and the potential for future development of the present study.

CHAPTER 2: SEASONAL CHANGES IN MICROBIAL AND HEAVY METAL QUALITY OF IRRIGATION WATER FROM SELECTED FARMS IN KZN

2.1 Introduction

Water is essential for all life forms, yet numerous people around the globe face a daily struggle, because of the shortage of water (Annan, 2005). Furthermore, large amounts of freshwater are being utilized for irrigation purposes, leading to an increase in the shortage of water supplies (UN, 2005). The scarcity of water and the energy costs faced by farmers compel them to make choices regarding the available source of water to use for irrigation purposes (Suslow *et al*., 2003). As a result, little attention is given to the quality of water being used for irrigation of agricultural produce (Gerba and Choi, 2006). The source of irrigation water, viz., rivers, streams, open canals, irrigation ditches, reservoirs, cisterns, rain barrels, groundwater and municipal supplies, ultimately affects the safety of the food product (Sapers, 2005). In South Africa, fresh produce farming is known to receive irrigation water from sources such as groundwater and surface water which irrigates approximately 24% and 76% of total irrigable area, respectively (Dennis and Nell, 2002). Irrigation waters have been directly implicated in contributing to several bacterial disease outbreaks world-wide (Hanning *et al*., 2009). Besides microbiological contaminants, irrigation waters may also be contaminated by persistent chemicals such as heavy metals (UN, 2007; The groundwater foundation, 2009).

The presence of microbial pathogens has been found to be associated with the use of contaminated irrigation water (Gast and Holt, 2000). Also, long storage of water has been shown to result in an increase in microbial pollution and many human health problems (Shaban and Malkawi, 2007). Islam *et al*. (2005) demonstrated the growth of *E. coli* 0157:H7 in carrots and onions, irrigated with water contaminated with this pathogen and the survival of *E. coli* 0157:H7 on the produce for more than two months as evident on the final produce (Islam *et al*., 2005). During the months of May to June of 2005, an outbreak of diarrhoeal disease had occurred amongst company workers in Copenhagen. These cases were reported from about 7 of 8 companies, which had received “food from the same catering kitchen”, with stool specimens from patients from two of these companies testing positive for *C. jejuni* (Mazick *et al*., 2006). To date, numerous outbreaks of bacterial disease associated with the consumption of fresh produce have been reported world-wide, however, although such outbreaks of food-borne disease in humans do occur in South Africa, these incidences are rarely reported (Smith *et al*., 2007). Therefore, it is important to monitor the microbial quality of irrigation waters and the subsequent fresh produce in order to detect the presence of potential pathogens, and thereby providing different control measures to prevent food-borne disease outbreaks in South Africa.

The long-term use of arsenic-contaminated groundwater was found to result in the contamination of paddy rice (Abedin *et al*., 2002). The excessive build-up of heavy metals in farming soils may result not only in the contamination of the environment, but may also lead to an increase in the amount of heavy metals taken up by crops. This may ultimately affect food quality and most importantly food safety (Muchuweti *et al*., 2006). Muchuweti *et al*. (2006) studied heavy metal concentrations in farm plots irrigated using “sewage sludge and sewage/sewage sludge admixtures” in Harare (Zimbabwe). It was found that the different crops irrigated with this water were heavily contaminated with cadmium (Cd), copper (Cu), lead (Pb) and Zinc (Zn). It was further evident that the degree of contamination was highest, in two of the crops, maize and *tsunga*, being used as a staple diet by the villagers in this region. Jackson *et al*. (2009) investigated the level of metal [aluminium (Al), copper (Cu), iron (Fe), lead (Pb), manganese (Mn), nickel (Ni) and zinc (Zn)] contamination in the Plankenburg and Diep Rivers of the Western Cape province of South Africa, used as a source of irrigation water, over a period of 12 and 9 months, respectively. The concentrations of most of these metals were found to be much higher than those of the recommended water quality guidelines, thus re-iterating the need for routine monitoring of these rivers (Jackson *et al*., 2009).

It has been shown that the metal concentrations in plant tissues, generally, increases with an increase in the concentrations of metals in the irrigation water and that the concentrations of these heavy metals in roots are typically higher compared to the metal concentrations in the leaves (Qadir *et al*., 2008). Recently, Arora *et al*. (2008) used atomic absorption spectrophotometry to evaluate the concentrations of different heavy metals, such as Fe, Mn, Cu and Zn, in vegetables that were irrigated with water from various sources. Fresh produce irrigated with wastewater were reported to accumulate heavy metals with concentrations (in mg/kg) ranging between: 116–378, 12–69, 5.2–16.8 and 22–46 for Fe, Mn, Cu and Zn, respectively (Arora *et al*., 2008). The study further revealed that high levels of heavy metals accumulated in the edible portions of food crops as a result of continuous irrigation of these crops with this water source.

Food-borne disease outbreaks are on the increase, due to the lack of satisfactory control measures; this has become an alarming global concern. Furthermore, this problem is expected to heighten in future, as more and more people are consuming minimally processed fresh produce, due to its nutritional value. Therefore, identifying the source of contamination, contaminants and the accumulative area of the plants would allow for the establishment of proper guidelines for the cultivation of fresh produce to reduce the chances of contamination. This study therefore investigated the microbial and chemical quality of different irrigation water used for cultivating fresh produce by local farmers in KwaZulu-Natal (KZN) as well as established a link between the quality of these irrigation waters and the fresh produce cultivated using these water sources. This study is important in order to make recommendations on how to deal with such problems, if this does indeed exist in KZN. Also, the outcome of this study is expected to shed more light on the extent of the microbial and chemical contamination of the irrigation water as well as the fresh produce plants in this region. To the best of our knowledge, little work on the effect of the microbial and chemical quality of irrigation waters on the quality of the final produce has been carried out in KZN.

2.2 Materials and methods

2.2.1 Description of sampling sites

Three different farms, designated A, B, C were used in this study. Farm A is 130 hectares large and located in Camperdown. The farm has two sources of irrigation water namely, river (A1) and borehole water (A2), which are used to irrigate the plants weekly on two separate plots. Dogs and cattle are allowed around the crop area. Workers don’t use gloves and there are no washing facilities for these workers. Crops planted are broccoli, spinach, jam tomatoes, Crisphead lettuce, cauliflower and cabbage. Crops are not washed and packed on site. Farm B is 60 hectares large and located in Cato ridge. The farm’s source of irrigation water is a mixture of borehole and dam water (B). Crops planted are broccoli, spinach, lettuce, cauliflower, chinese cabbage, parsley, bell pepper and red cabbage, and these crops are irrigated weekly. Crops are washed and packed on site. Farm C is 360 hectares large and located in Richmond. The farm’s source of irrigation water is dam water (C). Crops planted are oranges, cabbage (planted in winter) and jam tomatoes (planted in summer). Frequency of irrigation is weekly, or when dry twice a week. Crops are washed and packed on site. The irrigation water obtained from these farms was applied to crops via spray irrigation. The biogeographically location of the three farms cannot be revealed due to an agreement made with the owners’ of the respective farms.

2.2.2 Sample collection and processing

The following samples were collected monthly from the farms, for a period of 1 year: irrigation water, fruit and vegetables (roots, stems, leaves, edible portion) and soil. The stage of development of the fresh produce was noted at every sample collection. Four types of fresh produce, per irrigation water sample were analyzed, depending on the availability of crops onsite. All analyses were performed in triplicate.

Water samples were collected in sterile 5 L plastic containers and plant samples were first separated into different parts (roots, stems, leaves, edible portion) and then placed into sterile plastic bags. Soil samples were taken from 2.5 cm below the plant and then placed into sterile plastic bags. All the samples were transported in Styrofoam boxes containing ice packs and stored at 4 °C until the analyses began (Mukherjee *et al.*, 2004). Water samples were analyzed within 24 h of collection while plant and soil samples were analyzed within 48 h of collection. All the samples listed above were analyzed for the presence of commonly found food-borne bacterial pathogens and toxic heavy metals.

2.2.2.1 Bacterial analysis

Serial dilutions were prepared from each water sample using sterile distilled water. Hundred and fifty millilitres of the appropriate dilutions were filtered through a membrane filter (0.45 µm) and the membranes were placed onto selective media (Table 2.1). Each soil sample (10 g) was mixed with 90 ml of 0.1% peptone water (Merck) in a sterile beaker and placed on a shaker for 30 s. The plant samples were prepared for analyses according to Islam *et al.* (2004a). Ten grams (fresh weight) of each plant sample was homogenized with 90 ml of 0.1% peptone water using a blender. Serial 10-fold dilutions of the homogenized samples were then made, using 0.1% peptone water. One hundred microlitres of these dilutions were then spread plated onto different selective media and incubated appropriately to allow for growth of the respective presumptive bacterial pathogens (Table 2.1). The different bacterial countss were enumerated by counting the number of colonies per plate and these values were expressed as colony forming units (cfu) per 100 ml or gram of the sample.

Table 2.1: Selective media used in this study for the growth of specific organisms and their required growth conditions.

Specific organisms	**Medium**	**Growth conditions**	**Appearance on medium**	**References**
Campylobacter spp.	Columbia agar (Merck) supplemented with 5% lysed horse blood	37 °C, 48 h, under a microaerobic atmosphere	grey colonies	(Tholozan *et al*., 1999)
E. coli and Coliforms	coliform-chromo agar (Merck)	37 °C, 24 h	*E .coli* - blue to dark violet Coliforms - salmon to red	(Alonso *et al*., 1999)
Listeria monocytogenes	Agar Listeria Ottaviani and Agosti (ALOA, Fluka)	30 °C, 24 h	Appear surrounded with a distinct opaque halo-like precipitation zone	(Jantzen *et al*., 2006)
Salmonella spp. and *Shigella* spp.	*Salmonella-Shigella* agar (SS agar, Merck)	37 °C	*Salmonella* spp.- colourless colonies with black centres, *Shigella* spp. - colourless colonies	(Islam *et al*., 1997)

2.2.2.2 Heavy metal analysis

Water samples were filtered through a membrane (0.45 µm) and 50 µl of 70% nitric acid (Merck) was added to preserve the water samples for heavy metal analysis. All plant samples were first washed with distilled water to remove surface contaminants and then left to air dry for 24 h. The plant samples were dried in an oven (70-80 °C) for 24 h. The samples were mashed using a mortar and pestle and 0.5 g (dry weight) of the samples, weighed in crucibles. The samples were then digested using 3 ml of a mixture of 70% perchloric acid (Merck) and 70% nitric acid [1:4] and left to cool before filtering through Whatmann paper no. 42 (Arora *et al*., 2008). The solutions were brought up to 14 ml using double distilled water. Two grams of soil samples were added to 20 ml of 0.1M hydrochloric acid (Merck) and left to stand for 30 min and then filtered using Whatmann paper (no. 42) (Sabiene *et al*., 2004). All samples were analyzed in triplicate using inductively coupled plasma optical emission spectrophotometry (ICP-OES), for the presence of the following toxic heavy metals: arsenic (As), cadmium (Cd), mercury (Hg) and lead (Pb).

2.2.3 Statistical analysis

Student t-test was used to compare the means of microbial counts, from irrigation water, for the four seasons (spring, summer, autumn and winter) and one-way analysis of variance (ANOVA) were used to compare the means of these microbial counts for the different farms and seasons. The coefficient of correlation between microbial counts, seasons and farms were calculated by the Pearson correlations test. Statistical significance was set at P values of < 0.05 or <0.01.

2.3 Results

2.3.1 Bacterial analysis

2.3.1.1 Bacterial analysis of the irrigation water

Analysis of the different irrigation water samples (A1, A2, B, C), revealed that irrigation water sample B had the highest microbial load throughout the sampling period (Figures 2.1 and 2.2). The concentrations of the different presumptive bacterial pathogens namely, *Campylobacter* spp., coliforms, *E. coli*, *L. monocytogenes*, *Salmonella* spp. and *Shigella* spp. ranged from being not-detected to 2.25×10^3, 9.07×10^3, 5.94×10^3, 1.57×10^3, 5.59×10^3 and 14.53×10^3 cfu/100ml, respectively across all the water types tested. Presumptive coliforms and *Shigella* spp. were found at high concentrations in all water types tested except dam water, throughout the sampling period. During the spring period (months), a decrease in the microbial counts of all pathogens tested were observed in all water samples, except for the borehole water (A2) samples which had increased during this period (Figure 2.1b) and the dam water (C) samples, where an increase in presumptive *Campylobacter* spp. counts was observed (Figure 2.2b). Presumptive *Campylobacter* spp. was not detected in any of the water types tested except dam water, where a concentration as high as 2.25×10^3 cfu/100 ml was detected. The highest concentration of presumptive *L. monocytogenes* was detected in the borehole (A2) water sample at 1.57×10^3 cfu/100 ml in October 2009. In farm C, a trend was observed, whenever fresh produce were grown (winter and summer seasons) presumptive *Campylobacter* spp. counts decreased in the water sample but when no fresh produce were grown, presumptive *Campylobacter* spp. counts increased in the water sample. The microbial load of the irrigation waters (A1, A2 and B) increased during summer, in which the highest counts were evident. The most abundant bacteria present in these irrigation waters throughout the year were presumptive *Shigella* spp. (farms A1, A2 and B) and presumptive *Campylobacter* spp. counts (farm C).

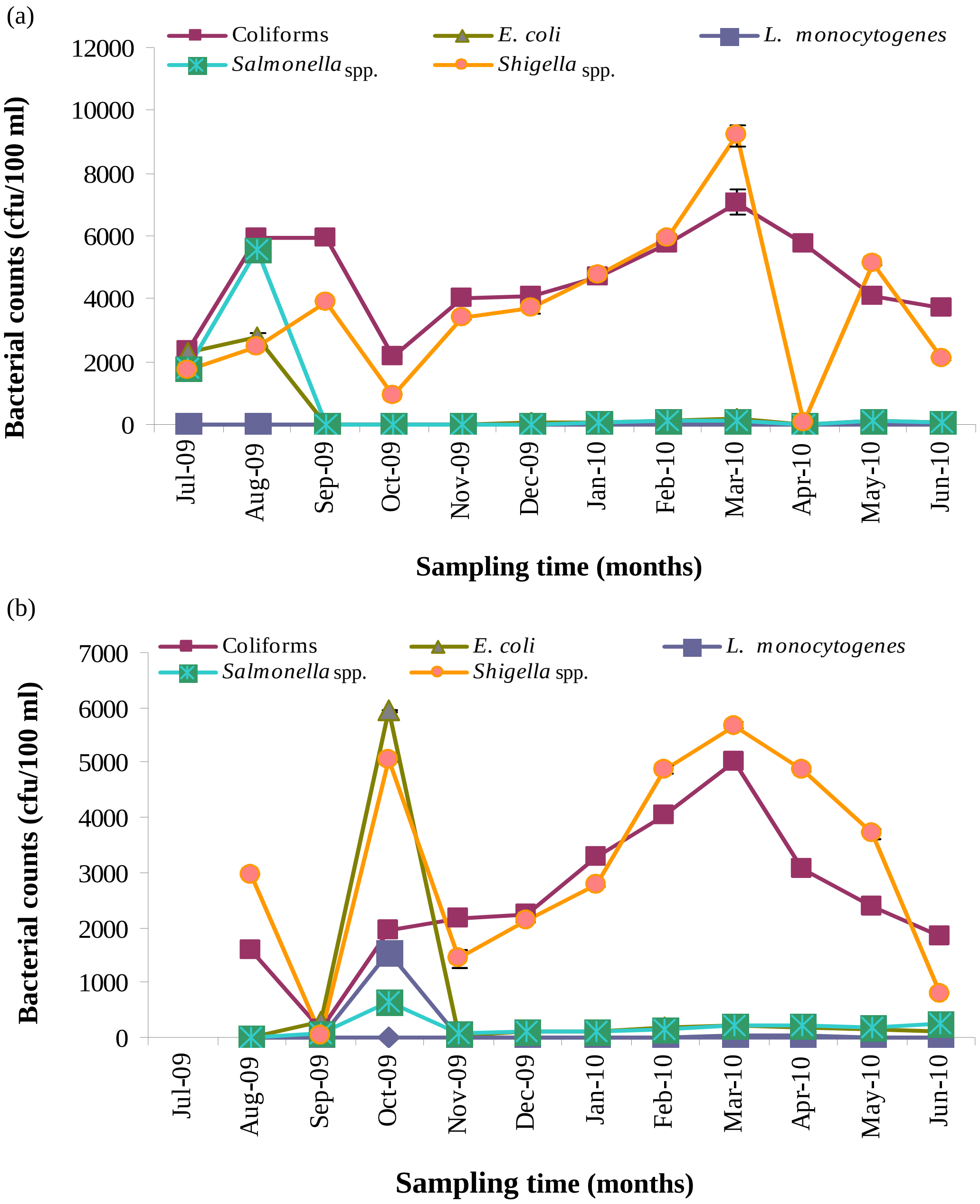
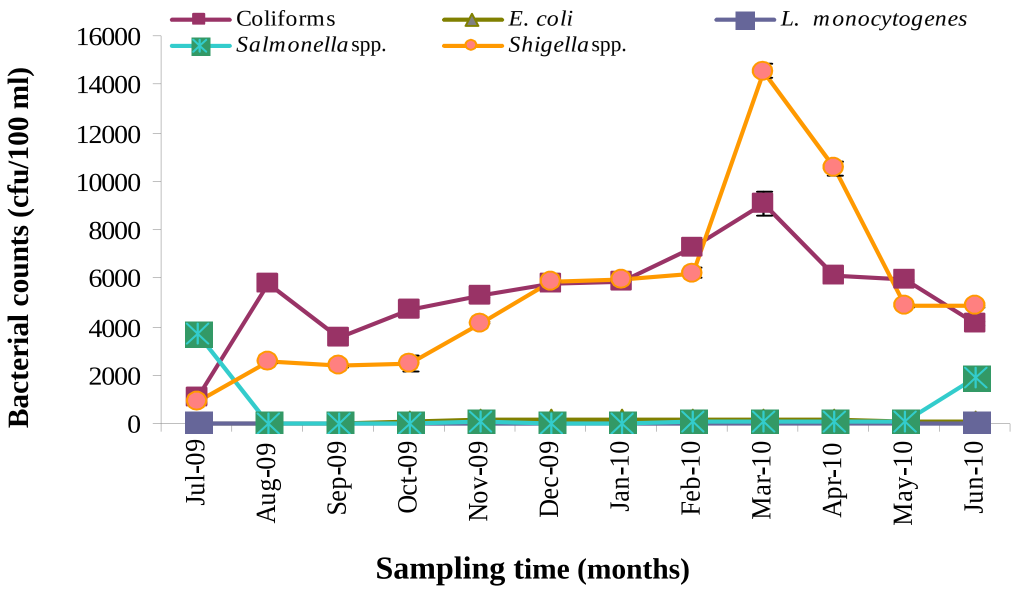


Figure 2.1: The microbial quality of irrigation waters collected from farm A1 (a) and A2 (b), over a one-year period.

(a)



(b)

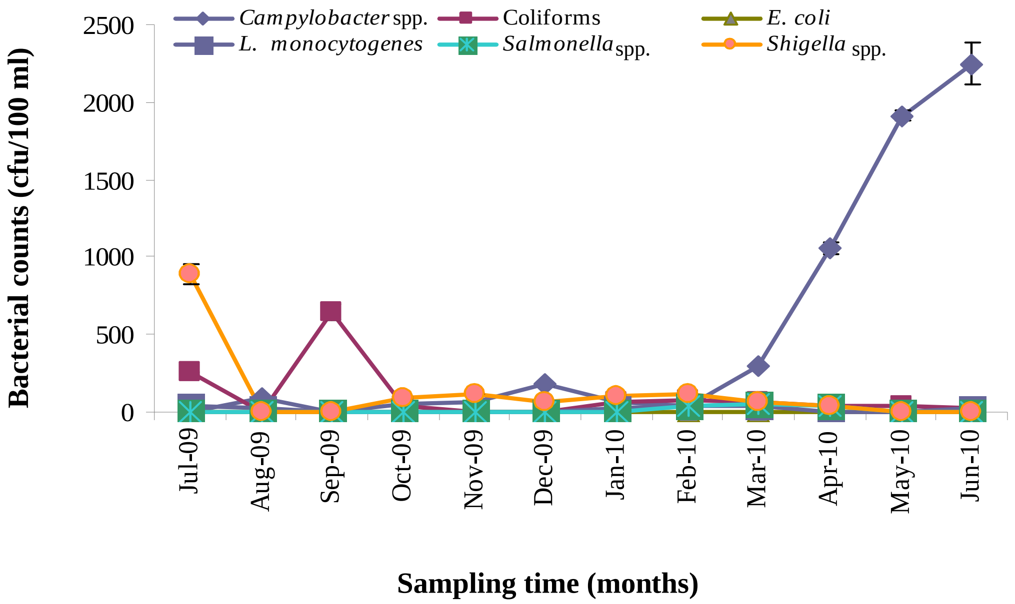


Figure 2.2: The microbial quality of irrigation waters collected from farm B (a) and C (b), over a one-year period.

Correlation matrices (significant at the 0.01 level) between the presumptive microbial pathogens present in the different water types throughout this study and the seasonal variations are represented in Table 2.2. *C. jejuni* showed negative, significant correlations with coliforms (*r* = - 0.355) and *Shigella* spp. (*r* = - 0.298). *E. coli* showed strong positive correlations with *L. monocytogenes* (*r* = 0.855) and *Salmonella* spp. (*r* = 0.462). In addition, strong positive significant correlations were observed between coliforms and *Shigella* spp. (*r* = 0.850). Positive correlations between the populations of coliforms, *Shigella* spp. and seasonal variations were found to be significant (p<0.01), however, negative correlations were established between seasonal variations and *Salmonella* spp.

Table 2.2: Correlation matrix of the presumptive microbial pathogens present in the different water types throughout this study and seasonal variations.

	***Campylobacter* spp.**	**Coliforms**	***E. coli***	***L. monocytogenes***	***Salmonella* spp.**	***Shigella* spp.**	**Seasonal variations**
C. jejuni	1						0.063^{ns}
Coliforms	$- 0.355^{*}$	1					0.312^{*}
E. coli	$- 0.089^{ns}$	0.033^{ns}	1				$- 0.164^{ns}$
L. monocytogenes	$- 0.043^{ns}$	$- 0.037^{ns}$	0.855^{*}	1			$- 0.028^{ns}$
***Salmonella* spp.**	$- 0.094^{ns}$	0.082^{ns}	0.462^{*}	0.043^{ns}	1		$- 0.341^{*}$
***Shigella* spp.**	$- 0.298^{*}$	0.850^{*}	0.088^{ns}	0.147^{ns}	$- 0.059^{ns}$	1	0.482^{*}

ns= not significant

[*] Correlation was significant at the 0.01 level (2-tailed)

2.3.1.2 Bacterial analysis of the fresh produce

2.3.1.2.1 Fresh produce from farm A1

The different types of fresh produce that were collected throughout this study included broccoli, bell pepper, cabbage, chinese cabbage, red cabbage, cauliflower, crisphead lettuce, jam tomatoes, parsley and spinach. Similar trends in the microbial quality of the fresh produce as observed in farm A1 (Tables 2.3-2.8), were observed throughout for the fresh produce collected from the different farms (Tables 2.9-2.26). It was apparent that the fresh produce with the highest microbial contamination was the leafy vegetables, such as lettuce and spinach, throughout the seasons. Presumptive *Campylobacter* spp., coliforms, *E. coli*, *L. monocytogenes*, *Shigella* spp. and *Salmonella* spp. were detected on different fresh produce throughout the sampling period, with presumptive *Campylobacter* spp., coliforms and *Shigella* spp. being the most abundant. The most abundant microbe in the soil samples collected was presumptive *Campylobacter* spp. Presumptive *Campylobacter* spp. and coliform populations were the most abundant in the spinach soil and jam tomato root samples at 2.93×10^6 and 2.08×10^6 cfu/g, in October 2009 and April 2010, respectively. Presumptive *E. coli* populations were the most abundant in the crisphead lettuce and spinach samples. Presumptive *L. monocytogenes* populations were the least abundant in the fresh produce tested, with the highest being recorded at 1.13×10^4 cfu/g in crisphead lettuce (January 2010). Presumptive *Salmonella* spp. was detected frequently in the edible portion of the spinach and crisphead lettuce plants. Presumptive *Shigella* spp. was found abundantly throughout the seasons with the highest recorded at 2.71×10^6 cfu/g in the broccoli sample (edible portion) collected in July 2009.

Table 2.3: The monthly variation in presumptive *Campylobacter* spp. counts (in cfu/g) on the different fresh produce collected from farm A1, over a one-year period.

(a)

Sampling date	Broccoli						Cabbage				Crisphead lettuce			
	Soil	Root	Stem	Leaf	Edible portion	Stage of growth*	Soil	Root	Edible portion	Stage of growth*	Soil	Root	Edible portion	Stage of growth*
Jul-09	14.1×10^{5}	3.1×10^{5}	4.7×10^{5}	5.57×10^{5}	3.18×10^{4}	6	6.1×10^{5}	13.83×10^{5}	3.1×10^{4}	9	3.53×10^{5}	3.1×10^{5}	4.57×10^{5}	8
Aug-09	4.27×10^{5}	11.23×10^{5}	1.05×10^{4}	4.8×10^{3}	6.6×10^{3}	5	3.2×10^{5}	3.43×10^{5}	7.03×10^{4}	5	4.17×10^{5}	9.83×10^{5}	1.81×10^{4}	3
Sep-09	ND	ND	ND	ND	ND	ND	3.33×10^{4}	2.2×10^{5}	8.13×10^{4}	8	4.5×10^{5}	3.47×10^{5}	4.2×10^{4}	4
Oct-09	2.04×10^{5}	6.3×10^{4}	N	2.5×10^{4}	1.08×10^{4}	3	18.83×10^{5}	25.17×10^{5}	2.05×10^{5}	6	2.07×10^{4}	4.27×10^{5}	1.03×10^{4}	5
Nov-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Dec-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Jan-10	ND	ND	ND	ND	ND	ND	1.99×10^{5}	2.72×10^{5}	3.5×10^{4}	6	2.99×10^{5}	2.98×10^{4}	2.08×10^{4}	8
Feb-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	2.42×10^{5}	3.03×10^{5}	3.2×10^{4}	4
Mar-10	ND	ND	ND	ND	ND	ND	1.86×10^{5}	2.65×10^{5}	4.83×10^{4}	6	2.46×10^{5}	4.17×10^{5}	3.33×10^{4}	5
Apr-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	3.07×10^{5}	3.03×10^{4}	2.15×10^{4}	8
May-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	3.03×10^{5}	2.71×10^{4}	1.97×10^{4}	8
Jun-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	6.4×10^{5}	3.57×10^{4}	2.75×10^{4}	8

(b)

Sampling date	Jam tomato						Spinach				
	Soil	Root	Stem	Leaf	Edible portion	Stage of growth*	Soil	Root	Stem	Edible portion	Stage of growth*
Jul-09	27.57×10^{5}	2.46×10^{5}	2.28×10^{5}	4.5×10^{5}	1.57×10^{4}	7	ND	ND	ND	ND	ND
Aug-09	1.11×10^{5}	18.1×10^{5}	1.11×10^{5}	3.03×10^{5}	7.87×10^{3}	4	ND	ND	ND	ND	ND
Sep-09	2.6×10^{5}	2.7×10^{5}	4.37×10^{4}	2.67×10^{4}	3.47×10^{4}	5	ND	ND	ND	ND	ND
Oct-09	ND	ND	ND	ND	ND	ND	29.33×10^{5}	27.57×10^{5}	9.13×10^{4}	10.83×10^{5}	13
Nov-09	ND	ND	ND	ND	ND	ND	7.03×10^{4}	11.07×10^{5}	6.5×10^{5}	8.07×10^{3}	6
Dec-09	ND	ND	ND	ND	ND	ND	9.3×10^{4}	11.8×10^{5}	9.57×10^{5}	3.33×10^{5}	9
Jan-10	ND	ND	ND	ND	ND	ND	8.23×10^{4}	4.73×10^{5}	3.63×10^{5}	2.04×10^{4}	12
Feb-10	ND	ND	ND	ND	ND	ND	1.43×10^{5}	11.43×10^{5}	3.23×10^{5}	9.13×10^{3}	6
Mar-10	ND	ND	ND	ND	ND	ND	2.57×10^{5}	13.43×10^{5}	2.76×10^{5}	1.46×10^{4}	8
Apr-10	2.89×10^{5}	3.03×10^{5}	2.97×10^{4}	2.85×10^{4}	4.37×10^{4}	5	1.23×10^{5}	8.8×10^{5}	2.83×10^{5}	7.73×10^{3}	5
May-10	2.73×10^{5}	2.94×10^{5}	3.23×10^{4}	3.4×10^{4}	2.87×10^{4}	7	2.44×10^{5}	12.17×10^{5}	2.76×10^{5}	1.25×10^{4}	8
Jun-10	ND	ND	ND	ND	ND	ND	2.84×10^{5}	15.8×10^{5}	5.70×10^{5}	3.67×10^{4}	12

ND = Not-determined as the fresh produce was not present at the time of sample collection; N = bacterial species was not detected; Stage of growth* = weeks

(a)

Table 2.4: The monthly variation in presumptive coliforms (in cfu/g) on the different fresh produce collected from farm A1, over a one-year period.

Sampling date	Broccoli						Cabbage				Crisphead lettuce			
	Soil	Root	Stem	Leaf	Edible portion	Stage of growth*	Soil	Root	Edible portion	Stage of growth*	Soil	Root	Edible portion	Stage of growth*
Jul-09	5.5×10^3	3.23×10^3	N	N	5.1×10^3	6	4.07×10^3	5.1×10^3	3.2×10^3	9	6.77×10^3	6.03×10^3	6.57×10^3	8
Aug-09	1.24×10^5	7.13×10^5	8.87×10^3	4.3×10^3	1.31×10^4	5	N	6.97×10^3	5.9×10^3	5	5.97×10^5	6×10^5	4.2×10^3	3
Sep-09	ND	ND	ND	ND	ND	ND	5.73×10^4	1.93×10^4	9.17×10^4	8	5.33×10^4	8.5×10^4	6.9×10^4	4
Oct-09	4.13×10^5	1.58×10^5	N	1.69×10^4	2×10^4	3	N	4.47×10^4	3.13×10^3	6	7.97×10^3	9.27×10^5	2.36×10^4	5
Nov-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Dec-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Jan-10	ND	ND	ND	ND	ND	ND	3.3×10^4	8.83×10^4	2.05×10^4	6	3.07×10^4	3.01×10^4	2.09×10^5	8
Feb-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	1.07×10^5	1.33×10^5	9×10^4	4
Mar-10	ND	ND	ND	ND	ND	ND	4.67×10^4	1.17×10^5	2.7×10^4	6	1.22×10^4	15.57×10^5	1.67×10^5	5
Apr-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	3.03×10^4	2.85×10^4	1.75×10^5	8
May-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	2.76×10^4	2.52×10^4	1.24×10^5	8
Jun-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	2.54×10^4	1.83×10^4	9.67×10^3	8

(b)

Sampling date	Jam tomato						Spinach				
	Soil	Root	Stem	Leaf	Edible portion	Stage of growth*	Soil	Root	Stem	Edible portion	Stage of growth*
Jul-09	5.37×10^3	N	N	N	N	7	ND	ND	ND	ND	ND
Aug-09	1.48×10^5	10.47×10^5	1.93×10^5	1.50×10^5	N	4	ND	ND	ND	ND	ND
Sep-09	9.73×10^5	19.33×10^5	8.47×10^4	20.2×10^5	2.18×10^4	5	ND	ND	ND	ND	ND
Oct-09	ND	ND	ND	ND	ND	ND	8.03×10^4	14.9×10^5	3.25×10^4	9.13×10^5	13
Nov-09	ND	ND	ND	ND	ND	ND	1.37×10^4	8.87×10^3	1.25×10^5	4.97×10^3	6
Dec-09	ND	ND	ND	ND	ND	ND	N	8.23×10^3	1.05×10^5	1.88×10^5	9
Jan-10	ND	ND	ND	ND	ND	ND	N	3.23×10^3	1.88×10^4	4.8×10^5	12
Feb-10	ND	ND	ND	ND	ND	ND	1.53×10^4	1.12×10^4	1.5×10^5	7.03×10^3	6
Mar-10	ND	ND	ND	ND	ND	ND	2.01×10^4	1.44×10^4	1.13×10^5	1.55×10^4	8
Apr-10	12.27×10^5	20.77×10^5	8.3×10^4	2.47×10^5	2.54×10^4	5	1.33×10^4	9.47×10^3	1.38×10^5	5.97×10^3	5
May-10	1.36×10^5	3.2×10^5	3.4×10^4	1.49×10^5	1.27×10^4	7	1.82×10^4	1.05×10^4	6.8×10^4	1.19×10^4	8
Jun-10	ND	ND	ND	ND	ND	ND	N	N	1.51×10^4	2.36×10^5	12

ND = Not-determined as the fresh produce was not present at the time of sample collection; N = bacterial species was not detected; Stage of growth* = weeks

Table 2.5: The monthly variation in presumptive *E. coli* counts (in cfu/g) on the different fresh produce collected from farm A1, over a one-year period.

(a)

Sampling date	Broccoli						Cabbage				Crisphead lettuce			
	Soil	**Root**	**Stem**	**Leaf**	**Edible portion**	**Stage of growth***	**Soil**	**Root**	**Edible portion**	**Stage of growth***	**Soil**	**Root**	**Edible portion**	**Stage of growth***
Jul-09	N	N	N	N	N	6	5.3×10^{3}	N	3.2×10^{3}	9	4.43×10^{3}	3.23×10^{3}	4.97×10^{3}	8
Aug-09	N	N	N	N	N	5	N	N	N	5	N	N	N	3
Sep-09	ND	ND	ND	ND	ND	ND	8.03×10^{3}	N	N	8	N	N	N	4
Oct-09	N	N	N	N	N	3	N	N	N	6	N	9.8×10^{3}	N	5
Nov-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Dec-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Jan-10	ND	ND	ND	ND	ND	ND	3.33×10^{4}	8.87×10^{3}	N	6	N	2.1×10^{5}	3.2×10^{5}	8
Feb-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	N	N	N	4
Mar-10	ND	ND	ND	ND	ND	ND	3.27×10^{4}	1.05×10^{4}	N	6	N	2.56×10^{5}	2.65×10^{5}	5
Apr-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	N	1.82×10^{5}	2.94×10^{4}	8
May-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	N	1.61×10^{5}	2.79×10^{5}	8
10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	N	1.35×10^{5}	2.34×10^{5}	8

(b)

Sampling date	Jam tomato						Spinach				
	Soil	**Root**	**Stem**	**Leaf**	**Edible portion**	**Stage of growth***	**Soil**	**Root**	**Stem**	**Edible portion**	**Stage of growth***
Jul-09	3.47×10^{3}	N	N	N	N	7	ND	ND	ND	ND	ND
Aug-09	N	N	N	N	N	4	ND	ND	ND	ND	ND
Sep-09	4.73×10^{3}	N	N	2.17×10^{5}	N	5	ND	ND	ND	ND	ND
Oct-09	ND	ND	ND	ND	ND	ND	N	N	N	N	13
Nov-09	ND	ND	ND	ND	ND	ND	N	N	N	N	6
Dec-09	ND	ND	ND	ND	ND	ND	N	N	3.77×10^{3}	2.06×10^{4}	9
Jan-10	ND	ND	ND	ND	ND	ND	N	N	9.4×10^{3}	4.23×10^{4}	12
Feb-10	ND	ND	ND	ND	ND	ND	N	N	N	N	6
Mar-10	ND	ND	ND	ND	ND	ND	N	N	N	9.67×10^{4}	8
Apr-10	5.63×10^{3}	N	N	2.61×10^{5}	N	5	N	N	N	N	5
May-10	6.23×10^{3}	N	N	2.72×10^{5}	N	7	N	N	N	1.03×10^{5}	8
Jun-10	ND	ND	ND	ND	ND	ND	N	N	1.52×10^{4}	5.63×10^{4}	12

ND = Not-determined as the fresh produce was not present at the time of sample collection; N = bacterial species was not detected; Stage of growth* = weeks

Table 2.6: The monthly variation in presumptive *L. monocytogenes* counts (in cfu/g) on the different fresh produce collected from farm A1, over a one-year period.

(a)

Sampling date	Broccoli						Cabbage				Crisphead lettuce			
	Soil	**Root**	**Stem**	**Leaf**	**Edible portion**	**Stage of growth***	**Soil**	**Root**	**Edible portion**	**Stage of growth***	**Soil**	**Root**	**Edible portion**	**Stage of growth***
Jul-09	N	N	N	N	N	6	5.3×10^3	N	N	9	5.77×10^3	N	4.03×10^3	8
Aug-09	3×10^3	3.33×10^3	N	N	N	5	N	N	N	5	N	N	N	3
Sep-09	ND	ND	ND	ND	ND	ND	N	N	N	8	N	N	N	4
Oct-09	N	N	N	N	N	3	N	N	N	6	N	N	N	5
Nov-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Dec-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Jan-10	ND	ND	ND	ND	ND	ND	N	N	N	6	4.73×10^3	9.53×10^3	1.13×10^4	8
Feb-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	N	N	N	4
Mar-10	ND	ND	ND	ND	ND	ND	N	N	N	6	N	N	N	5
Apr-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	3.07×10^3	3.47×10^3	8.6×10^3	8
May-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	N	N	6.77×10^3	8
Jun-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	N	5.53×10^3	N	8

(b)

Sampling date	Jam tomato						Spinach				
	Soil	**Root**	**Stem**	**Leaf**	**Edible portion**	**Stage of growth***	**Soil**	**Root**	**Stem**	**Edible portion**	**Stage of growth***
Jul-09	N	N	N	N	N	7	ND	ND	ND	ND	ND
Aug-09	N	N	N	N	N	4	ND	ND	ND	ND	ND
Sep-09	N	N	N	N	N	5	ND	ND	ND	ND	ND
Oct-09	ND	ND	ND	ND	ND	ND	N	N	N	N	13
Nov-09	ND	ND	ND	ND	ND	ND	N	N	N	N	6
Dec-09	ND	ND	ND	ND	ND	ND	N	N	N	3.2×10^3	9
Jan-10	ND	ND	ND	ND	ND	ND	N	N	N	6.87×10^3	12
Feb-10	ND	ND	ND	ND	ND	ND	N	N	N	N	6
Mar-10	ND	ND	ND	ND	ND	ND	N	N	N	N	8
Apr-10	3.13×10^3	3.3×10^3	N	N	N	5	N	N	N	N	5
May-10	3×10^3	N	N	N	N	7	N	N	N	N	8
Jun-10	ND	ND	ND	ND	ND	ND	N	N	N	5.33×10^3	12

ND = Not-determined as the fresh produce was not present at the time of sample collection; N = bacterial species was not detected; Stage of growth* = weeks

Table 2.7: The monthly variation in presumptive *Salmonella* spp. counts (in cfu/g) on the different fresh produce collected from farm A1, over a one-year period.

(a)

Sampling date	Broccoli						Cabbage				Crisphead lettuce			
	Soil	**Root**	**Stem**	**Leaf**	**Edible portion**	**Stage of growth***	**Soil**	**Root**	**Edible portion**	**Stage of growth***	**Soil**	**Root**	**Edible portion**	**Stage of growth***
Jul-09	3.13×10^{3}	N	N	N	N	6	N	3.3×10^{4}	N	9	N	6.27×10^{3}	N	8
Aug-09	N	N	N	N	N	5	N	N	N	5	N	N	N	3
Sep-09	ND	ND	ND	ND	ND	ND	4.37×10^{3}	N	N	8	N	N	N	4
Oct-09	N	N	N	N	N	3	N	N	N	6	N	N	N	5
Nov-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Dec-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Jan-10	ND	ND	ND	ND	ND	ND	1.07×10^{4}	3.23×10^{3}	N	6	N	3.4×10^{3}	6.77×10^{3}	8
Feb-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	3.13×10^{3}	N	N	4
Mar-10	ND	ND	ND	ND	ND	ND	1.32×10^{4}	5.27×10^{3}	3.27×10^{3}	6	N	6.73×10^{3}	3.53×10^{3}	5
Apr-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	N	N	4.67×10^{3}	8
May-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	N	N	5.07×10^{3}	8
Jun-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	N	6.23×10^{3}	6.67×10^{3}	8

(b)

Sampling date	Jam tomato						Spinach				
	Soil	**Root**	**Stem**	**Leaf**	**Edible portion**	**Stage of growth***	**Soil**	**Root**	**Stem**	**Edible portion**	**Stage of growth***
Jul-09	4.37×10^{3}	N	N	N	N	7	ND	ND	ND	ND	ND
Aug-09	N	N	N	N	N	4	ND	ND	ND	ND	ND
Sep-09	N	N	N	N	N	5	ND	ND	ND	ND	ND
Oct-09	ND	ND	ND	ND	ND	ND	N	8.57×10^{3}	N	N	13
Nov-09	ND	ND	ND	ND	ND	ND	N	N	9.47×10^{3}	N	6
Dec-09	ND	ND	ND	ND	ND	ND	N	N	N	3.23×10^{3}	9
Jan-10	ND	ND	ND	ND	ND	ND	N	N	N	3.57×10^{4}	12
Feb-10	ND	ND	ND	ND	ND	ND	N	N	N	1.53×10^{4}	6
Mar-10	ND	ND	ND	ND	ND	ND	N	N	N	2.25×10^{4}	8
Apr-10	N	N	N	N	N	5	N	N	N	1.34×10^{4}	5
May-10	3.3×10^{3}	N	N	N	N	7	N	N	N	2.56×10^{4}	8
Jun-10	ND	ND	ND	ND	ND	ND	N	N	N	8.27×10^{4}	12

ND = Not-determined as the fresh produce was not present at the time of sample collection; N = bacterial species was not detected; Stage of growth* = weeks

(a) **Table 2.8:** The monthly variation in presumptive *Shigella* spp. counts (in cfu/g) on the different fresh produce collected from farm A1, over a one-year period.

Sampling date	Broccoli						Cabbage				Crisphead lettuce			
	Soil	**Root**	**Stem**	**Leaf**	**Edible portion**	**Stage of growth***	**Soil**	**Root**	**Edible portion**	**Stage of growth***	**Soil**	**Root**	**Edible portion**	**Stage of growth***
Jul-09	8.77×10^4	3.33×10^4	15.57×10^5	2.6×10^4	27.07×10^5	6	8.93×10^5	3.43×10^5	4.27×10^5	9	1.49×10^5	6.23×10^3	3.73×10^5	8
Aug-09	2.71×10^4	2.43×10^5	N	3.37×10^4	3.1×10^4	5	6.13×10^4	2.49×10^5	3.17×10^3	5	1.8×10^4	2×10^4	6.1×10^3	3
Sep-09	ND	ND	ND	ND	ND	ND	4.07×10^4	3.27×10^5	3.17×10^5	8	1.96×10^4	2.56×10^4	7.5×10^3	4
Oct-09	4.53×10^5	5.4×10^4	N	N	N	3	2.16×10^4	N	4.37×10^3	6	N	8×10^4	N	5
Nov-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Dec-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Jan-10	ND	ND	ND	ND	ND	ND	4.1×10^4	3.73×10^4	4.63×10^3	6	N	2.01×10^4	8.67×10^5	8
Feb-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	3.3×10^4	4.2×10^4	2.97×10^4	4
Mar-10	ND	ND	ND	ND	ND	ND	6.13×10^4	5.3×10^4	7.37×10^3	6	6.37×10^3	9.1×10^4	9.53×10^3	5
Apr-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	N	1.84×10^4	6.7×10^5	8
May-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	N	1.62×10^4	5.33×10^5	8
Jun-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	N	1.33×10^4	2.95×10^5	8

(b)

Sampling date	Jam tomato						Spinach				
	Soil	**Root**	**Stem**	**Leaf**	**Edible portion**	**Stage of growth***	**Soil**	**Root**	**Stem**	**Edible portion**	**Stage of growth***
Jul-09	6.37×10^5	13.47×10^5	16.4×10^5	6.63×10^3	1.02×10^5	7	ND	ND	ND	ND	ND
Aug-09	3.9×10^4	8.27×10^4	3.5×10^3	4.57×10^3	3.07×10^3	4	ND	ND	ND	ND	ND
Sep-09	1.34×10^5	2.96×10^4	2.87×10^4	3.07×10^4	N	5	ND	ND	ND	ND	ND
Oct-09	ND	ND	ND	ND	ND	ND	1.28×10^4	3.67×10^5	3.13×10^3	6.17×10^4	13
Nov-09	ND	ND	ND	ND	ND	ND	N	N	N	3.57×10^3	6
Dec-09	ND	ND	ND	ND	ND	ND	N	N	N	8.93×10^3	9
Jan-10	ND	ND	ND	ND	ND	ND	1.52×10^4	3.37×10^5	N	2.11×10^5	12
Feb-10	ND	ND	ND	ND	ND	ND	N	N	N	8.47×10^3	6
Mar-10	ND	ND	ND	ND	ND	ND	N	N	N	2.54×10^4	8
Apr-10	1.73×10^5	3.17×10^4	2.83×10^4	6.5×10^4	N	5	N	N	N	7.17×10^3	5
May-10	1.71×10^5	3.23×10^4	2.76×10^4	6.07×10^4	N	7	N	N	N	2.25×10^4	8
Jun-10	ND	ND	ND	ND	ND	ND	1.13×10^4	1.99×10^5	N	1.35×10^5	12

ND = Not-determined as the fresh produce was not present at the time of sample collection; N = bacterial species was not detected; Stage of growth* = weeks

2.3.2 Heavy metal analysis

2.3.2.1 Irrigation water

Heavy metals (As, Cd, Hg, Pb) were detected in all the irrigation water samples tested, with Pb most detected and Cd, the least. The highest concentrations (mg/L) of As, Cd, Hg and Pb in the water samples were 0.028 (B), 0.027 (B), 0.057 (A1), 0.040 (C), respectively (Table 2.27). All the heavy metals, except Hg, were found at high concentrations, in these water samples, in July 2009 (winter) however, Hg was found at high concentrations in June 2010 (winter). A reduction of 61% of Pb concentrations in river water (farm A1) and a reduction of 43% of Hg concentrations in borehole water (farm A2) was observed during winter and spring (August 2009 to September 2009). During December 2009 to June 2010, Cd was not detected in majority of the water samples collected from the different farms. The mixture of dam and borehole water (farm B) as well as dam water (farm C) had the highest heavy metal content compared to the other water types tested.

Table 2.27: Heavy metal concentration in irrigation water samples obtained from different farms in KZN, over a one-year period [a].

Heavy metals (mg/L)	**Sampling date**											
	Jul-09	**Aug-09**	**Sep-09**	**Oct-09**	**Nov-09**	**Dec-09**	**Jan-10**	**Feb-10**	**Mar-10**	**Apr-10**	**May-10**	**Jun-10**
Arsenic (As)												
A1	0.014±0.001	0.004±0.000	0.003±0.002	N	0.004±0.002	0.009±0.001	N	N	0.008	0.010±0.004	0.007	N
A2	0.017±0.001	0.004±0.001	0.007±0.003	N	0.010	0.008±0.004	N	N	0.005	0.014	0.001	N
B	0.028±0.000	0.001±0.001	0.002±0.001	N	0.005±0.003	0.007	0.021	0.005	0.006	0.011±0.002	N	N
C	0.010±0.001	0.001±0.001	N	N	0.012±0.005	0.006±0.002	0.014	0.025	0.005±0.004	0.012±0.003	N	N
Cadmium (Cd)												
A1	0.015±0.000	N	0.003±0.002	0.003±0.000	0.006±0.001	N	N	N	N	0.001±0.000	0.003	N
A2	0.017±0.001	N	0.007±0.003	0.003±0.000	0.006±0.001	N	N	N	N	0.001±0.000	N	N
B	0.027±0.001	0.003±0.001	0.002±0.001	0.003±0.001	0.008±0.001	N	N	N	N	0.001±0.000	N	N
C	0.014±0.002	0.003±0.001	N	0.004±0.000	0.007±0.000	N	0.001	0.003	N	0.001	0.001	N
Mercury (Hg)												
A1	0.006±0.001	0.015±0.002	0.005±0.003	0.014±0.002	0.008±0.002	N	N	0.016±0.000	0.004±0.003	0.022±0.005	N	0.057±0.018
A2	0.007±0.001	0.014±0.003	0.008±0.001	0.014±0.002	0.007±0.002	N	N	0.016±0.001	0.011±0.006	0.013±0.002	N	0.034±0.012
B	0.033±0.001	0.020±0.003	0.028±0.013	0.016±0.004	0.013±0.003	N	N	0.017±0.001	0.007±0.003	0.015±0.002	N	0.038±0.003
C	0.043±0.013	0.026±0.002	0.022±0.001	0.014±0.001	0.015±0.002	N	N	0.017±0.002	0.006±0.006	0.018±0.002	N	0.040±0.006
Lead (Pb)												
A1	N	0.018±0.001	0.007±0.000	0.027±0.001	0.015±0.001	0.005±0.003	0.018±0.002	0.009±0.000	N	0.008±0.006	0.005±0.001	0.011
A2	N	0.018±0.002	0.003±0.003	0.025±0.002	0.016±0.001	0.002±0.001	0.017±0.001	0.007±0.004	N	0.005±0.001	N	N
B	0.037±0.001	0.001	0.019±0.002	0.025±0.000	0.016±0.002	0.006	0.016±0.001	0.005±0.002	N	0.005±0.002	0.005±0.001	N
C	0.040±0.006	N	0.015±0.003	0.026±0.003	0.017±0.001	0.007±0.000	0.020±0.002	0.011±0.008	N	0.005±0.002	0.004±0.001	N

N = heavy metal was not detected; [a] values are averages ± standard deviations (*n* = 3) and the detection limits for heavy metals (mg/L) were as follows: As (0.0053), Cd (0.0025), Hg (0.001), Pb (0.0042)

2.3.2.2 Fresh produce

The standards for the required limit of heavy metals in fresh produce could not be found, thus heavy metal concentrations found in the field samples were compared to the limits found for irrigation water. The heavy metal analysis of fresh produce samples collected from farm A1 (Tables 2.28-2.31), showed similar trends to that observed from the other farm samples tested (Appendix B, Tables B1-B14) and revealed Hg and Pb to be the most abundant heavy metals detected in the fresh produce and soil samples. The highest concentrations of Hg and Pb were detected in the jam tomato soil and root at 0.079 and 0.225 mg/L, respectively during the winter period (July 2009-August 2009) (Tables 2.30-2.31). Of the heavy metals tested, Hg was present at concentrations higher than the recommended limit. It was evident that the soil samples taken from beneath the plants had accumulated high concentrations of heavy metals, such as the concentration of Hg in the jam tomato soil during April 2010 (0.021 mg/L) (Table 2.30). The root of the plants were noted to accumulate the highest concentration of heavy metals while the edible portion of the fresh produce itself was shown to contain high concentrations of Hg, for example, the edible portion of the cabbage plant had accumulated 0.124 mg/L of Hg in October 2009 (Table 2.30). The highest concentration of heavy metals were found during the winter period July 2009 and June 2010, such as the concentration of As in the spinach plant (edible portion) which had increased by 32%, during autumn and winter (May to June 2010) (Table 2.28) and the concentration of Pb in this produce had increased by 44%, during this period (Table 2.31). Crisphead lettuce, cabbage and spinach were shown to contain the highest concentrations of heavy metals compared to the edible portions of the other fresh produce tested. This was seen in most of the produce collected from the different farms.

Table 2.28: Concentrations of As (mg/L) in fresh produce samples collected from farm A1, over a one-year period [a].

Samples	Sampling date											
	Jul-09	**Aug-09**	**Sep-09**	**Oct-09**	**Nov-09**	**Dec-09**	**Jan-10**	**Feb-10**	**Mar-10**	**Apr-10**	**May-10**	**Jun-10**
Broccoli soil	0.016±0.001	0.009±0.000	ND	N	ND	ND	ND	ND	ND	ND	ND	ND
ccoli root	0.020±0.002	0.004±0.000	ND	0.005	ND	ND	ND	ND	ND	ND	ND	ND
Broccoli stem	0.02±0.006	0.005±0.001	ND	N	ND	ND	ND	ND	ND	ND	ND	ND
Broccoli leaf	0.019±0.001	0.004±0.000	ND	N	ND	ND	ND	ND	ND	ND	ND	ND
Broccoli	0.015±0.001	0.004±0.000	ND	N	ND	ND	ND	ND	ND	ND	ND	ND
Cabbage soil	0.016±0.001	0.009±0.001	0.008±0.006	N	ND	ND	N	ND	0.006	ND	ND	ND
Cabbage root	0.016±0.002	0.003±0.000	0.002	N	ND	ND	0.022±0.024	ND	0.006±0.003	ND	ND	ND
Cabbage	0.016±0.001	0.003±0.002	N	N	ND	ND	0.016	ND	N	ND	ND	ND
Crisphead lettuce soil	0.017±0.00	0.015±0.012	0.014±0.012	N	ND	ND	N	N	0.006	0.016±0.001	N	N
Crisphead lettuce root	0.015±0.00	0.004±0.001	N	N	ND	ND	0.009	0.003	0.013±0.002	0.015±0.006	0.013±0.007	0.018±0.002
Crisphead lettuce	0.034±0.03	0.004±0.001	0.004	0.005	ND	ND	0.022±0.017	0.007	0.005±0.003	0.028±0.004	0.019±0.004	0.025±0.002
Jam tomato soil	0.016±0.001	0.007±0.002	0.014±0.003	ND	ND	ND	ND	ND	ND	0.016	0.003	ND
Jam tomato root	0.034±0.023	0.005±0.001	N	ND	ND	ND	ND	ND	ND	0.021±0.004	0.013±0.005	ND
Jam tomato stem	0.026±0.002	0.004±0.001	N	ND	ND	ND	ND	ND	ND	0.014±0.008	0.010±0.005	ND
Jam tomato leaf	0.017±0.001	0.008±0.004	N	ND	ND	ND	ND	ND	ND	0.009±0.002	0.017±0.002	ND
Jam tomato	0.016±0.002	0.004±0.001	0.001	ND	ND	ND	ND	ND	ND	0.024±0.014	0.008	ND
Spinach soil	ND	ND	ND	N	0.003	N	N	N	0.003	0.003±0.003	0.004	0.014±0.004
Spinach root	ND	ND	ND	N	0.013	N	N	0.006±0.005	0.005	0.007±0.002	0.024±0.003	0.085±0.003
Spinach stem	ND	ND	ND	N	0.005±0.003	0.005	0.020±0.012	0.005±0.003	0.006±0.001	N	0.013±0.004	0.008±0.002
Spinach	ND	ND	ND	N	0.005	0.006	0.011±0.006	0.007±0.004	0.007±0.001	0.007±0.003	0.019±0.005	0.025±0.003

ND = Not-determined, as the fresh produce was not present at the time of sample collection; N = heavy metal was not detected; [a] values are averages ± standard deviations (*n* = 3) and the detection limits for heavy metals (mg/L) were as follows: As (0.0053), Cd (0.0025), Hg (0.001), Pb (0.0042)

Table 2.29: Concentrations of Cd (mg/L) in fresh produce samples collected from farm A1, over a one-year period [a].

Samples	**Sampling date**											
	Jul-09	**Aug-09**	**Sep-09**	**Oct-09**	**Nov-09**	**Dec-09**	**Jan-10**	**Feb-10**	**Mar-10**	**Apr-10**	**May-10**	**Jun-10**
Broccoli soil	0.017±0.001	0.006±0.001	ND	0.005±0.000	ND	ND	ND	ND	ND	ND	ND	ND
Broccoli root	0.013±0.001	0.001±0.001	ND	0.004±0.000	ND	ND	ND	ND	ND	ND	ND	ND
Broccoli stem	0.02±0.007	N	ND	0.004±0.000	ND	ND	ND	ND	ND	ND	ND	ND
Broccoli leaf	0.013±0.001	N	ND	0.004±0.001	ND	ND	ND	ND	ND	ND	ND	ND
Broccoli	0.015±0.001	N	ND	0.006±0.001	ND	ND	ND	ND	ND	ND	ND	ND
Cabbage soil	0.017±0.002	0.003±0.001	0.013±0.006	0.006±0.001	ND	ND	0.001±0.001	ND	0.001±0.000	ND	ND	ND
Cabbage root	0.014±0.001	N	0.002	0.004±0.001	ND	ND	0.001±0.001	ND	N	ND	ND	ND
Cabbage	0.014±0.000	N	N	0.004±0.000	ND	ND	0.001±0.000	ND	N	ND	ND	ND
Crisphead lettuce soil	0.017±0.001	0.011±0.011	0.014±0.012	0.007±0.000	ND	ND	0.002±0.000	0.002±0.001	0.002±0.000	0.001±0.000	0.009±0.002	N
Crisphead lettuce root	0.015±0.000	0.001±0.001	N	0.005±0.000	ND	ND	0.002	N	N	0.001±0.000	N	N
Crisphead lettuce	0.034±0.032	N	0.004	0.004±0.001	ND	ND	0.005±0.000	N	N	0.007±0.001	0.001	0.003±0.002
Jam tomato soil	0.017±0.001	0.003	0.014±0.003	ND	ND	ND	ND	ND	ND	N	0.006±0.000	ND
Jam tomato root	0.031±0.031	0.002±0.001	N	ND	ND	ND	ND	ND	ND	0.003±0.001	0.003	ND
Jam tomato stem	0.012±0.005	0.001±0.001	N	ND	ND	ND	ND	ND	ND	0.002±0.001	0.001	ND
Jam tomato leaf	0.015±0.000	0.006±0.005	N	ND	ND	ND	ND	ND	ND	0.003±0.001	0.002	ND
Jam tomato	0.016±0.001	N	0.001	ND	ND	ND	ND	ND	ND	0.004±0.001	0.001	ND
Spinach soil	ND	ND	ND	0.006±0.001	0.008±0.001	N	0.006±0.001	N	0.003±0.001	0.004±0.002	0.007±0.005	0.009±0.000
Spinach root	ND	ND	ND	0.004±0.000	0.008±0.001	N	0.002±0.001	N	0.002	0.003±0.001	0.001	0.005±0.001
Spinach stem	ND	ND	ND	0.004±0.000	0.007±0.000	N	0.002±0.001	0.014	0.001±0.001	N	0.001	0.001±0.000
Spinach	ND	ND	ND	0.004±0.001	0.007±0.000	N	0.005±0.001	N	0.001±0.000	0.002±0.001	0.004	0.003±0.001

ND = Not-determined, as the fresh produce was not present at the time of sample collection; N = heavy metal was not detected; [a] values are averages ± standard deviations (*n* = 3) and the detection limits for heavy metals (mg/L) were as follows: As (0.0053), Cd (0.0025), Hg (0.001), Pb (0.0042)

Table 2.30: Concentrations of Hg (mg/L) in fresh produce samples collected from farm A1, over a one-year period [a].

Samples	Sampling date											
	Jul-09	Aug-09	Sep-09	Oct-09	Nov-09	Dec-09	Jan-10	Feb-10	Mar-10	Apr-10	May-10	Jun-10
Broccoli soil	0.006±0.001	0.015±0.002	ND	0.018±0.001	ND	ND	ND	ND	ND	ND	ND	ND
Broccoli root	N	0.019±0.005	ND	0.029±0.070	ND	ND	ND	ND	ND	ND	ND	ND
Broccoli stem	0.005±0.001	0.039±0.036	ND	0.042±0.084	ND	ND	ND	ND	ND	ND	ND	ND
Broccoli leaf	N	0.021±0.005	ND	0.052±0.001	ND	ND	ND	ND	ND	ND	ND	ND
Broccoli	0.005±0.001	0.034±0.026	ND	0.012±0.049	ND	ND	ND	ND	ND	ND	ND	ND
Cabbage soil	0.006±0.001	0.015±0.003	0.007±0.001	0.013±0.003	ND	ND	N	ND	0.001	ND	ND	ND
Cabbage root	0.004±0.003	0.019±0.001	0.027±0.002	0.098±0.002	ND	ND	N	ND	0.002±0.001	ND	ND	ND
Cabbage	0.003±0.001	0.014±0.008	0.024±0.001	0.124±0.006	ND	ND	N	ND	N	ND	ND	ND
Crisphead lettuce soil	0.005±0.001	0.012±0.001	0.012±0.007	0.017±0.023	ND	ND	N	0.016±0.001	N	0.020±0.001	N	0.053±0.005
Crisphead lettuce root	0.005±0.001	0.023±0.007	0.024±0.001	0.077±0.034	ND	ND	0.005	0.022±0.006	0.002	0.020±0.001	N	N
Crisphead lettuce	0.005±0.001	0.02±0.006	0.022±0.001	0.018±0.001	ND	ND	0.019±0.007	0.021±0.008	0.002±0.001	0.029±0.003	N	N
Jam tomato soil	0.009±0.001	0.011±0.003	0.005±0.002	ND	ND	ND	ND	ND	ND	0.021±0.002	N	ND
Jam tomato root	0.005	0.079±0.080	0.024±0.001	ND	ND	ND	ND	ND	ND	0.032±0.001	N	ND
Jam tomato stem	N	0.026±0.006	0.029±0.002	ND	ND	ND	ND	ND	ND	0.036±0.003	N	ND
Jam tomato leaf	0.010±0.003	0.067±0.047	0.025±0.002	ND	ND	ND	ND	ND	ND	0.027±0.001	N	ND
Jam tomato	0.026±0.012	0.023±0.009	0.023±0.001	ND	ND	ND	ND	ND	ND	0.038±0.002	N	ND
Spinach soil	ND	ND	ND	0.009±0.001	0.007±0.001	N	N	0.017±0.001	N	0.020±0.001	N	N
Spinach root	ND	ND	ND	0.033±0.003	0.011±0.001	N	N	0.018±0.001	0.003±0.001	0.020±0.003	N	N
Spinach stem	ND	ND	ND	0.069±0.025	0.010±0.001	N	0.003	0.019±0.001	0.004±0.002	N	N	N
Spinach	ND	ND	ND	0.018±0.001	0.010±0.006	N	0.010±0.001	0.018±0.001	0.004±0.001	0.026±0.002	N	N

ND = Not-determined, as the fresh produce was not present at the time of sample collection; N = heavy metal was not detected; [a] values are averages ± standard deviations (*n* = 3) and the detection limits for heavy metals (mg/L) were as follows: As (0.0053), Cd (0.0025), Hg (0.001), Pb (0.0042)

Table 2.31: Concentrations of Pb (mg/L) in fresh produce samples collected from farm A1, over a one-year period [a].

Samples	**Sampling date**											
	Jul-09	**Aug-09**	**Sep-09**	**Oct-09**	**Nov-09**	**Dec-09**	**Jan-10**	**Feb-10**	**Mar-10**	**Apr-10**	**May-10**	**Jun-10**
Broccoli soil	0.132±0.043	0.129±0.009	ND	0.029±0.008	ND	ND	ND	ND	ND	ND	ND	ND
Broccoli root	N	0.024±0.003	ND	0.052±0.015	ND	ND	ND	ND	ND	ND	ND	ND
Broccoli stem	N	0.028±0.011	ND	0.029±0.001	ND	ND	ND	ND	ND	ND	ND	ND
Broccoli leaf	N	0.023±0.002	ND	0.025±0.027	ND	ND	ND	ND	ND	ND	ND	ND
Broccoli	N	0.029±0.005	ND	0.172±0.039	ND	ND	ND	ND	ND	ND	ND	ND
Cabbage soil	0.087±0.055	0.204±0.003	0.143±0.015	0.096±0.002	ND	ND	0.073±0.004	ND	0.064±0.009	ND	ND	ND
Cabbage root	0.003	0.028±0.008	0.049±0.020	0.073±0.004	ND	ND	0.031±0.018	ND	0.008±0.006	ND	ND	ND
Cabbage	N	0.021±0.011	0.014±0.002	0.029±0.012	ND	ND	0.017±0.001	ND	N	ND	ND	ND
Crisphead lettuce soil	0.154±0.023	0.13±0.013	0.12±0.026	0.044±0.004	ND	ND	0.058±0.003	0.092±0.016	0.045±0.011	0.011	0.153±0.031	0.163±0.003
Crisphead lettuce root	N	0.026±0.005	0.025±0.002	0.083±0.001	ND	ND	0.022	0.011±0.003	0.006	0.035±0.004	0.003±0.002	0.014±0.004
Crisphead lettuce	N	0.016±0.003	0.013±0.001	0.025±0.014	ND	ND	0.026±0.002	0.009±0.001	N	0.034±0.011	0.006±0.002	0.017±0.002
Jam tomato soil	0.225±0.057	0.139±0.058	0.116±0.014	ND	ND	ND	ND	ND	ND	0.004	0.098±0.015	ND
Jam tomato root	N	0.038±0.010	0.022±0.002	ND	ND	ND	ND	ND	ND	0.019±0.003	0.043±0.004	ND
Jam tomato stem	N	0.024±0.001	0.02±0.006	ND	ND	ND	ND	ND	ND	0.007±0.002	0.011±0.002	ND
Jam tomato leaf	N	0.043±0.008	0.02±0.001	ND	ND	ND	ND	ND	ND	0.023±0.014	0.016±0.004	ND
Jam tomato	0.012±0.01	0.032±0.015	0.017±0.002	ND	ND	ND	ND	ND	ND	0.016±0.004	0.006±0.001	ND
Spinach soil	ND	ND	ND	0.201±0.083	0.095±0.017	0.081±0.017	0.065±0.009	0.088±0.008	0.057±0.038	0.011±0.002	0.145±0.032	0.153±0.005
Spinach root	ND	ND	ND	0.041±0.010	0.053±0.021	N	0.045±0.016	0.010±0.006	0.009±0.006	0.021±0.011	0.032±0.003	0.061±0.004
Spinach stem	ND	ND	ND	0.029±0.002	0.018±0.002	0.010±0.003	0.020±0.003	0.015±0.014	0.025	N	0.004±0.002	0.007±0.000
Spinach	ND	ND	ND	0.026±0.001	0.020±0.003	0.011±0.007	0.025±0.001	0.016±0.006	N	0.021±0.008	0.045±0.003	0.065±0.003

ND = Not-determined, as the fresh produce was not present at the time of sample collection; N = heavy metal was not detected; [a] values are averages ± standard deviations (*n* = 3) and the detection limits for heavy metals (mg/L) were as follows: As (0.0053), Cd (0.0025), Hg (0.001), Pb (0.0042)

2.4 Discussion

Fresh produce is consumed daily by many people world-wide and as the demand for fresh produce continues to increase, the need for freshwater sources for irrigation also increases. This can be very problematic for water sources that come into contact with areas where there are large confined animal operations or a large number of grazing animals, as this water can contain high microbial loads due to contamination from such regions (Hanning *et al*., 2009). Water commonly used for food crop irrigation is usually not treated and is therefore likely to contain a high microbial load (Stine *et al*., 2005). The source as well as the quality of different irrigation waters dictates the level of microbial contamination of the fresh produce. Microbes that have been implicated in different food-borne illnesses, including *E. coli*, *Salmonella* spp. and *Shigella* spp., have also been associated with contaminated irrigation water (Gast and Holt, 2000).

The guideline limit for the incidence of *E. coli* in irrigation water is 2×10^3 cfu/100 ml (DWAF, 1996). Presumptive *E. coli* was detected above this limit in the irrigation water from farm A1 and A2, with the highest counts being recorded in October 2009 at 5940 cfu/100 ml (farm A2). Presumptive *Salmonella* spp., *Shigella* spp. and coliforms were abundant in the irrigation waters sampled with the highest presumptive *Shigella* spp. counts observed in the mixed water sample (B) with a concentration of approximately, 1.45×10^4 cfu/100ml, in March 2010. It was evident that the irrigation water samples tested (except dam water) were heavily contaminated with presumptive *Shigella* spp. and coliforms, with a significant positive correlation between these microorganisms being established ($p<0.01$). Presumptive *Campylobacter* spp. was only detected in water sample C (dam water), and the population increased when the irrigation water was not in use, and the dam was allowed to be stagnant. This allowed for the growth and survival of *Campylobacter* spp., being a microaerophile (Moore *et al*., 2005), since stagnant waters limit the entrance of oxygen. Furthermore, temperature and solute concentration govern the water solubility of oxygen. In spring, the lowest microbial counts were evident in the water samples tested (except sample C) while the microbial load of the irrigation water samples were highest during the summer months. Majority of the water samples tested from farms A1, A2 and B showed high levels of these presumptive pathogens, during summer. However, the irrigation water from farm C (dam water), had high concentrations of presumptive *Campylobacter* spp., with the highest concentrations recorded from April to June 2010. It has been noted that the water quality depends partially on land use and how these water resources are managed as well as protected (Rose *et al*., 2001), as some pathogenic microorganisms may survive longer in water or soil when conditions are optimal than what has been considered to be the norm. The attachment of pathogenic bacteria to surface areas is important in their survival as they can integrate themselves into biofilms and thus, be protected from harsh conditions (Toze, 1997). It has been previously recognized that seasonal effects may have an impact on the survival of pathogens, with the different seasons in the year having a direct effect on the contaminated state of fresh produce by way of a change in the climatic temperature, rainfall, farming practices (such as fertiliser application) (Hall *et al*., 2002), and changes in the percent of ultra-violet irradiation (Griffin *et al*., 1999) etc. Also, the frequency of irrigation is affected by seasonal temperatures, therefore when fresh produce surfaces are in direct contact with the irrigation water, an increase in the microbial load as well as a higher threat of contamination can be expected (Stine *et al*., 2005).

The availability of water in the soil can lead to an increase in the microbial population in plant tissues as a result of higher turgor of plants, higher plant transpiration rate and subsequent moisture accumulation on the leaf surface (Fonseca, 2006; Coelho *et al*., 2005). This could explain why the highest concentrations of microbial contamination were obtained in summer, in most of the produce obtained from the different farms. High microbial counts were evident in most of the plants tested, with presumptive *Campylobacter* spp. and *Shigella* spp. being the most abundant microorganisms. However, presumptive *Campylobacter* spp. was not detected in the irrigation waters (A1, A2, B) throughout the study but was evident in the fresh produce and in the soil. The reason for this could be due to the microaerophilic nature of this organism (Moore *et al*., 2005), and conditions may not have favoured its proliferation in these different water sources (A1, A2, B) and thus, this organism could have entered into the viable but non-culturable (VBNC) state (Sardessai, 2005). Also, it must be noted that irrigation water was applied to these crops at least once a week and these plants may have accumulated the microorganisms from these waters. The leafy vegetables such as lettuce and spinach had the most microbial contamination as compared to broccoli and jam tomatoes. Crops that are grown closer to the ground are much more susceptible to contamination since they can come into direct contact with the contaminant, through splashed soil or manure during irrigation (Hanning *et al*., 2009). Leafy vegetables with large leaf surface areas have also been shown to have a high water holding capacity and are therefore at a greater risk of contamination during irrigation (Ilic *et al*., 2009).

It was observed that the produce was more heavily contaminated towards maturation, which is a concern. The area of the plant that had accumulated the highest concentrations of the pathogens tested was the root, and this could be as a result of the rhizosphere of the plant, which is in close proximity to the soil. The microbial load is greater in the rhizosphere as compared to the bulk soil, which is partially due to the release of root exudates organic carbon (soluble sugars, amino acids and phenols) and root turnover. Bacterial counts in this area could be about 23 times higher than in the soil area as previously reported (Godley, 2004; Newman, 1985). The high microbial load observed in the soil samples in this study, could be linked to the quality of irrigation water, as these waters are applied to the plants, hence compromising the quality of the soil. The fresh produce had accumulated higher microbial concentrations over time, as a result of repeated exposure to these contaminated water sources. Ait Melloul and Hassani (1999) investigated the use of untreated wastewater for irrigating crops in Morocco and observed that crops irrigated with untreated wastewater, showed a higher rate of salmonellosis in the children of agricultural workers (39%) compared to children of non-agriculturalists (25%). It was apparent that a relationship between the consumption of produce irrigated with untreated and treated wastes existed, although no attempt was made to determine the association. Chambers *et al.* (2002) established that contaminated water employed for irrigation, spraying, or the washing of produce to be eaten raw may increase the risk of disease. Fonseca (2006) evaluated iceberg lettuce "for yield, microbial population, and post-harvest quality either following different irrigation termination (IT) schedules or before and after a rainfall event". Lettuce that had received late IT (4 d before harvest) was reported to have higher aerobic bacteria counts and lower quality than plants that had been subjected to early IT (16 d before harvest). It was further observed that the microbial counts increased when the time between the last irrigation and harvest was shorter (Fonseca, 2006).

All water samples tested complied with the recommended limits of 0.1, 0.1 and 0.5 mg/L for As, Cd and Pb, respectively in irrigation water (Ayers and Westcot, 1985). However, all the samples tested exceeded the recommended limit of 0.001 mg/L for Hg set by the FAO and WHO (2007); with a concentration as high as 0.043 mg/L detected in dam water during the month of July 2009. The detection of heavy metals in the plant despite their absence in the irrigation water used on the farm may be due the fact that the quality of the irrigation water changes with time, as evidenced by this study. Thus, the samples may have been collected when the metals were diluted out in the samples. Sharma *et al*. (2007) evaluated heavy metal contamination in irrigation water, soil and palak (edible portion) samples during the summer and winter seasons in Varanasi, India. The heavy metal content of the irrigation water used was found to be within the recommended limits for all the heavy metals tested except for Cd. The Cd content for the soil and the edible portion of the fresh produce was also above the limit during summer (Sharma *et al.*, 2007). Similar results were obtained in this study as the Hg content of the soil and the fresh produce samples was above the recommended limit. Roychowdhury *et al*. (2005) revealed that the level of As in groundwater resulted in an increase in the concentration of this heavy metal in the soil and the plants tested. The root of the plant was shown to have taken up the most As as compared to the stem and leaves of the plant (Roychowdhury *et al*., 2005). This corroborates the finding of the present study as the roots of the plant were shown to accumulate the highest concentrations of heavy metals.

A direct link between the quality of irrigation water and that of the fresh produce was evident in this study, as the highest concentrations of the microorganisms were detected in both the plant and irrigation waters during summer. Furthermore, a link between the heavy metal quality of the irrigation waters and the fresh produce was observed as heavy metal concentrations were the highest in July 2009 and June 2010 in both irrigation water and the fresh produce tested. It is important to note that numerous factors may have influenced the quality of the irrigation waters (such as dust, soil, bird droppings, wild and domestic animals etc. (Beuchat, 2002)), therefore, it is important to constantly monitor the water sources, as it may have serious implications on consumers through the consumption of the produce irrigated with contaminated water. Also, since farmers may not possess the knowledge of how to prevent such problems, proper guidelines and recommendations should be put in place in order to prevent the possible risk of fresh produce contamination and hence disease outbreaks as a result of poor water qualities in South Africa and in particular, KwaZulu-Natal province.

2.3.1.2.2 Fresh produce from farm A2

The microbial analysis of the fresh produce collected from farm A2 (Tables 2.9-2.14), had revealed that the presumptive *Campylobacter* spp. and coliform counts were found abundantly throughout the seasons with the highest concentrations being observed in the spinach stem and crisphead lettuce root samples at 2.81×10^6 and 2.2×10^6 cfu/g in winter and spring, respectively. Presumptive *E. coli*, *L. monocytogenes* and *Salmonella* spp. were not detected in cauliflower samples collected from this farm. Of the spinach samples tested from farm A2, presumptive *E. coli* was not detected, however, presumptive *L. monocytogenes* and *Salmonella* spp. were detected in the spinach soil and spinach root samples at 3.05×10^3 and 3.27×10^4 cfu/g in October and July 2009, respectively. Presumptive *Shigella* spp. was found abundantly throughout the seasons with the highest concentration being recorded at 1.93×10^6 cfu/g in the broccoli leaf during January 2010. The fresh produce that harboured the highest concentrations of these organisms were crisphead lettuce and spinach, while broccoli and jam tomatoes had the least. The highest bacterial counts were recorded in the summer season. The following trends were observed in samples collected from the different farms: (a) the accumulative area of the plant for these presumptive bacterial pathogens were the root of the plant and the fresh produce itself as these organisms where mostly concentrated in these parts; (b) the younger the plant the less it was prone to contamination by these microorganisms and the older the plant the more the level of contamination; and (c) the accumulative area of the plant for these microbes was affected by the stage of development of the plant, because in the initial stages of development the most contaminated area was the root and overtime it had spread to other parts of the plant, such as with the presence of presumptive *E. coli* in the crisphead lettuce plant (farm A2), when the plant was 5 weeks into development, the bacteria were not detected in the plant system, however, when the plant was 8 weeks old, the bacteria were detected throughout the plant system with contamination having spread to the fresh produce itself (lettuce) at 3.63×10^3 cfu/g (Table 2.11). The soil sample, which was collected from beneath the plant also, showed a high microbial load.

(a) **Table 2.9**: The monthly variation in presumptive *Campylobacter* spp. counts (in cfu/g) on the different fresh produce collected from farm A2, over a one-year period.

Sampling date	Broccoli						Cabbage				Crisphead lettuce			
	Soil	**Root**	**Stem**	**Leaf**	**Edible portion**	**Stage of growth***	**Soil**	**Root**	**Edible portion**	**Stage of growth***	**Soil**	**Root**	**Edible portion**	**Stage of growth***
Jul-09	2.69×10^5	2.66×10^5	2.9×10^5	3.57×10^5	ND	8	2.04×10^5	9.77×10^5	1.56×10^4	6	3.23×10^5	2.08×10^5	6.8×10^5	7
Aug-09	7.17×10^4	4.1×10^5	8.73×10^3	6.7×10^4	5.9×10^4	6	19×10^5	2.18×10^5	6.43×10^4	7	3.93×10^5	12.17×10^5	7.1×10^4	6
Sep-09	6.77×10^4	11.93×10^5	2.05×10^4	2.53×10^5	2.19×10^4	7	1.77×10^5	12.57×10^5	17.97×10^5	11	9.73×10^4	21.6×10^5	7.17×10^4	5
Oct-09	ND	ND	ND	ND	ND	ND	7.27×10^4	1.52×10^5	8.23×10^4	8	1.09×10^4	1.05×10^5	6.9×10^5	7
Nov-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	7.03×10^5	4.17×10^5	4.07×10^3	5
Dec-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	8.7×10^5	4.17×10^5	4.77×10^4	8
Jan-10	9.13×10^4	8.83×10^5	2.01×10^4	2.53×10^5	1.97×10^4	7	ND	ND	ND	ND	8.27×10^5	6.43×10^5	2.95×10^4	8
Feb-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Mar-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Apr-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	8.67×10^5	4.73×10^5	4.9×10^3	5
May-10	ND	ND	ND	ND	ND	ND	1.15×10^5	3.03×10^5	1.15×10^4	3	9.63×10^5	8.67×10^5	3.83×10^4	8
Jun-10	1.27×10^5	15.63×10^5	2.47×10^4	2.93×10^5	2.43×10^4	9	2.35×10^5	13.63×10^5	2.03×10^4	6	ND	ND	ND	ND

(b)

Sampling date	Spinach					Cauliflower				
	Soil	**Root**	**Stem**	**Edible portion**	**Stage of growth***	**Soil**	**Root**	**Leaf**	**Edible portion**	**Stage of growth***
Jul-09	4.47×10^4	15.3×10^5	28.1×10^5	25.5×10^5	7	ND	ND	ND	ND	ND
Aug-09	ND	ND	ND	ND	ND	6.1×10^5	3.7×10^5	6.27×10^5	9.27×10^4	6
Sep-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Oct-09	4.67×10^4	1.52×10^5	3.13×10^4	1.21×10^4	5	ND	ND	ND	ND	ND
Nov-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Dec-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Jan-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Feb-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Mar-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Apr-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
May-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Jun-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND

ND = Not-determined as the fresh produce was not present at the time of sample collection; Stage of growth* = weeks

(a)

Table 2.10: The monthly variation in presumptive coliforms (in cfu/g) on the different fresh produce collected from farm A2, over a one-year period.

Sampling date	Broccoli						Cabbage				Crisphead lettuce			
	Soil	Root	Stem	Leaf	Edible portion	Stage of growth*	Soil	Root	Edible portion	Stage of growth*	Soil	Root	Edible portion	Stage of growth*
Jul-09	N	N	N	N	ND	8	N	N	N	6	N	N	N	7
Aug-09	4.3×10^3	1.09×10^5	9.03×10^3	9.13×10^3	1.05×10^4	6	N	4.63×10^4	5.2×10^3	7	4.47×10^3	12.13×10^5	2.92×10^4	6
Sep-09	8.27×10^3	11.83×10^5	3.1×10^4	20.17×10^5	3.5×10^4	7	3.7×10^4	1.73×10^5	8.17×10^4	11	8.67×10^5	21.97×10^5	3.47×10^4	5
Oct-09	ND	ND	ND	ND	ND	ND	5.97×10^3	1.27×10^4	N	8	3.3×10^5	9.37×10^5	8.17×10^5	7
Nov-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	4.67×10^5	3.05×10^5	9.13×10^4	5
Dec-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	3.33×10^4	1.04×10^4	3.63×10^5	8
Jan-10	5.87×10^3	6.77×10^5	N	26.27×10^5	4.97×10^4	7	ND	ND	ND	ND	2.9×10^4	1.22×10^4	4.8×10^5	8
Feb-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Mar-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Apr-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	5.27×10^5	3.17×10^5	1.05×10^5	5
May-10	ND	ND	ND	ND	ND	ND	3.07×10^3	1.16×10^4	N	3	2.55×10^4	1.03×10^4	1.57×10^5	8
Jun-10	3.2×10^3	8.3×10^4	N	4.6×10^3	5.67×10^3	9	N	N	N	6	ND	ND	ND	ND

(b)

Sampling date	Spinach					Cauliflower				
	Soil	Root	Stem	Edible portion	Stage of growth*	Soil	Root	Leaf	Edible portion	Stage of growth*
Jul-09	N	N	N	N	7	ND	ND	ND	ND	ND
Aug-09	ND	ND	ND	ND	ND	7.37×10^3	3.93×10^4	3.67×10^5	11.1×10^5	6
Sep-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Oct-09	3.9×10^4	1.58×10^5	3.87×10^3	7.13×10^3	5	ND	ND	ND	ND	ND
Nov-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Dec-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Jan-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Feb-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Mar-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Apr-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
May-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Jun-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND

ND = Not-determined as the fresh produce was not present at the time of sample collection; N = bacterial species was not detected; Stage of growth* = weeks

Table 2.11: The monthly variation in presumptive *E. coli* counts (in cfu/g) on the different fresh produce collected from farm A2, over a one-year period.

(a)

Sampling date	Broccoli						Cabbage				Crisphead lettuce			
	Soil	**Root**	**Stem**	**Leaf**	**Edible portion**	**Stage of growth***	**Soil**	**Root**	**Edible portion**	**Stage of growth***	**Soil**	**Root**	**Edible portion**	**Stage of growth***
Jul-09	N	N	N	N	ND	8	N	N	N	6	N	N	N	7
Aug-09	N	N	N	N	N	6	N	N	N	7	N	N	N	6
Sep-09	N	N	N	N	N	7	N	N	N	11	N	1.79×10^4	N	5
Oct-09	ND	ND	ND	ND	ND	ND	N	N	4.13×10^4	8	4.33×10^3	N	N	7
Nov-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	N	N	N	5
Dec-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	N	N	3.63×10^3	8
Jan-10	N	3.3×10^3	N	3.4×10^3	N	7	ND	ND	ND	ND	7.53×10^4	1.88×10^4	9.23×10^5	8
Feb-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Mar-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Apr-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	3.07×10^3	N	N	5
May-10	ND	ND	ND	ND	ND	ND	N	N	N	3	3.17×10^4	N	3.3×10^5	8
Jun-10	N	N	N	N	N	9	N	N	N	6	ND	ND	ND	ND

(b)

Sampling date	Spinach					Cauliflower				
	Soil	**Root**	**Stem**	**Edible portion**	**Stage of growth***	**Soil**	**Root**	**Leaf**	**Edible portion**	**Stage of growth***
Jul-09	N	N	N	N	7	ND	ND	ND	ND	ND
Aug-09	ND	ND	ND	ND	ND	N	N	N	N	6
Sep-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Oct-09	N	N	N	N	5	ND	ND	ND	ND	ND
Nov-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Dec-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Jan-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Feb-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Mar-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Apr-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
May-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Jun-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND

ND = Not-determined as the fresh produce was not present at the time of sample collection; N = bacterial species was not detected; Stage of growth* = weeks

Table 2.12: The monthly variation in presumptive *L. monocytogenes* counts (in cfu/g) on the different fresh produce collected from farm A2, over a one-year period.

(a)

Sampling date	Broccoli						Cabbage				Crisphead lettuce			
	Soil	**Root**	**Stem**	**Leaf**	**Edible portion**	**Stage of growth***	**Soil**	**Root**	**Edible portion**	**Stage of growth***	**Soil**	**Root**	**Edible portion**	**Stage of growth***
Jul-09	3.63×10^{3}	3.97×10^{3}	N	N	ND	8	4.13×10^{3}	N	N	6	N	N	N	7
Aug-09	3.23×10^{3}	N	N	N	N	6	N	N	N	7	N	N	N	6
Sep-09	3.17×10^{3}	N	N	N	N	7	3.6×10^{3}	4.23×10^{3}	N	11	N	N	N	5
Oct-09	ND	ND	ND	ND	ND	ND	N	N	N	8	N	N	N	7
Nov-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	N	N	N	5
Dec-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	N	N	3.17×10^{3}	8
Jan-10	3.67×10^{3}	N	N	3.23×10^{3}	N	7	ND	ND	ND	ND	N	N	3.9×10^{3}	8
Feb-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Mar-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Apr-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	N	N	3.03×10^{3}	5
May-10	ND	ND	ND	ND	ND	ND	N	N	N	3	N	N	3×10^{3}	8
Jun-10	4.5×10^{3}	5.37×10^{3}	N	N	N	9	4.13×10^{3}	N	N	6	ND	ND	ND	ND

(b)

Sampling date	Spinach					Cauliflower				
	Soil	**Root**	**Stem**	**Edible portion**	**Stage of growth***	**Soil**	**Root**	**Leaf**	**Edible portion**	**Stage of growth***
Jul-09	N	N	N	N	7	ND	ND	ND	ND	ND
Aug-09	ND	ND	ND	ND	ND	N	N	N	N	6
Sep-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Oct-09	3.05×10^{3}	N	N	N	5	ND	ND	ND	ND	ND
Nov-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Dec-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Jan-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Feb-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Mar-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Apr-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
May-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Jun-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND

ND = Not-determined as the fresh produce was not present at the time of sample collection; N = bacterial species was not detected; Stage of growth* = weeks

Table 2.13: The monthly variation in presumptive *Salmonella* spp. counts (in cfu/g) on the different fresh produce collected from farm A2, over a one-year period.

(a)

Sampling date	Broccoli						Cabbage				Crisphead lettuce			
	Soil	**Root**	**Stem**	**Leaf**	**Edible portion**	**Stage of growth***	**Soil**	**Root**	**Edible portion**	**Stage of growth***	**Soil**	**Root**	**Edible portion**	**Stage of growth***
Jul-09	N	N	N	N	ND	8	N	6.77×10^{4}	N	6	N	5.27×10^{3}	N	7
Aug-09	N	N	N	N	N	6	N	N	N	7	N	N	N	6
Sep-09	N	N	N	N	N	7	4.93×10^{3}	N	1.43×10^{4}	11	N	N	N	5
Oct-09	ND	ND	ND	ND	ND	ND	N	N	N	8	N	N	N	7
Nov-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	3.23×10^{4}	9.77×10^{3}	N	5
Dec-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	N	3.27×10^{3}	3.45×10^{3}	8
Jan-10	3.23×10^{3}	N	N	4.67×10^{3}	N	7	ND	ND	ND	ND	N	3.1×10^{3}	2.55×10^{4}	8
Feb-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Mar-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Apr-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	6.27×10^{4}	1.35×10^{4}	N	5
May-10	ND	ND	ND	ND	ND	ND	3.63×10^{3}	N	N	3	N	N	2.04×10^{4}	8
Jun-10	N	N	N	N	N	9	3.57×10^{3}	7.6×10^{4}	N	6	ND	ND	ND	ND

(b)

Sampling date	Spinach					Cauliflower				
	Soil	**Root**	**Stem**	**Edible portion**	**Stage of growth***	**Soil**	**Root**	**Leaf**	**Edible portion**	**Stage of growth***
Jul-09	N	3.27×10^{4}	N	N	7	ND	ND	ND	ND	ND
Aug-09	ND	ND	ND	ND	ND	N	N	N	N	6
Sep-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Oct-09	N	N	N	N	5	ND	ND	ND	ND	ND
Nov-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Dec-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Jan-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Feb-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Mar-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Apr-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
May-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Jun-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND

ND = Not-determined as the fresh produce was not present at the time of sample collection; N = bacterial species was not detected; Stage of growth* = weeks

Table 2.14: The monthly variation in presumptive *Shigella* spp. counts (in cfu/g) on the different fresh produce collected from farm A2, over a one-year period.

(a)

Sampling date	Broccoli						Cabbage				Crisphead lettuce			
	Soil	Root	Stem	Leaf	Edible portion	Stage of growth*	Soil	Root	Edible portion	Stage of growth*	Soil	Root	Edible portion	Stage of growth*
Jul-09	5×10^{4}	N	6.53×10^{4}	N	ND	8	1.28×10^{5}	3.06×10^{4}	3.5×10^{4}	6	1.56×10^{4}	7.03×10^{3}	9.1×10^{3}	7
Aug-09	2.85×10^{4}	7.03×10^{4}	N	N	3.03×10^{3}	6	8.17×10^{3}	3.13×10^{4}	1×10^{5}	7	5.2×10^{3}	1.04×10^{5}	8.27×10^{4}	6
Sep-09	1.05×10^{4}	5.13×10^{5}	N	10.2×10^{5}	1×10^{4}	7	3.13×10^{4}	3.4×10^{3}	2.86×10^{4}	11	6.3×10^{4}	10.03×10^{5}	6.47×10^{4}	5
Oct-09	ND	ND	ND	ND	ND	ND	5.5×10^{3}	8.2×10^{3}	N	8	1.04×10^{4}	5.87×10^{3}	3.33×10^{4}	7
Nov-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	3.87×10^{5}	1.36×10^{5}	N	5
Dec-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	3.03×10^{5}	9.03×10^{4}	3.1×10^{3}	8
Jan-10	N	N	N	19.33×10^{5}	15.13×10^{5}	7	ND	ND	ND	ND	3.87×10^{5}	8.27×10^{4}	2.11×10^{5}	8
Feb-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Mar-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Apr-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	2.85×10^{5}	8.4×10^{4}	3.03×10^{3}	5
May-10	ND	ND	ND	ND	ND	ND	1.3×10^{4}	5×10^{3}	N	3	2.05×10^{5}	7.13×10^{4}	3×10^{3}	8
Jun-10	9.37×10^{3}	3.13×10^{5}	N	3.63×10^{5}	5.5×10^{3}	9	8.77×10^{4}	2.55×10^{4}	2.98×10^{4}	6	ND	ND	ND	ND

(b)

Sampling date	Spinach					Cauliflower				
	Soil	Root	Stem	Edible portion	Stage of growth*	Soil	Root	Leaf	Edible portion	Stage of growth*
Jul-09	1.73×10^{5}	7.67×10^{4}	4.33×10^{3}	5.27×10^{3}	7	ND	ND	ND	ND	ND
Aug-09	ND	ND	ND	ND	ND	8.37×10^{4}	3.1×10^{5}	N	3.13×10^{5}	6
Sep-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Oct-09	3.67×10^{3}	3.23×10^{4}	N	N	5	ND	ND	ND	ND	ND
Nov-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Dec-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Jan-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Feb-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Mar-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Apr-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
May-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Jun-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND

ND = Not-determined as the fresh produce was not present at the time of sample collection; N = bacterial species was not detected; Stage of growth* = weeks

2.3.1.2.3 Fresh produce from farm B

The fresh produce collected from this farm had the highest bacterial load. The microbial analysis of the fresh produce collected from farm B (Tables 2.15-2.23) showed that presumptive *Campylobacter* spp., coliforms and *Shigella* spp. counts were found to be the most abundant microorganisms in the fresh produce plants tested. The highest concentration of presumptive *Campylobacter* spp. was 2.97×10^6 cfu/g detected in the crisphead lettuce soil during the winter period (August 2009). Presumptive *E. coli* was not detected in broccoli, cabbage, chinese cabbage and red cabbage collected from farm B, throughout the study, however, it was detected in bell pepper, spinach and parsley. In crisphead lettuce (edible portion), presumptive *E. coli* was detected only at 3.43×10^3 cfu/g, during the winter period (June 2010). Presumptive *L. monocytogenes* was not detected in the chinese cabbage and parsley samples collected from farm B, and this presumptive pathogen crisphead lettuce (edible portion) at a high concentration of 4.27×10^3 cfu/g, during the winter period (June 2010). Presumptive *Salmonella* spp. was not detected on cabbage, chinese cabbage, red cabbage and parsley collected from farm B, throughout this study. However, these bacteria were detected on crisphead lettuce and bell pepper samples as well as in broccoli leaf and the spinach leaf (edible portion). Presumptive *Shigella* spp. was detected in the edible portion of spinach at a concentration of 1.52 cfu/g in January 2010. The soil samples were shown to be heavily contaminated by presumptive *Campylobacter* spp. counts.

Table 2.15: The monthly variation in presumptive *Campylobacter* spp. counts (in cfu/g) on the different fresh produce collected from farm B, over a one-year period.

(a)

Sampling date	Cabbage				Chinese cabbage				Red cabbage			
	Soil	**Root**	**Edible portion**	**Stage of growth***	**Soil**	**Root**	**Edible portion**	**Stage of growth***	**Soil**	**Root**	**Edible portion**	**Stage of growth***
Jul-09	ND	ND	ND	ND	ND	ND	ND	ND	3.1×10^{4}	5×10^{5}	1.58×10^{4}	10
Aug-09	ND	ND	ND	ND	ND	ND	ND	ND	8.37×10^{4}	8.97×10^{3}	5.27×10^{3}	5
Sep-09	ND	ND	ND	ND	1.14×10^{4}	9.73×10^{4}	4.4×10^{3}	8	3.07×10^{4}	8.6×10^{4}	5.13×10^{3}	6
Oct-09	ND	ND	ND	ND	ND	ND	ND	ND	1.46×10^{5}	1.27×10^{5}	1.29×10^{4}	8
Nov-09	1.42×10^{4}	4.13×10^{4}	4.67×10^{4}	5	ND	ND	ND	ND	ND	ND	ND	ND
Dec-09	3.2×10^{5}	3.1×10^{4}	1.13×10^{4}	7	ND	ND	ND	ND	ND	ND	ND	ND
Jan-10	2.06×10^{5}	1.21×10^{5}	2.18×10^{4}	10	ND	ND	ND	ND	ND	ND	ND	ND
Feb-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Mar-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Apr-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
May-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Jun-10	2.86×10^{5}	1.86×10^{5}	4.07×10^{4}	12	ND	ND	ND	ND	ND	ND	ND	ND

(b)

Sampling date	Broccoli						Crisphead lettuce				Spinach				
	Soil	**Root**	**Stem**	**Leaf**	**Edible portion**	**Stage of growth***	**Soil**	**Root**	**Edible portion**	**Stage of growth***	**Soil**	**Root**	**Stem**	**Edible portion**	**Stage of growth***
Jul-09	6.77×10^{5}	15.17×10^{5}	N	N	N	12	4.83×10^{5}	6.5×10^{4}	3.13×10^{3}	8	1.23×10^{5}	13.97×10^{5}	2.1×10^{5}	6.87×10^{3}	9
Aug-09	22.37×10^{5}	8.43×10^{4}	3.33×10^{4}	11.73×10^{5}	23.2×10^{5}	8	2.97×10^{4}	2.81×10^{4}	14.23×10^{5}	9	10.13×10^{5}	20.77×10^{5}	5.27×10^{4}	20.53×10^{5}	11
Sep-09	3.3×10^{4}	1.91×10^{5}	5.23×10^{3}	1.72×10^{4}	3.17×10^{4}	10	ND	ND	ND	ND	4.27×10^{3}	4.4×10^{4}	3.4×10^{4}	6.17×10^{3}	12
Oct-09	ND	ND	ND	ND	ND	ND	4.37×10^{5}	25.3×10^{5}	8.07×10^{5}	6	1.49×10^{5}	4.93×10^{3}	ND	7.13×10^{4}	6
Nov-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	6.7×10^{5}	11.43×10^{5}	5.97×10^{4}	6.2×10^{4}	5
Dec-09	ND	ND	ND	ND	ND	ND	5.13×10^{5}	3.67×10^{5}	7.9×10^{4}	4	9.77×10^{4}	3.87×10^{4}	1×10^{4}	3.67×10^{5}	7
Jan-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	5.5×10^{5}	12.4×10^{5}	3.37×10^{4}	2.07×10^{5}	5
Feb-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Mar-10	3.73×10^{4}	2.27×10^{5}	8.27×10^{3}	2.39×10^{4}	4.03×10^{4}	10	ND	ND	ND	ND	ND	ND	ND	ND	ND
Apr-10	ND	ND	ND	ND	ND	ND	4.57×10^{5}	3.8×10^{5}	6.4×10^{4}	4	ND	ND	ND	ND	ND
May-10	ND	ND	ND	ND	ND	ND	3.93×10^{5}	3.1×10^{5}	5.8×10^{4}	5	ND	ND	ND	ND	ND
Jun-10	ND	ND	ND	ND	ND	ND	6.8×10^{5}	9.27×10^{5}	1.08×10^{5}	8	ND	ND	ND	ND	ND

ND = Not-determined as the fresh produce was not present at the time of sample collection; N = bacterial species was not detected; Stage of growth* = weeks

Table 2.16: The monthly variation in presumptive *Campylobacter* spp. counts (a) and presumptive coliforms (b) on parsley and bell pepper collected from farm B, over a one-year period.

(a)

Sampling date	Presumptive *Campylobacter* spp. counts (cfu/g)										
	Parsley					**Bell pepper**					
	Soil	**Root**	**Stem**	**Edible portion**	**Stage of growth***	**Soil**	**Root**	**Stem**	**Leaf**	**Edible portion**	**Stage of growth***
Jul-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Aug-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Sep-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Oct-09	3.15×10^{5}	3.2×10^{4}	5.47×10^{3}	4.37×10^{4}	8	ND	ND	ND	ND	ND	ND
Nov-09	6.4×10^{4}	6.83×10^{5}	5.3×10^{4}	6.73×10^{3}	6	ND	ND	ND	ND	ND	ND
Dec-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Jan-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Feb-10	ND	ND	ND	ND	ND	2.95×10^{4}	2.34×10^{4}	N	2.78×10^{4}	1.22×10^{4}	5
Mar-10	ND	ND	ND	ND	ND	3.27×10^{4}	2.44×10^{4}	5.4×10^{3}	3.53×10^{4}	1.56×10^{4}	11
Apr-10	ND	ND	ND	ND	ND	3.13×10^{4}	2.21×10^{4}	3.3×10^{3}	5.53×10^{4}	2.37×10^{4}	15
May-10	ND	ND	ND	ND	ND	3.02×10^{4}	2.23×10^{4}	3.4×10^{3}	3.7×10^{4}	1.34×10^{4}	12
Jun-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND

(b)

Sampling date	Presumptive coliforms (cfu/g)										
	Parsley					**Bell pepper**					
	Soil	**Root**	**Stem**	**Edible portion**	**Stage of growth***	**Soil**	**Root**	**Stem**	**Leaf**	**Edible portion**	**Stage of growth***
Jul-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Aug-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Sep-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Oct-09	N	8.77×10^{4}	1.02×10^{5}	1.05×10^{4}	8	ND	ND	ND	ND	ND	ND
Nov-09	8.1×10^{3}	14×10^{5}	7.3×10^{3}	2.01×10^{4}	6	ND	ND	ND	ND	ND	ND
Dec-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Jan-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Feb-10	ND	ND	ND	ND	ND	2.96×10^{4}	3.07×10^{4}	N	3.5×10^{4}	6.5×10^{3}	5
Mar-10	ND	ND	ND	ND	ND	4.57×10^{4}	1.14×10^{5}	N	7.3×10^{4}	8.27×10^{3}	11
Apr-10	ND	ND	ND	ND	ND	3.2×10^{4}	1.3×10^{5}	3.03×10^{3}	6.5×10^{4}	9.67×10^{3}	15
May-10	ND	ND	ND	ND	ND	3.2×10^{4}	1.23×10^{4}	N	6.87×10^{4}	5.93×10^{3}	12
Jun-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND

ND = Not-determined as the fresh produce was not present at the time of sample collection; N = bacterial species was not detected; Stage of growth* = weeks

(a)

Table 2.17: The monthly variation in presumptive coliforms (in cfu/g) on the different fresh produce collected from farm B, over a one-year period.

Sampling date	**Cabbage**				**Chinese cabbage**				**Red cabbage**			
	Soil	**Root**	**Edible portion**	**Stage of growth***	**Soil**	**Root**	**Edible portion**	**Stage of growth***	**Soil**	**Root**	**Edible portion**	**Stage of growth***
Jul-09	ND	ND	ND	ND	ND	ND	ND	ND	3.4×10^{3}	14.73×10^{5}	1.14×10^{4}	10
Aug-09	ND	ND	ND	ND	ND	ND	ND	ND	N	8.83×10^{3}	N	5
Sep-09	ND	ND	ND	ND	4.63×10^{3}	2.97×10^{4}	6.13×10^{3}	8	N	1.44×10^{4}	1.01×10^{4}	6
Oct-09	ND	ND	ND	ND	ND	ND	ND	ND	N	8.73×10^{4}	1.92×10^{4}	8
Nov-09	1.08×10^{5}	4.63×10^{4}	4.3×10^{4}	5	ND	ND	ND	ND	ND	ND	ND	ND
Dec-09	5×10^{3}	N	5.43×10^{4}	7	ND	ND	ND	ND	ND	ND	ND	ND
Jan-10	6.33×10^{4}	7.07×10^{4}	8.07×10^{4}	10	ND	ND	ND	ND	ND	ND	ND	ND
Feb-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Mar-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Apr-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
May-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Jun-10	3.37×10^{4}	4.73×10^{4}	4.8×10^{4}	12	ND	ND	ND	ND	ND	ND	ND	ND

(b)

Sampling date	**Broccoli**						**Crisphead lettuce**				**Spinach**				
	Soil	**Root**	**Stem**	**Leaf**	**Edible portion**	**Stage of growth***	**Soil**	**Root**	**Edible portion**	**Stage of growth***	**Soil**	**Root**	**Stem**	**Edible portion**	**Stage of growth***
Jul-09	N	N	N	N	N	12	3.23×10^{3}	9.33×10^{3}	N	8	1.36×10^{4}	1.4×10^{4}	N	6.93×10^{3}	9
Aug-09	27.47×10^{5}	6.33×10^{4}	2.39×10^{4}	18.77×10^{5}	4.93×10^{5}	8	29.7×10^{5}	29.13×10^{5}	12.97×10^{5}	9	23.47×10^{5}	25.87×10^{5}	6.5×10^{4}	23.47×10^{5}	11
Sep-09	5.33×10^{3}	3.67×10^{4}	5.27×10^{3}	N	6.07×10^{3}	10	ND	ND	ND	ND	N	3.97×10^{3}	1.75×10^{4}	N	12
Oct-09	ND	ND	ND	ND	ND	ND	1.17×10^{5}	20.47×10^{5}	2.32×10^{5}	6	N	6.33×10^{5}	ND	6.43×10^{4}	6
Nov-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	7.67×10^{5}	22.03×10^{5}	1.39×10^{5}	2.07×10^{4}	5
Dec-09	ND	ND	ND	ND	ND	ND	4×10^{4}	1.82×10^{4}	N	4	7.83×10^{3}	3.77×10^{4}	3.5×10^{3}	7.07×10^{5}	7
Jan-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	11.13×10^{5}	24.5×10^{5}	1.7×10^{5}	2.08×10^{5}	5
Feb-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Mar-10	6.47×10^{3}	7.33×10^{4}	N	3.47×10^{3}	7.37×10^{3}	10	ND	ND	ND	ND	ND	ND	ND	ND	ND
Apr-10	ND	ND	ND	ND	ND	ND	3.03×10^{4}	1.62×10^{4}	N	4	ND	ND	ND	ND	ND
May-10	ND	ND	ND	ND	ND	ND	3.27×10^{4}	1.55×10^{4}	N	5	ND	ND	ND	ND	ND
Jun-10	ND	ND	ND	ND	ND	ND	5.07×10^{3}	1.28×10^{4}	6.53×10^{4}	8	ND	ND	ND	ND	ND

ND = Not-determined as the fresh produce was not present at the time of sample collection; N = bacterial species was not detected; Stage of growth* = weeks

(a)

Table 2.18: The monthly variation in presumptive *E. coli* counts (in cfu/g) on the different fresh produce collected from farm B, over a one-year period.

Sampling date	Cabbage				Chinese cabbage				Red cabbage			
	Soil	Root	Edible portion	Stage of growth*	Soil	Root	Edible portion	Stage of growth*	Soil	Root	Edible portion	Stage of growth*
Jul-09	ND	ND	ND	ND	ND	ND	ND	ND	N	N	N	10
Aug-09	ND	ND	ND	ND	ND	ND	ND	ND	N	N	N	5
Sep-09	ND	ND	ND	ND	N	N	N	8	N	N	N	6
Oct-09	ND	ND	ND	ND	ND	ND	ND	ND	N	N	N	8
Nov-09	N	N	N	5	ND	ND	ND	ND	ND	ND	ND	ND
Dec-09	N	N	N	7	ND	ND	ND	ND	ND	ND	ND	ND
Jan-10	N	N	N	10	ND	ND	ND	ND	ND	ND	ND	ND
Feb-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Mar-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Apr-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
May-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Jun-10	N	N	N	12	ND	ND	ND	ND	ND	ND	ND	ND

(b)

Sampling date	Broccoli						Crisphead lettuce				Spinach				
	Soil	Root	Stem	Leaf	Edible portion	Stage of growth*	Soil	Root	Edible portion	Stage of growth*	Soil	Root	Stem	Edible portion	Stage of growth*
Jul-09	N	N	N	N	N	12	N	N	N	8	N	N	N	N	9
Aug-09	N	N	N	N	N	8	N	N	N	9	N	N	N	N	11
Sep-09	N	N	N	N	N	10	ND	ND	ND	ND	N	N	N	N	12
Oct-09	ND	ND	ND	ND	ND	ND	N	N	N	6	N	N	ND	7.9×10^4	6
Nov-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	N	N	3.23×10^3	7.17×10^3	5
Dec-09	ND	ND	ND	ND	ND	ND	N	N	N	4	N	N	N	N	7
Jan-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	N	5.43×10^3	3.03×10^3	1.21×10^4	5
Feb-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Mar-10	N	N	N	N	N	10	ND	ND	ND	ND	ND	ND	ND	ND	ND
Apr-10	ND	ND	ND	ND	ND	ND	N	N	N	4	ND	ND	ND	ND	ND
May-10	ND	ND	ND	ND	ND	ND	N	N	N	5	ND	ND	ND	ND	ND
Jun-10	ND	ND	ND	ND	ND	ND	N	N	3.43×10^3	8	ND	ND	ND	ND	ND

ND = Not-determined as the fresh produce was not present at the time of sample collection; N = bacterial species was not detected; Stage of growth* = weeks

Table 2.19: The monthly variation in presumptive *E. coli* (a) and *L. monocytogenes* (b) counts on parsley and bell pepper collected from farm B, over a one-year period.

(a)

Sampling date	**Presumptive *E. coli* counts (cfu/g)**										
	Parsley					**Bell pepper**					
	Soil	**Root**	**Stem**	**Edible portion**	**Stage of growth***	**Soil**	**Root**	**Stem**	**Leaf**	**Edible portion**	**Stage of growth***
Jul-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Aug-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Sep-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Oct-09	N	N	N	4.37×10^4	8	ND	ND	ND	ND	ND	ND
Nov-09	N	N	N	N	6	ND	ND	ND	ND	ND	ND
Dec-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Jan-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Feb-10	ND	ND	ND	ND	ND	N	N	N	N	N	5
Mar-10	ND	ND	ND	ND	ND	N	N	N	N	3.2×10^3	11
Apr-10	ND	ND	ND	ND	ND	N	N	N	3.17×10^3	5.27×10^3	15
May-10	ND	ND	ND	ND	ND	N	N	N	N	N	12
Jun-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND

(b)

Sampling date	**Presumptive *L. monocytogenes* counts (cfu/g)**										
	Parsley					**Bell pepper**					
	Soil	**Root**	**Stem**	**Edible portion**	**Stage of growth***	**Soil**	**Root**	**Stem**	**Leaf**	**Edible portion**	**Stage of growth***
Jul-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Aug-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Sep-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Oct-09	N	N	N	N	8	ND	ND	ND	ND	ND	ND
Nov-09	N	N	N	N	6	ND	ND	ND	ND	ND	ND
Dec-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Jan-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Feb-10	ND	ND	ND	ND	ND	N	N	N	N	N	5
Mar-10	ND	ND	ND	ND	ND	N	N	N	N	3.57×10^3	11
Apr-10	ND	ND	ND	ND	ND	N	N	N	N	4.63×10^3	15
May-10	ND	ND	ND	ND	ND	N	N	N	N	3.73×10^3	12
Jun-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND

ND = Not-determined as the fresh produce was not present at the time of sample collection; N = bacterial species was not detected; Stage of growth* = weeks

Table 2.20: The monthly variation in presumptive *L. monocytogenes* counts (in cfu/g) on the different fresh produce collected from farm B, over a one-year period.

(a)

Sampling date	Cabbage				Chinese cabbage				Red cabbage			
	Soil	Root	Edible portion	Stage of growth*	Soil	Root	Edible portion	Stage of growth*	Soil	Root	Edible portion	Stage of growth*
Jul-09	ND	ND	ND	ND	ND	ND	ND	ND	3.03×10^{3}	N	3.37×10^{3}	10
Aug-09	ND	ND	ND	ND	ND	ND	ND	ND	N	N	N	5
Sep-09	ND	ND	ND	ND	N	N	N	8	3.13×10^{3}	N	N	6
Oct-09	ND	ND	ND	ND	ND	ND	ND	ND	N	N	N	8
Nov-09	3.1×10^{3}	3.93×10^{3}	N	5	ND	ND	ND	ND	ND	ND	ND	ND
Dec-09	N	N	N	7	ND	ND	ND	ND	ND	ND	ND	ND
Jan-10	3.07×10^{3}	4.53×10^{3}	N	10	ND	ND	ND	ND	ND	ND	ND	ND
Feb-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Mar-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Apr-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
May-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Jun-10	6.27×10^{3}	9.23×10^{3}	N	12	ND	ND	ND	ND	ND	ND	ND	ND

(b)

Sampling date	Broccoli						Crisphead lettuce				Spinach				
	Soil	Root	Stem	Leaf	Edible portion	Stage of growth*	Soil	Root	Edible portion	Stage of growth*	Soil	Root	Stem	Edible portion	Stage of growth*
Jul-09	8.97×10^{3}	4.77×10^{3}	N	N	N	12	1.36×10^{4}	5.97×10^{3}	N	8	N	N	N	N	9
Aug-09	N	N	N	N	N	8	3.2×10^{3}	4.73×10^{3}	N	9	N	N	N	N	11
Sep-09	3.17×10^{3}	3.05×10^{3}	N	N	N	10	ND	ND	ND	ND	N	N	N	N	12
Oct-09	ND	ND	ND	ND	ND	ND	N	N	N	6	N	N	ND	N	6
Nov-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	N	N	N	N	5
Dec-09	ND	ND	ND	ND	ND	ND	N	N	N	4	N	3.2×10^{3}	N	N	7
Jan-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	N	N	N	3.13×10^{3}	5
Feb-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Mar-10	4.03×10^{3}	4.5×10^{3}	N	3.27×10^{3}	N	10	ND	ND	ND	ND	ND	ND	ND	ND	ND
Apr-10	ND	ND	ND	ND	ND	ND	N	N	N	4	ND	ND	ND	ND	ND
May-10	ND	ND	ND	ND	ND	ND	N	N	N	5	ND	ND	ND	ND	ND
Jun-10	ND	ND	ND	ND	ND	ND	1.25×10^{4}	4.53×10^{3}	4.27×10^{3}	8	ND	ND	ND	ND	ND

ND = Not-determined as the fresh produce was not present at the time of sample collection; N = bacterial species was not detected; Stage of growth* = weeks

Table 2.21: The monthly variation in presumptive *Salmonella* spp. counts (in cfu/g) on the different fresh produce collected from farm B, over a one-year period.

(a)

Sampling date	Cabbage				Chinese cabbage				Red cabbage			
	Soil	**Root**	**Edible portion**	**Stage of growth***	**Soil**	**Root**	**Edible portion**	**Stage of growth***	**Soil**	**Root**	**Edible portion**	**Stage of growth***
Jul-09	ND	ND	ND	ND	ND	ND	ND	ND	N	N	N	10
Aug-09	ND	ND	ND	ND	ND	ND	ND	ND	N	N	N	5
Sep-09	ND	ND	ND	ND	N	N	N	8	N	N	N	6
Oct-09	ND	ND	ND	ND	ND	ND	ND	ND	N	N	N	8
Nov-09	N	N	N	5	ND	ND	ND	ND	ND	ND	ND	ND
Dec-09	N	N	N	7	ND	ND	ND	ND	ND	ND	ND	ND
Jan-10	N	N	N	10	ND	ND	ND	ND	ND	ND	ND	ND
Feb-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Mar-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Apr-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
May-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Jun-10	N	N	N	12	ND	ND	ND	ND	ND	ND	ND	ND

(b)

Sampling date	Broccoli						Crisphead lettuce				Spinach				
	Soil	**Root**	**Stem**	**Leaf**	**Edible portion**	**Stage of growth***	**Soil**	**Root**	**Edible portion**	**Stage of growth***	**Soil**	**Root**	**Stem**	**Edible portion**	**Stage of growth***
Jul-09	N	N	N	N	N	12	N	N	N	8	N	N	N	N	9
Aug-09	N	N	N	N	N	8	N	N	N	9	N	N	N	N	11
Sep-09	N	N	N	N	N	10	ND	ND	ND	ND	N	N	N	N	12
Oct-09	ND	ND	ND	ND	ND	ND	N	N	N	6	N	N	N	N	6
Nov-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	N	N	N	N	5
Dec-09	ND	ND	ND	ND	ND	ND	6.37×10^{3}	N	N	4	N	N	N	N	7
Jan-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	N	N	N	3.67×10^{3}	5
Feb-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Mar-10	N	N	N	3.13×10^{3}	N	10	ND	ND	ND	ND	ND	ND	ND	ND	ND
Apr-10	ND	ND	ND	ND	ND	ND	5.37×10^{3}	N	N	4	ND	ND	ND	ND	ND
May-10	ND	ND	ND	ND	ND	ND	N	N	3.33×10^{3}	5	ND	ND	ND	ND	ND
Jun-10	ND	ND	ND	ND	ND	ND	N	N	5.37×10^{3}	8	ND	ND	ND	ND	ND

ND = Not-determined as the fresh produce was not present at the time of sample collection; N = bacterial species was not detected; Stage of growth* = weeks

Table 2.22: The monthly variation in presumptive *Salmonella* spp. (a) and *Shigella* spp. (b) counts on parsley and bell pepper collected from farm B, over a one-year period.

(a)

Sampling date	**Presumptive *Salmonella* spp. counts (cfu/g)**										
	Parsley					**Bell pepper**					
	Soil	**Root**	**Stem**	**Edible portion**	**Stage of growth***	**Soil**	**Root**	**Stem**	**Leaf**	**Edible portion**	**Stage of growth***
Jul-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Aug-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Sep-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Oct-09	N	N	N	N	8	ND	ND	ND	ND	ND	ND
Nov-09	N	N	N	N	6	ND	ND	ND	ND	ND	ND
Dec-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Jan-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Feb-10	ND	ND	ND	ND	ND	N	N	N	3.27×10^{3}	N	5
Mar-10	ND	ND	ND	ND	ND	N	N	N	3.73×10^{3}	3.13×10^{3}	11
Apr-10	ND	ND	ND	ND	ND	N	N	N	3.23×10^{3}	3.37×10^{3}	15
May-10	ND	ND	ND	ND	ND	N	N	N	3.87×10^{3}	3.37×10^{3}	12
Jun-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND

(b)

Sampling date	**Presumptive *Shigella* spp. counts (cfu/g)**										
	Parsley					**Bell pepper**					
	Soil	**Root**	**Stem**	**Edible portion**	**Stage of growth***	**Soil**	**Root**	**Stem**	**Leaf**	**Edible portion**	**Stage of growth***
Jul-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Aug-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Sep-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Oct-09	N	1.16×10^{4}	N	5.07×10^{4}	8	ND	ND	ND	ND	ND	ND
Nov-09	3.97×10^{3}	3.4×10^{5}	N	N	6	ND	ND	ND	ND	ND	ND
Dec-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Jan-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Feb-10	ND	ND	ND	ND	ND	1.55×10^{5}	2.91×10^{4}	N	2.42×10^{5}	9.07×10^{3}	5
Mar-10	ND	ND	ND	ND	ND	1.35×10^{5}	7.03×10^{4}	N	2.64×10^{5}	1.75×10^{4}	11
Apr-10	ND	ND	ND	ND	ND	1.29×10^{5}	5.23×10^{4}	N	2.41×10^{5}	2.55×10^{4}	15
May-10	ND	ND	ND	ND	ND	1.22×10^{5}	7.57×10^{4}	N	2.72×10^{5}	1.85×10^{4}	12
Jun-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND

ND = Not-determined as the fresh produce was not present at the time of sample collection; N = bacterial species was not detected; Stage of growth* = weeks

Table 2.23: The monthly variation in presumptive *Shigella* spp. counts (in cfu/g) on the different fresh produce collected from farm B, over a one-year period.

(a)

Sampling date	Cabbage				Chinese cabbage				Red cabbage			
	Soil	**Root**	**Edible portion**	**Stage of growth***	**Soil**	**Root**	**Edible portion**	**Stage of growth***	**Soil**	**Root**	**Edible portion**	**Stage of growth***
Jul-09	ND	ND	ND	ND	ND	ND	ND	ND	1.55×10^{5}	14.03×10^{5}	8.33×10^{4}	10
Aug-09	ND	ND	ND	ND	ND	ND	ND	ND	3.43×10^{3}	3.73×10^{3}	3.37×10^{3}	5
Sep-09	ND	ND	ND	ND	N	1.64×10^{5}	3.6×10^{3}	8	N	7.2×10^{4}	N	6
Oct-09	ND	ND	ND	ND	ND	ND	ND	ND	N	2.87×10^{4}	N	8
Nov-09	6.8×10^{5}	1.17×10^{5}	8.5×10^{3}	5	ND	ND	ND	ND	ND	ND	ND	ND
Dec-09	4.17×10^{3}	3.13×10^{3}	4.3×10^{4}	7	ND	ND	ND	ND	ND	ND	ND	ND
Jan-10	5.13×10^{5}	1.95×10^{5}	1.05×10^{4}	10	ND	ND	ND	ND	ND	ND	ND	ND
Feb-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Mar-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Apr-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
May-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Jun-10	3.1×10^{5}	1.54×10^{5}	8.53×10^{3}	12	ND	ND	ND	ND	ND	ND	ND	ND

(b)

Sampling date	Broccoli						Crisphead lettuce				Spinach				
	Soil	**Root**	**Stem**	**Leaf**	**Edible portion**	**Stage of growth***	**Soil**	**Root**	**Edible portion**	**Stage of growth***	**Soil**	**Root**	**Stem**	**Edible portion**	**Stage of growth***
Jul-09	1.99×10^{5}	25.17×10^{5}	1.98×10^{5}	2.47×10^{5}	N	12	3.93×10^{5}	23.77×10^{5}	1.49×10^{4}	8	1.2×10^{5}	13.03×10^{5}	N	9.57×10^{3}	9
Aug-09	3.2×10^{4}	1.56×10^{5}	N	5.83×10^{3}	3.13×10^{3}	8	3.5×10^{3}	4.4×10^{4}	2.15×10^{4}	9	2.61×10^{4}	2.6×10^{5}	N	1.43×10^{4}	11
Sep-09	N	2.96×10^{5}	3.27×10^{3}	N	7.43×10^{3}	10	ND	ND	ND	ND	N	4.07×10^{3}	N	4.4×10^{3}	12
Oct-09	ND	ND	ND	ND	ND	ND	N	2.35×10^{5}	8.03×10^{4}	6	N	7.4×10^{5}	ND	1.07×10^{4}	6
Nov-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	2.87×10^{5}	6×10^{5}	4.7×10^{3}	3.35×10^{3}	5
Dec-09	ND	ND	ND	ND	ND	ND	1.05×10^{4}	2.92×10^{4}	N	4	3.63×10^{3}	1.52×10^{4}	3.23×10^{3}	3.2×10^{4}	7
Jan-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	2.52×10^{5}	5.3×10^{5}	5.5×10^{3}	1.52×10^{5}	5
Feb-10	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Mar-10	6.37×10^{3}	3.5×10^{5}	N	9.33×10^{3}	1.15×10^{4}	10	ND	ND	ND	ND	ND	ND	ND	ND	ND
Apr-10	ND	ND	ND	ND	ND	ND	9.47×10^{3}	2.69×10^{4}	N	4	ND	ND	ND	ND	ND
May-10	ND	ND	ND	ND	ND	ND	9.23×10^{3}	2.54×10^{4}	N	5	ND	ND	ND	ND	ND
Jun-10	ND	ND	ND	ND	ND	ND	2.92×10^{5}	20.77×10^{5}	1.23×10^{4}	8	ND	ND	ND	ND	ND

ND = Not-determined as the fresh produce was not present at the time of sample collection; N = bacterial species was not detected; Stage of growth* = weeks

2.3.1.2.4 Fresh produce from farm C

The microbial analysis of the fresh produce samples collected from farm C (Tables 2.24-2.26), revealed that presumptive *Campylobacter* spp. and coliforms were found to be abundant with the highest concentrations observed in the cabbage (edible portion) and jam tomato soil samples at 1.94×10^6 and 2.55×10^6 cfu/g in August 2009 and January 2010, respectively. Presumptive *E. coli* and *Salmonella* spp. counts were only detected in the cabbage soil and the jam tomato leaf samples at 6.3×10^3 (July 2009) and 3.3×10^3 (February 2009) cfu/g, respectively. Presumptive *L. monocytogenes* was not detected in the edible portions of the fresh produce sampled. Presumptive *Shigella* spp. was found to be abundant in the fresh produce, with the highest concentration being recorded at 2.94×10^6 cfu/g in the cabbage sample (edible portion) during winter (July 2009). No fresh produce samples were available for collection from farm C from March 2010-June 2010, as this farmer specialises in orange farming.

Table 2.24: The monthly variation in presumptive *Campylobacter* spp. (a) and presumptive coliform (b) counts on the different fresh produce collected from farm C, over a one-year period.

(a)

Sampling date	Presumptive *Campylobacter* spp. counts (cfu/g)									
	Cabbage				Jam tomato					
	Soil	Root	Edible portion	Stage of growth*	Soil	Root	Stem	Leaf	Edible portion	Stage of growth*
Jul-09	5.07×10^{4}	1.48×10^{5}	6.23×10^{3}	12	ND	ND	ND	ND	ND	ND
Aug-09	6.57×10^{4}	14.27×10^{5}	19.37×10^{5}	6	ND	ND	ND	ND	ND	ND
Sep-09	3.07×10^{3}	3.37×10^{4}	3.27×10^{3}	8	ND	ND	ND	ND	ND	ND
Oct-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Nov-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Dec-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Jan-10	ND	ND	ND	ND	3.23×10^{3}	1.55×10^{4}	N	3.07×10^{3}	ND	ND
Feb-10	ND	ND	ND	ND	N	9.67×10^{3}	N	3.63×10^{3}	5.57×10^{3}	6

(b)

Sampling date	Presumptive coliforms (cfu/g)									
	Cabbage				Jam tomato					
	Soil	Root	Edible portion	Stage of growth*	Soil	Root	Stem	Leaf	Edible portion	Stage of growth*
Jul-09	6.33×10^{3}	1.35×10^{4}	6.53×10^{4}	12	ND	ND	ND	ND	ND	ND
Aug-09	17.97×10^{5}	7.07×10^{5}	4.27×10^{5}	6	ND	ND	ND	ND	ND	ND
Sep-09	3.13×10^{4}	3.23×10^{3}	N	8	ND	ND	ND	ND	ND	ND
Oct-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Nov-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Dec-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Jan-10	ND	ND	ND	ND	25.47×10^{5}	16.17×10^{5}	4.33×10^{3}	3.5×10^{4}	ND	ND
Feb-10	ND	ND	ND	ND	23.33×10^{5}	3.13×10^{5}	N	1.86×10^{5}	N	6

ND = Not-determined as the fresh produce was not present at the time of sample collection; N = bacterial species was not detected; Stage of growth* = weeks

(a) **Table 2.25:** The monthly variation in presumptive *E. coli* (a) and *L. monocytogenes* (b) counts on the different fresh produce collected from farm C, over a one-year period.

Sampling date	**Presumptive *E. coli* counts (cfu/g)**									
	Cabbage				**Jam tomato**					
	Soil	**Root**	**Edible portion**	**Stage of growth***	**Soil**	**Root**	**Stem**	**Leaf**	**Edible portion**	**Stage of growth***
Jul-09	6.3×10^{3}	N	N	12	ND	ND	ND	ND	ND	ND
Aug-09	N	N	N	6	ND	ND	ND	ND	ND	ND
Sep-09	N	N	N	8	ND	ND	ND	ND	ND	ND
Oct-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Nov-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Dec-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Jan-10	ND	ND	ND	ND	N	N	N	N	ND	ND
Feb-10	ND	ND	ND	ND	N	N	N	N	N	6

(b)

Sampling date	**Presumptive *L. monocytogenes* counts (cfu/g)**									
	Cabbage				**Jam tomato**					
	Soil	**Root**	**Edible portion**	**Stage of growth***	**Soil**	**Root**	**Stem**	**Leaf**	**Edible portion**	**Stage of growth***
Jul-09	N	3.13×10^{3}	N	12	ND	ND	ND	ND	ND	ND
Aug-09	7.27×10^{3}	6.57×10^{3}	N	6	ND	ND	ND	ND	ND	ND
Sep-09	N	N	N	8	ND	ND	ND	ND	ND	ND
Oct-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Nov-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Dec-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Jan-10	ND	ND	ND	ND	6.2×10^{3}	7.37×10^{3}	N	N	ND	ND
Feb-10	ND	ND	ND	ND	6.3×10^{3}	5.27×10^{3}	N	3.57×10^{3}	N	6

ND = Not-determined as the fresh produce was not present at the time of sample collection; N = bacterial species was not detected; Stage of growth* = weeks

Table 2.26: The monthly variation in presumptive *Salmonella* spp. (a) and *Shigella* spp. (b) counts on the different fresh produce collected from farm C, over a one-year period.

(a)

Sampling date	Presumptive *Salmonella* spp. counts (cfu/g)									
	Cabbage				**Jam tomato**					
	Soil	**Root**	**Edible portion**	**Stage of growth***	**Soil**	**Root**	**Stem**	**Leaf**	**Edible portion**	**Stage of growth***
Jul-09	N	N	N	12	ND	ND	ND	ND	ND	ND
Aug-09	N	N	N	6	ND	ND	ND	ND	ND	ND
Sep-09	N	N	N	8	ND	ND	ND	ND	ND	ND
Oct-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Nov-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Dec-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Jan-10	ND	ND	ND	ND	N	N	N	N	ND	ND
Feb-10	ND	ND	ND	ND	N	N	N	3.3×10^{3}	N	6

(b)

Sampling date	Presumptive *Shigella* spp. counts (cfu/g)									
	Cabbage				**Jam tomato**					
	Soil	**Root**	**Edible portion**	**Stage of growth***	**Soil**	**Root**	**Stem**	**Leaf**	**Edible portion**	**Stage of growth***
Jul-09	4×10^{3}	7.83×10^{5}	29.4×10^{5}	12	ND	ND	ND	ND	ND	ND
Aug-09	1.96×10^{4}	4.2×10^{3}	N	6	ND	ND	ND	ND	ND	ND
Sep-09	6.23×10^{4}	3.4×10^{4}	8.9×10^{3}	8	ND	ND	ND	ND	ND	ND
Oct-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Nov-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Dec-09	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Jan-10	ND	ND	ND	ND	6.83×10^{4}	5.17×10^{4}	3.87×10^{3}	9.27×10^{3}	ND	ND
Feb-10	ND	ND	ND	ND	1.98×10^{5}	3.13×10^{5}	3.23×10^{3}	1.89×10^{4}	6.43×10^{3}	6

ND = Not-determined as the fresh produce was not present at the time of sample collection; N = bacterial species was not detected; Stage of growth* = weeks

CHAPTER 3: EFFECT OF *P. aeruginosa* ON THE UPTAKE OF BACTERIAL PATHOGENS FROM SOIL TO THE FINAL PRODUCE

3.1 Introduction

An increase in the consumption of fresh minimally processed produce has renewed interest on the role of the microenvironment of the fresh produce on produce safety, since this environment can either assist or obstruct food safety, affecting production and the persistence of pathogens on plants. This understanding may assist in developing novel technologies in order to improve post-harvest treatment and handling of fresh produce. This is important as pathogens on produce play a significant role in causing food-borne illnesses world-wide (Aruscavage *et al.*, 2006).

The plant rhizosphere is a "major soil ecological environment for plant-microbe interactions" involving the colonization of various microbes in and around the roots of the developing plant. This colonization can either result in associative, symbiotic, neutralistic or parasitic relations depending on the nutrient status of the plant in the soil environment (Sindhu *et al.*, 2002). Microorganisms may be associated in two ways; in one situation two organisms benefit mutually from each other, in the other, the presence of the organism or its products may be detrimental to the growth of the other organism. An example of the latter is when a bacterium uses an antagonistic action towards another (Rettger, 1905). The occurrence of antagonists in the soil environment may play a role in the reduction of numbers of human pathogens (Johannessen *et al.*, 2005). Janisiewicz *et al.* (1999) found a decrease in the survival of *Escherichia coli* O157:H7 in unpasteurized cider as compared to that of sterilized apple juice. It was suggested that the decrease in survival of this pathogen may have resulted from interactions with natural populations (Janisiewicz *et al.*, 1999), which could be as a result of competition.

Competition occurs when microorganisms try to acquire the same resource from their environment (Prescott *et al.*, 2005). If an organism is able to grow rapidly, this is a competitive advantage, because this organism is able to establish dominance at the time when nutrient levels are high or when there are fewer nutrients that remain. Also, competitors who have very efficient modes of nutrient uptake or have the ability to produce antimicrobial compounds, have this competitive advantage (Beattie and Lindow 1994; Aruscavage *et al.*, 2006). Schuenzel and Harrison (2002) found that approximately 3% of epiphytes that had been isolated from produce contained inhibitory compounds that were effective against one or more of the subsequent pathogens: *Staphylococcus aureus*, *E. coli* O157, *Salmonella* spp. and *L. monocytogenes*. It was further shown that the isolates from shredded lettuce were more likely to produce inhibitory compounds that were successful against all four pathogens tested. Most of the inhibitory epiphytes were shown to be gram-negative, with the highest percentage comprising of Pseudomonads (Schuenzel and Harrison, 2002; Aruscavage *et al.*, 2006). However, many epiphytic bacteria may support the growth of "immigrants" in establishing themselves and some plant pathogens can actually support the survival of members of the Enterobacteriaceae on produce (Cooley *et al*., 2006; Wells and Butterfield, 1997).

Gram (1993) assessed the antibacterial effects of 209 *Pseudomonas* isolates from rotten iced fish as well as freshly caught fish using target organisms in agar diffusion assays. Approximately, a third of the strains inhibited the growth of either one or many of the target microbes tested namely, *E. coli*, *Shewanella putrefaciens*, *Aeromonas sobria*, *Pseudomonas fluorescens*, *L. monocytogenes* and *S. aureus*. It was found that this inhibitory action was more distinct among the strains that produced siderophores; also the presence of iron was found to eliminate the antibacterial action of two-thirds of the inhibitory strains. It was suggested that the "siderophore-mediated competition for iron may explain the inhibitory activity of these strains" (Gram, 1993). This shows that certain *Pseudomonas* spp. may have inhibitory action towards human pathogens such as *E. coli* and *L. monocytogenes.* Johannessen *et al*. (2005) established the inhibitory effect of *Pseudomonas* spp. on the growth of *E. coli* O157:H7 *in vitro.* It was assumed that these Pseudomonads may have an antagonistic effect on the pathogens present in the soil. However, it was observed that over time the pathogen (*E. coli* O157:H7) was able to persist in the soil environment (Johannessen *et al*., 2005). Since these Pseudomonads have been shown to inhibit human pathogens, it is important to test their effect on the uptake of food-borne pathogens to the fresh produce, as well as determine the optimal conditions for the inhibition. These optimal conditions could provide a means of removing/reducing the bacterial contamination of fresh produce through inhibiting food-borne pathogens in soil and thereby preventing their uptake into the fresh produce, and thus provide an alternative to the global problem of food-borne disease outbreaks.

3.2 Materials and methods

3.2.1 Sample collection and isolate purification

Irrigation water samples were collected from a farm in Camperdown and analysed for the presence of different microbial pathogens using the membrane filtration technique as described in chapter 2 and the membranes were placed onto different selective media and incubated appropriately for the growth of selected presumptive pathogens (Table 2.1) in addition, enumeration of presumptive *P. aeruginosa* was conducted on cephaloridine fucidin cetrimide (CFC, Oxoid) agar which was incubated at 25 °C for 48 h. After incubation, presumptive *P. aeruginosa* were identified by both pigmented and non-pigmented colonies that formed on the CFC plates (Jeppesen and Jeppesen, 2003). A representative colony from each plate was selected and purified on Plate count agar (PCA) (Merck).

3.2.2 Confirmation and identification of isolates

3.2.2.1 Biochemical tests

The following biochemical tests were performed on each of the purified isolates: Indole, Methyl-Red, Voges-Proskauer, and Citrate utilization collectively known as the IMViC test. In addition, catalase and oxidase tests were also performed on the isolates (Clesceri *et al*., 2002).

3.2.2.2 DNA isolation, PCR amplification of 16S rRNA and analysis

The genomic DNA of the selected isolates were extracted using the ZR Fungal/Bacterial DNA Kit^{TM} (Zymo Research) and the 16S rRNA genes of these isolates were amplified using the universal primer sets 63F (5’-CAGGCCTAACACATGCAAGTC-3’) and 1387R (5’-GGCGGWGTGTACAAGGC-3’) (Marchesi *et al.*, 1998). Each reaction mixture (25 µl) contained 2.5 µl of 10 × PCR buffer, 1 µl of 25 mM $MgCl_2$, 1 µl each of the forward and reverse primers (10 µM), 1 µl of 1 mM deoxynucleoside triphosphate (dNTPs), 0.5 U of SuperTherm *Taq* DNA polymerase (Southern Cross Biotech), 1 µl of template DNA (0.6 ng/µl, standardized using a Nanodrop) and 17 µl of sterile double-distilled water. PCR was performed using the PE Applied Biosystems GeneAmp PCR System 9700 (Perkin-Elmer). The PCR cycling conditions were as follows: initial denaturation at 95 °C for 5 min followed by 30 cycles of annealing and extension at 95 °C for 1 min, 55 °C for 1 min, 72 °C for 1.5 min and a final extension at 72 °C for 5 min. The amplicons were analyzed by electrophoresis on 1% (w/v) agarose (SeaKem) gels in 1 × TAE buffer at a voltage of 90 V for 90 min. After electrophoresis, the gel was stained in 0.5 μg/ml ethidium bromide (Sigma) for 20 min and visualized by UV transillumination (Chemi-Genius^2 BioImaging System, Syngene). The 16S rRNA PCR products were sent to Inqaba Biotech for sequencing and were subsequently subjected to a BLAST search.

3.2.3 Inhibition assays

The effect of *P. aeruginosa* on the pathogens was determined using inhibition testing following a method by Johannessen *et al*. (2005), but modified to include an agar diffusion assay. Presumptive *P. aeruginosa* and the presumptive pathogens were grown individually in tryptone soy broth (TSB) (Merck) for 24 h at 25 °C with shaking and at 37 °C without shaking, respectively. A lawn of the presumptive pathogens were made by placing 1 ml of a 10^6 cfu/ml TSB culture in a 9 cm petri dish onto which molten plate count agar (9 ml) was poured. After the agar had solidified, wells (5mm in diameter) were punched into the agar and 100 µl of a 10^7 cfu/ml presumptive *Pseudomonas* culture was used to fill the wells on the agar plates. Each isolate was spotted three times on each of two agar plates, giving six repetitions for each isolate. The plate count agar plates were incubated at 25 °C for 24 h and the diameter of the zones of inhibition were measured (mm). If inhibiting activity was observed at 25 °C, it was also tested at 10 °C, 15 °C (incubated for 1 week each), 20 °C (incubated for 3 days), and 30 °C (incubated for 24 h) (Johannessen *et al*., 2005; Schuenzel and Harrison, 2002). The minimum inhibitory concentration (MIC), the effect of pH as well as the effect of varying concentrations of iron (Fe) was also tested using the method described above but with the following modifications. To determine the MIC, varying concentrations of presumptive *P. aeruginosa* (10^4, 10^5, 10^6, 10^7, and 10^8) were used. The effect of pH on inhibition was tested using PCA at pH 5, 7 and 9. The effect of iron (Fe) on inhibition was determined using PCA supplemented with 1, 3 and 5% Fe.

3.2.4 Greenhouse study

Potting soil was purchased from Top Nursery at Westville KwaZulu-Natal (KZN) and placed in 9 pots, which was used to plant 4 week old butter lettuce seedlings obtained from a farm in KZN. These seedlings were watered twice a week with household water. At week 8, the plants were surface sterilized using 70% (v/v) ethanol for 1 min and then used for 3 different experimental set-ups in the greenhouse of the Department of Microbiology at the University of KwaZulu-Natal (UKZN) (Westville campus) as follows: experiment 1: autoclaved soil (at 121 °C for 3 × 60 min), experiment 2: non-autoclaved soil and experiment 3: autoclaved soil as described above. All experiments were set-up in triplicate. To all experiments, *L. monocytogenes* was added at 10^6 cfu/ml. *P. aeruginosa* was added only to the soil of experiments 1 and 2 at 10^7 cfu/ml. Experiment 3, did not receive any *Pseudomonas* culture as this served as the negative control. The autoclaved soil was analyzed prior to spiking and no contamination was evident. One hundred microlitres of culture was added per gram of soil (dry weight) and homogenization was achieved by mixing the soil with a large sterile spoon. The cultures were added on the first day of the experiment (week 0) and the plants and the subsequent soil were analyzed for the presence of *L. monocytogenes.* The soil was also analyzed for the presence of *P. aeruginosa*. These analyses were performed weekly for four weeks by blending 10 g of the lettuce leaves in 0.1% peptone water (100 ml), while 5 grams of the soil was added to 45 ml of 0.1% peptone water. Thereafter, appropriate dilution series were carried out using 0.1% peptone water (Islam *et al*., 2004a) and 0.1 ml of these dilutions was spread plated onto ALOA (Table 2.1) and CFC agar, for each sample. Dilutions of the non-autoclaved soil samples were also spread plated onto PCA, and incubated for 72 h at 25 °C, for the growth of total heterotrophic bacteria (THB). The plants were irrigated with sterile distilled water throughout the experiment, other than day 0. The temperature of the soil was taken at every sampling period, using a thermometer. A 0.5 g soil sample was taken at every sampling period and the genomic DNA was extracted using the Ultraclean^{TM} soil DNA Kit (Mo Bio Laboratories, Inc) for molecular detection of *P. aeruginosa* and *L. monocytogenes* using denaturing gradient gel electrophoresis as described below.

3.2.4.1 PCR amplification of V3 to V5 region

PCR for bacterial 16S rRNA genes were performed using the universal primer set for denaturing gradient gel electrophoresis (DGGE), F341-GC (CCTACGGGAGGCAGCAG) with a 5′ GC-clamp: CGCCCGCCGCGCCCCGCGCCC GTCCCGCCGCCCCCGCCCG and R907 (CCGTCAATTCMTTTGAGTTT) (Casamayor *et al.*, 2002). A GC-clamp was attached to the forward primer in order to prevent the complete separation of the strands during DGGE (Muyzer *et al.*, 1993). For PCR, 2 μl DNA extract (0.6 ng/μl) was added to the PCR reaction mixture (50 μl) containing 5 μl of 10 × PCR-buffer, 2 μl of 25 mM $MgCl_2$, 2.5 μl each of F341-GC and 907R (10 μM), 5 μl of 2 mM dNTPs, 30.5 μl sterile double-distilled water and 0.5 U of SuperTherm *Taq* DNA polymerase (Southern Cross Biotech). PCR was performed using the GeneAmp PCR System (Version 2.25, Perkin Elmer). A modified form (Muyzer *et al.*, 1993) of the touchdown thermal profile technique (Watanabe *et al.*, 1998) was used: an initial denaturation (94 °C, 5 min), followed by annealing via 10 cycles of 94 °C for 1 min; 65 °C for 1 min with a decrease in temperature of 1 °C per cycle; and 72 °C for 3 min. This was followed by 20 cycles of 94 °C for 1 min; 55 °C for 1 min; 72 °C for 3 min and a final 5 min extension step at 72 °C. The amplification of the correct product size of 585 bp was confirmed by electrophoresis in a 2% (w/v) agarose gel in a 1 × TAE running buffer with a voltage of 90 V for 120 min. After electrophoresis, the gel was stained in 0.5 μg/ml ethidium bromide and visualized by UV transillumination (Chemi-Genius^2 BioImaging System, Syngene).

3.2.4.2 Denaturing gradient gel electrophoresis (DGGE)

PCR amplicons were separated by DGGE using the D-Code Universal Mutation Detection System (BioRad) (Muyzer and Smalla, 1998). Firstly, 0% and 100% denaturing solutions were prepared, filtered through 0.45 μm pore size GN-6 Metricel membrane filters (Pall, 47 mm) and stored in brown bottles at 4°C. The DGGE gel was cast by preparing 20 ml each of low (30%) and high (60%) density solutions containing 20 μl TEMED (BioRad) and 200 μl of 10% ammonium persulphate, for gradient formation. The density solutions were applied through the gradient delivery system to cast the perpendicular 6% acrylamide DGGE gels (dimensions: 200 mm by 200 mm by 1 mm). Prior to sample loading, a pre-run was performed at a constant voltage of 150 V at 60 °C for 30 min in order to aid the sample migration out of the wells during the electrophoretic run. Following the pre-run, samples were loaded into the gel (3 µl gel loading buffer : 10 µl PCR product) and DGGE was conducted at a constant voltage of 60 V in 1 × TAE buffer at 60 °C for 16 h. After electrophoresis, the gel was stained in 0.5 µg/ml ethidium bromide (BioRad) for 20 min, destained in 1 × TAE buffer for a further 20 min and thereafter visualized by UV transillumination (Chemi-Genius^2 BioImaging System, Syngene). Dominant bands were excised from the gel, washed with ddH_2O and left overnight in ddH_2O. These samples were then PCR amplified using the F341 and R907 primer sets (without the GC-clamp) with the PCR conditions and visualization of bands as stated in section 3.2.4.1. These products were then sent to Inqaba Biotech for sequencing and were then subjected to a BLAST search to confirm the organism represented by the bands.

3.3 Results

3.3.1 Confirmation and identification of isolates

3.3.1.1 Biochemical characterization

Of the representative colonies chosen from the different plates, some of the isolates displayed biochemical test reactions that confirmed the presumptive identity of the food-borne pathogens tested for (Table 3.1).

Table 3.1: The Gram stain and biochemical test results of the presumptive pathogens isolated from irrigation water.

Isolate	**Gram reaction**	**Biochemical tests**						**Presumptive identity**
		Indole	**Methyl-Red**	**Voges-proskauer**	**Citrate**	**Catalase**	**Oxidase**	
1	(-) rods	-	-	-	+	+	+	*Pseudomonas aeruginosa*
2	(+) cocci	-	+	+	-	+	-	*Listeria monocytogenes*
3	(-) rods	+	+	-	-	+	-	*Escherichia coli*
4	(-) rods	-	-	+	+	+	-	*Salmonella* spp.
5	(-) rods	-	+	-	-	+	-	*Shigella* spp.

+ = positive result; - = negative result

3.3.1.2 BLAST search of the 16S rRNA gene sequences of the iisolates

The BLAST search confirmed the identity of the following bacterial isolates: *E. coli*, *L. monocytogenes*, *P. aeruginosa*, *Salmonella* spp. and *Shigella* spp. (Table 3.2). These isolates were then used in the subsequent inhibition assay.

Table 3.2: BLAST search results of the 16s rDNA gene sequences of the bacterial isolates.

Isolate	Organism	% Identity	E-value	Accession Number
1	*E. coli*	99	0	HM371196.1
2	*Listeria monocytogenes*	99	0	FJ774256.1
3	*Pseudomonas aeruginosa*	100	0	FN645737.1
4	*Salmonella* spp.	100	0	FN356961.1
5	*Shigella* spp.	100	0	HQ398233.1

3.3.2 Factors affecting the inhibitory activity of *P. aeruginosa* on *L. monocytogenes*

Of all the isolates tested only *L. monocytogenes* was inhibited by *P. aeruginosa*. The inhibitory effect of various concentrations of *P. aeruginosa* on *L. monocytogenes* was investigated (Table 3.3). It was observed that *P. aeruginosa* only inhibited *L. monocytogenes* at concentrations higher than that of the latter organism. Greater zones of inhibition were also observed as the concentration of *P. aeruginosa* increases, with the MIC found to be 10^7 cfu/ml.

Table 3.3: The effect of various concentrations of *P. aeruginosa* on the growth of *L. monocytogenes* [a].

Concentrations of *P. aeruginosa* (cfu/ml)	Diameter of zone of inhibition (mm)
10^4	N
10^5	N
10^6	N
10^7	11.5±0.55
10^8	14±0.00

N= no inhibition was observed; [a] values are averages ± standard deviations (*n* = 3)

Temperature was shown to play a role in the inhibition process (Figure 3.1), as the results indicated that an increase in temperature was directly proportional to the zone of inhibition obtained. Only the 30°C temperature showed an increase in activity of 2.4%. The effect of pH on the inhibiting activity of *P. aeruginosa* was also investigated and pH 7 showed greatest inhibiting activity. Interesting to note, however, was that inhibition had occurred throughout acidic (pH 5) and basic (pH 9) pH's, with an increase in the zones of inhibition in the latter (Figure 3.2). It was observed that the conditions which had allowed for the greatest increase in activity (2.4%) was at 30 °C with a pH of 7. When Fe was added, even at the lowest oncentration of 1%, no inhibition was observed (Table 3.4).

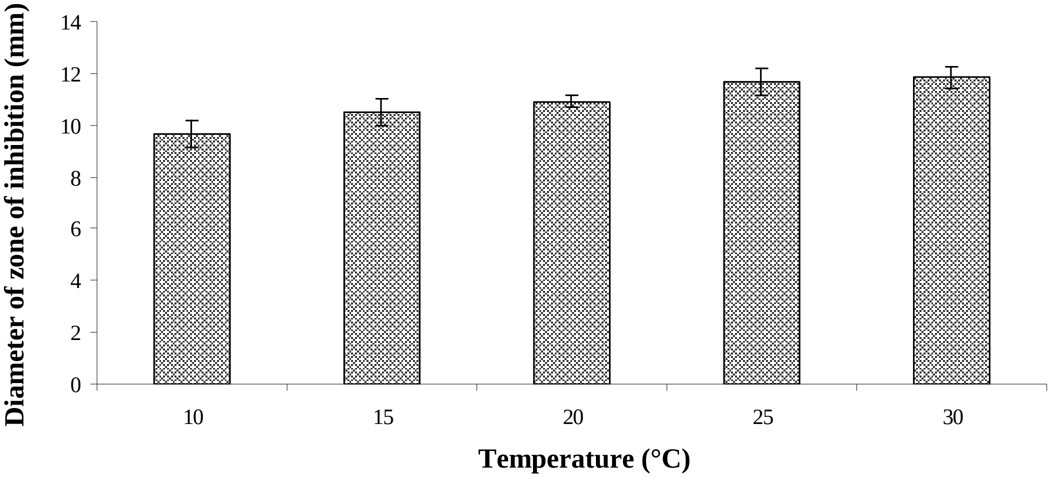


Figure 3.1: The effect of temperature on the inhibitory activity of *P. aeruginosa* against *L. monocytogenes* (Bars indicate an average of 3 values and error bars indicate the standard deviations).

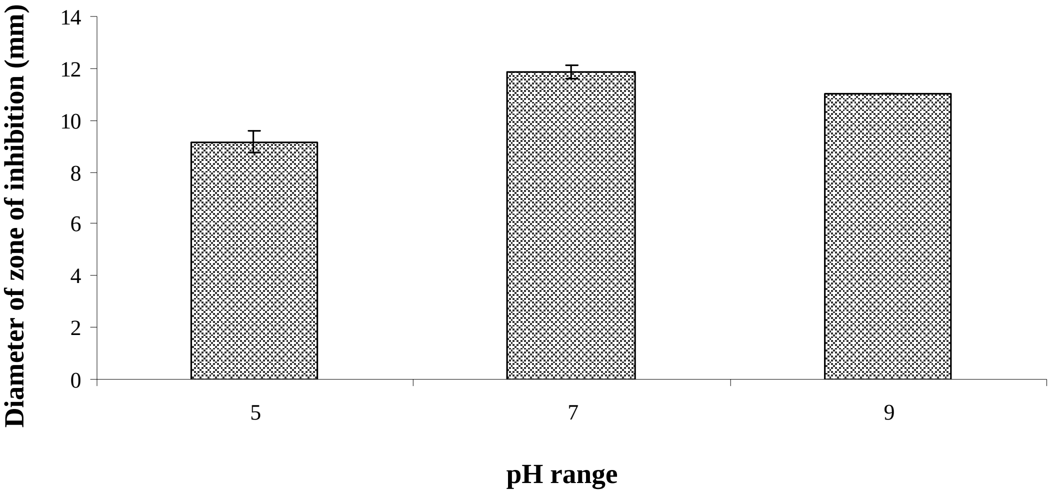


Figure 3.2: The effect of pH on the inhibitory activity of *P. aeruginosa* against *L. monocytogenes* (Bars indicate an average of 3 values and error bars indicate the standard deviations).

Table 3.4: The effect of various concentrations of Fe on the inhibiting activity of *P. aeruginosa* against *L. monocytogenes*.

Fe concentration (%)	**Diameter of zone of inhibition (mm)**
1	N
2	N
3	N

N= no inhibition was observed

3.3.3 The inhibitory effect of *P. aeruginosa* on the uptake of *L. monocytogenes* by the lettuce plant

A greenhouse study was performed in order to determine whether *P. aeruginosa*, when present at high concentrations could inhibit *L. monocytogenes*, such that it would not enter the fresh produce, itself. Temperature of the soil in the greenhouse environment was approximately ±33 °C throughout the study. In the lettuce planted in non-autoclaved soil (Figure 3.3), a decrease in the concentration of *L. monocytogenes* was observed in the soil from weeks 0-2, after which this pathogen was not detected in the soil. Also, this pathogen was not detected in the lettuce leaves of this setup. The THB and *P. aeruginosa* in soil had decreased by 13.77 and 7.8 log cfu/g from weeks 0-4, respectively.

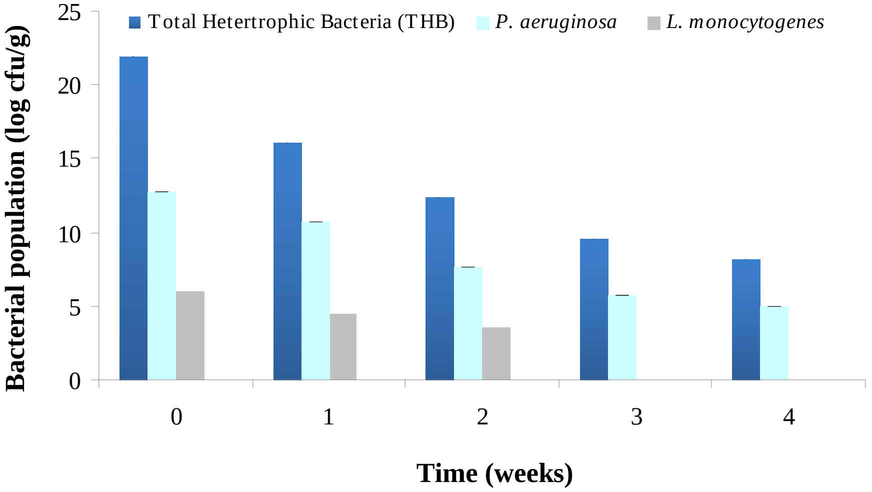


Figure 3.3: Time-course survival of *L. monocytogenes* and *P. aeruginosa* in non-autoclaved soil (Bars indicate an average of 3 values and error bars indicate the standard deviations).

In the lettuce planted in autoclaved soil, (Figure 3.4), *P. aeruginosa* and *L. monocytogenes* were detected in the soil from weeks 0-4. From weeks 2-4, *L.*

monocytogenes was detected in the lettuce plant itself, which shows that this pathogen had been taken up into the plant from the soil. At week 4, *L. monocytogenes* was detected in the lettuce plant at 4.47 cfu/g.

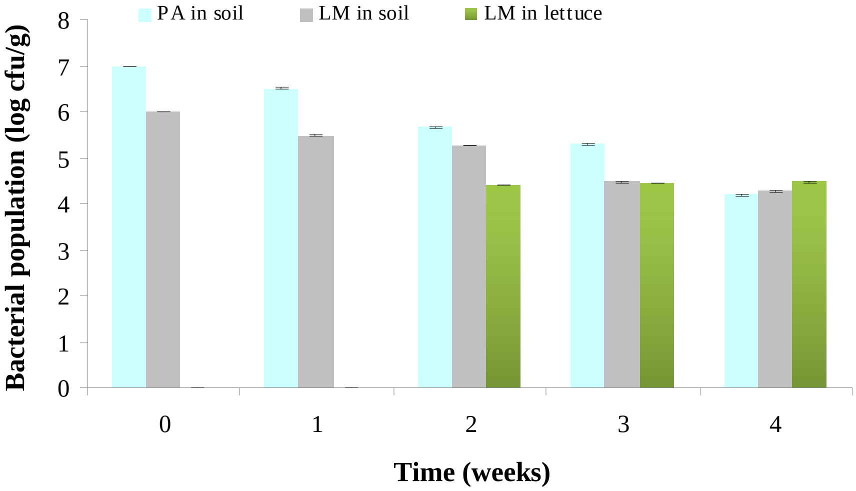


Figure 3.4: Time-course survival of *L. monocytogenes* (LM) and *P. aeruginosa* (PA) in autoclaved soil and the uptake of *L. monocytogenes* into the lettuce plant (Bars indicate an average of 3 values and error bars indicate the standard deviations).

The lettuce plant which served as the negative control (Figure 3.5) was only spiked with *L. monocytogenes* in autoclaved soil. *L. monocytogenes* was able to survive in the soil through the duration of the experiment, and this pathogen was detected in the lettuce leaves from week 1 with an increase of approximately 2.18 log cfu/g from weeks 1-4. *L. monocytogenes* was detected in the fresh produce (week 4) at 5.72 log cfu/g, which is 5.72 (Figure 3.3) and 1.25 (Figure 3.4) log cfu/g higher than that detected in the other greenhouse experiments.

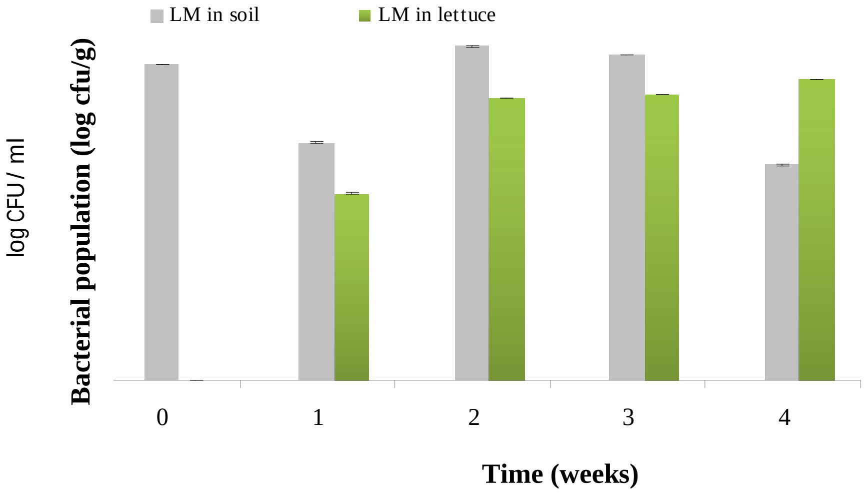


Figure 3.5: Time-course survival of *L. monocytogenes* (LM) in autoclaved soil and its subsequent uptake into the lettuce plant (Bars indicate an average of 3 values and error bars indicate the standard deviations).

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3.3.3.1 DGGE profiles depicting the presence of *P. aeruginosa* and *L. monocytogenes* in the soil (greenhouse study)

DGGE was used for the detection of the presence of *L. monocytogenes* and *P. aeruginosa* in the soil, to further confirm the results from the plate count. Lanes a1, b1 were the *L. monocytogenes* positive control and lane a12 was the *P. aeruginosa* positive control. The predominant bands (A1, A2) were excised, PCR amplified and sent for sequencing. These bands were confirmed to be *L. monocytogenes* (A1) and *P. aeruginosa* (A2), as in Table 3.2 at 97 and 100% identity, respectively. Figure 3.3 revealed that *L. monocytogenes* was not detected from week 2 onwards on laboratory media but its DNA was detected using DGGE (Figure 3.6 a2-a6). It should be noted that DGGE did confirm a decrease in the concentrations of *L. monocytogenes* and *P. aeruginosa* over time, as the DNA extract of all samples were standardised to 0.6 ng/µl (Figure 3.6 a2-a6). The DGGE profiles (Figure 3.6 a7-a11) of the autoclaved soil setup, showed a decrease in *L. monocytogenes* in the soil from week 2, which could indicate that this organism was taken up by the plant at week 2, as evidenced by culturable methods (Figure 3.4).

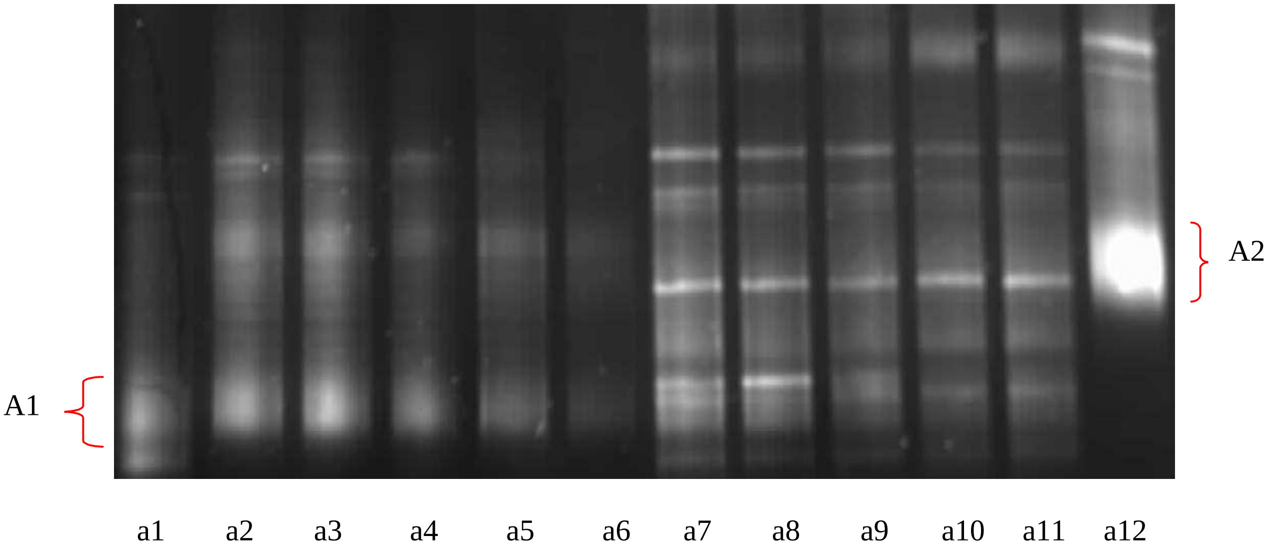


Figure 3.6: DGGE profiles of the 16S rRNA gene fragments of the soil collected over a 5 week period from the green house experiments: Lanes: a2, a3, a4, a5 and a6 represent weeks 0, 1, 2, 3 and 4 of the non-autoclaved soil study and a7, a8, a9, a10 and a11 represent weeks 0, 1, 2, 3 and 4 of the autoclaved soil study and Lanes a1 and a12 represent the *L. monocytogenes* and *P. aeruginosa* positive controls, respectively.

DGGE profiles of the soil from the negative control setup (Figure 3.7) revealed that the concentration of *L. monocytogenes* was low at week 1, therefore suggesting that this organism was taken up by the lettuce plant (as confirmed by the plate counts (Figure 3.5)). After week 1 the concentrations of *L. monocytogenes* had increased in the soil as depicted by the DGGE profiles of the soil.

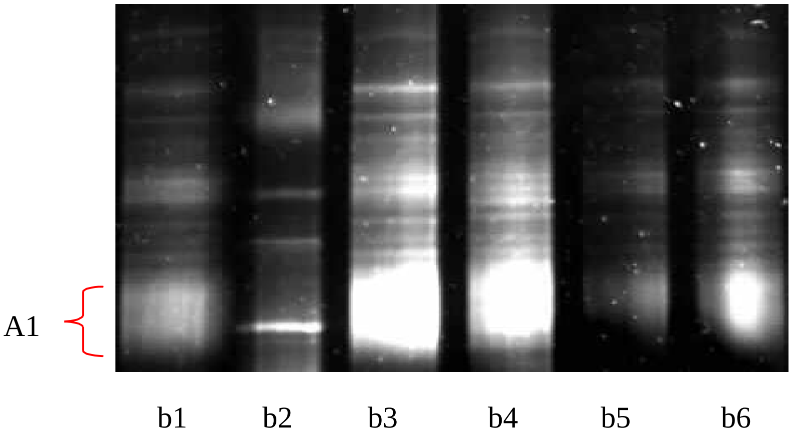


Figure 3.7: DGGE profiles of the 16S rRNA gene fragments of the soil collected over a 5 week period from the green house experiments: Lanes: b2, b3, b4, b5 and b6 represent weeks 0, 1, 2, 3 and 4 of the control setup and lane 1 represents the *L. monocytogenes* positive control.

Using DGGE profiles, it was confirmed that a higher concentration of *P. aeruginosa* in the non-autoclaved soil of the lettuce plant was able to inhibit *L. monocytogenes* over time (Figure 3.6 a2-a6). Also, the DGGE profiles of the lettuce plant which had autoclaved soil (Figure 3.6 a7-a11), had shown this inhibition but the concentration of *L. monocytogenes* in this set of DGGE profiles was much higher than that in Figure 3.6 (a2-a6). The DGGE profiles of the negative control setup (Figure 3.7) had shown the highest concentration of *L. monocytogenes* at week 4 (Figure 3.7 b6) as this setup was not spiked with the inhibiting bacteria, *P. aeruginosa*.

3.4 Discussion

Numerous food-borne disease outbreaks have been linked to the consumption of fresh minimally processed produce contaminated with pathogenic microbes (Chang and Fang, 2007). Bacterial pathogens including, *E. coli* 0157:H7 (Francis *et al*., 1999) *Salmonella* spp. (Herikstad *et al*., 2002) and *Shigella* spp. (Islam *et al*., 1993) have been previously linked to disease outbreaks due to the consumption of contaminated produce. In addition, *L. monocytogenes* is ubiquitous in the environment, especially in soil and plant matter, therefore its presence in fresh produce that are grown in close association with the soil is possible (Beuchat and Ryu 1997; Brackett, 1999a; Udompijitkul *et al*., 2007). It has previously been reported that certain *Pseudomonas* spp. may possess an inhibitory action towards different human pathogens, such as *E. coli* and *L. monocytogenes* (Johannessen *et al*., 2005).

Pseudomonads are present in such high numbers in environments such as soil because these microbes are able to utilize various natural and xenobiotic compounds as sources of carbon, nitrogen, sulphur and phosphorus (Molina *et al*., 2000). It has been recognized that certain *Pseudomonas* spp. may promote plant growth either "indirectly by suppressing pathogens, or directly through the secretion of phytohormones and vitamins or by increasing the mineral uptake by plants" (Sharma *et al*., 2003). Fluorescent pseudomonads have been reported to increase crop productivity when seed inoculated and also to decrease the numbers of harmful microbes under pot house as well as field conditions (Bakker *et al*., 1991; Loper and Buyer, 1991; Sindhu *et al*., 2002). These strains of *Pseudomonas* display antagonistic activity and are able to suppress the establishment and survival of pathogens due to the production of antibiotics (Sindhu *et al*., 2002).

Some Pseudomonads, such as *Pseudomonas syringae,* are able to survive in highly colonized environments, such as soil, because they may act as antagonists to other bacterial species (Janisiewski *et al*., 1999). Elevated concentrations of *P. syringae* have been recognized as an antagonist to *E. coli* O157 in plant wounds, probably competing for the same sources of carbon and energy. It was observed that when *P. syringae* was not present, *E. coli* O157 increased in concentration as compared to when these microbes were co-inoculated (Janisiewski *et al*., 1999). In the present study, *P. aeruginosa* suppressed the growth of *L. monocytogenes* at 10 °C, 15 °C, 20 °C, 25 °C and 30 °C, with the zones of inhibition being much higher at 25 °C and 30 °C (Figure 3.1). However, *P. aeruginosa* did not display any inhibitory action towards the other pathogens tested (Table 3.1). The reason for the inhibitory action of *P. aeruginosa* towards *L. monocytogenes* could be due to the possibility that these two organisms may have been competing for the same resource. It was also evident that the minimal concentration of *P. aeruginosa* required for inhibition was 10^7 cfu/ml (MIC) (Table 3.3) with a greater inhibitory effect observed with increasing concentrations of *P. aeruginosa*. Gram *et al.* (1999), in their investigation of the effect of *Pseudomonas fluorescens* on *Vibrio anguillarum* concluded that the antagonist must be present at significantly higher concentrations than the pathogen for inhibition to occur and that the degree of inhibition increases with an increase in the concentration of the antagonist which corroborates the findings of the current study.

The effect of pH and iron concentration on the inhibitory activity of *P. aeruginosa* against *L. monocytogenes* was also tested by varying the pH and iron concentration in the medium used. It was found that inhibition was highest at neutral pH compared to acidic and basic pHs, with the lowest inhibition observed at acidic pH of 5. In the presence of the various concentrations of iron tested, no inhibition of *L. monocytogenes* was observed. This was previously observed by Gram *et al.* (1999), who found that no zones of inhibition of *V. anguillarum* by *P. fluorescens* was observed in media that was supplemented with iron, as in the case of this study. The reason for the lack of inhibition by *P. aeruginosa* could be because inhibition only occurs under iron-limiting conditions through the production of siderophores, which deprive the pathogen of iron. This production of siderophores is a virulence factor in many microorganisms, such as members of the family *Enterobacteriaceae* and *Pseudomonas aeruginosa* and *Vibrio anguillarum* (Gram *et al.*, 1999; Crosa, 1980; Wooldridge and Williams, 1993).

Since the pseudomonads are present in high concentrations in the soil environment, they ought to out-compete the added pathogen because pseudomonads are highly adapted to the soil environment (Johannessen *et al.*, 2005). This was confirmed in this study (Figure 3.3, non-autoclaved soil) as the concentration of *P. aeruginosa* at week 0 was at a concentration of 12.81 log cfu/g of soil which was approximately twice the concentration of *L. monocytogenes* (6 log cfu/g of soil). From week 0 to 2, the numbers of the THB, *P. aeruginosa* and *L. monocytogenes* had decreased in the soil. The decrease in the concentration of *L. monocytogenes* could probably be due to the antagonistic action of the *P. aeruginosa*, since this organism was present at a much higher initial concentration. The decrease (log 5.16 cfu/g) in the population of *P. aeruginosa* in the soil could probably be due to the fact that some *Pseudomonas* spp., such as *P. fluorescens*, *P. aeruginosa* and *P. syringe* are known to enter into a viable but non-culturable (VBNC) state when conditions are not favourable (Cook and Bolster, 2007). Bunker *et al*. (2004) revealed that *P. fluorescens* cells could remain in the VBNC state in the soil for about a year. Numerous bacterial species, most importantly, human pathogens, have been known to act in response to different environmental stresses by way of entry into a novel physiological state. In this state, the cells are viable but due to this change they are no longer culturable using standard laboratory methods (Oliver, 2009). *L. monocytogenes* was not detected from week 2 and it was assumed that the growth of this organism was inhibited by *P. aeruginosa*. However, DGGE analysis of the samples proved otherwise because *L. monocytogenes*, not detected by standard laboratory plating, was detected using DGGE suggesting that this organism could have also adopted the VBNC state as a survival mechanism, as *L. monocytogenes* as been previously shown to enter this state (Besnard *et al*., 2000). Favourable conditions could have resulted from a lack of inhibitory action by *P. aeruginosa*. *P. aeruginosa* was still detected at week 4 at 5.01 log cfu/g of soil. During this experiment no uptake of *L. monocytogenes* into the plant itself was detected, which could probably be due to the presence of high concentrations of *P. aeruginosa* in the soil that had inhibited the *L. monocytogenes* present in the soil, thereby preventing its subsequent uptake into the plant. Johannessen *et al*. (2005) revealed that the *Pseudomonas* spp. that inhibited the growth of *E. coli* O157:H7 *in vitro* were actually present in the soil shaken off the roots of the lettuce plant. It must be noted, however, that even though *L. monocytogenes* was not taken up by the plant, the soil (as evidenced by DGGE), suggests that this organism could over time be taken up by the plant, when conditions become favourable.

In the greenhouse study (Figure 3.4, autoclaved soil), *P. aeruginosa* was added to the soil at a minimal inhibitory concentration (7 log cfu/ml) and *L. monocytogenes* was added at 6 log cfu/g of soil. From week 1 to week 4, there was a decrease in the concentrations of *P. aeruginosa* and *L. monocytogenes* that was observed at 2.81 and 1.73 log cfu/g of soil, respectively. Also *L. monocytogenes* was detected in the lettuce plant at 4.41 log cfu/g at week 2. It can therefore be assumed that because *P. aeruginosa* was present initially at the MIC, *L. monocytogenes* was able to survive and enter the edible portion of the lettuce plant. From week 2 to week

4, there was a gradual decrease in the concentration of *P. aeruginosa* in the soil; however, the concentrations of *L. monocytogenes* in the soil remained fairly constant, from week 3. In the lettuce plant, *L. monocytogenes* concentration was constant at approximately 4.4 log cfu/g from week 2. De Roever (1998) suggested that the development, survivability and the inactivation of microbes found on fresh produce is dependant on the interaction of the following factors: the character and capability of the organisms present, the physiological status of the plant tissue as well as its natural resistance towards microbial metabolic processes, the characteristics of the surrounding environment of the plant tissue (for example pH, water activity, etc.), and the effect of food practices and processes on the microbial numbers or plant metabolism.

The decrease in the concentration of *L. monocytogenes* in the soil, of the control setup (Figure 3.5), from 6 to 4.51 log cfu/g (week 0 to 1) could be due to the uptake of this pathogen into the fresh produce and the lack of inhibitory action from the pseudomonads. From week 2 to 4, there was an inversely proportional relationship which was evident between the concentration of *L. monocytogenes* in the soil and in the lettuce plant, with an increase in the latter. At week 4, the concentration of *L. monocytogenes* in the lettuce plant was 5.72 log cfu/g, this concentration was higher than that observed in the other 2 experiments. This is expected since *P. aeruginosa* was not present in this experiment and *L. monocytogenes* was therefore able to grow and flourish. The control wells in the DGGE gels had multiple bands, however, the predominate bands (A1 and A2) were sequenced and these were identified as *L. monocytogenes* and *P. aeruginosa*, respectively. The reason for the multiple bands of the pure cultures, could be that multiple copies of the same gene could be present in these organisms. Similarly, Nicolaisen and Ramsing (2002) found that a pure culture of *N. multiformis* produced four bands. It was stated that multiple gene copies and the formation of heteroduplexes during the last PCR cycles is a limitation when using the complexity of a DGGE band patterns to assess the biodiversity present in a sample (Nicolaisen and Ramsing, 2002).

When comparing the different setups in the greenhouse study (Figures 3.3, 3.4 and 3.5) the presence of *P. aeruginosa* in the soil does indeed have an impact on the uptake of a potential pathogen such as *L. monocytogenes* into the fresh produce, as confirmed by DGGE. This is important, as the presence of pathogens on fresh produce could have detrimental effects in terms of consumer safety. When *Pseudomonas* concentrations are abundant, as in soils, there is the potential for these bacteria to inhibit or suppress the growth of others. However, when *P. aeruginosa* was present at the MIC, *L. monocytogenes* was able to survive and when *P. aeruginosa* was not present in the soil *L. monocytogenes* was able to flourish in the soil and the fresh produce, itself. However, it must be noted that even though there was a decrease in the survival of *L. monocytogenes* in the soil, due to the inhibitory action of *P. aeruginosa* in the soil, the possibility of one surviving cell of *L. monocytogenes*, is still a concern, as this pathogen has been previously shown to cause major outbreaks **(**Schlech *et al.*, 1983) of disease due to the consumption of fresh produce (CDC, 1995; De Roever, 1998). This study has indicated that food-borne pathogens such as *L. monocytogenes* can be inhibited and prevented from being taken up into fresh produce, therefore this area of research shows promise for further future investigation.

CHAPTER 4: EFFECT OF DIFFERENT POST-HARVEST TREATMENT METHODS ON THE QUALITY OF THE FRESH PRODUCE

4.1 Introduction

Fresh fruit and vegetables are capable of harbouring food-borne pathogens and when consumed could result in numerous disease outbreaks world-wide (Core, 2005). These food-borne disease outbreaks have been linked to pathogenic bacteria such as *Salmonella* spp., *E. coli* and *L. monocytogenes* (Hassenberg and Idler, 2005; CDC, 1997; Burnett and Beuchat, 2000; Khadre *et al.*, 2001; Tauxe, 2002). As mentioned in the previous Chapter of this dissertation, pre-harvest contamination of fruit and vegetables is associated with the quality of the irrigation water (De Roever, 1998). However, contamination may also occur due to the unsanitary handling of fresh produce by farm workers which may result in its direct contamination (Janisiewicz *et al*., 1999). Since there is a great possibility of fresh produce contamination either by way of pre-harvest and/or post-harvest, it is important that proper washing procedures be in place to reduce or eliminate the threat of food-borne disease outbreaks.

Washing as well as sanitizing treatments have been shown to improve product safety by reducing microbial populations (Sapers, 2001). Chlorination has been used in routine washing steps, to treat post-harvest cooling water (Suslow, 1997). Chlorine has been used for decades in sanitation programs, primarily as either sodium or calcium hypochlorite (Suslow, 1997). The fresh-cut industry has used chlorine as a disinfectant in order to assure the safety of their produce. However, eliminating chlorine from the disinfection process is becoming a trend because of the concerns that are associated with its effectiveness and the environmental and health risks associated with the formation of carcinogenic halogenated disinfection by-products (Olmez and Kretzschmar, 2009). Also, the inhibitory effect of chlorine solutions on microbial cells is dependant on the amount of free chlorine in solution, therefore these chlorine washing solutions must be routinely checked (WHO, 1998). An alternative disinfectant solution, such as hydrogen peroxide has been recognized as being safe for food applications since it produces no residue because it is rapidly decomposed by an enzyme, catalase, to water and oxygen. Hydrogen peroxide has previously been reported to cause a significant reduction in the population of *Salmonella* spp. (Ukuku, 2004). Hydrogen peroxide at 0.5 % concentrations has been shown to inhibit the “development of postharvest decay” that is caused by numerous fungi

(Bachmann and Earles, 2000). Besides commercially used washing methods for fresh produce, consumers use house-hold washing methods such as washing with tap water or salt solution to remove contaminants from fresh produce. Hassenberg and Idler (2005) compared the effect of washing with tap water and/or ozone on the microbial load of fresh produce and observed a 1-log bacterial reduction, using tap water alone. In addition to using an appropriate method for disinfection the storage temperature of the produce may cause a reduction in the quality of the mature produce.

High storage temperatures for fresh produce have been shown to promote the growth of various microbes, which in turn promotes the spoilage of the produce (Carlin *et al*., 1995; Hassenberg and Idler, 2005). However, refrigeration is and has been the key method for controlling the rate of deterioration of fresh produce by means of reducing the respiration rate of the produce and by slowing the growth rate of spoilage microbes (Fonseca and Rushing, 2006; Cameron *et al*., 1994; King and Bolin, 1989). However, some pathogenic microbes such as, *L. monocytogenes* are able to survive at refrigeration temperatures (Carlin *et al*., 1995; Hassenberg and Idler, 2005). Chang and Fang (2007) showed the survivability of *E. coli* O157:H7 at 4 ºC for 10-12 days on shredded lettuce and therefore this pathogen poses an extreme threat to human health.

Post-harvest handling is the final stage in the processing of fresh produce. Therefore, a level of freshness must be maintained (Bachmann and Earles, 2000). It is not only important to find a post-harvest treatment method that can reduce the microbial contamination of the fresh produce but also this method must be able to maintain its effectiveness through storage by refrigeration. Also, it is important that after treatment the freshness and quality of the produce, be as it was when it was harvested. The objective of this study was, therefore, to determine the influence that different post-harvest treatment methods may have on the microbial quality as well as the nutritional quality of the fresh produce.

4.2 Materials and Methods

4.2.1 Sample collection

Four different fresh produce (Broccoli, Cabbage, Crisphead lettuce and Spinach) were collected from a farm in Camperdown, KZN, in sterile plastic bags and transported in a Styrofoam boxes containing icepacks. The samples were kept at 4 °C until required for processing (±1 h) (Mukherjee *et al.*, 2004).

4.2.2 Treatment of the produce

Approximately 250 g of the samples were placed in sterile beakers and washed with 5 L of solution using household methods as well as commercially used methods, except for the non-aqueous treatment method where 250 g of the samples were exposed to Ultra-Violet (UV) light.

4.2.2.1 Household methods

i) Samples were washed in tap water for 120 s (Vina *et al.*, 2007; Hassenberg and Idler, 2005).

ii) Samples were subjected to a household treatment by adding a handful of salt (5 g) to 1 L of water.

4.2.2.2 Commercially used methods

i) The chlorine solution for treatment was prepared using commercial sodium hypochlorite (6.15%), which was adjusted to pH 6 using HCl. The samples were dipped into this solution at a concentration of 40 µl/L for 3 min (Fonseca and Rushing, 2006).

ii) Blanching was performed by immersing the samples in boiling water (100 °C) for 1 min followed by quick submersion in cold water (4 °C) for 1 min (Vina *et al.*, 2007), even though this method is not applied industrially to cabbage, lettuce and spinach, it was included in this study in order to compare the effectiveness of all the treatment methods on different types of produce.

iii) The samples were washed with 5% H_2O_2 (Merck) (which was prepared from a 30% stock solution by dilution with sterilized distilled water). This solution was used to wash the produce by agitation for 5 min (Ukuku, 2004).

4.2.2.3 Non-aqueous method

UV light was employed for 3 min using a fluorescent lamp (30 W) with UV emission at 254 nm. The bulb was placed 15 cm above the samples (Fonseca and Rushing, 2006).

An unwashed control was also included in this experiment. After treatment, 15 g of the samples as well as the control were placed in sterile polyethylene bags. These bags were placed in a 4 °C cold room for 6 days. The following analyses were performed on the fresh produce samples at day 0 (before refrigeration) and day 6 (after refrigeration).

4.2.3 Analysis of the fresh produce samples

4.2.3.1 Microbiological analysis

The plant samples were prepared for analyses according to Islam *et al*. (2004a). Ten grams of each plant sample was homogenized with 90 ml of 0.1% peptone water (Islam *et al*., 2004a) using a blender. This served as the 10^{-1} dilution, from where subsequent serial dilutions were carried out using 0.1% peptone water. One hundred microlitres of the dilutions were then spread plated onto different selective media and incubated appropriately as indicated in Table 2.1). Appropriate dilutions were also spread plated onto nutrient agar, which was incubated at 25 °C for 72 h (Hassenberg and Idler, 2005), in order to enumerate total heterotrophic bacteria. The different bacterial populations as well as total population were enumerated by counting the number of colonies per plate and these values were expressed as colony forming units (cfu) per gram of the sample.

4.2.3.2 pH testing

This was performed by cutting the surface of the fresh produce and a pH indicator strip (Merck, Germany) was placed on the cut surface and left for approximately 2 min, and the colour code displayed was compared to the pH of standards (pH 0-14).

4.2.3.3 Vitamin C (Ascorbic acid) content

Ascorbic acid (AA) content of the fresh produce samples was determined using ascorbic acid test strips (Merck). This was used according to manufacturer's instructions for the determination of the AA content of vegetables.

4.2.3.4 Determination of total carbohydrate

The total carbohydrate content of the fresh produce was determined using the method of Sadasivam and Manickum (1996). One hundred milligrams of the sample (dry weight) were placed a boiling tube and 5 ml of 2.5N HCl (Merck) was added. This was hydrolysed by keeping it in a boiling water bath for 3 h, after which the tubes were allowed to cool to room temperature. This was neutralised with solid sodium carbonate (Merck) until the effervescence had ceased and then made up to 100 ml with distilled water and centrifuged at 5000 rpm for 10 min. One millilitre aliquots of the supernatant were taken for analysis. The working standards (0.05, 0.1, 0.2, 0.4, 0.6, 0.8 and 1 mg/ml) were prepared from glucose and made up to 1 ml by adding distilled water. Then 4 ml of the ice cold anthrone reagent was added to all the test tubes and heated for 8 min in a boiling water bath. The tubes were cooled quickly on ice and the green to dark green colour developed was read at 630 nm and used to generate a standard curve from which the amount of carbohydrate present in the sample was calculated using the following formula:

Amount of carbohydrate present in 100 mg of the sample
= (mg of glucose/volume of test sample) × 100

4.2.3.5 Estimation of reducing sugar by the dinitrosalicyclic acid method (DNS)

The sugars were extracted from 100 mg of the sample with hot 80% ethanol (Merck) twice, using 5 ml at each time. The supernatant was then collected and water allowed to evaporate by keeping it on a water bath at 80 °C for 10 min before dissolving the extracted sugar in 10 ml sterile distilled water. Three millilitres of the DNS reagent was then added to 3 ml of the extract, in a test tube and the tube contents heated in a boiling water bath for 5 min. While the contents of the test tube was still warm, 1 ml of 40% Rochelle salt solution was added. This was then cooled and the intensity of the dark red colour that had developed was read at 510 nm. A series of standards was also run using glucose to generate a standard curve from which the amount of reducing sugars present in the sample was calculated (Sadasivam and Manickum, 1996).

4.2.3.6 Estimation of total chlorophyll, chlorophyll *a* and chlorophyll *b* content

One gram of finely cut sample was placed in a mortar. The tissue was then ground to a pulp with a pestle, after the addition of 20 ml of 80% acetone (Merck). This was then centrifuged (5000 rpm for 5 min) and the supernatant was transferred into a 100 ml volumetric flask. Thereafter, the residue was ground again using 20 ml of 80% acetone, centrifuged as before, and the supernatant added to the flask. This procedure was repeated until the residue was colourless. The mortar and pestle was then washed with 80% acetone and this was added to the flask. The volume was then made up to 100 ml with 80% acetone. The absorbance of the solution was read at 645, 663 and 652 nm against the solvent (80% acetone) blank. The amount of chlorophyll present in the extract was estimated using the following equations (Sadasivam and Manickum, 1996):

mg total chlorophyll/g tissue = $20.2\,(A_{645}) + 8.02\,(A_{663}) \times (V/(1000 \times W)$

mg chlorophyll *a*/g tissue = $12.7\,(A_{663}) - 2.69\,(A_{645}) \times (V/(1000 \times W)$

mg chlorophyll b/g tissue = $22.9\,(A_{645}) - 4.68\,(A_{663})\,(V/(1000 \times W)$

Where A = absorbance at specific wavelengths

V = final volume of chlorophyll extract in 80% acetone (100 ml)
W = fresh weight of tissue extracted

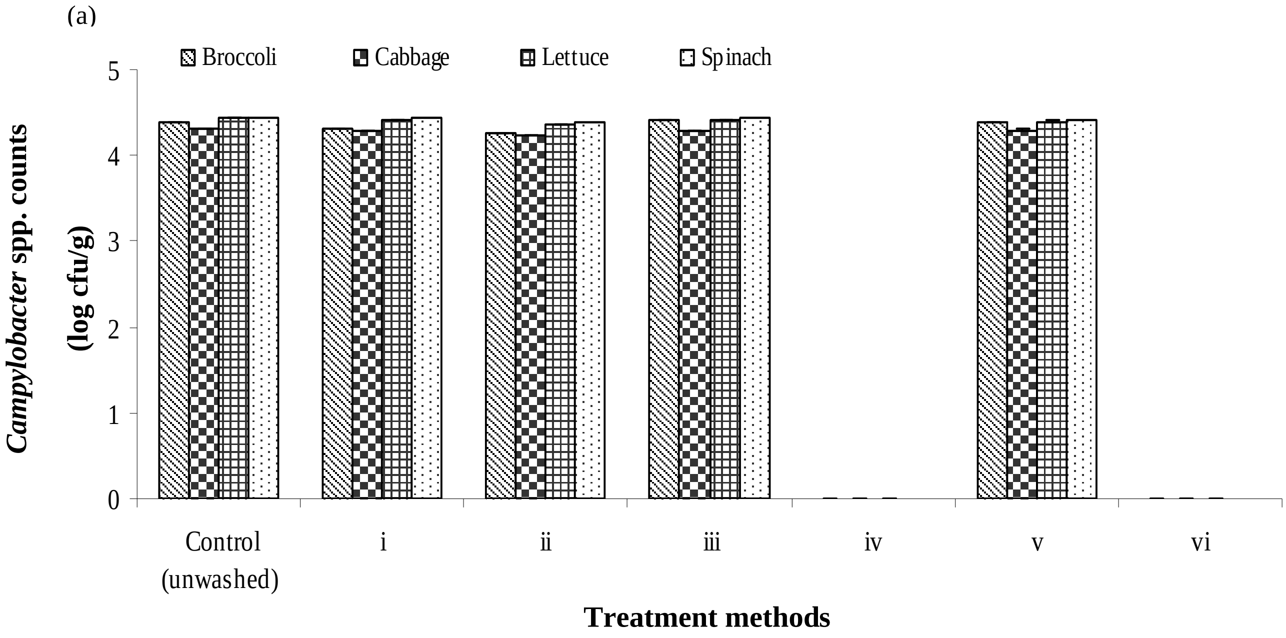
4.2.3.7 Sensory test (8 people)

The sensory quality of the treated fresh produce was analyzed by a randomly selected member sensory panel (8 persons). Personnel were required to evaluate changes in visual quality, texture, freshness, colour, off-odours, tissue damage and decay of the fresh produce. Overall visual quality was evaluated for gloss, freshness and colour uniformity and intensity. Samples were scored on an interval hedonic scale (Allende and Artes, 2003) where the extremes and centre of the interval were represented as follows: 0 = dislike extremely, no characteristic of the product, 5 = neither like nor dislike, limit of acceptance from the consumer's point of view, and 10 = like extremely, very characteristic of the product. The other characteristics such as colour and texture were evaluated on a 5-point scale where 5 = full characteristic of the product, 2.5 = moderate and 0 = no characteristic. Defects of the product such as off-odours, decay and tissue damage were evaluated as follows: 5 = severe, 2.5 = moderate and 0 = absence (Martinez-Sanchez *et al.*, 2006). The consumers used in the sensory evaluation were not well-trained members.

4.3 Results

4.3.1 Microbiological analysis

Presumptive *Campylobacter* spp. population was detected in all the fresh produce samples before refrigeration (Figure 4.1 a) except in the blanched and UV treated samples, in which this microorganism was not detected even after refrigeration. The population of presumptive *Campylobacter* spp. in the controls (unwashed produce) was similar to that of the tap water, NaCl, chlorine and hydrogen peroxide treated fresh produce, before refrigeration. Presumptive *Campylobacter* spp. counts had increased after refrigeration (Figure 4.1 b); in majority of the fresh produce samples tested. An increase in presumptive *Campylobacter* spp. counts of 14% was observed in the tap water treated spinach sample, after refrigeration, as compared to the control. The concentration of presumptive *Campylobacter* spp. in the NaCl treated samples was similar to that in the chlorine and hydrogen peroxide treated fresh produce samples, after refrigeration.



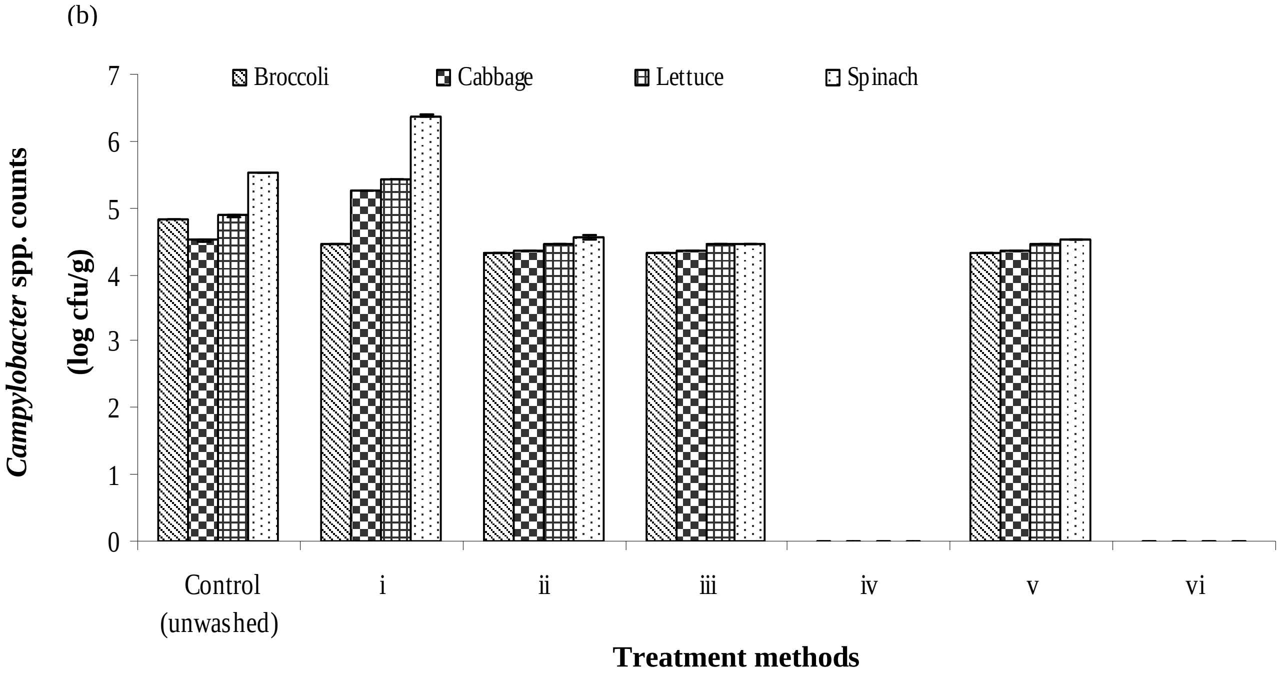


Figure 4.1: Counts of presumptive *Campylobacter* spp. in the fresh produce treated with different agents (i = tap water, ii = NaCl, iii = chlorine, iv = blanching, v = H_2O_2, vi = UV); a = before refrigeration at day 0 and b = after refrigeration at day 6 (Bars indicate an average of 3 values and error bars indicate the standard deviations).

Presumptive coliforms (Figures 4.2) were not detected in the treated cabbage samples before and after refrigeration, with the exception of the tap water treated cabbage sample which had a concentration of 3.53 log cfu/g, after refrigeration. Before refrigeration (Figure 4.2 a), presumptive coliforms were not detected in the blanched and hydrogen peroxide treated fresh produce samples, however, after refrigeration (Figure 4.2 b), these organisms were detected in the blanched broccoli sample at 3.5 log cfu/g and in all the hydrogen peroxide treated fresh produce samples, except the cabbage treated sample. An increase in presumptive coliform counts was observed after refrigeration in majority of the fresh produce samples tested. Presumptive coliform counts of all the tap water treated fresh produce were higher than that of the controls, after refrigeration. Decreases in presumptive coliform counts ranging from 26-21%, as compared to the control, were observed in the chlorine, blanched, hydrogen peroxide and UV treated broccoli samples, after refrigeration.

Presumptive *E. coli* (Figure 4.3) was not detected in the unwashed broccoli control as well as in the treated broccoli samples before and after refrigeration as well as in any of the blanched hydrogen peroxide and UV treated fresh produce samples at day 0, and the blanched and UV treated fresh produce samples at day 6. At day 0 (Figure 4.3 a), presumptive *E. coli* was not detected in any of the cabbage samples tested, however, at day 6 (Figure 4.3 b) it was detected in the control as well as the treated samples (i, ii, iii, v). The highest presumptive *E. coli* counts of 4.88 log cfu/g was observed in the NaCl treated cabbage, after refrigeration. Presumptive *E. coli* was observed in the lettuce (Figure 4.3 a) control as well as in the tap water and NaCl treated samples at day 0. However, in the chlorine and hydrogen peroxide treated lettuce, presumptive *E. coli* was only evident after refrigeration of the treated produce. A similar trend to that of lettuce was observed in the spinach samples tested, however, the presence of presumptive *E. coli* in the chlorine treated sample was observed at day 0 and day 6 with an increase of 21% observed in the latter.

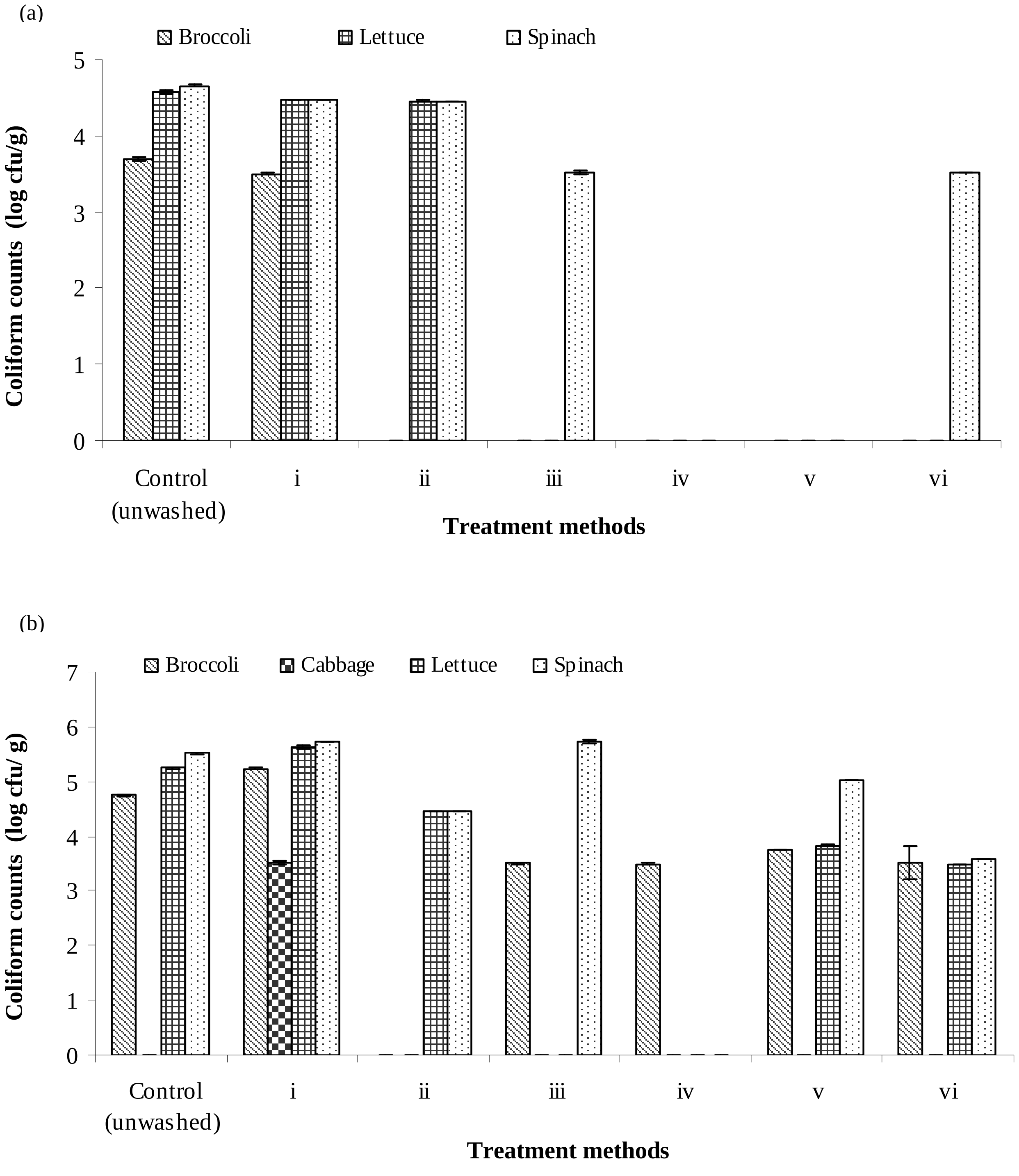


Figure 4.2: Counts of presumptive coliforms in the fresh produce treated with different agents (i = tap water, ii = NaCl, iii = chlorine, iv = blanching, v = H_2O_2, vi = UV); a = before refrigeration at day 0 and b = after refrigeration at day 6 (Bars indicate an average of 3 values and error bars indicate the standard deviations).

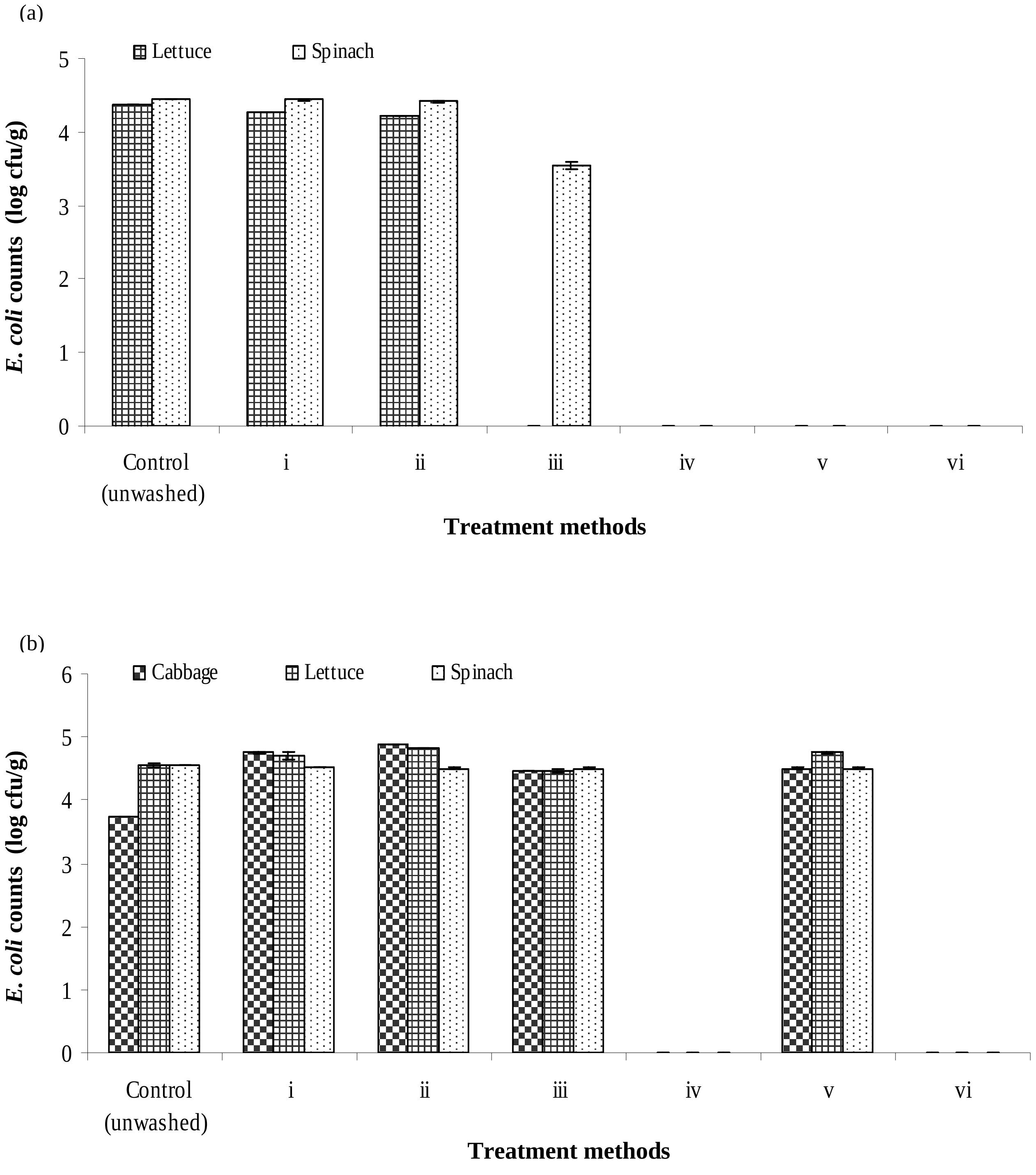


Figure 4.3: Counts of presumptive *E. coli* in the fresh produce treated with different agents (i = tap water, ii = NaCl, iii = chlorine, iv = blanching, v = H_2O_2, vi = UV); a = before refrigeration at day 0 and b = after refrigeration at day 6 (Bars indicate an average of 3 values and error bars indicate the standard deviations).

Presumptive *L. monocytogenes* (Figures 4.4-4.5) was not detected in any of the fresh produce sampled at day 0, except the spinach samples. After refrigeration of the chlorine treated and blanched cabbage samples, presumptive *L. monocytogenes* was detected at 3.51 and 3.53 log cfu/g, respectively. A similar trend was observed in the lettuce treated samples as *L. monocytogenes* was only detected in the chlorine treated (iii) and blanched (iv) lettuce samples at 3.51 and 3.79 log cfu/g. When comparing the concentrations of presumptive *L. monocytogenes* in the control spinach with the other treated spinach samples at day 0 (Figure 4.4), the UV treated spinach sample was 0.11 log cfu/g lower than the control. Presumptive *L. monocytogenes* was detected in the blanched spinach sample, after refrigeration (Figure 4.5), at 3.74 log cfu/g which was 0.12 log cfu/g higher than the control.

Presumptive *Salmonella* spp. was not detected in any of the broccoli samples tested before and after refrigeration. Of all the cabbage samples tested presumptive *Salmonella* spp. were only detected in the blanched cabbage sample after refrigeration at 3.5 log cfu/g (Figure 4.6 b). The presence of presumptive *Salmonella* spp. in the tap water and NaCl treated samples were similar to the control before and after washing. Presumptive *Salmonella* spp. was not detected in the hydrogen peroxide and UV treated lettuce samples and this organism was detected in the blanched lettuce sample after refrigeration. Presumptive *Salmonella* spp. counts was detected in the chlorine treated sample before refrigeration at 3.5 log cfu/g (Figure 4.6 a). *Salmonella* spp. was not detected in the UV treated spinach sample throughout the experiment, even after refrigeration. A similar pattern with the presence of presumptive *Salmonella* spp. in the control, tap water and NaCl treated lettuce samples was observed with the spinach sample, except that the presence of this organism was approximately, 1 log cfu/g higher in the treated spinach samples. Presumptive *Salmonella* spp. was detected in the chlorine treated spinach sample only after refrigeration at 4.34 log cfu/g. This organism was present in the blanched spinach on day 6 at 3.97 log cfu/g. Also the largest reduction (compared to the control) of presumptive *Salmonella* spp. in spinach was seen with the hydrogen peroxide wash method at 1.3 log cfu/g after 6 days of refrigeration, however, UV treatment was shown to be the most effective method as presumptive *Salmonella* spp. was not detected in these treated samples before and after refrigeration.

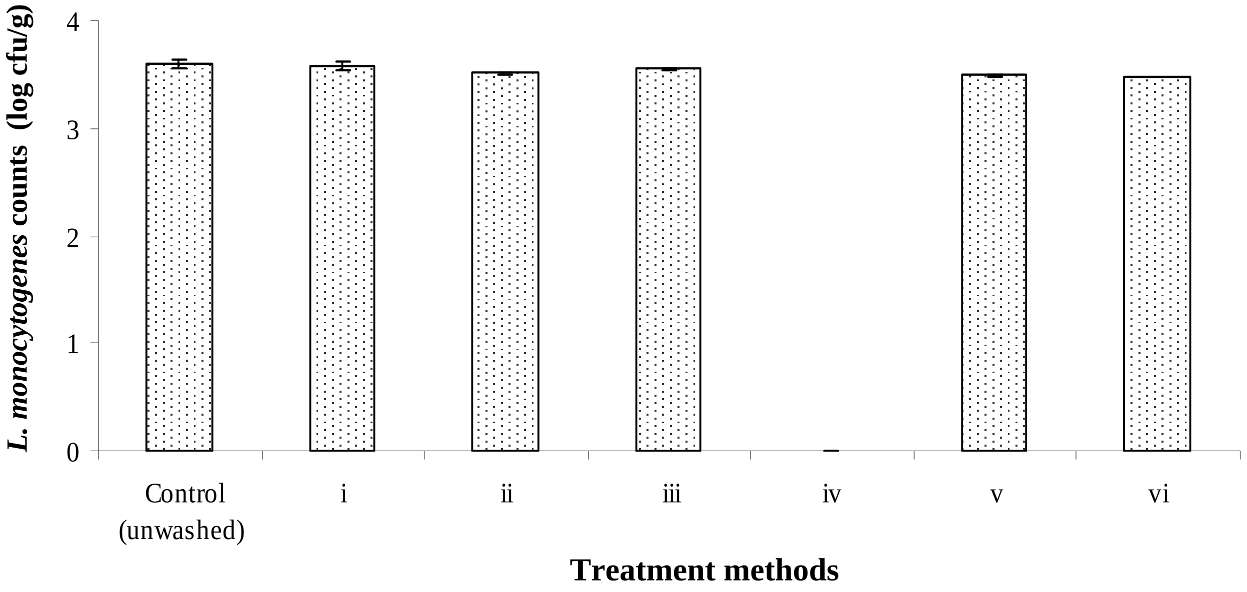


Figure 4.4: Counts of presumptive *L. monocytogenes* in spinach samples treated with different agents (i = tap water, ii = NaCl, iii = chlorine, iv = blanching, v = H_2O_2, vi = UV) before refrigeration at day 0 (Bars indicate an average of 3 values and error bars indicate the standard deviations).

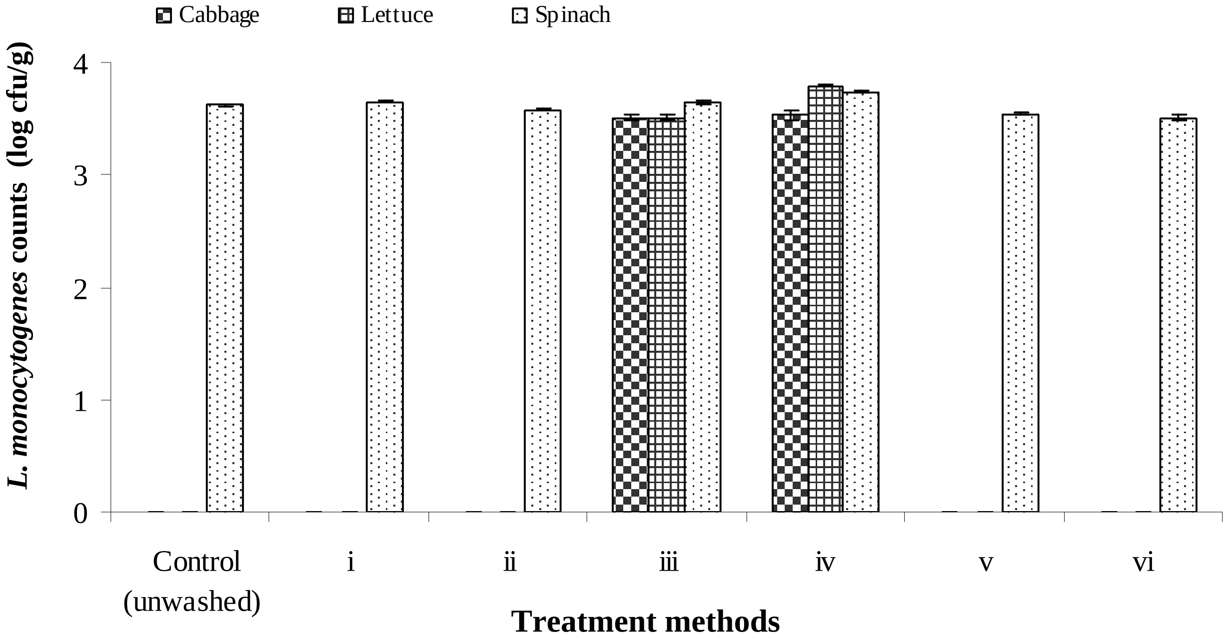


Figure 4.5: Counts of presumptive *L. monocytogenes* in the fresh produce treated with different agents (i = tap water, ii = NaCl, iii = chlorine, iv = blanching, v = H_2O_2, vi = UV) after refrigeration at day 6 (Bars indicate an average of 3 values and error bars indicate the standard deviations).

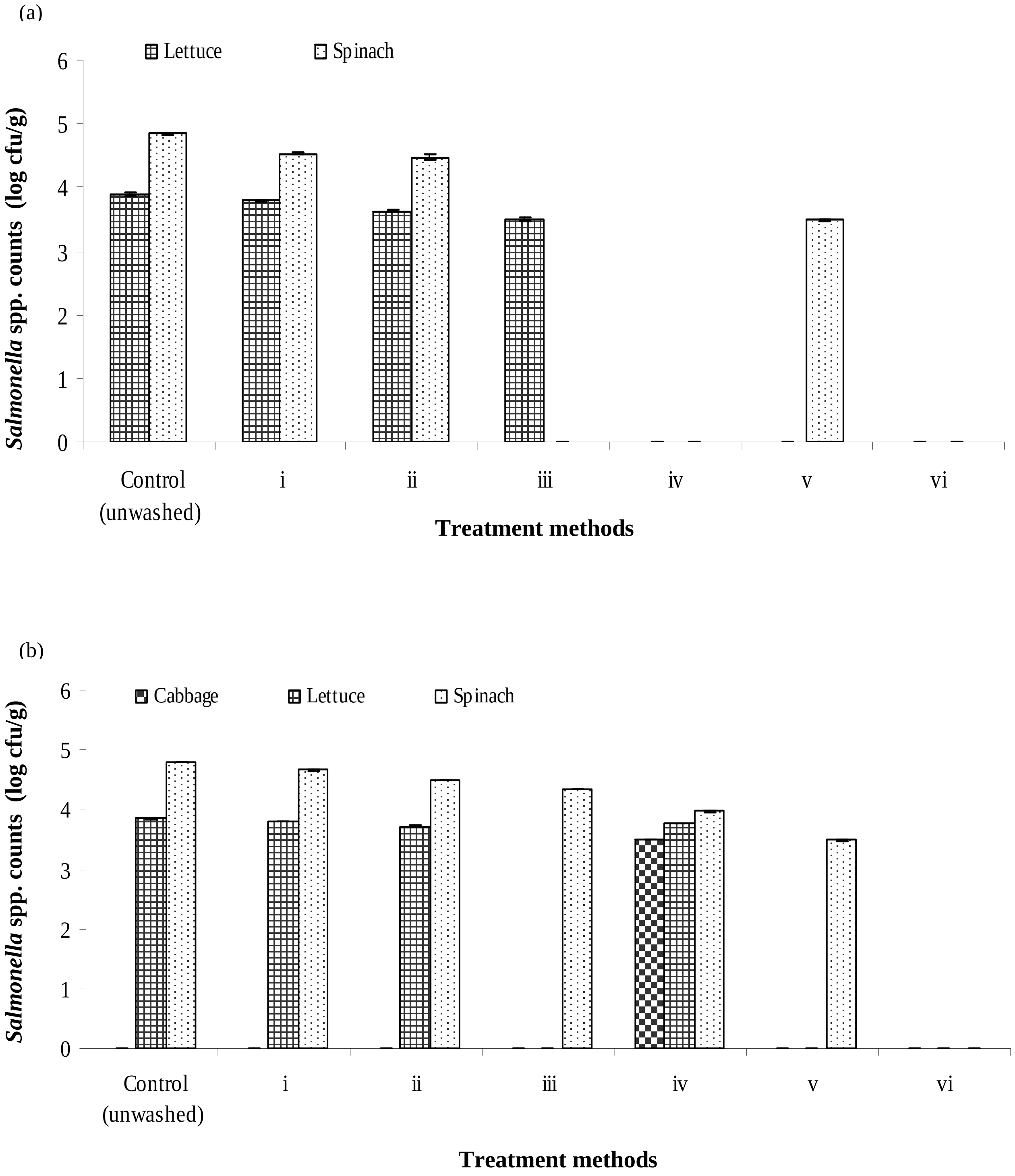
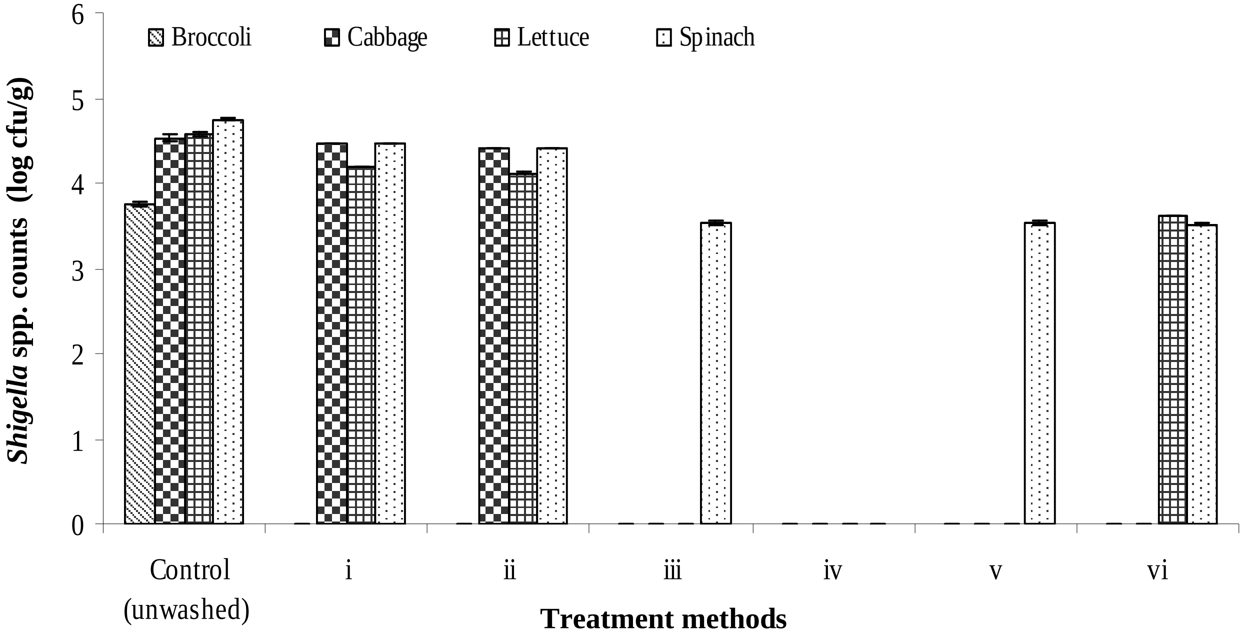


Figure 4.6: Counts of presumptive *Salmonella* spp. in the fresh produce treated with different agents (i = tap water, ii = NaCl, iii = chlorine, iv = blanching, v = H_2O_2, vi = UV); a = before refrigeration at day 0 and b = after refrigeration at day 6 (Bars indicate an average of 3 values and error bars indicate the standard deviations).

Presumptive *Shigella* spp. were only detected in the broccoli control on day 0 at 3.75 log cfu/g (Figure 4.7 a). At day 6, an increase of presumptive *Shigella* spp. in the control of approximately 0.65 log cfu/g was observed. Presumptive *Shigella* spp. was not detected in the blanched and hydrogen peroxide treated broccoli samples at day 6, however, the concentration of presumptive *Shigella* spp. in the control at day 6 was similar to that found in the tap water and NaCl treated broccoli samples. The highest reduction in counts of presumptive *Shigella* spp. in broccoli was observed using the chlorine, blanched and hydrogen peroxide treatment methods, before refrigeration. Presumptive *Shigella* spp. was not detected in the blanched and UV treated cabbage samples. Presumptive *Shigella* spp. was detected in the control, tap water and NaCl treated cabbage samples on day 0 at 4.5 log cfu/g. These counts were maintained after refrigeration of the tap water and NaCl treated samples, however, the control showed an increased of 0.93 log cfu/g after refrigeration (Figure 4.7 b). Presumptive *Shigella* spp. was detected in the chlorine and hydrogen peroxide treated cabbage samples at 4.13 and 3.53 log cfu/g, respectively, on day 6. In the lettuce samples (Figure 4.7 a), presumptive *Shigella* spp. was present on day 0 in the control, tap water, NaCl and in the UV treated samples, however, on day 6 this organism was not detected in the UV treated sample. Presumptive *Shigella* spp. was found to be abundant in the control spinach as well as in all the treated spinach samples on day 0; however, this organism was not detected in the blanched spinach sample.

(a)



(b)

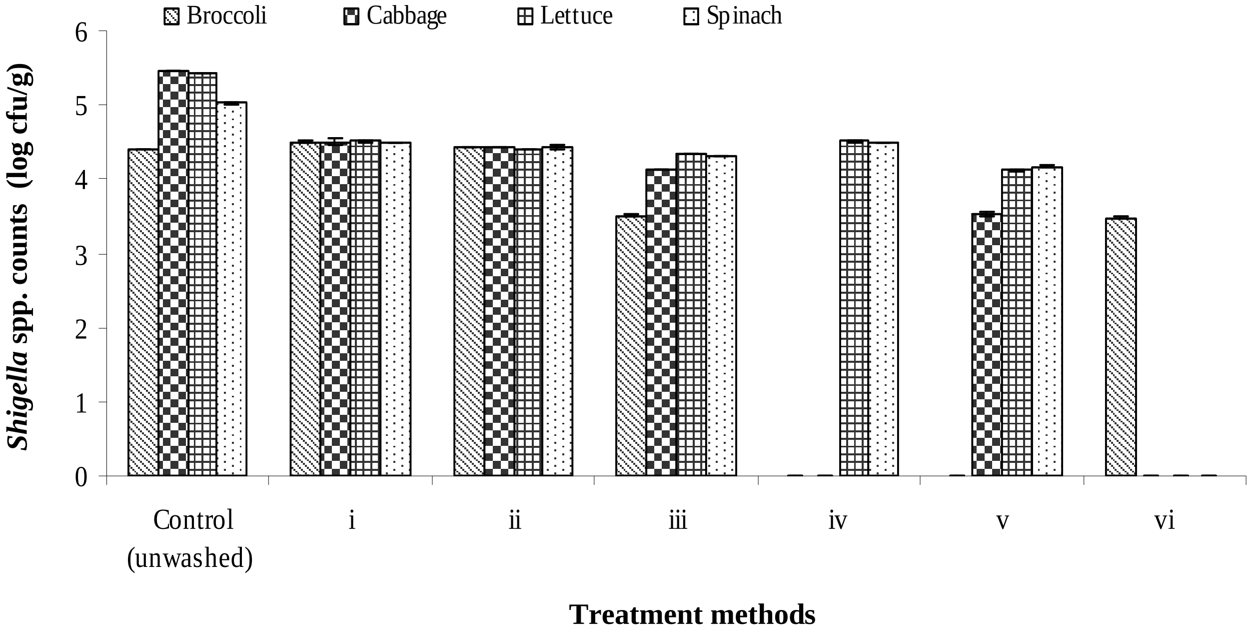


Figure 4.7: Counts of presumptive *Shigella* spp. in the fresh produce treated with different agents (i = tap water, ii = NaCl, iii = chlorine, iv = blanching, v = H_2O_2, vi = UV); a = before refrigeration at day 0 and b = after refrigeration at day 6 (Bars indicate an average of 3 values and error bars indicate the standard deviations).

An overall increase in THB counts of the broccoli samples were observed after refrigeration for 6 days (Figure 4.8 b). However, no bacteria were detected in blanched broccoli samples at day 0, however, at day 6 the THB were detected at 3.59 log cfu/g. At day 0 (Figure 4.8 a), the highest and lowest THB counts of the broccoli samples were present in the control and the chlorine treated samples at 5.53 and 3.54 log cfu/g, respectively. After refrigeration, the highest and lowest THB counts were present in the tap water treated and the blanched broccoli sample at 7.49 and 3.59 log cfu/g, respectively (Figure 4.8 b). A very similar trend was observed with the cabbage and lettuce samples tested. THB counts were evident at day 0 in all the spinach samples tested except the blanched spinach sample, however, THB counts were detected at 6.41 log cfu/g after refrigeration (Figure 4.8 b). Before refrigeration, the chlorine and hydrogen peroxide treated samples had the highest reduction of THB counts as compared to the control, however, no THB were detected in the blanched samples. The THB in all the treated samples as well as the control had increased after refrigeration, except for the NaCl treated spinach samples were the THB counts had decreased by 1.64 log cfu/g, after refrigeration.

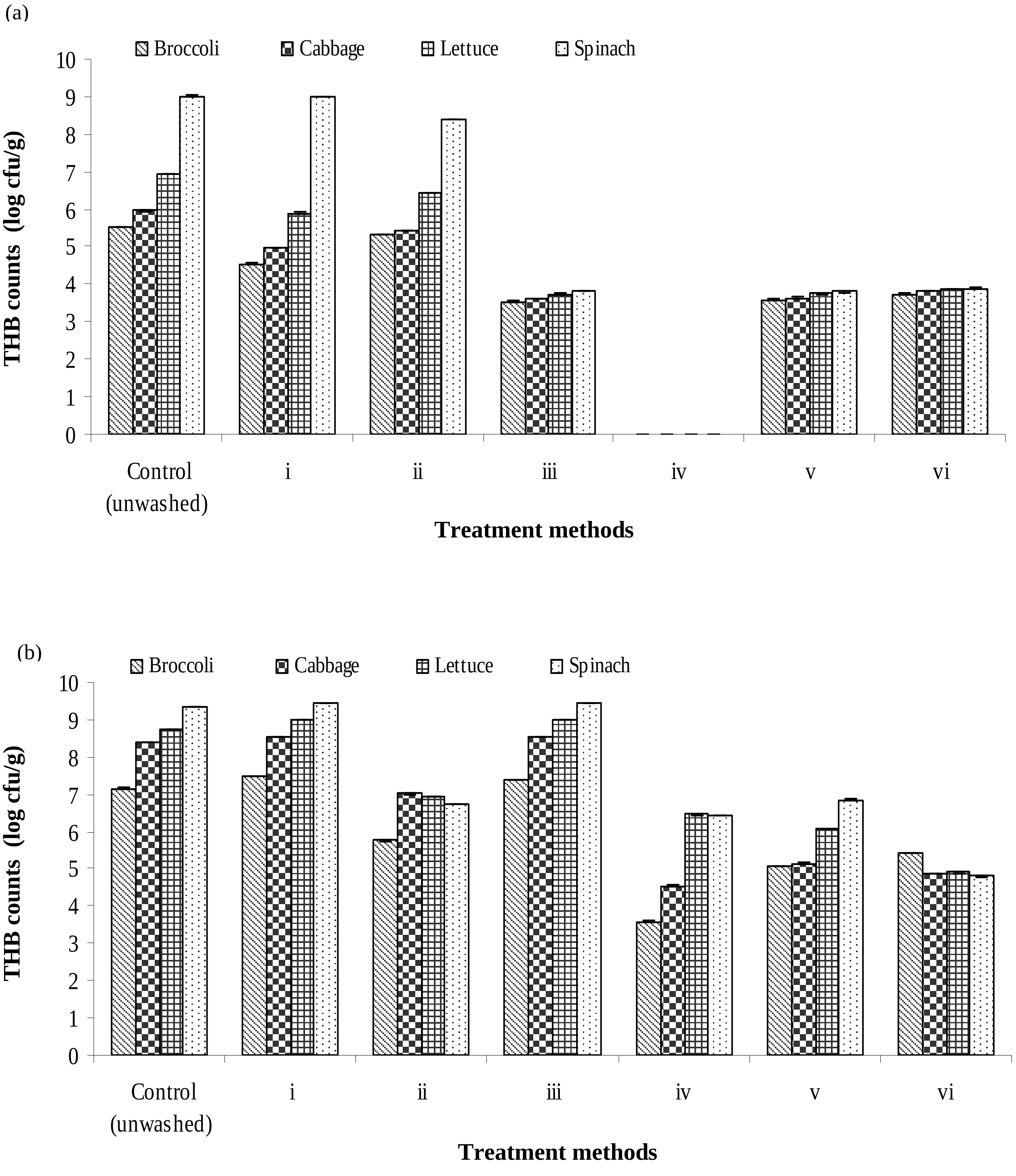


Figure 4.8: Counts of total heterotrophic bacteria (THB) in the fresh produce treated with different agents (i = tap water, ii = NaCl, iii = chlorine, iv = blanching, v = H_2O_2, vi = UV); a = before refrigeration at day 0 and b = after refrigeration at day 6 (Bars indicate an average of 3 values and error bars indicate the standard deviations).

4.3.2 pH and vitamin C (AA) content of the produce

At day 0, the pH of all the fresh produce samples tested (broccoli, cabbage, lettuce and spinach) ranged between 6-7, and remained constant after refrigeration at day 6. At day 0, the AA content was approximately 2000 mg/L for all the broccoli samples, 700 mg/L for the cabbage samples, 200 mg/L for the lettuce samples and 300 mg/L for the spinach samples. At day 6, after refrigeration, the AA content of all the broccoli samples tested had decreased by 1000 mg/L and the AA content of the cabbage samples remained constant, with an increase in the AA content of the UV treated cabbage sample of approximately 300 mg/L. A similar trend was observed with the AA content of the UV treated lettuce and spinach samples, with an increase of 300 and 200 mg/L being observed, respectively. The AA content of the rest of the lettuce and spinach samples had remained constant.

4.3.3 Total carbohydrate content analysis

At day 0, a decrease of 9.66 mg/g of the total carbohydrate (mg/g) (Table 4.1) content was observed in the blanched broccoli sample; however at day 6, this sample had the highest total carbohydrate content of 75.71 mg/g of all the other blanched samples. A decrease in the total carbohydrate content was observed in all the broccoli samples tested at day 6. The total carbohydrate content of the cabbage samples tested showed no major differences at day 0, however, at day 6, an overall decrease was observed. A trend was observed between the UV treated, blanched cabbage and lettuce samples, an increase in the total carbohydrate content, compared to the control, of 13.82 and 4.83 mg/g was observed with the UV treated cabbage and lettuce samples (day 6), respectively. A decrease in the total carbohydrate content, compared to the control, of 9.22 and 16.67 mg/g was observed with the blanched cabbage and lettuce samples (day 6), respectively. For the spinach samples no differences in the total carbohydrate content was observed but at day 6 the total carbohydrate content of the blanched spinach sample had decreased by 13.61 mg/g compared to the control. An overall decrease in the total carbohydrate content of all the samples tested was observed after refrigeration.

Table 4.1: The effect of different washing methods on the total carbohydrate content of the treated fresh produce, before (day 0) and after refrigeration (day 6) [a].

Sample Treatment	Broccoli		Cabbage		Lettuce		Spinach	
	Total carbohydrate (mg/g)							
	0	6 days	0	6 days	0	6 days	0	6 days
Control (Unwashed)	87.56±0.66	66.27±2.01	86.02±2.74	60.13±2.01	73.73±9.49	46.08±0.66	74.83±7.80	51.57±2.12
i	85.80±8.34	65.17±1.32	84.70±1.66	64.52±0.66	78.12±2.01	48.06±1.74	78.56±2.01	52.67±1.74
ii	86.68±1.37	74.39±4.11	83.17±1.01	66.93±1.66	79.88±1.66	44.77±3.48	79.00±1.74	51.57±1.01
iii	88.22±5.39	73.07±1.97	80.54±10.50	66.27±0.76	78.56±4.38	48.50±4.76	79.22±2.12	53.76±3.74
iv	77.90±10.50	75.71±1.14	84.49±3.25	50.91±1.52	80.32±2.63	29.41±2.74	78.34±2.37	37.96±1.01
v	86.60±0.76	71.98±1.01	84.05±2.49	65.17±0.66	78.34±5.70	48.28±4.85	78.78±2.12	52.67±1.14
vi	84.70±2.49	77.90±1.66	84.70±4.02	73.95±1.37	77.46±1.01	50.91±1.01	76.80±1.01	52.67±1.32

i = tap water, ii = NaCl, iii = chlorine, iv = blanching, v = H_2O_2, vi = UV; [a] values are averages ± standard deviations (*n* = 3)

4.3.4 Reducing sugar concentrations of the fresh produce

The effect of the different washing methods on the concentration of reducing sugars (Table 4.2) present in fresh produce was determined at day 0 and after refrigeration at day 6. At day 0, no major differences were observed in terms of the reducing sugar quality of the four fresh produce after treatment, as compared to the control. At day 6, there was an overall decrease in the reducing sugar quality of all the fresh produce tested, including the control. The blanched broccoli sample had the highest concentration of reducing sugars at day 6, 18.21 µg/g higher than the control. Of the cabbage samples tested, at day 6, the blanched cabbage sample had a lower reducing sugar concentration as compared to the control of about 33.03 µg/g. A similar trend was observed with that of the spinach sample tested, with a 60.18 µg/g decrease in the reducing sugar content of the blanched spinach sample as compared to the control.

Table 4.2: The effect of different washing methods on the concentration of reducing sugars in the treated fresh produce, before (day 0) and after refrigeration (day 6) [a].

Sample	Broccoli		Cabbage		Lettuce		Spinach	
Treatment	Reducing sugar (μg/g)							
	0	6 days	0	6 days	0	6 days	0	6 days
Control (Unwashed)	473.30±1.41	426.39±1.60	453.24±1.60	438.12±0.53	446.76±0.93	436.57±1.60	455.09±1.85	441.20±2.45
i	468.98±6.68	420.83±5.16	456.33±4.38	437.19±2.33	443.36±2.98	437.19±3.74	453.86±2.33	437.50±1.85
ii	472.07±3.85	426.08±1.07	456.94±2.78	441.51±4.18	449.54±6.07	433.80±3.34	454.78±1.93	439.66±3.85
iii	469.91±4.24	426.39±0.93	455.09±1.85	441.20±0.93	444.91±3.34	435.34±1.93	457.87±1.85	443.67±0.53
iv	470.52±8.60	444.60±1.93	454.78±3.85	405.09±0.93	442.44±3.74	434.41±1.07	454.17±1.60	381.02±4.63
v	474.54±7.23	437.19±1.41	456.02±2.45	436.57±2.45	446.76±2.45	390.59±7.87	455.71±2.14	436.57±1.85
vi	472.38±1.93	443.67±2.83	458.80±5.78	443.06±1.85	445.52±1.41	435.34±1.07	457.56±4.38	441.82±1.07

i = tap water, ii = NaCl, iii = chlorine, iv = blanching, v = H_2O_2, vi = UV; [a] values are averages ± standard deviations (*n* = 3)

4.3.5 Total chlorophyll, chlorophyll *a* and chlorophyll *b* contents of the produce

The total chlorophyll content (Table 4.3) of the broccoli, cabbage, lettuce and spinach controls, before refrigeration, was 185, 117, 344 and 1063 μg/g, respectively. The total chlorophyll content of the broccoli samples tested were similar to the control, except the blanched broccoli had the highest chlorophyll contents, with an increase of 8.47 μg/g (total chlorophyll) observed in the blanched broccoli sample, before refrigeration, as compared to the control. No differences were observed in the total chlorophyll contents of the cabbage, lettuce and spinach samples at day 0. All UV treated samples showed an increase in the total chlorophyll content, as compared to the control at day 0. An overall decrease in the total chlorophyll and chlorophyll *b* contents were evident in all the samples tested after refrigeration. The chlorophyll *b* content of the blanched broccoli and UV treated broccoli samples were the highest before (50.38 and 46.39 μg/g) and after refrigeration (22.89 and14.07 μg/g), respectively. A decrease in the chlorophyll *a* contents of the broccoli and cabbage samples was observed after refrigeration, however, an increase was noted with the lettuce and spinach samples after refrigeration. The chlorophyll *b* content of the blanched cabbage sample increased after refrigeration by 10.55 μg/g. A similar increase in the chlorophyll *b* content after refrigeration was observed with that of the blanched lettuce and spinach samples. The UV treated spinach samples also showed an increase in the chlorophyll *b* content of 2.75 μg/g, after refrigeration.

Table 4.3: The effect of different treatment methods on the quality of chlorophylls found in four different fresh produce after treatment (day 0) and after refrigeration (day 6) [a].

Samples	Treatment	Total chlorophyll (µg/g)		Chlorophyll a (µg/g)		Chlorophyll b (µg/g)	
		0	6 days	0	6 days	0	6 days
Broccoli	Control (Unwashed)	184.58±3.05	139.42±7.58	142.01±1.01	126.00±5.02	42.61±2.25	13.45±2.94
	i	182.79±2.46	132.30±3.15	149.59±2.75	122.82±2.42	33.24±3.06	9.51±1.99
	ii	180.79±6.85	137.82±0.86	145.27±5.32	123.46±6.09	35.56±4.85	14.39±6.80
	iii	186.33±9.15	137.28±1.63	141.32±6.90	122.61±0.58	45.05±6.29	14.70±1.05
	iv	193.05±3.42	138.35±3.21	142.72±2.46	124.3±0.77	50.38±1.73	14.07±3.46
	v	189.41±6.43	139.43±0.62	145.04±5.62	123.70±2.90	44.41±10.57	15.76±3.24
	vi	191.96±5.94	150.99±4.07	145.62±2.77	128.13±3.71	46.39±3.90	22.89±1.73
Cabbage	Control (Unwashed)	116.75±4.47	104.67±2.62	109.74±1.26	102.17±2.54	7.03±4.68	2.52±0.95
	i	119.05±7.34	105.61±1.52	107.60±1.53	102.51±1.98	11.47±6.92	3.12±1.62
	ii	122.52±9.34	99.17±1.42	113.10±4.28	96.94±1.28	9.44±5.17	2.25±1.40
	iii	119.72±4.87	110.18±1.07	107.51±2.55	102.81±1.55	12.23±2.50	7.39±1.66
	iv	118.09±5.10	125.77±3.65	111.86±2.79	109.00±1.23	6.25±2.33	16.80±4.82
	v	119.57±4.23	104.54±1.75	110.74±1.91	100.81±2.69	8.85±2.93	3.75±1.85
	vi	123.74±2.22	110.86±2.43	111.56±2.13	102.72±0.52	12.20±1.15	8.15±2.06
Lettuce	Control (Unwashed)	344.09±4.05	302.79±1.20	290.58±1.71	286.44±2.75	53.59±4.94	16.40±2.12
	i	342.53±3.44	292.83±1.00	276.56±1.81	286.85±4.33	66.05±5.22	6.03±3.73
	ii	337.15±7.33	292.57±0.84	277.27±3.31	284.13±4.86	59.95±5.08	8.49±5.10
	iii	331.63±4.29	294.72±0.40	276.63±5.44	285.22±2.09	55.07±1.64	9.55±2.01
	iv	330.02±1.16	310.04±6.25	276.39±1.16	290.98±4.93	53.70±3.53	19.11±1.97
	v	339.96±4.26	296.74±4.58	280.57±2.48	284.95±2.31	59.46±6.31	11.84±2.73
	vi	346.83±1.45	295.53±2.45	278.74±3.57	284.19±2.22	68.17±3.33	11.39±2.43
Spinach	Control (Unwashed)	1062.67±9.69	1016.49±3.45	967.27±6.05	971.58±3.54	95.61±11.33	45.09±4.53
	i	1054.76±7.44	101.72±9.59	960.52±6.63	968.22±2.63	94.44±7.90	42.68±12.21
	ii	1064.17±9.22	1014.07±2.25	963.86±6.44	970.07±5.77	100.52±8.03	44.19±6.50
	iii	1054.72±1.62	1011.66±5.30	969.71±6.82	968.56±2.26	85.22±6.20	43.28±7.51
	iv	1053.65±8.03	1032.38±10.25	968.01±3.96	969.01±1.24	85.84±4.62	63.56±9.94
	v	1051.23±5.82	1013.12±7.32	966.50±2.68	972.03±5.54	84.94±4.84	41.27±7.14
	vi	1069.80±4.57	1037.20±0.62	968.16±6.49	974.33±1.41	101.85±6.02	63.06±1.74

i = tap water, ii = NaCl, iii = chlorine, iv = blanching, v = H_2O_2, vi = UV; [a] values are averages ± standard deviations (*n* = 3)

4.3.6 Sensory evaluation of the treated produce

In terms of the sensory evaluation (Appendix B, Tables B92-B103), it was noted that no major differences in the sensory quality was observed with that of the control, and all treatment methods except the blanching method before and after refrigeration. It was evident from the evaluation that the blanched samples had a greater intensity of colour compared to the other treated samples and the control. The quality of the broccoli blanched sample was rated higher than all the other treated broccoli samples, in terms of freshness, colour intensity, texture and gloss of the product (before and after refrigeration). The blanched cabbage samples, had received the lowest scores at day 6, in terms of its texture and freshness, and off-odours were reported as moderate. Also, the hydrogen peroxide treated cabbage samples should a moderate colour intensity as compared to the control which showed a full characteristic of the product after refrigeration. The blanched lettuce and spinach samples, off-odours and tissue damage were severe, and these produce were rated as having no characteristic of the product.

3.4 Discussion

Fresh produce has been known to contribute significantly to the healthy lifestyle of individuals, but several disease outbreaks have proven that even though the produce are very important to consumers they could harbour pathogens that could be extremely hazardous to the consumers (Core, 2005). Therefore, washing and sanitizing treatments are important for the removal or inactivation of pathogens (Sapers, 2001). The response of microbes to washing and sanitizing treatments depends partly on the conditions of contamination that affect the attachment and survival of these microbes on fresh produce surfaces (Sapers, 2001).

It was evident from this study that effective removal of presumptive *Campylobacter* spp. from fresh produce was achieved by the blanching and UV treatment methods (Figure 4.1). The most effective method in the removal of coliforms from fresh produce was the blanching method (Figure 4.2). However, this method did not completely remove coliforms from the broccoli treated sample as these bacteria were still detected after refrigeration. These microorganisms could have entered into a viable but non-culturable (VBNC) state during the blanching process (before refrigeration) and became culturable under favourable conditions, after refrigeration. The most effective method in reducing the concentrations of presumptive *E. coli* (Figure 4.3) from fresh produce was blanching and UV treatment method. Presumptive *L. monocytogenes* (Figure 4.4 b) still remained abundant after day 6 in all the spinach samples tested, with no differences recorded when compared to the control after refrigeration. Refrigeration has been used as a means of controlling the spoilage of produce throughout the years by retarding the growth of microorganisms (Cameron *et al*., 1994). However, this study has revealed otherwise as in majority of the samples tested the bacterial counts had increased after refrigeration. The most effective method in reducing the microbial counts of presumptive *Salmonella* spp. (Figure 4.6) was UV light. Previous studies have shown that cells of this pathogen are able to attach to fresh produce and form biofilms, thereby protecting them from harsh washing conditions during post-harvest treatment (Core, 2005). Presumptive *Shigella* spp. (Figure 4.7) was found abundantly in all fresh produce tested. UV treatment was found to be the most effective in reducing presumptive *Shigella* spp. from fresh produce. The treatment which caused the greatest reduction in THB counts (Figure 4.8) in the fresh produce sampled was UV treatment. Overall, it can be seen that UV treatment was the most effective post-harvest treatment method in reducing the microbial content of fresh produce. Fonseca and Rushing (2006), demonstrated the effect of chlorine, ozone and UV-light on fresh-cut watermelon cubes, with the latter treatment method being non-aqueous. It was evident that the aqueous treatment methods used were not effective in reducing microbial load as compared to the non-aqueous method, UV-light. The quality of the cubes was also lower when the aqueous methods were used (Fonseca and Rushing, 2006). UV treatment has also been shown to reduce the microbial populations in fresh processed vegetables (Allende and Artes, 2003; Lemoine *et al*., 2007).

In the present study, hydrogen peroxide was shown to be more effective in reducing the microbial population than the chlorine treatment method. Chlorinated water is currently being used, industrially, in packing-houses for the purpose of sanitizing fresh produce in order to reduce the post harvest decay of fresh produce (Nunes and Emond, 1998). Hypochlorite treatments at pH 6 were reported to significantly reduce the microbial load of fresh-cut muskmelons stored at 2 ºC. However, chlorine solutions have been found to be less effective or completely ineffective against *L. monocytogenes* (Ayhan *et al*., 1998; Beuchat and Brackett, 1990).

In this study, although the overall effectiveness of chlorine treatment was lacking, it was nevertheless effective against presumptive coliforms in the cabbage and lettuce samples. Even though chlorine treatment is widely known, the potential hazards that have been associated with chlorine reaction by-products as well as issues regarding the disposal of waste waters, have led to evaluation of other possible methods for fresh produce disinfection (Suslow, 1997). Adams *et al*. (1989) reported a 92.4% reduction of the lettuce leaf microflora after washing with tap water. Han *et al*. (2000) suggested that water washing alone is not sufficient to remove bacteria that are tightly attached to injured surfaces of vegetables, in this case green peppers. In the current study it was also apparent that the washing of fresh produce with tap water alone was less effective than chlorine for the removal of microbes. Hassenberg and Idler (2005) found that, after six days of storage at 4 °C, a four log increase in the microbial counts occurred after the initial treatment of washing with tap water. Similarly, an increase in the microbial load in majority of the tap water washed samples had been observed, after refrigeration. The NaCl treatment method showed a similar pattern to the tap water treated samples throughout this study. These increases could probably be due to the fact that these products may have become more perishable as they have been subjected to additional physical stress (as in the blanched cabbage, lettuce and spinach samples) and furthermore, processing of fresh produce has been shown to promote faster microbial degradation of the produce in comparison with the raw product (Lemoine *et al.*, 2007).

The pH of all the fresh produce ranged from 6-7 before and after refrigeration. This is in accordance with Bolin and Huxsoll (1991) who reported a constant pH during the storage of cut lettuce at 2 °C for 21 days. Hassenberg and Idler (2005) reported that the vitamin C and sugar contents of lettuce were not affected by treatment methods. This corroborates the findings in the present study, as AA content (Vitamin C) of the samples tested did not change compared to the control at day 0. However, at day 6, the AA content of all the broccoli samples had decreased by 50%. Lemoine *et al*. (2007) showed a similar reduction of about 50% in the AA content of broccoli samples in both the control and treated (UV) florets during storage. However, an increase was observed in the AA content of the UV treated cabbage, lettuce and spinach samples after refrigeration, compared to the control produce. Furthermore, blanching did not change the AA content of the fresh produce compared to the control. Vina *et al*. (2007) also reported that blanching for 1 min did not cause significant changes in the AA content of brussel sprouts. These authors also reported a reduction in the sugar contents during storage (Hassenberg and Idler, 2005). The reduced sugar content was also observed by Lopez-Galvez *et al.* (1997) who reported between 12% and 20% reduction during 15 days of storage at 5 °C. Similarly, a decrease in the concentrations of reducing sugar and total carbohydrate content was observed after refrigeration for all the produce tested, in this study. Lemoine *et al*. (2007) showed that the total sugar content of broccoli florets also diminished during storage and no significant differences were observed between the control and the UV-C treated samples. Reducing sugar content also decreased in both the control and the UV-C treated florets during storage, with a lower decrease in the treated florets. The lower levels of reducing sugars in the control samples could be attributed to their higher respiratory activity, particularly at the end of storage (Lemoine *et al*., 2007). In this study, the UV treated broccoli samples showed a lower loss of reducing sugars after refrigeration. Furthermore, a major decrease in the reducing sugar and total carbohydrate content was observed in the blanched samples (cabbage, lettuce, spinach) after refrigeration. This could probably be due to the loss of texture that these produce suffered during the blanching process.

Cruciferous vegetables including broccoli and cabbage are rich in chlorophyll (Olivera *et al*., 2008). This was shown in the present study as all the fresh produce tested showed high total chlorophyll levels. Lemoine *et al*. (2007) reported that the chlorophyll content of the untreated broccoli controls were approximately 180 µg/g. Similarly, in the current study, the total chlorophyll content of broccoli, cabbage, lettuce and spinach were approximately 185, 117, 344 and 1063 µg/g, respectively. The UV treated fresh produce samples showed higher chlorophyll levels compared to the controls after refrigeration; this is in accordance with the findings of Lemoine *et al*. (2007) who proved that UV treatment delayed yellowing and chlorophyll degradation of fresh produce (broccoli) during storage. Blanching (1 min) of fresh produce in the present study did not affect the initial chlorophyll content of the samples, corroborating the findings of Vina *et al*. (2007) that blanching for 1 min and 3 min did not significantly affect the initial chlorophyll content of the samples (brussel sprouts). However, the broccoli blanched samples showed, an increase in the initial chlorophyll contents. Again, Lisiewska *et al*. (2004) observed that the blanching of dill (*Anethum graveolens* L.) leaves for 30 s in water at 94-96 °C, did not significantly affect the content of chlorophyll *a* and *b*. Several researchers have shown that the thermal inactivation of enzymes achieved by blanching limits the degradation of chlorophylls (Vina *et al*., 2007).

Although hydrogen peroxide treatment of the samples did in some cases decrease microbial counts, the limitation with using hydrogen peroxide as a treatment method is its effect on product colour as it causes bleaching or browning of the produce (Parish *et al*., 2003). This was evident in this study as the cabbage treated sample showed signs of browning after refrigeration. Blanching is a process that has been designed to inactivate the enzymes involved in off-flavours and odours and to achieve the stabilization of the texture and nutritional quality of the fresh produce as well as the destruction of microorganisms, but since blanching is a heat treatment, changes that are associated with thermal processing can be expected. "These include loss of turgor in cells, due to thermal destruction of membrane integrity and partial degradation of cell wall polymer" (Olivera *et al*., 2008; Bahceci, 2005). Olivera *et al*. (2008) showed that blanching caused a significant reduction in the firmness of the fresh product, which was greater than 80%. This was observed for the cabbage, lettuce and spinach samples tested, which showed a great loss of firmness and texture, after blanching. The effect of heat treatments on the colour of fresh produce has been studied intensely. It has been noted that during the initial part of the heating process, an increase in green colour occurred (Olivera *et al*., 2008; Tijskens *et al*., 2001).

This was observed during the blanching process as green intense colours were evident, for all the blanched produce.

Sapers (2001) found that the effectiveness of the washing method depends on the time interval between the contamination event and the time of washing. In addition, when bacteria attach to the surfaces of fresh produce they are likely to move into pores, indentations or irregularities found on the surfaces of produce, thus limiting there exposure to the washing treatment (Seo and Frank, 1999). Kroupitski *et al*. (2009) revealed that the incubation of *gfp*-tagged *S. enterica* with lettuce leaves in the light, resulted in the aggregation of bacteria near open stomata and invasion into the inner leaf tissue. Han *et al*. (2000) noted that *E. coli* O157:H7 appeared to not have penetrated the intact surface of the vegetable (green pepper) but however, this bacterium seemed to attach to coarse, porous and injured surfaces. Sapers (2001) noted that it must be realised that the conventional washing technology was developed in order to remove soil from the fresh produce and not microorganisms, and even with the use of newer sanitizing agents “improvements in efficacy have been incremental”.

Of all the post-harvest treatment methods tested, tap water and NaCl treated produce (household methods) had the highest microbial loads. Therefore, it is important that fresh produce be treated for such contamination before reaching the consumer. Of the methods used for industrial application, UV treatment proved to be most effective for bacterial removal or reduction from the fresh produce, as well as maintaining its effectiveness through storage (refrigeration for 6 days). This method has also increased the characteristics of the fresh produce such as chlorophyll content, compared to the control. UV treatment in this study resulted in a large decrease in microbial counts but using this method, few bacterial colonies were still detected in some cases. Therefore, even this method may not be adequate to ensure the safety of the product (Fonseca and Rushing, 2006). Therefore it is imperative that further studies be conducted in order to optimise such potential methods such as the use of UV light, for application throughout the fresh produce industry, with the objective of preventing or reducing world-wide outbreaks of disease as a result of consumption of contaminated produce.

CHAPTER 5: GENERAL DISCUSSION AND CONCLUSION

5.1 The research in perspective

Water is an essential life resource and the main constituent of almost all life forms constituting more than 60% by volume in most animals and plants (Pidwirny, 2006). Water is also the most essential component throughout the growth and harvesting of fresh produce in the agriculture industry (Sapers, 2005). This industry is the single largest user of freshwater on a global scale (FAO, 1996). However, with the current global water crisis, freshwater sources are scarce (European commission, 2002).

South African demands on the already scarce water resources are increasing (DEAT, 1999). The sources of irrigation water that are used in South Africa include dams, rivers, ground water, reservoirs, industrial effluents and municipal supplies (SAWQG, 1996). South Africa depends on surface water resources for most of its irrigation requirements; however, this country is semi-arid with less than 9% of its rainfall available as surface water. It has been noted that the water sources for the northern part of South Africa are fully development and utilised, while the opposite is true for the South-Eastern regions of the country. This has lead to the exploitation of every major river in this country (NWRS, 2004; Midgley *et al.*, 2005). In South Africa, about 33% of the nation's waters are used for the irrigation of crops (Backeberg, 1996). This could create problems because if this water becomes contaminated to hazardous levels, there would be no alternative resource available due to it being wide-spread (Barnes *et al.*, 2007). The huge demand for freshwater in the fresh produce industry (FAO, 1996), together with other costs, forces farmers to use all available water resources (Suslow *et al.*, 2003). In many parts of South Africa, river water is used without any treatment. These waters also receive most of the nations treated sewage and therefore may contain high concentrations of microorganisms (DWAF, 1996; WHO 2002). Some of the contamination of rivers could also be caused by illegal dumping of industrial wastes, resulting in high concentrations of microorganisms and heavy metals (Barnes *et al.*, 2007). Therefore, the use of water for irrigation can be a major source of human pathogens that contaminates fresh produce (Sapers, 2005).

The consumption of fresh produce has increased over the years because of the changes in dietary habits of consumers (Barnes *et al.*, 2007). Fresh produce provides most of the daily vitamins, fibre and mineral requirements for humans (Johnston *et al*., 2006). Even though these products are nutritious, they can become contaminated by human pathogens, resulting in food-borne illnesses. Food-borne illnesses following the consumption of contaminated foodstuffs have been recognized and documented for centuries (DOH, 2007). The causes of food-borne illnesses include viruses, bacteria, parasites, toxins, metals and prions. Symptoms of these illnesses range from mild gastroenteritis to life-threatening neurologic, hepatic, and renal syndromes (Mead *et al*., 1999). Bacteria are the major causes of these food-borne illnesses, followed by viruses and parasites (DOH, 2007). *Salmonella* spp. has been associated with food-borne illnesses in the United States, with an increasing amount of outbreaks linked to contaminated produce (Hanning *et al*., 2009). A large outbreak caused by the consumption of verotoxin-producing *E. coli* contaminated lettuce had occurred in Sweden, in 2005 (Soderstrom *et al*., 2008). The threat of pathogenic microbes is of a serious concern, as certain strains of bacteria such as *E. coli* and *Salmonella* spp., have a very low infectious dose (Fratamico and Strobaugh, 1998). Furthermore, Kaferstein (2003) reported about 1.5 billion cases of diarrhoea, causing approximately 1.8 million deaths in children younger than 5 years of age, 70% of which were attributed to food-borne contaminants (Kaferstein, 2003; Dlamini, 2008).

In recent years, there have been numerous reported incidents of food-borne diseases in South Africa (DOH, 2007), for example, Smith *et al*. (2007) reported an outbreak of food-borne disease amongst school teachers at Rob Ferreira High School in White River, Mpumalanga, in December 2006. This outbreak was reported to have occurred after these teachers had consumed food that was prepared by the school kitchen. The causative agent of this outbreak was identified as *Salmonella enterica* serotype Virchow (*Salmonella* Virchow). It was further explained that outbreaks of food-borne disease in humans are common in South Africa, but these incidences are rarely reported (Smith *et al*., 2007). Some of the major reasons for this underreporting include the lack of efficient food-borne surveillance and that the South African legislation requires that food-borne outbreaks only be reported if the same doctor or health facility observes four or more of the same case (Dlamini, 2008).

In order to prevent or reduce such outbreaks of disease, it is important to monitor possible sources of contamination such as the quality of irrigation waters especially in the KwaZulu-Natal (KZN) province, as limited work has been performed in this area. It was evident throughout a year of sampling from four different farms in

KZN that the microbial and heavy metal quality of irrigation waters (river, borehole, dam, mixture of borehole and dam waters) are of a concern. Presumptive *E. coli* exceeded the DWAF (1996) guideline limit of 2×10^3 cfu/100 ml for *E. coli* in irrigation water, during the winter and spring periods. Presumptive *Salmonella* spp., *Shigella* spp. and coliforms were found at high concentrations in the waters sampled. Coliforms are known to be facultative anaerobic, Gram-negative, non-spore forming rods that have the ability to ferment lactose with gas formation within 48 h at 35 °C. These organisms are commonly used as bacterial indicators of sanitary quality of food and water, and are considered as an indicator of microbial pollution (Halablab *et al.*, 2010). The presence of faecal coliforms indicates that pathogens may be present as a result of faecal contamination by human or animal (Vishwanathan and Kaur, 2001). One of the chief sources of faecal contamination of natural waters is the un-serviced informal settlements that are established near rivers (Barnes *et al.*, 2007). Of the water sources tested in this study, dam water was the only one which had low concentrations of the presumptive pathogens. However from April to June 2010 the population of presumptive *Campylobacter* spp. in this water type was high.

It must be noted that water quality is affected by both natural processes as well as human activities. Usually, the quality of natural waters vary from place to place, and depend on seasonal changes, climatic changes and the types of soils, rocks and surfaces through which it moves. A range of human activities, such as agricultural activities, urban and industrial developments, mining and recreation, may considerably alter the quality of these natural waters and changes the water use potential (WQM, 2010). Furthermore, microbial populations have irregular activities in water bodies, meaning that their concentration can change independently of the original amount added to the water body due to various processes, including growth, decay, settling and chemical reactions (van Niekerk, 2000).

The consumption of lettuce and other leafy crops contaminated by poor quality irrigation water and manure, were shown to be causes of outbreaks of enterohemorrhagic *E. coli* O157:H7 (Islam *et al.*, 2005). This demonstrates that contaminated irrigation water could contaminate fresh produce and hence be a health hazard. The fresh produce samples tested in the present study showed an increase in microbial counts in summer, when the concentration of these presumptive bacterial populations had also increased in the irrigation waters. These findings provide evidence for a direct link between contaminated irrigation water and contaminated fresh produce, suggesting that irrigation water could be a major cause of the accumulation of contaminants on the fresh produce itself. The fresh produce was shown to be contaminated with presumptive *Campylobacter* spp., coliforms and *Shigella* spp. The area of the plant that had accumulated the highest concentrations of the microorganisms tested was the roots and edible portions of the plant. It was also found that contamination seemed to increase closer to harvest. Halablab *et al*. (2010) assessed the microbiological quality of fresh vegetables that were collected from different regions in the Bekaa Valley. Approximately sixty-three vegetable samples (lettuce, parsley and *Malva*), that were irrigated with Litani River water in the Bekaa Valley (Lebanon), and other control samples were assessed for their microbial load. Lettuce samples had significantly higher microbial loads, including coliforms, *E. coli* and *S. aureus*, than the parsley samples collected from different locations in the Bekaa Valley (Halablab *et al*., 2010). This is in agreement with the findings from the present study that the leafy produce, such as crisphead lettuce and spinach, had a higher microbial load compared to the other produce tested such as parsley.

Pollution by metals and organic compounds, such as pesticides, has been receiving increasing attention since serious cases of health impacts to humans and animals have occurred throughout the world through the unrestrained exposure to these pollutants (WQM, 2010). This type of pollution may be associated with specific industries or activities such as mining (WQM, 2010). Therefore, besides monitoring the microbial quality of these water types, it was necessary to evaluate the heavy metal content of these waters and that of the subsequent produce. In the water samples tested in this study, mercury (Hg) exceeded the recommended limit of 0.001 mg/L for drinking water established by the FAO and WHO. Arsenic (As), cadmium (Cd) and lead (Pb) were found to be within the recommended limits for irrigation water of 0.1, 0.1 and 0.5 mg/L, respectively (Ayers and Westcot, 1985). The heavy metal content of fresh produce were compared to the limits set for drinking water (Hg) and irrigation water (As, Cd, Pb). It was found that the majority of the fresh produce had accumulated Hg at levels above 0.001 mg/L, while As, Cd and Pb fell below the limit. It is recognized that the pH of natural waters is largely determined by geological and atmospheric influences. Also, freshwater sources in South Africa are somewhat well-buffered. However, human-induced acidification that results from industrial effluents, mine drainage and acid precipitation may cause a lowering of the pH, leading to mobilisation of metal elements such as Fe, Al, Cd, Co, Cu, Hg, Mn, Ni, Pb and Zn (DEAT, 1999). Although the concentrations of the different heavy metals were lower in water as compared to the fresh produce, the heavy metals could have accumulated in the fresh produce plant because of repeated exposure to the contaminated irrigation water and hence the fresh produce had higher concentrations of heavy metals as compared to the waters tested. The area of the plant which had accumulated high concentrations of heavy metals was the roots of the plant. The soil was also shown to contain higher heavy metal concentrations as compared to the plant.

Water has been suspected to be a major threat for the contamination of fresh produce because the produce is exposed to this water during irrigation as well as during the application of pesticides (Johnston *et al*., 2006; Barnes *et al*., 2007). Therefore, it is important to find strategies to effectively remove food-borne pathogens from the fresh produce during pre-harvest. In this study, the inhibitory effect of *Pseudomonas* spp. on different food-borne bacteria was evaluated. Several studies have been conducted previously to explore the antagonistic effect of native soil microflora on human pathogens (Johannessen *et al*., 2005). In this study, it was found that only *L. monocytogenes,* and not *E. coli, Salmonella* spp., and *Shigella* spp., was inhibited by *Pseudomonas* spp. Increased inhibition was influenced by increasing concentrations of *P. aeruginosa*, an increase in temperature, and at neutral pH. In the presence of iron, however, no inhibition of *L. monocytogenes* was detected. It was observed in the greenhouse experiments conducted, that a high concentration (12.81 log cfu/g) of *P. aeruginosa* in the soil, resulted in the inhibition of *L. monocytogenes,* thus preventing its uptake into the lettuce plant. However, at the minimal inhibitory concentration of 10^7 cfu/ml, *L. monocytogenes* was only slightly inhibited and was still taken up by the plant. The absence of *P. aeruginosa* in the soil (Control), resulted in the uptake of *L. monocytogenes* into the plant as early as week 1, compared to the other experiments where *L. monocytogenes* was not detected in the lettuce plant in non-autoclaved soil and detected at week 2 in the lettuce plant in autoclaved soil, that were spiked with *P. aeruginosa*. It was observed that *P. aeruginosa* did inhibit *L. monocytogenes* in the soil, thus preventing its uptake into the plant. However, the use of *P. aeruginosa* for inhibiting food-borne pathogens before their uptake into the edible portion of the plant was limited as this organism was found to be inhibitory to only one of the pathogens tested. Even though this organism had inhibited *L. monocytogenes* in the soil by week 3 (non-autoclaved soil) as evidenced by the spread-plating technique, the presence of *L. monocytogenes* was detected by denaturing gradient gel electrophoresis (DGGE), although at low concentrations, indicating that *L. monocytogenes* was probably non-culturable by standard techniques and could have entered into the VBNC state (Besnard *et al.*, 2000). Hence, when conditions became favourable, this organism could have proliferated in the soil and hence be taken up into the plant. However, results from the current study suggested that the use of *Pseudomonas* spp. in the food safety area of study may offer the means for the biological control of food-borne microbial pathogens without subjecting these fresh produce to different treatment methods which may alter the quality of the fresh produce. However, what must also be considered is the practicality and feasibility of using a biosafety level 2 pathogen (*P. aeruginosa*) to control other biosafety level 2 pathogens (for example *Listeria monocytogenes*). This does not seem practical; however, future studies should be employed in order to detect other potential biocontrol agents, which can alleviate the problem of food safety, but rather curb it.

Another strategy for reducing the level of contamination is the removal of pathogens from fresh produce at maturation, before consumption, through the use of post-harvest treatment methods. It is also important to note that consumers perception of good quality food is that it should look good and be firm. Although their purchases are based on the texture (feel of it) and appearance, their repeated purchases are based on eating quality (flavour) (Kader, 2002). Therefore, wash methods should remove microbial pathogens from the fresh produce as well as preserve the quality of the fresh produce. Household (tap water and NaCl) and industrial (chlorine, blanching, hydrogen peroxide and UV treatment) treatment methods were tested for their efficiency in the removal of presumptive pathogens from fresh produce. Of the household methods tested, NaCl was shown to be more effective in reducing microbial loads on fresh produce, than the use of tap water alone. Tap water has been used for years for the removal of pathogens from fresh produce; however, the efficacy of this type of treatment in eliminating or reducing naturally occurring microbial pathogens on fresh produce is limited (Brackett, 1999b). Chlorine treatment is still the most widely used washing method for reduction or elimination of pathogens in the fresh produce industry because of their easy use and the low costs associated with their use. However, its efficacy is limited to about 1 or 2 log reductions (Udompijitkul *et al.*, 2007). The efficacy of chlorine for the removal of pathogens from the fresh produce was also found to have limitations in this study, as chlorine treatment was not effective in reducing some of the presumptive pathogens tested. Seo and Frank (1999) showed that the disinfectant's effectiveness depends on the accessibility between the active sanitizing agent and the target microbes. Microorganisms may hide in cracks, crevices and stomata or penetrate into interior structures and may be protected from the action of these disinfectants. It has been noted that the hydrophobicity of microbial cells may aid in their protection against the penetration of disinfectants and may also facilitate the attachment to the epidermal layer of plant tissue (Burnett and Beuchat, 2001; Udompijitkul *et al*., 2007). The blanching method lowered the textural quality of the cabbage, lettuce and spinach samples tested. Of all the treatment methods tested, Ultra-Violet (UV) treatment was shown to be the most effective treatment method for the reduction of presumptive pathogens (even after refrigeration) from fresh produce while still retaining the same chlorophyll, vitamin C, reducing sugar and total carbohydrate content as compared to the unwashed product. In some cases, an increase in some of the nutritional content tested for was seen with the UV treatment method, such as the chlorophyll *b* content of broccoli samples, which was 3.78 and 9.44 µg/g higher than the control, before and after refrigeration, respectively.

As shown above, both the pre- and post-harvest removal methods are limited in terms of their ability to completely remove the presumptive pathogens from fresh produce. Moreover, the post-harvest processing of fresh produce, in some instances, promotes faster microbial degradation of the products in comparison with the raw commodities, since they are subjected to additional stress (Brackett, 1987; Lemoine *et al*., 2007). Because of these limitations with post-harvest washing technologies, it is preferable, to try and avoid microbial contamination of fruit and vegetables, wherever possible by following good agricultural and manufacturing practices rather than to depend on decontamination technologies (Sapers, 2001). A way to apply this would be to prevent or limit the exposure of fresh produce to sources of contamination. As reported previously by Beuchat and Ryu (1997) and shown in the current study, irrigation water can be a major source of contamination of fresh produce. The treatment of water with chlorine as a disinfectant has been used for years. However, the downfall with this treatment method is that a constant supply of chlorine is needed as liquid bleach degrades over time (Burch and Thomas, 1998). Therefore, it can be recommended that adopting proper decontamination methods (such as routine monitoring of irrigation water sources, removal of wild and domestic animals from farming plots, the use of gloves by the farm workers, etc.) for the removal of pathogens from irrigation water before its application to crops could be a solution to the growing concern regarding food safety. However, cognizance should be taken of the cost implications of such measures.

5.2 Potential for future development of the study

The viable but non-culturable state (VBNC) in bacteria has been elucidated over the last three decades. VBNC bacteria represent a part of the bacterial population that cannot grow on standard laboratory media (Besnard *et al*., 2000) and are a major concern as they remain potentially pathogenic under favourable growth conditions (Ravel *et al*., 1995). Standard laboratory media was used in this study and therefore VBNC bacteria could not be detected. Processes such as resuscitation should be employed when determining the effect of treatment methods in removing potential bacterial pathogens from fresh produce in order to account for the presence of bacteria only after refrigeration. The current study was limited as the wrong method was used for quantifying *Campylobacter* spp. and therefore accurate results could not be expected. Albeit culture independent approaches are attractive due to their speedy results but also limited in that nucleic acids is targeted as the presence of microbial DNA will not cause food borne illnesses but the presence of viable cells. Hence, cultural methods are required and can deliver sound results if appropriate methods are used. In addition, DGGE was limited in this study as heteroduplexes formed during the late PCR cycles, resulting in multiple banding on DGGE gels (Nicolaisen and Ramsing, 2002).

Further research is needed for the detection of the presence of VBNC bacteria in South African water sources and also post-harvest studies should include this aspect as different disinfectants could cause this state in bacteria. Also further research should pay attention to the effect of *Pseudomonas* spp. on food-borne pathogens and the effect of UV treatment on the microbial load of fresh produce, as these methods have shown promise for the removal of pathogens from fresh produce, in this study. The presence of viral pathogens in irrigation water and fresh produce in KZN should also be investigated in order to expand the information obtained in the current study. Furthermore, in order to mitigate against threats to food safety, strategies to decontaminate irrigation water should be researched for implementation in agricultural practice.

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APPENDIX A: REAGENTS

a) *Biochemical Tests*

i. Indole

Test reagent: Kovac’s (10 drops per tube) (Fluka)

Isoamyl alcohol (Merck)	150	ml
Para-dimethylaminobenzaldehyde (Merck)	10	g
HCl (conc) (Merck)	50	ml

ii. Methyl-Red

MR-VP broth (Merck)

Methyl red (Merck)	0.05	g
95% ethyl alcohol	150	ml
distilled water (bring up)	250	ml

iii. Voges-Proskauer

Barrits A: 40% solution of potassium hydroxide

Potassium hydroxide (Merck)	40	g
Distilled water (bring up)	100	ml

Barrits B: 5% α-naphthol in absolute ethyl alcohol

α-naphthol	5	g
Absolute ethyl alcohol (bring up)	100	ml

iv. Citrate

Simmons citrate agar (Oxoid)	23	g
Distilled water	1000	ml

v. Catalase

3% hydrogen peroxide (3 to 4 drops was added per slant)

Hydrogen peroxide	3	ml
Distilled water	97	ml

vi. Oxidase

Tetramethyl-p-phenylenediamine dihydrochloride	1	g

Distilled water	500	µl

b) *Anthrone reagent*

Anthrone (Fluka)	200	mg
Ice cold 95% H_2SO_4	100	ml

c) *Dinitrosalicyclic acid (DNS) method*

i. DNS reagent

Dinitrosalicyclic acid	1	g
Crystalline phenol (Merck)	200	mg
Sodium sulphite (Merck)	50	mg
1% NaOH	100	ml

ii. 40% Rochelle salt

Potassium sodium tartrate (Merck)	40	g
Distilled water (bring up)	100	ml

d) *0.5 M Disodium ethylenediaminetetraacetate (EDTA)*

EDTA (Saarchem)	186.12	g
Double distilled water (bring up)	1000	ml
pH adjustment (sodium hydroxide pellets ~20 g)	pH 8	

e) *50 × Tris-acetate EDTA buffer (TAE)*

Tris base	242	g
Glacial acetic acid (Merck)	57.1	ml
0.5 M EDTA (pH 8)	100	ml
Double distilled water (bring up)	1000	ml
pH adjustment (sodium hydroxide pellets/glacial acetic acid)	pH 8	

f) *Denaturing solution (0%)*

40% Acrylamide/bisacrylamide (BioRad)	15	ml
50 × TAE buffer (pH 8) (BioRad)	2	ml
Double distilled water	83	ml

g) ***Denaturing solution (100%)***

40% Acrylamide/bisacrylamide	15	ml
50 × TAE buffer (pH 8)	2	ml
40% (v/v) Deionized formamide (BioRad)	40	ml
7 M Urea (BioRad)	42	g
Double distilled water (bring up)	100	ml

APPENDIX B: NUMERICAL DATA

Table B1: Concentrations of As (mg/L) in fresh produce samples collected from farm A2, over a one-year period [a].

Samples	**Sampling period (months)**											
	Jul-09	**Aug-09**	**Sep-09**	**Oct-09**	**Nov-09**	**Dec-09**	**Jan-10**	**Feb-10**	**Mar-10**	**Apr-10**	**May-10**	**Jun-10**
Broccoli soil	0.015±0.001	0.018±0.004	0.011±0.002	ND	ND	ND	0.082±0.052	ND	ND	ND	ND	0.026±0.002
	0.015±0.001	0.004±0.000	N	ND	ND	ND	0.018±0.004	ND	ND	ND	ND	0.028±0.002
Broccoli stem	0.015±0.000	0.007±0.002	N	ND	ND	ND	0.088	ND	ND	ND	ND	0.023±0.002
Broccoli leaf	0.016±0.001	0.005±0.001	N	ND	ND	ND	0.023±0.030	ND	ND	ND	ND	0.012±0.001
Broccoli	ND	0.008±0.004	0.002	ND	ND	ND	0.006	ND	ND	ND	ND	0.009±0.000
Cabbage soil	0.015±0.001	0.024±0.017	0.034±0.043	N	ND	ND	ND	ND	ND	ND	0.012±0.003	0.012±0.001
Cabbage root	0.015±0.002	0.004±0.008	N	N	ND	ND	ND	ND	ND	ND	0.013±0.003	0.026±0.005
Cabbage	0.015±0.001	0.012±0.002	N	N	ND	ND	ND	ND	ND	ND	0.014±0.006	0.019±0.004
Crisphead lettuce soil	0.016±0.001	0.008±0.001	0.012±0.002	N	0.009±0.002	0.002	0.025±0.005	ND	ND	0.018±0.009	0.018±0.001	ND
Crisphead lettuce root	0.016±0.000	0.004±0.001	N	0.002	0.004±0.005	0.004	0.065±0.055	ND	ND	0.007±0.003	0.019±0.002	ND
Crisphead lettuce	0.020±0.001	0.004	N	0.001	N	N	0.016±0.021	ND	ND	0.009±0.004	0.016±0.006	ND
Spinach soil	0.015±0.001	ND	ND	N	ND	ND	ND	ND	ND	ND	ND	ND
Spinach root	0.016±0.001	ND	ND	N	ND	ND	ND	ND	ND	ND	ND	ND
Spinach stem	0.016±0.001	ND	ND	0.007	ND	ND	ND	ND	ND	ND	ND	ND
Spinach	0.017±0.002	ND	ND	N	ND	ND	ND	ND	ND	ND	ND	ND
Cauliflower soil	ND	0.009±0.001	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Cauliflower root	ND	0.005±0.001	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Cauliflower leaf	ND	0.003±0.001	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Cauliflower	ND	0.005±0.002	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND

ND = Not-determined, as the fresh produce was not present at the time of sample collection; N = heavy metal was not detected; [a] values are averages ± standard deviations (*n* = 3) and the detection limits for heavy metals (mg/L) were as follows: As (0.0053), Cd (0.0025), Hg (0.001), Pb (0.0042)

Table B2: Concentrations of Cd (mg/L) in fresh produce samples collected from farm A2, over a one-year period [a].

Samples	**Sampling period (months)**											
	Jul-09	**Aug-09**	**Sep-09**	**Oct-09**	**Nov-09**	**Dec-09**	**Jan-10**	**Feb-10**	**Mar-10**	**Apr-10**	**May-10**	**Jun-10**
Broccoli soil	0.016±0.001	0.013±0.004	0.011±0.002	ND	ND	ND	0.006±0.001	ND	ND	ND	ND	0.006±0.000
Broccoli root	0.015±0.000	N	N	ND	ND	ND	0.004±0.004	ND	ND	ND	ND	0.017±0.001
Broccoli stem	0.015±0.001	0.004	N	ND	ND	ND	0.006±0.000	ND	ND	ND	ND	0.001
Broccoli leaf	0.015±0.000	0.002±0.002	N	ND	ND	ND	0.006±0.000	ND	ND	ND	ND	0.011±0.002
Broccoli	ND	0.005	N	ND	ND	ND	0.002±0.001	ND	ND	ND	ND	0.006±0.001
Cabbage soil	0.016±0.001	0.020±0.017	0.034±0.043	0.007±0.002	ND	ND	ND	ND	ND	ND	0.007±0.001	0.008±0.002
Cabbage root	0.014±0.001	N	N	0.004±0.001	ND	ND	ND	ND	ND	ND	0.005	0.006±0.002
Cabbage	0.017±0.001	0.011	N	0.004±0.000	ND	ND	ND	ND	ND	ND	N	0.001±0.000
Crisphead lettuce soil	0.017±0.001	0.006±0.002	0.012±0.002	0.008±0.002	0.007±0.000	N	0.004±0.001	ND	ND	0.001±0.000	0.006±0.001	ND
Crisphead lettuce root	0.014±0.000	N	N	0.004±0.000	0.007±0.001	N	0.005±0.001	ND	ND	0.001±0.000	0.002±0.001	ND
Crisphead lettuce	0.019±0.001	0.001	N	0.004±0.001	0.008±0.000	N	0.004±0.001	ND	ND	0.001±0.000	N	ND
Spinach soil	0.016±0.001	ND	ND	0.005±0.001	ND	ND	ND	ND	ND	ND	ND	ND
Spinach root	0.015±0.000	ND	ND	0.005±0.001	ND	ND	ND	ND	ND	ND	ND	ND
Spinach stem	0.014±0.001	ND	ND	0.004±0.001	ND	ND	ND	ND	ND	ND	ND	ND
Spinach	0.017±0.001	ND	ND	0.004±0.000	ND	ND	ND	ND	ND	ND	ND	ND
Cauliflower soil	ND	0.007±0.001	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Cauliflower root	ND	0.002	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Cauliflower leaf	ND	N	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Cauliflower	ND	0.003±0.001	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND

ND = Not-determined, as the fresh produce was not present at the time of sample collection; N = heavy metal was not detected; [a] values are averages ± standard deviations (*n* = 3) and the detection limits for heavy metals (mg/L) were as follows: As (0.0053), Cd (0.0025), Hg (0.001), Pb (0.0042)

Table B3: Concentrations of Hg (mg/L) in fresh produce samples collected from farm A2, over a one-year period [a].

Samples	**Sampling period (months)**											
	Jul-09	**Aug-09**	**Sep-09**	**Oct-09**	**Nov-09**	**Dec-09**	**Jan-10**	**Feb-10**	**Mar-10**	**Apr-10**	**May-10**	**Jun-10**
Broccoli soil	0.006±0.001	0.020±0.006	0.007±0.001	ND	ND	ND	0.008±0.004	ND	ND	ND	ND	N
Broccoli root	0.006±0.001	0.028±0.020	0.023±0.001	ND	ND	ND	0.002	ND	ND	ND	ND	N
Broccoli stem	0.006±0.001	0.026±0.007	0.024±0.000	ND	ND	ND	0.008±0.003	ND	ND	ND	ND	N
Broccoli leaf	0.005±0.002	0.027±0.011	0.023±0.002	ND	ND	ND	0.009±0.001	ND	ND	ND	ND	N
Broccoli	ND	0.025±0.009	0.024±0.002	ND	ND	ND	N	ND	ND	ND	ND	N
Cabbage soil	0.006±0.001	0.019±0.001	0.008±0.001	0.013±0.000	ND	ND	ND	ND	ND	ND	N	N
Cabbage root	N	0.022±0.004	0.023±0.001	0.018±0.004	ND	ND	ND	ND	ND	ND	N	N
Cabbage	0.007±0.001	0.025±0.008	N	0.017±0.001	ND	ND	ND	ND	ND	ND	N	N
Crisphead lettuce soil	0.007±0.000	0.017±0.004	0.007±0.002	0.023±0.010	0.010±0.001	N	0.005±0.001	ND	ND	0.023±0.003	N	ND
Crisphead lettuce root	0.004±0.001	0.030±0.020	0.024±0.002	0.015±0.001	0.010±0.002	N	0.009±0.002	ND	ND	0.029±0.004	N	ND
Crisphead lettuce	0.004±0.001	0.022±0.009	0.025±0.003	0.017±0.001	0.018±0.004	0.009	0.004±0.002	ND	ND	0.028±0.002	N	ND
Spinach soil	0.006±0.001	ND	ND	0.015±0.002	ND	ND	ND	ND	ND	ND	ND	ND
Spinach root	0.004±0.002	ND	ND	0.019±0.003	ND	ND	ND	ND	ND	ND	ND	ND
Spinach stem	0.004±0.001	ND	ND	0.016±0.000	ND	ND	ND	ND	ND	ND	ND	ND
Spinach	0.006±0.001	ND	ND	0.014±0.001	ND	ND	ND	ND	ND	ND	ND	ND
Cauliflower soil	ND	0.014±0.002	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Cauliflower root	ND	0.036±0.016	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Cauliflower leaf	ND	0.050±0.053	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Cauliflower	ND	0.023±0.005	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND

ND = Not-determined, as the fresh produce was not present at the time of sample collection; N = heavy metal was not detected; [a] values are averages ± standard deviations (*n* = 3) and the detection limits for heavy metals (mg/L) were as follows: As (0.0053), Cd (0.0025), Hg (0.001), Pb (0.0042)

Table B4: Concentrations of Pb (mg/L) in fresh produce samples collected from farm A1, over a one-year period [a].

Samples	**Sampling period (months)**											
	Jul-09	**Aug-09**	**Sep-09**	**Oct-09**	**Nov-09**	**Dec-09**	**Jan-10**	**Feb-10**	**Mar-10**	**Apr-10**	**May-10**	**Jun-10**
Broccoli soil	0.036±0.020	0.279±0.072	0.109±0.018	ND	ND	ND	0.056±0.006	ND	ND	ND	ND	0.074±0.001
Broccoli root	N	0.018±0.002	0.058±0.014	ND	ND	ND	0.036±0.016	ND	ND	ND	ND	0.039±0.003
Broccoli stem	N	0.030±0.003	0.013±0.002	ND	ND	ND	0.022±0.001	ND	ND	ND	ND	0.010±0.002
Broccoli leaf	0.032	0.024±0.002	0.020±0.005	ND	ND	ND	0.026±0.002	ND	ND	ND	ND	0.012±0.002
Broccoli	-	0.024±0.007	0.014±0.001	ND	ND	ND	0.023±0.003	ND	ND	ND	ND	0.008±0.001
Cabbage soil	0.018±0.007	0.126±0.016	0.068±0.020	0.100±0.008	ND	ND	ND	ND	ND	ND	0.039±0.008	0.051±0.005
Cabbage root	N	0.023±0.006	0.025±0.015	0.076±0.020	ND	ND	ND	ND	ND	ND	0.047±0.008	0.053±0.003
Cabbage	N	0.024±0.005	N	0.033±0.008	ND	ND	ND	ND	ND	ND	0.006±0.002	0.007±0.001
Crisphead lettuce soil	0.005±0.017	0.111±0.003	0.098±0.018	0.077±0.004	0.042±0.012	0.034±0.014	0.050±0.003	ND	ND	0.009±0.004	0.052±0.012	ND
Crisphead lettuce root	N	0.026±0.009	0.029±0.003	0.054±0.004	0.045±0.011	0.051±0.006	0.026±0.005	ND	ND	0.016±0.008	0.022±0.008	ND
Crisphead lettuce	N	0.019±0.002	0.020±0.002	0.044±0.027	0.028±0.001	0.021±0.001	0.026±0.004	ND	ND	0.010±0.002	0.008±0.003	ND
Spinach soil	0.061±0.011	ND	ND	0.087±0.003	ND	ND	ND	ND	ND	ND	ND	ND
Spinach root	N	ND	ND	0.056±0.008	ND	ND	ND	ND	ND	ND	ND	ND
Spinach stem	N	ND	ND	0.028±0.002	ND	ND	ND	ND	ND	ND	ND	ND
Spinach	N	ND	ND	0.029±0.001	ND	ND	ND	ND	ND	ND	ND	ND
Cauliflower soil	ND	0.144±0.021	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Cauliflower root	ND	0.031±0.006	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Cauliflower leaf	ND	0.015±0.003	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Cauliflower	ND	0.026±0.005	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND

ND = Not-determined, as the fresh produce was not present at the time of sample collection; N = heavy metal was not detected; [a] values are averages ± standard deviations (*n* = 3) and the detection limits for heavy metals (mg/L) were as follows: As (0.0053), Cd (0.0025), Hg (0.001), Pb (0.0042)

Table B5: Concentrations of As (mg/L) in fresh produce samples collected from farm B, over a one-year period [a].

Samples	**Sampling period (months)**											
	Jul-09	Aug-09	Sep-09	Oct-09	Nov-09	Dec-09	Jan-10	Feb-10	Mar-10	Apr-10	May-10	Jun-10
Broccoli soil	0.033±0.002	0.033±0.017	0.032±0.003	ND	ND	ND	ND	ND	0.012	ND	ND	ND
Broccoli root	0.028±0.001	0.007±0.001	N	ND	ND	ND	ND	ND	0.016±0.013	ND	ND	ND
Broccoli stem	0.027±0.001	0.003±0.001	N	ND	ND	ND	ND	ND	0.004	ND	ND	ND
Broccoli leaf	0.027±0.001	0.006±0.007	N	ND	ND	ND	ND	ND	0.004±0.003	ND	ND	ND
Broccoli	0.029±0.002	0.003±0.002	0.018±0.001	ND	ND	ND	ND	ND	0.011±0.002	ND	ND	ND
Red cabbage soil	0.041±0.001	0.029±0.013	N	N	ND	ND	ND	ND	ND	ND	ND	ND
Red cabbage stem	0.028±0.001	0.005±0.001	N	N	ND	ND	ND	ND	ND	ND	ND	ND
Red cabbage	0.028±0.001	0.001±0.000	0.015±0.001	0.001	ND	ND	ND	ND	ND	ND	ND	ND
Cabbage soil	ND	ND	ND	ND	0.020±0.010	0.012	0.041±0.027	ND	ND	ND	ND	0.020±0.003
Cabbage root	ND	ND	ND	ND	0.016±0.004	0.029±0.036	0.011	ND	ND	ND	ND	0.026±0.003
Cabbage	ND	ND	ND	ND	0.012±0.009	0.008±0.005	0.056±0.055	ND	ND	ND	ND	0.013±0.004
Crisphead lettuce soil	0.032±0.001	0.026±0.003	ND	0.012	ND	0.001±0.001	ND	ND	ND	0.009±0.008	0.002±0.001	0.027±0.001
Crisphead lettuce root	0.028±0.001	0.006±0.001	ND	0.004	ND	0.011±0.001	ND	ND	ND	0.013±0.004	0.026±0.008	0.016±0.001
Crisphead lettuce	0.028±0.000	0.004±0.002	ND	N	ND	0.009±0.005	ND	ND	ND	0.012±0.009	0.012±0.003	0.018±0.006
Spinach soil	0.015±0.001	0.023±0.002	N	N	0.008±0.006	0.009	0.017±0.001	ND	ND	0.004±0.002	ND	ND
Spinach root	0.016±0.000	0.001	N	0.003	0.013±0.003	0.009±0.002	0.018±0.001	ND	ND	0.005±0.000	ND	ND
Spinach stem	0.016±0.001	0.001	0.012±0.010	ND	0.008±0.004	0.003	0.016±0.009	ND	ND	N	ND	ND
Spinach	0.017±0.001	0.001±0.001	N	N	0.003	0.003	0.013±0.005	ND	ND	0.007±0.003	ND	ND

ND = Not-determined, as the fresh produce was not present at the time of sample collection; N = heavy metal was not detected; [a] values are averages ± standard deviations ($n = 3$) and the detection limits for heavy metals (mg/L) were as follows: As (0.0053), Cd (0.0025), Hg (0.001), Pb (0.0042)

Table B6: Concentrations of As (mg/L) in Chinese cabbage, Parsley and Spinach samples collected from farm B, over a one-year period [a].

Samples	**Sampling period (months)**											
	Jul-09	Aug-09	Sep-09	Oct-09	Nov-09	Dec-09	Jan-10	Feb-10	Mar-10	Apr-10	May-10	Jun-10
Chinese cabbage soil	ND	ND	N	ND	ND	ND	ND	ND	ND	ND	ND	ND
Chinese cabbage root	ND	ND	N	ND	ND	ND	ND	ND	ND	ND	ND	ND
Chinese cabbage	ND	ND	0.014±0.002	ND	ND	ND	ND	ND	ND	ND	ND	ND
Parsley soil	ND	ND	ND	N	0.014±0.004	ND	ND	ND	ND	ND	ND	ND
Parsley root	ND	ND	ND	0.011±0.010	0.029	ND	ND	ND	ND	ND	ND	ND
Parsley stem	ND	ND	ND	N	0.012±0.002	ND	ND	ND	ND	ND	ND	ND
Parsley	ND	ND	ND	N	N	ND	ND	ND	ND	ND	ND	ND
Bell Pepper soil	ND	ND	ND	ND	ND	ND	ND	N	0.010	0.016±0.003	0.020±0.004	ND
Bell Pepper root	ND	ND	ND	ND	ND	ND	ND	0.012	0.009	0.011±0.003	0.021±0.006	ND
Bell Pepper stem	ND	ND	ND	ND	ND	ND	ND	0.002	0.008±0.007	0.013±0.001	0.016±0.006	ND
Bell Pepper leaf	ND	ND	ND	ND	ND	ND	ND	0.007	0.005±0.003	0.012±0.003	0.023±0.006	ND
Bell Pepper	ND	ND	ND	ND	ND	ND	ND	0.012	0.006±0.004	0.020±0.004	0.018±0.005	ND

ND = Not-determined, as the fresh produce was not present at the time of sample collection; N = heavy metal was not detected; [a] values are averages ± standard deviations (*n* = 3) and the detection limits for heavy metals (mg/L) were as follows: As (0.0053), Cd (0.0025), Hg (0.001), Pb (0.0042)

Table B7: Concentrations of Cd (mg/L) in fresh produce samples collected from farm B, over a one-year period [a].

Samples	**Sampling period (months)**											
	Jul-09	**Aug-09**	**Sep-09**	**Oct-09**	**Nov-09**	**Dec-09**	**Jan-10**	**Feb-10**	**Mar-10**	**Apr-10**	**May-10**	**Jun-10**
Broccoli soil	0.035±0.003	0.036±0.018	0.032±0.003	ND	ND	ND	ND	ND	0.017±0.002	ND	ND	ND
Broccoli root	0.027±0.000	0.010±0.000	N	ND	ND	ND	ND	ND	0.001	ND	ND	ND
Broccoli stem	0.027±0.000	0.004±0.001	N	ND	ND	ND	ND	ND	0.001±0.000	ND	ND	ND
Broccoli leaf	0.027±0.000	0.008±0.007	N	ND	ND	ND	ND	ND	0.011±0.001	ND	ND	ND
Broccoli	0.027±0.000	0.004±0.001	0.018±0.001	ND	ND	ND	ND	ND	N	ND	ND	ND
Red cabbage soil	0.041±0.001	0.035±0.015	N	0.005±0.003	ND	ND	ND	ND	ND	ND	ND	ND
Red cabbage stem	0.027±0.000	0.010±0.001	N	0.008±0.002	ND	ND	ND	ND	ND	ND	ND	ND
Red cabbage	0.026±0.001	0.003±0.000	0.015±0.001	0.005±0.001	ND	ND	ND	ND	ND	ND	ND	ND
Cabbage soil	ND	ND	ND	ND	0.012±0.003	0.002	0.006±0.000	ND	ND	ND	ND	0.017±0.001
Cabbage root	ND	ND	ND	ND	0.007±0.001	N	0.002±0.001	ND	ND	ND	ND	0.003±0.000
Cabbage	ND	ND	ND	ND	0.007±0.000	N	0.005±0.001	ND	ND	ND	ND	0.006±0.000
Crisphead lettuce soil	0.032±0.001	0.033±0.003	ND	0.007±0.002	ND	0.007±0.002	ND	ND	ND	0.004±0.001	0.021±0.005	0.033±0.001
Crisphead lettuce root	0.027±0.001	0.010±0.000	ND	0.005±0.001	ND	N	ND	ND	ND	0.001±0.001	0.002	0.005±0.001
Crisphead lettuce	0.027±0.000	0.004±0.001	ND	0.006±0.001	ND	N	ND	ND	ND	0.001±0.000	0.003±0.002	0.001
Spinach soil	0.030±0.001	0.026±0.001	N	0.010±0.004	0.017±0.014	N	0.005±0.000	ND	ND	0.002±0.000	ND	ND
Spinach root	0.027±0.000	0.003±0.000	N	0.006±0.001	0.007±0.001	N	0.002±0.001	ND	ND	0.002±0.000	ND	ND
Spinach stem	0.027±0.000	0.003±0.001	0.012±0.010	ND	0.007±0.000	N	0.004±0.001	ND	ND	N	ND	ND
Spinach	0.027±0.000	0.003±0.001	N	0.006±0.001	0.007±0.000	N	0.005±0.000	ND	ND	0.002±0.001	ND	ND

ND = Not-determined, as the fresh produce was not present at the time of sample collection; N = heavy metal was not detected; [a] values are averages ± standard deviations (*n* = 3) and the detection limits for heavy metals (mg/L) were as follows: As (0.0053), Cd (0.0025), Hg (0.001), Pb (0.0042)

Table B8: Concentrations of Cd (mg/L) in Chinese cabbage, Parsley and Spinach samples collected from farm B, over a one-year period [a].

Samples	**Sampling period (months)**											
	Jul-09	**Aug-09**	**Sep-09**	**Oct-09**	**Nov-09**	**Dec-09**	**Jan-10**	**Feb-10**	**Mar-10**	**Apr-10**	**May-10**	**Jun-10**
Chinese cabbage soil	ND	ND	N	ND	ND	ND	ND	ND	ND	ND	ND	ND
Chinese cabbage root	ND	ND	N	ND	ND	ND	ND	ND	ND	ND	ND	ND
Chinese cabbage	ND	ND	0.014±0.002	ND	ND	ND	ND	ND	ND	ND	ND	ND
Parsley soil	ND	ND	ND	0.016±0.003	0.022±0.005	ND	ND	ND	ND	ND	ND	ND
Parsley root	ND	ND	ND	0.006±0.001	0.008±0.001	ND	ND	ND	ND	ND	ND	ND
Parsley stem	ND	ND	ND	0.009±0.004	0.007±0.001	ND	ND	ND	ND	ND	ND	ND
Parsley	ND	ND	ND	0.006±0.001	0.008±0.001	ND	ND	ND	ND	ND	ND	ND
Bell Pepper soil	ND	ND	ND	ND	ND	ND	ND	0.026±0.005	0.021±0.003	0.001±0.000	0.021±0.006	ND
Bell Pepper root	ND	ND	ND	ND	ND	ND	ND	N	0.002	0.002±0.000	0.003±0.001	ND
Bell Pepper stem	ND	ND	ND	ND	ND	ND	ND	N	0.001	0.002±0.001	0.002	ND
Bell Pepper leaf	ND	ND	ND	ND	ND	ND	ND	0.008	0.002	0.001±0.000	0.003±0.003	ND
Bell Pepper	ND	ND	ND	ND	ND	ND	ND	N	0.001±0.001	0.005±0.002	0.003±0.001	ND

ND = Not-determined, as the fresh produce was not present at the time of sample collection; N = heavy metal was not detected; [a] values are averages ± standard deviations (*n* = 3) and the detection limits for heavy metals (mg/L) were as follows: As (0.0053), Cd (0.0025), Hg (0.001), Pb (0.0042)

Table B9: Concentrations of Hg (mg/L) in fresh produce samples collected from farm B, over a one-year period [a].

Samples	**Sampling time (months)**											
	Jul-09	**Aug-09**	**Sep-09**	**Oct-09**	**Nov-09**	**Dec-09**	**Jan-10**	**Feb-10**	**Mar-10**	**Apr-10**	**May-10**	**Jun-10**
Broccoli soil	0.033±0.001	0.063	0.005±0.001	ND	ND	ND	ND	ND	0.001	ND	ND	ND
Broccoli root	0.034±0.001	0.034±0.001	N	ND	ND	ND	ND	ND	0.006	ND	ND	ND
Broccoli stem	0.034±0.000	0.041±0.004	N	ND	ND	ND	ND	ND	0.004±0.001	ND	ND	ND
Broccoli leaf	0.034±0.000	0.032±0.001	N	ND	ND	ND	ND	ND	0.007±0.001	ND	ND	ND
Broccoli	0.034±0.001	0.033±0.002	0.004±0.001	ND	ND	ND	ND	ND	0.002	ND	ND	ND
Red cabbage soil	0.034±0.001	0.025±0.002	N	0.010±0.001	ND	ND	ND	ND	ND	ND	ND	ND
Red cabbage stem	0.033±0.001	0.035±0.003	N	0.018±0.004	ND	ND	ND	ND	ND	ND	ND	ND
Red cabbage	0.032±0.001	0.026±0.001	0.003±0.002	0.018±0.001	ND	ND	ND	ND	ND	ND	ND	ND
Cabbage soil	ND	ND	ND	ND	0.010±0.002	N	0.007±0.001	ND	ND	ND	ND	N
Cabbage root	ND	ND	ND	ND	0.010±0.001	0.021±0.006	N	ND	ND	ND	ND	N
Cabbage	ND	ND	ND	ND	0.019±0.002	N	0.006±0.001	ND	ND	ND	ND	N
Crisphead lettuce soil	0.034±0.000	0.024±0.002	ND	0.009±0.002	ND	N	ND	ND	ND	0.023±0.001	N	N
Crisphead lettuce root	0.032±0.001	0.035±0.001	ND	0.015±0.001	ND	0.046	ND	ND	ND	0.027±0.001	N	N
Crisphead lettuce	0.034±0.001	0.026±0.001	ND	0.017±0.002	ND	0.043±0.010	ND	ND	ND	0.026±0.002	N	0.050±0.001
Spinach soil	0.034±0.001	0.031±0.002	N	0.024±0.016	0.010±0.001	N	0.009±0.001	ND	ND	0.014±0.001	ND	ND
Spinach root	0.033±0.001	0.029±0.002	0.021	0.016±0.001	0.017±0.002	N	N	ND	ND	0.016±0.000	ND	ND
Spinach stem	0.033±0.001	0.035±0.010	0.011±0.005	ND	0.020±0.003	0.007	0.009±0.001	ND	ND	N	ND	ND
Spinach	0.034±0.001	0.030±0.002	0.026±0.002	0.018±0.002	0.011±0.002	N	0.043±0.032	ND	ND	0.026±0.002	ND	ND

ND = Not-determined, as the fresh produce was not present at the time of sample collection; N = heavy metal was not detected; [a] values are averages ± standard deviations (*n* = 3) and the detection limits for heavy metals (mg/L) were as follows: As (0.0053), Cd (0.0025), Hg (0.001), Pb (0.0042)

Table B10: Concentrations of Hg (mg/L) in Chinese cabbage, Parsley and Spinach samples collected from farm B, over a one-year period [a].

Samples	Sampling period (months)											
	Jul-09	**Aug-09**	**Sep-09**	**Oct-09**	**Nov-09**	**Dec-09**	**Jan-10**	**Feb-10**	**Mar-10**	**Apr-10**	**May-10**	**Jun-10**
Chinese cabbage soil	ND	ND	N	ND	ND	ND	ND	ND	ND	ND	ND	ND
Chinese cabbage root	ND	ND	N	ND	ND	ND	ND	ND	ND	ND	ND	ND
Chinese cabbage	ND	ND	0.005±0.001	ND	ND	ND	ND	ND	ND	ND	ND	ND
Parsley soil	ND	ND	ND	0.011±0.002	0.009±0.002	ND	ND	ND	ND	ND	ND	ND
Parsley root	ND	ND	ND	0.016±0.001	0.011±0.001	ND	ND	ND	ND	ND	ND	ND
Parsley stem	ND	ND	ND	0.016±0.002	0.016±0.002	ND	ND	ND	ND	ND	ND	ND
Parsley	ND	ND	ND	0.023±0.002	0.033±0.009	ND	ND	ND	ND	ND	ND	ND
Bell Pepper soil	ND	ND	ND	ND	ND	ND	ND	0.017±0.001	N	0.021±0.002	N	ND
Bell Pepper root	ND	ND	ND	ND	ND	ND	ND	0.023±0.009	0.002±0.002	0.021±0.001	N	ND
Bell Pepper stem	ND	ND	ND	ND	ND	ND	ND	0.017±0.001	0.014±0.010	0.029±0.006	N	ND
Bell Pepper leaf	ND	ND	ND	ND	ND	ND	ND	0.018±0.001	0.001	0.023±0.002	N	ND
Bell Pepper	ND	ND	ND	ND	ND	ND	ND	0.019±0.003	0.005±0.001	0.044±0.003	N	ND

ND = Not-determined, as the fresh produce was not present at the time of sample collection; N = heavy metal was not detected; [a] values are averages ± standard deviations (*n* = 3) and the detection limits for heavy metals (mg/L) were as follows: As (0.0053), Cd (0.0025), Hg (0.001), Pb (0.0042)

Table B11: Concentrations of Pb (mg/L) in fresh produce samples collected from farm B, over a one-year period [a].

Samples	Sampling period (months)											
	Jul-09	**Aug-09**	**Sep-09**	**Oct-09**	**Nov-09**	**Dec-09**	**Jan-10**	**Feb-10**	**Mar-10**	**Apr-10**	**May-10**	**Jun-10**
Broccoli soil	0.192±0.023	0.526±0.336	0.221±0.012	ND	ND	ND	ND	ND	0.100±0.012	ND	ND	ND
Broccoli root	0.040±0.004	0.045±0.006	N	ND	ND	ND	ND	ND	0.037±0.019	ND	ND	ND
Broccoli stem	0.040±0.001	0.004±0.002	N	ND	ND	ND	ND	ND	0.006	ND	ND	ND
Broccoli leaf	0.043±0.006	0.004	N	ND	ND	ND	ND	ND	0.055±0.005	ND	ND	ND
Broccoli	0.037±0.004	0.003	0.360±0.010	ND	ND	ND	ND	ND	0.001	ND	ND	ND
Red cabbage soil	0.132±0.016	0.135±0.103	N	0.185±0.021	ND	ND	ND	ND	ND	ND	ND	ND
Red cabbage stem	0.041±0.002	0.036±0.008	N	0.168±0.061	ND	ND	ND	ND	ND	ND	ND	ND
Red cabbage	0.039±0.008	0.003	0.318±0.006	0.037±0.019	ND	ND	ND	ND	ND	ND	ND	ND
Cabbage soil	ND	ND	ND	ND	0.070±0.033	0.056±0.026	0.043±0.005	ND	ND	ND	ND	0.166±0.002
Cabbage root	ND	ND	ND	ND	0.030±0.010	0.013±0.007	0.042±0.014	ND	ND	ND	ND	0.031±0.004
Cabbage	ND	ND	ND	ND	0.023±0.006	0.015±0.006	0.022±0.005	ND	ND	ND	ND	0.037±0.002
Crisphead lettuce soil	0.160±0.011	0.213±0.009	ND	0.145±0.011	ND	0.180±0.022	ND	ND	ND	0.010±0.002	0.157±0.003	0.140±0.005
Crisphead lettuce root	0.043±0.008	0.041±0.006	ND	0.038±0.004	ND	0.008±0.004	ND	ND	ND	0.033±0.019	0.017±0.005	0.044±0.003
Crisphead lettuce	0.038±0.001	0.010±0.009	ND	0.027±0.002	ND	0.006±0.004	ND	ND	ND	0.033±0.009	0.014±0.005	0.027
Spinach soil	0.078±0.006	0.172±0.009	N	0.150±0.012	0.069±0.004	0.030±0.024	0.031±0.004	ND	ND	0.008±0.001	ND	ND
Spinach root	0.036±0.002	0.028	0.013	0.047±0.011	0.048±0.017	0.026±0.002	0.028±0.010	ND	ND	0.014±0.001	ND	ND
Spinach stem	0.043±0.005	0.004	0.007±0.006	N	0.029±0.013	0.027±0.019	0.021±0.005	ND	ND	N	ND	ND
Spinach	0.043±0.009	0.004±0.003	0.015±0.001	0.028±0.003	0.042±0.024	0.032±0.024	0.025±0.002	ND	ND	0.021±0.008	ND	ND

ND = Not-determined, as the fresh produce was not present at the time of sample collection; N = heavy metal was not detected; [a] values are averages ± standard deviations ($n = 3$) and the detection limits for heavy metals (mg/L) were as follows: As (0.0053), Cd (0.0025), Hg (0.001), Pb (0.0042)

Table B12: Concentrations of Pb (mg/L) in Chinese cabbage, Parsley and Spinach samples collected from farm B, over a one-year period [a].

Samples	Sampling period (months)										
	Jul-09	**Aug-09**	**Sep-09**	**Oct-09**	**Nov-09**	**Dec-09**	**Jan-10**	**Feb-10**	**Mar-10**	**Apr-10**	**May-10**	**Jun-10**
Chinese cabbage soil	ND	ND	N	ND	ND	ND	ND	ND	ND	ND	ND	ND
Chinese cabbage root	ND	ND	N	ND	ND	ND	ND	ND	ND	ND	ND	ND
Chinese cabbage	ND	ND	0.269±0.064	ND	ND	ND	ND	ND	ND	ND	ND	ND
Parsley soil	ND	ND	ND	0.221±0.066	0.156±0.048	ND	ND	ND	ND	ND	ND	ND
Parsley root	ND	ND	ND	0.031±0.006	0.039±0.011	ND	ND	ND	ND	ND	ND	ND
Parsley stem	ND	ND	ND	0.038±0.006	0.017±0.003	ND	ND	ND	ND	ND	ND	ND
Parsley	ND	ND	ND	0.029±0.004	0.068±0.025	ND	ND	ND	ND	ND	ND	ND
Bell Pepper soil	ND	ND	ND	ND	ND	ND	ND	0.155±0.006	0.092±0.011	0.005±0.002	0.115±0.016	ND
Bell Pepper root	ND	ND	ND	ND	ND	ND	ND	0.011±0.010	0.010±0.014	0.036±0.009	0.036±0.003	ND
Bell Pepper stem	ND	ND	ND	ND	ND	ND	ND	0.008±0.002	0.005	0.016±0.007	0.014±0.004	ND
Bell Pepper leaf	ND	ND	ND	ND	ND	ND	ND	0.010±0.001	N	0.006±0.001	0.029±0.008	ND
Bell Pepper	ND	ND	ND	ND	ND	ND	ND	0.009±0.006	0.002±0.001	0.019±0.002	0.026±0.006	ND

ND = Not-determined, as the fresh produce was not present at the time of sample collection; N = heavy metal was not detected; [a] values are averages ± standard deviations (*n* = 3) and the detection limits for heavy metals (mg/L) were as follows: As (0.0053), Cd (0.0025), Hg (0.001), Pb (0.0042)

Table B13: Concentrations of As (a) and Cd (b) (mg/L) in fresh produce samples collected from farm C, over a one-year period [a].

(a)

Samples	**Sampling period (months)**							
	Jul-09	**Aug-09**	**Sep-09**	**Oct-09**	**Nov-09**	**Dec-09**	**Jan-10**	**Feb-10**
Cabbage soil	0.010±0.001	0.034±0.033	N	ND	ND	ND	ND	ND
Cabbage root	0.006±0.000	0.001±0.001	N	ND	ND	ND	ND	ND
Cabbage	0.007±0.001	0.001±0.001	0.010±0.003	ND	ND	ND	ND	ND
Jam tomato soil	ND	ND	ND	ND	ND	ND	N	0.008
Jam tomato root	ND	ND	ND	ND	ND	ND	0.013±0.004	0.012
Jam tomato stem	ND	ND	ND	ND	ND	ND	0.099±0.044	0.004
Jam tomato leaf	ND	ND	ND	ND	ND	ND	0.007±0.003	0.010±0.001
Jam tomato	ND	ND	ND	ND	ND	ND	ND	0.011

(b)

Samples	**Sampling period (months)**							
	Jul-09	**Aug-09**	**Sep-09**	**Oct-09**	**Nov-09**	**Dec-09**	**Jan-10**	**Feb-10**
Cabbage soil	0.014±0.001	0.040±0.038	N	ND	ND	ND	ND	ND
Cabbage root	0.011±0.001	0.002±0.001	N	ND	ND	ND	ND	ND
Cabbage	0.011±0.001	0.003±0.001	0.010±0.003	ND	ND	ND	ND	ND
Jam tomato soil	ND	ND	ND	ND	ND	ND	0.006±0.001	0.009±0.003
Jam tomato root	ND	ND	ND	ND	ND	ND	0.002±0.000	N
Jam tomato stem	ND	ND	ND	ND	ND	ND	0.006±0.001	N
Jam tomato leaf	ND	ND	ND	ND	ND	ND	0.002±0.000	N
Jam tomato	ND	ND	ND	ND	ND	ND	ND	0.009±0.000

ND = Not-determined, as the fresh produce was not present at the time of sample collection; N = heavy metal was not detected; [a] values are averages ± standard deviations (*n* = 3) and the detection limits for heavy metals (mg/L) were as follows: As (0.0053), Cd (0.0025), Hg (0.001), Pb (0.0042)

Table B14: Concentrations of Hg (a) and Pb (b) (mg/L) in fresh produce samples collected from farm C, over a one-year period [a].

(a)

Samples	Sampling period (months)							
	Jul-09	**Aug-09**	**Sep-09**	**Oct-09**	**Nov-09**	**Dec-09**	**Jan-10**	**Feb-10**
Cabbage soil	0.036±0.001	0.026±0.001	0.022±0.002	ND	ND	ND	ND	ND
Cabbage root	0.033±0.001	0.027±0.003	0.024±0.002	ND	ND	ND	ND	ND
Cabbage	0.034±0.001	0.026±0.001	0.021±0.003	ND	ND	ND	ND	ND
Jam tomato soil	ND	ND	ND	ND	ND	ND	N	0.019±0.002
Jam tomato root	ND	ND	ND	ND	ND	ND	0.015±0.003	0.018±0.001
Jam tomato stem	ND	ND	ND	ND	ND	ND	0.012±0.002	0.024±0.009
Jam tomato leaf	ND	ND	ND	ND	ND	ND	0.002	0.018±0.001
Jam tomato	ND	ND	ND	ND	ND	ND	ND	0.016±0.001

(b)

Samples	Sampling period (months)							
	Jul-09	**Aug-09**	**Sep-09**	**Oct-09**	**Nov-09**	**Dec-09**	**Jan-10**	**Feb-10**
Cabbage soil	0.132±0.011	0.070±0.013	0.015±0.003	ND	ND	ND	ND	ND
Cabbage root	0.035±0.002	0.016±0.002	0.025±0.010	ND	ND	ND	ND	ND
Cabbage	0.041±0.008	N	0.233±0.065	ND	ND	ND	ND	ND
Jam tomato soil	ND	ND	ND	ND	ND	ND	0.071±0.005	0.111±0.025
Jam tomato root	ND	ND	ND	ND	ND	ND	0.026±0.002	0.051±0.019
Jam tomato stem	ND	ND	ND	ND	ND	ND	0.029±0.004	0.007±0.005
Jam tomato leaf	ND	ND	ND	ND	ND	ND	0.020±0.002	0.004±0.002
Jam tomato	ND	ND	ND	ND	ND	ND	ND	0.097±0.004

ND = Not-determined, as the fresh produce was not present at the time of sample collection; N = heavy metal was not detected; [a] values are averages ± standard deviations (*n* = 3) and the detection limits for heavy metals (mg/L) were as follows: As (0.0053), Cd (0.0025), Hg (0.001), Pb (0.0042)

Table B15: The microbial analysis of irrigation water collected from farm A1 during a one-year period.

Sampling period	Replicates	Presumptive microbial pathogens (cfu/100 ml)					
		Campylobacter spp.	Coliforms	*E. coli*	*L. monocytogenes*	*Salmonella* spp.	*Shigella* spp.
Jul-09	1	N	2300.00	2300.00	N	1780.00	1740.00
	2	N	2320.00	2220.00	N	1820.00	1700.00
	3	N	2360.00	2260.00	N	1740.00	1740.00
	average	-	2326.67	2260.00	-	1780.00	1726.67
	SD	-	30.55	40.00	-	40.00	23.09
Aug-09	1	N	5940.00	2600.00	30.00	5680.00	2420.00
	2	N	5980.00	2840.00	30.00	5560.00	2460.00
	3	N	5920.00	2820.00	32.00	5540.00	2500.00
	average	-	5946.67	2753.33	30.67	5593.33	2460.00
	SD	-	30.55	133.17	1.15	75.72	40.00
Sep-09	1	N	5960.00	N	N	N	3960.00
	2	N	5920.00	N	N	N	3920.00
	3	N	5960.00	N	N	N	3900.00
	average	-	5946.67	-	-	-	3926.67
	SD	-	23.09	-	-	-	30.55
Oct-09	1	N	2020.00	N	N	N	800.00
	2	N	2060.00	N	N	N	1160.00
	3	N	2420.00	N	N	N	900.00
	average	-	2166.67	-	-	-	953.33
	SD	-	220.30	-	-	-	185.83
Nov-09	1	N	4000.00	N	N	N	3400.00
	2	N	4060.00	N	N	N	3320.00
	3	N	4060.00	N	N	N	3460.00
	average	-	4040.00	-	-	-	3393.33
	SD	-	34.64	-	-	-	70.24
Dec-09	1	N	4220.00	30.00	N	N	3620.00
	2	N	3920.00	31.00	N	N	3580.00
	3	N	4180.00	34.00	N	N	3860.00
	average	-	4106.67	31.67	-	-	3686.67
	SD	-	162.89	2.08	-	-	151.44
Jan-10	1	N	4600.00	68.00	N	30.00	4820.00
	2	N	4660.00	69.00	N	32.00	4800.00
	3	N	4820.00	79.00	N	32.00	4640.00
	average	-	4693.33	72.00	-	31.33	4753.33
	SD	-	113.72	6.08	-	1.15	98.66
Feb-10	1	N	5780.00	123.00	30.00	102.00	5920.00
	2	N	5800.00	121.00	34.00	110.00	5980.00

Table B15/ Cont.

Sampling period	Replicates	Presumptive microbial pathogens (cfu/100 ml)					
		Campylobacter spp.	Coliforms	*E. coli*	*L. monocytogenes*	*Salmonella* spp.	*Shigella* spp.
	3	N	5740.00	123.00	31.00	113.00	5960.00
	average	-	5773.33	122.33	31.67	108.33	5953.33
	SD	-	30.55	1.15	2.08	5.69	30.55
Mar-10	1	N	7200.00	171.00	59.00	130.00	8800.00
	2	N	6600.00	174.00	63.00	135.00	9400.00
	3	N	7400.00	180.00	64.00	138.00	9400.00
	average	-	7066.67	175.00	62.00	134.33	9200.00
	SD	-	416.33	4.58	2.65	4.04	346.41
Apr-10	1	N	5740.00	1.51	0.41	0.94	82.00
	2	N	5720.00	1.54	0.42	0.96	86.00
	3	N	5740.00	1.59	0.41	0.97	94.00
	average	-	5733.33	1.55	0.41	0.96	87.33
	SD	-	11.55	0.04	0.01	0.02	6.11
May-10	1	N	4060.00	135.00	32.00	110.00	5120.00
	2	N	4100.00	138.00	34.00	113.00	5100.00
	3	N	4160.00	131.00	35.00	113.00	5100.00
	average	-	4106.67	134.67	33.67	112.00	5106.67
	SD	-	50.33	3.51	1.53	1.73	11.55
Jun-10	1	N	3700.00	80.00	30.00	71.00	2100.00
	2	N	3720.00	78.00	30.00	73.00	2180.00
	3	N	3660.00	82.00	31.00	73.00	2040.00
	average	-	3693.33	80.00	30.33	72.33	2106.67
	SD	-	30.55	2.00	0.58	1.15	70.24

N = microbial pathogen was not detected; SD = standard deviation

Table B16: The microbial analysis of irrigation water collected from farm A2 during a one-year period.

Sampling period	Replicates	Presumptive microbial pathogens (cfu/100 ml)					
		***Campylobacter* spp.**	**Coliforms**	***E. coli***	***L. monocytogenes***	***Salmonella* spp.**	***Shigella* spp.**
Jul-09	1	N	N	N	N	N	N
	2	N	N	N	N	N	N
	3	N	N	N	N	N	N
	average	-	-	-	-	-	-
	SD	-	-	-	-	-	-
Aug-09	1	N	1610.00	N	N	N	2980.00
	2	N	1550.00	N	N	N	2970.00
	3	N	1600.00	N	N	N	2960.00
	average	-	1586.67	-	-	-	2970.00
	SD	-	32.15	-	-	-	10.00
Sep-09	1	N	147.00	281.00	N	71.00	45.00
	2	N	155.00	276.00	N	72.00	46.00
	3	N	152.00	275.00	N	70.00	48.00
	average	-	151.33	277.33	-	71.00	46.33
	SD	-	4.04	3.21	-	1.00	1.53
Oct-09	1	N	2020.00	5960.00	1500.00	600.00	5020.00
	2	N	1980.00	5920.00	1580.00	720.00	5120.00
	3	N	1860.00	5940.00	1620.00	N	5000.00
	average	-	1953.33	5940.00	1566.67	660.00	5046.67
	SD	-	83.27	20.00	61.10	84.85	64.29
Nov-09	1	N	2020.00	45.00	N	80.00	1460.00
	2	N	2180.00	40.00	N	76.00	1420.00
	3	N	2260.00	41.00	N	77.00	1400.00
	average	-	2153.33	42.00	-	77.67	1426.67
	SD	-	122.20	2.65	-	2.08	30.55
Dec-09	1	N	2200.00	90.00	N	91.00	2000.00
	2	N	2380.00	92.00	N	94.00	2100.00
	3	N	2180.00	90.00	N	94.00	2340.00
	average	-	2253.33	90.67	-	93.00	2146.67
	SD	-	110.15	1.15	-	1.73	174.74
Jan-10	1	N	3200.00	120.00	N	121.00	2700.00
	2	N	3240.00	125.00	N	120.00	2840.00
	3	N	3420.00	123.00	N	126.00	2760.00
	average	-	3286.67	122.67	-	122.33	2766.67
	SD	-	117.19	2.52	-	3.21	70.24
Feb-10	1	N	4020.00	159.00	N	157.00	4820.00
	2	N	4040.00	167.00	N	154.00	4840.00

Table B16/ Cont.

Sampling period	Replicates	Presumptive microbial pathogens (cfu/100 ml)					
		Campylobacter spp.	Coliforms	*E. coli*	*L. monocytogenes*	*Salmonella* spp.	*Shigella* spp.
	3	N	4040.00	168.00	N	155.00	4900.00
	average	-	4033.33	164.67	-	155.33	4853.33
	SD	-	11.55	4.93	-	1.53	41.63
Mar-10	1	N	5020.00	200.00	30.00	221.00	5740.00
	2	N	5060.00	221.00	33.00	220.00	5700.00
	3	N	5000.00	205.00	30.00	220.00	5600.00
	average	-	5026.67	208.67	31.00	220.33	5680.00
	SD	-	30.55	10.97	1.73	0.58	72.11
Apr-10	1	N	3020.00	187.00	30.00	201.00	4820.00
	2	N	3040.00	187.00	30.00	201.00	4860.00
	3	N	3100.00	181.00	31.00	208.00	4940.00
	average	-	3053.33	185.00	30.33	203.33	4873.33
	SD	-	41.63	3.46	0.58	4.04	61.10
May-10	1	N	2400.00	131.00	N	187.00	3740.00
	2	N	2360.00	131.00	N	186.00	3700.00
	3	N	2340.00	133.00	N	184.00	3660.00
	average	-	2366.67	131.67	-	185.67	3700.00
	SD	-	30.55	1.15	-	1.53	40.00
Jun-10	1	N	1860.00	94.00	N	247.00	900.00
	2	N	1840.00	93.00	N	249.00	760.00
	3	N	1860.00	98.00	N	251.00	720.00
	average	-	1853.33	95.00	-	249.00	793.33
	SD	-	11.55	2.65	-	2.00	94.52

N = microbial pathogen was not detected; SD = standard deviation

Table B17: The microbial analysis of irrigation water collected from farm B during a one-year period.

Sampling period	**Replicates**	**Presumptive microbial pathogens (cfu/100 ml)**					
		***Campylobacter* spp.**	**Coliforms**	***E. coli***	***L. monocytogenes***	***Salmonella* spp.**	***Shigella* spp.**
Jul-09	1	N	1040.00	N	N	3680.00	840.00
	2	N	1100.00	N	N	3720.00	900.00
	3	N	1120.00	N	N	3780.00	940.00
	average	-	1086.67	-	-	3726.67	893.33
	SD	-	41.63	-	-	50.33	50.33
Aug-09	1	N	5620.00	31.00	N	36.00	2560.00
	2	N	5820.00	30.00	N	33.00	2500.00
	3	N	5800.00	35.00	N	30.00	2600.00
	average	-	5746.67	32.00	-	33.00	2553.33
	SD	-	110.15	2.65	-	3.00	50.33
Sep-09	1	N	3520.00	N	N	N	2320.00
	2	N	3620.00	N	N	N	2380.00
	3	N	3600.00	N	N	N	2420.00
	average	-	3580.00	-	-	-	2373.33
	SD	-	52.92	-	-	-	50.33
Oct-09	1	N	4720.00	92.00	N	N	2080.00
	2	N	4600.00	98.00	N	N	2620.00
	3	N	4780.00	89.00	N	N	2660.00
	average	-	4700.00	93.00	-	-	2453.33
	SD	-	91.65	4.58	-	-	323.93
Nov-09	1	N	5200.00	143.00	N	67.00	4100.00
	2	N	5280.00	140.00	N	70.00	4200.00
	3	N	5360.00	141.00	N	73.00	4120.00
	average	-	5280.00	141.33	-	70.00	4140.00
	SD	-	80.00	1.53	-	3.00	52.92
Dec-09	1	N	5960.00	140.00	N	30.00	5860.00
	2	N	5740.00	141.00	N	31.00	5860.00
	3	N	5720.00	143.00	N	30.00	5960.00
	average	-	5806.67	141.33	-	30.33	5893.33
	SD	-	133.17	1.53	-	0.58	57.74
Jan-10	1	N	5940.00	161.00	N	35.00	6000.00
	2	N	5820.00	165.00	N	30.00	5940.00
	3	N	5900.00	171.00	N	32.00	5960.00
	average	-	5886.67	165.67	-	32.33	5966.67
	SD	-	61.10	5.03	-	2.52	30.55
Feb-10	1	N	7000.00	190.00	N	65.00	6200.00
	2	N	7400.00	193.00	N	60.00	6400.00

Table B17/ Cont.

Sampling period	Replicates	Presumptive microbial pathogens (cfu/100 ml)					
		Campylobacter spp.	Coliforms	*E. coli*	*L. monocytogenes*	*Salmonella* spp.	*Shigella* spp.
	3	N	7400.00	195.00	N	63.00	6000.00
	average	-	7266.67	192.67	-	62.67	6200.00
	SD	-	230.94	2.52	-	2.52	200.00
Mar-10	1	N	9000.00	201.00	101.00	75.00	14800.00
	2	N	9600.00	200.00	103.00	76.00	14600.00
	3	N	8600.00	206.00	103.00	78.00	14200.00
	average	-	9066.67	202.33	102.33	76.33	14533.33
	SD	-	503.32	3.21	1.15	1.53	305.51
Apr-10	1	N	6000.00	184.00	87.00	70.00	10200.00
	2	N	6200.00	185.00	88.00	71.00	10600.00
	3	N	6200.00	187.00	81.00	65.00	10800.00
	average	-	6133.33	185.33	85.33	68.67	10533.33
	SD	-	115.47	1.53	3.79	3.21	305.51
May-10	1	N	5960.00	97.00	35.00	105.00	4900.00
	2	N	5900.00	96.00	34.00	110.00	4940.00
	3	N	5900.00	94.00	31.00	113.00	4800.00
	average	-	5920.00	95.67	33.33	109.33	4880.00
	SD	-	34.64	1.53	2.08	4.04	72.11
Jun-10	1	N	4020.00	75.00	30.00	1900.00	4800.00
	2	N	4160.00	74.00	31.00	1860.00	4900.00
	3	N	4180.00	70.00	31.00	1860.00	4820.00
	average	-	4120.00	73.00	30.67	1873.33	4840.00
	SD	-	87.18	2.65	0.58	23.09	52.92

N = microbial pathogen was not detected; SD = standard deviation

Table B18: The microbial analysis of irrigation water collected from farm C during a one-year period.

Sampling period	**Replicates**	**Presumptive microbial pathogens (cfu/ 100 ml)**					
		***Campylobacter* spp.**	**Coliforms**	***E. coli***	***L. monocytogenes***	***Salmonella* spp.**	***Shigella* spp.**
Jul-09	1	N	249.00	N	35.00	N	860.00
	2	N	255.00	N	38.00	N	960.00
	3	N	253.00	N	40.00	N	840.00
	average	-	252.33	-	37.67	-	886.67
	SD	-	3.06	-	2.52	-	64.29
Aug-09	1	90.00	N	N	30.00	N	N
	2	91.00	N	N	31.00	N	N
	3	89.00	N	N	30.00	N	N
	average	90.00	-	-	30.33	-	-
	SD	1.00	-	-	0.58	-	-
Sep-09	1	N	700.00	N	N	N	N
	2	N	620.00	N	N	N	N
	3	N	600.00	N	N	N	N
	average	-	640.00	-	-	-	-
	SD	-	52.92	-	-	-	-
Oct-09	1	45.00	31.00	N	N	N	95.00
	2	49.00	35.00	N	N	N	91.00
	3	51.00	N	N	N	N	86.00
	average	48.33	33.00	-	-	-	90.67
	SD	3.06	2.83	-	-	-	4.51
Nov-09	1	60.00	N	N	N	N	116.00
	2	69.00	N	N	N	N	121.00
	3	62.00	N	N	N	N	118.00
	average	63.67	-	-	-	-	118.33
	SD	4.73	-	-	-	-	2.52
Dec-09	1	176.00	N	N	N	N	65.00
	2	178.00	N	N	N	N	69.00
	3	169.00	N	N	N	N	66.00
	average	174.33	-	-	-	-	66.67
	SD	4.73	-	-	-	-	2.08
Jan-10	1	70.00	59.00	N	30.00	N	105.00
	2	68.00	63.00	N	31.00	N	108.00
	3	63.00	65.00	N	33.00	N	100.00
	average	67.00	62.33	-	31.33	-	104.33
	SD	3.61	3.06	-	1.53	-	4.04
Feb-10	1	35.00	78.00	N	43.00	39.00	120.00
	2	33.00	84.00	N	44.00	38.00	121.00

Table B18/ Cont.

Sampling period	**Replicates**	**Presumptive microbial pathogens (cfu/100 ml)**					
		***Campylobacter* spp.**	**Coliforms**	***E. coli***	***L. monocytogenes***	***Salmonella* spp.**	***Shigella* spp.**
	3	32.00	82.00	N	46.00	34.00	118.00
	average	33.33	81.33	-	44.33	37.00	119.67
	SD	1.53	3.06	-	1.53	2.65	1.53
Mar-10	1	298.00	67.00	N	34.00	45.00	59.00
	2	298.00	69.00	N	32.00	49.00	60.00
	3	297.00	72.00	N	32.00	54.00	64.00
	average	297.67	69.33	-	32.67	49.33	61.00
	SD	0.58	2.52	-	1.15	4.51	2.65
Apr-10	1	1040.00	45.00	N	N	31.00	41.00
	2	1100.00	41.00	N	N	35.00	41.00
	3	1020.00	48.00	N	N	38.00	40.00
	average	1053.33	44.67	-	-	34.67	40.67
	SD	41.63	3.51	-	-	3.51	0.58
May-10	1	1940.00	31.00	N	N	N	N
	2	1920.00	33.00	N	N	N	N
	3	1880.00	33.00	N	N	N	N
	average	1913.33	32.33	-	-	-	-
	SD	30.55	1.15	-	-	-	-
Jun-10	1	2100.00	30.00	N	30.00	N	N
	2	2360.00	31.00	N	32.00	N	N
	3	2280.00	30.00	N	31.00	N	N
	average	2246.67	30.33	-	31.00	-	-
	SD	133.17	0.58	-	1.00	-	-

N = microbial pathogen was not detected; SD = standard deviation

Table B19: Counts obtained for presumptive *Campylobacter* spp. in different fresh produce collected from farm A1, over a one-year period.

Sampling period	Samples	*Campylobacter* spp. counts (cfu/g)				
		A	B	C	Average	SD
Jul-09	Broccoli soil	1420000	1350000	1460000	1410000.00	55677.64
	Broccoli root	300000	320000	310000	310000.00	10000.00
	Broccoli stem	450000	460000	500000	470000.00	26457.51
	Broccoli leaf	550000	580000	540000	556666.67	20816.66
	Broccoli	28900 37000	28800 32000	29300 35000	31833.33	3492.09
	Cabbage soil	630000	620000	580000	610000.00	26457.51
	Cabbage root	1380000	1400000	1370000	1383333.33	15275.25
	Cabbage	25000 39000	24700 38000	24500 35000	31033.33	7027.56
	Crisphead lettuce soil	380000	330000	350000	353333.33	25166.11
	Crisphead lettuce root	320000	310000	300000	310000.00	10000.00
	Crisphead lettuce	470000	450000	450000	456666.67	11547.01
	Jam tomato soil	2790000	2760000	2720000	2756666.67	35118.85
	Jam tomato root	244000	249000	245000	246000.00	2645.75
	Jam tomato stem	220000	239000	225000	228000.00	9848.86
	Jam tomato leaf	460000	460000	430000	450000.00	17320.51
	Jam tomato	15500	15600	16000	15700.00	264.58
Aug-09	Broccoli soil	450000	420000	450000	426666.67	20816.66
	Broccoli root	1150000	1120000	1100000	1123333.33	25166.11
	Broccoli stem	10000	11500	10100	10533.33	838.65
	Broccoli leaf	5000	4400	5000	4800.00	346.41
	Broccoli	6500	6900	6400	6600.00	264.58
	Cabbage soil	330000	320000	310000	320000.00	10000.00
	Cabbage root	310000	330000	390000	343333.33	41633.32
	Cabbage	69000	79000	63000	70333.33	8082.90
	Crisphead lettuce soil	430000	420000	400000	416666.67	15275.25
	Crisphead lettuce root	990000	960000	1000000	983333.33	20816.66
	Crisphead lettuce	18300	18000	18000	18100.00	173.21
	Jam tomato soil	110000	113000	110000	111000.00	1732.05
	Jam tomato root	1850000	1780000	1800000	1810000.00	36055.51
	Jam tomato stem	112000	111000	110000	111000.00	1000.00
	Jam tomato leaf	300000	310000	300000	303333.33	5773.50
	Jam tomato	8100	7500	8000	7866.67	321.46
Sep-09	Cabbage soil	34000	31000	35000	33333.33	2081.67
	Cabbage root	223000	217000	220000	220000.00	3000.00
	Cabbage	85000	78000	81000	81333.33	3511.88
	Crisphead lettuce soil	450000	490000	410000	450000.00	40000.00
	Crisphead lettuce root	340000	370000	330000	346666.67	20816.66
	Crisphead lettuce	40000	40000	40000	42000.00	3464.10
	Jam tomato soil	263000	257000	260000	260000.00	3000.00
	Jam tomato root	266000	273000	271000	270000.00	3605.55
	Jam tomato stem	38000	54000	39000	43666.67	8962.89
	Jam tomato leaf	25600 31000	25200	25100	26725.00	2858.18
	Jam tomato	40000 29800	31000	38000	34700.00	5055.69
Oct-09	Broccoli soil	205000	206000	200000	203666.67	3214.55
	Broccoli root	65000	64000	60000	63000.00	2645.75
	Broccoli leaf	25500	24600	25000	25033.33	450.92
	Broccoli	10500	11900	10000	10800.00	984.89
	Cabbage soil	1940000	1800000	1910000	1883333.33	73711.15

Table B19/ Cont.

Sampling period	Samples	*Campylobacter* spp. counts (cfu/g)				
		A	B	C	Average	SD
	Cabbage root	2540000	2500000	2510000	2516666.67	20816.66
	Cabbage	200000	209000	205000	204666.67	4509.25
	Crisphead lettuce soil	21100	21000	20100	20733.33	550.76
	Crisphead lettuce root	450000	420000	410000	426666.67	20816.66
	Crisphead lettuce	9800	11200	10000	10333.33	757.19
	Spinach soil	2930000	2900000	2970000	2933333.33	35118.85
	Spinach root	2760000	2700000	2810000	2756666.67	55075.71
	Spinach stem	91000	93000	90000	91333.33	1527.53
	Spinach	1100000	1090000	1060000	1083333.33	20816.66
Nov-09	Spinach soil	76000	65000	70000	70333.33	5507.57
	Spinach root	1060000	1210000	1050000	1106666.67	89628.86
	Spinach stem	610000	630000	710000	650000.00	52915.03
	Spinach	7900	8000	8300	8066.67	208.17
Dec-09	Spinach soil	96000	93000	90000	93000.00	3000.00
	Spinach root	1230000	1210000	1100000	1180000.00	70000.00
	Spinach stem	970000	940000	960000	956666.67	15275.25
	Spinach	330000	360000	310000	333333.33	25166.11
Jan-10	Cabbage soil	198000	198000	201000	199000.00	1732.05
	Cabbage root	271000	268000	276000	271666.67	4041.45
	Cabbage	38000	36000	31000	35000.00	3605.55
	Crisphead lettuce soil	296000	310000	293000	299250.00	7455.42
	Crisphead lettuce root	29800	29800	29700	29766.67	57.74
	Crisphead lettuce	21100	21300	20000	20800.00	700.00
	Spinach soil	83000	84000	80000	82333.33	2081.67
	Spinach root	490000	470000	460000	473333.33	15275.25
	Spinach stem	350000	390000	350000	363333.33	23094.01
	Spinach	20700	20000	20600	20433.33	378.59
Feb-10	Crisphead lettuce soil	240000	241000	245000	242000.00	2645.75
	Crisphead lettuce root	310000	300000	300000	303333.33	5773.50
	Crisphead lettuce	30000	31000	35000	32000.00	2645.75
	Spinach soil	142000	145000	141000	142666.67	2081.67
	Spinach root	1150000	1130000	1150000	1143333.33	11547.01
	Spinach stem	320000	340000	310000	323333.33	15275.25
	Spinach	9000	9300	9100	9133.33	152.75
Mar-10	Cabbage soil	185000	186000	187000	186000.00	1000.00
	Cabbage root	265000	265000	264000	264666.67	577.35
	Cabbage	49000	50000	46000	48333.33	2081.67
	Crisphead lettuce soil	244000	250000	243000	245666.67	3785.94
	Crisphead lettuce root	420000	420000	410000	416666.67	5773.50
	Crisphead lettuce	32000	35000	33000	33333.33	1527.53
	Spinach soil	260000	254000	256000	256666.67	3055.05
	Spinach root	1360000	1350000	1320000	1343333.33	20816.66
	Spinach stem	274000	279000	276000	276333.33	2516.61
	Spinach	14600	14800	14300	14566.67	251.66
Apr-10	Crisphead lettuce soil	310000	310000	300000	306666.67	5773.50
	Crisphead lettuce root	31000	30000	30000	30333.33	577.35
	Crisphead lettuce	21500	21600	21500	21533.33	57.74
	Jam tomato soil	283000 284000	300000	287000	288500.00	7852.81
	Jam tomato root	310000	300000	300000	303333.33	5773.50
	Jam tomato stem	29800	29400	29800	29666.67	230.94
	Jam tomato leaf	28500	28200	28800	28500.00	300.00
	Jam tomato	45000	41000	45000	43666.67	2309.40
	Spinach soil	122000	124000	124000	123333.33	1154.70

Table B19/ Cont.

Sampling period	**Samples**	***Campylobacter* spp. counts (cfu/g)**				
		A	**B**	**C**	**Average**	**SD**
	Spinach root	890000	850000	900000	880000.00	26457.51
	Spinach stem	284000	281000	285000	283333.33	2081.67
	Spinach	7600	7700	7900	7733.33	152.75
May-10	Crisphead lettuce soil	300000	310000	300000	303333.33	5773.50
	Crisphead lettuce root	26800	27600	26900	27100.00	435.89
	Crisphead lettuce	19700	19500	19800	19666.67	152.75
	Jam tomato soil	276000	267000	275000	272666.67	4932.88
	Jam tomato root	294000	291000	297000	294000.00	3000.00
	Jam tomato stem	33000	34000	30000	32333.33	2081.67
	Jam tomato leaf	34000	33000	35000	34000.00	1000.00
	Jam tomato	28800	28700	28700	28733.33	57.74
	Spinach soil	243000	244000	244000	243666.67	577.35
	Spinach root	1200000	1240000	1210000	1216666.67	20816.66
	Spinach stem	275000	278000	275000	276000.00	1732.05
	Spinach	12800	12800	12000	12533.33	461.88
Jun-10	Crisphead lettuce soil	680000	610000	630000	640000	36055.51
	Crisphead lettuce root	32000	37000	38000	35666.67	3214.55
	Crisphead lettuce	27300	27400	27900	27533.33	321.46
	Spinach soil	283000	284000	284000	283666.67	577.35
	Spinach root	1580000	1580000	1580000	158000	0.00
	Spinach stem	540000	590000	580000	570000	26457.51
	Spinach	35000	38000	37000	36666.67	1527.53

SD = standard deviation; cfu/g = colony forming units per gram of sample

Table B20: Counts obtained for presumptive coliforms in different fresh produce collected from farm A1, over a one-year period.

Sampling period	Samples	Coliforms (cfu/g)				
		A	B	C	Average	SD
Jul-09	Broccoli soil	6000	4900	5600	5500.00	556.78
	Broccoli root	3500	3200	3000	3233.33	251.66
	Broccoli	5400	4800	5100	5100.00	300.00
	Cabbage soil	3800	4300	4100	4066.67	251.66
	Cabbage root	5400	4900	5000	5100.00	264.58
	Cabbage	3000	3100	3500	3200.00	264.58
	Crisphead lettuce soil	6400	6900	7000	6766.67	321.46
	Crisphead lettuce root	6100	6200	5800	6033.33	208.17
	Crisphead lettuce	6800	6500	6400	6566.67	208.17
	Jam tomato soil	5400	5300	5400	5366.67	57.74
Aug-09	Broccoli soil	120000	132000	121000	124333.33	6658.33
	Broccoli root	740000	690000	710000	713333.33	25166.11
	Broccoli stem	9800	8700	8100	8866.67	862.17
	Broccoli leaf	4900	3800	4200	4300.00	556.78
	Broccoli	13300	13000	12900	13066.67	208.17
	Cabbage root	6700	7000	7200	6966.67	251.66
	Cabbage	6200	5800	5700	5900.00	264.58
	Crisphead lettuce soil	600000	590000	600000	596666.67	5773.50
	Crisphead lettuce root	590000	600000	610000	600000.00	10000.00
	Crisphead lettuce	4200	4500	3900	4200.00	300.00
	Jam tomato soil	145000	148000	151000	148000.00	3000.00
	Jam tomato root	1060000	1010000	1070000	1046666.67	32145.50
	Jam tomato stem	197000	190000	193000	193333.33	3511.88
	Jam tomato leaf	150000	152000	149000	150333.33	1527.53
Sep-09	Cabbage soil	56000	56000	60000	57333.33	2309.40
	Cabbage root	19900	18600	19500	19333.33	665.83
	Cabbage	90000	92000	93000	91666.67	1527.53
	Crisphead lettuce soil	54000	53000	53000	53333.33	577.35
	Crisphead lettuce root	88000	81000	86000	85000.00	3605.55
	Crisphead lettuce	74000	66000	67000	69000.00	4358.90
	Jam tomato soil	950000	970000	1000000	973333.33	25166.11
	Jam tomato root	1980000	1970000	1850000	1933333.33	72341.78
	Jam tomato stem	80000	83000	91000	84666.67	5686.24
	Jam tomato leaf	195000	203000	208000	202000.00	6557.44
	Jam tomato	21900	22600	20900	21800.00	854.40
Oct-09	Broccoli soil	400000	430000	410000	413333.33	15275.25
	Broccoli root	160000	154000	161000	158333.33	3785.94
	Broccoli leaf	16600	16900	17200	16900.00	300.00
	Broccoli	20100	20400	19600	20033.33	404.15
	Cabbage root	48000	41000	45000	44666.67	3511.88
	Cabbage	3000	3100	3300	3133.33	152.75
	Crisphead lettuce soil	7900	8200	7800	7966.67	208.17
	Crisphead lettuce root	930000	950000	900000	926666.67	25166.11
	Crisphead lettuce	24000	23900	22900	23600.00	608.28
	Spinach soil	80000	83000	78000	80333.33	2516.61
	Spinach root	1450000	1510000	1450000	1490000.00	34641.02
	Spinach stem	34000 35000	29800	31000	32450.00	2451.53
	Spinach	910000	900000	930000	913333.33	15275.25
Nov-09	Spinach soil	13100	14500	13600	13733.33	709.46
	Spinach root	9000	8900	8700	8866.67	152.75
	Spinach stem	120000	139000	116000	125000.00	12288.21
	Spinach	5000	5100	4800	4966.67	152.75

Table B20/ Cont.

Sampling period	Samples	Coliforms (cfu/g)				
		A	B	C	Average	SD
Dec-09	Spinach root	8000	8100	8600	8233.33	321.46
	Spinach stem	100000	105000	111000	105333.33	5507.57
	Spinach	193000	187000	184000	188000.00	4582.58
Jan-10	Cabbage soil	30000	36000	33000	33000.00	3000.00
	Cabbage root	90000	87000	88000	88333.33	1527.53
	Cabbage	20300	20400	20900	20533.33	321.46
	Crisphead lettuce soil	30000	31000	31000	30666.67	577.35
	Crisphead lettuce root	29800 29700	29700	31000	30050.00	635.09
	Crisphead lettuce	198000	209000	220000	209000.00	11000.00
	Spinach root	3100	3000	3600	3233.33	321.46
	Spinach stem	19000	19100	18400	18833.33	378.59
	Spinach	450000	490000	500000	480000.00	26457.51
Feb-10	Crisphead lettuce soil	105000	110000	106000	107000.00	2645.75
	Crisphead lettuce root	135000	133000	132000	133333.33	1527.53
	Crisphead lettuce	89000	91000	90000	90000.00	1000.00
	Spinach soil	15400	15500	15100	15333.33	208.17
	Spinach root	11000	11000	11500	11166.67	288.68
	Spinach stem	145000	155000	150000	150000.00	5000.00
	Spinach	7100	7000	7000	7033.33	57.74
Mar-10	Cabbage soil	45000	48000	47000	46666.67	1527.53
	Cabbage root	118000	116000	117000	117000.00	1000.00
	Cabbage	26500	26800	27600	26966.67	568.62
	Crisphead lettuce soil	12300	12400	12000	12233.33	208.17
	Crisphead lettuce root	1560000	1550000	1560000	1556666.67	5773.50
	Crisphead lettuce	167000	166000	169000	167333.33	1527.53
	Spinach soil	20100	20000	20300	20133.33	152.75
	Spinach root	14300	14200	14700	14400.00	264.58
	Spinach stem	110000	113000	116000	113000.00	3000.00
	Spinach	15700	15500	15400	15533.33	152.75
Apr-10	Crisphead lettuce soil	30000	30000	31000	30333.33	577.35
	Crisphead lettuce root	28700	28400	28400	28500.00	173.21
	Crisphead lettuce	174000	176000	175000	175000.00	1000.00
	Jam tomato soil	1240000	1230000	1210000	1226666.67	15275.25
	Jam tomato root	2050000	2070000	2110000	2076666.67	30550.50
	Jam tomato stem	84000	83000	82000	83000.00	1000.00
	Jam tomato leaf	245000	247000	248000	246666.67	1527.53
	Jam tomato	25100	25600	25500	25400.00	264.58
	Spinach soil	13100	13600	13300	13333.33	251.66
	Spinach root	9400	9500	9500	9466.67	57.74
	Spinach stem	137000	138000	138000	137666.67	577.35
	Spinach	5900	6000	6000	5966.67	57.74
May-10	Crisphead lettuce soil	27600	27500	27700	27600.00	100.00
	Crisphead lettuce root	25100	25200	25300	25200.00	100.00
	Crisphead lettuce	121000	123000	127000	123666.67	3055.05
	Jam tomato soil	131000	138000	139000	136000.00	4358.90
	Jam tomato root	300000	350000	310000	320000.00	26457.51
	Jam tomato stem	30000	37000	35000	34000.00	3605.55
	Jam tomato leaf	151000	148000	147000	148666.67	2081.67
	Jam tomato	12800	12600	12800	12733.33	115.47
	Spinach soil	18000	18400	18300	18233.33	208.17
	Spinach root	10200	10800	10500	10500.00	300.00
	Spinach stem	68000	68000	68000	68000.00	0.00
	Spinach	11800	11900	11900	11866.67	57.74

Table B20/ Cont.

Sampling period	**Samples**	**Coliforms (cfu/g)**				
		A	**B**	**C**	**Average**	**SD**
Jun-10	Crisphead lettuce soil	25300	25500	25500	25433.33	115.47
	Crisphead lettuce root	18400	18200	18300	18300.00	100.00
	Crisphead lettuce	10200	9500	9300	9666.67	472.58
	Spinach stem	15200	15100	15000	15100.00	100.00
	Spinach	236000	237000	235000	236000.00	1000.00

SD = standard deviation; cfu/g = colony forming units per gram of sample

Table B21: Counts obtained for presumptive *E. coli* in different fresh produce collected from farm A1, over a one-year period.

Sampling period	Samples	*E. coli* counts (cfu/g)				
		A	B	C	Average	SD
Jul-09	Cabbage soil	5400	5400	5100	5300.00	173.21
	Cabbage	3000	3100	3500	3200.00	264.58
	Crisphead lettuce soil	4500	4600	4200	4433.33	208.17
	Crisphead lettuce root	3200	3500	3000	3233.33	251.66
	Crisphead lettuce	5400	4700	4800	4966.67	378.59
	Jam tomato soil	3400	3300	3700	3466.67	208.17
Sep-09	Cabbage soil	8000	8300	7800	8033.33	251.66
	Jam tomato soil	4500	4600	5100	4733.33	321.46
	Jam tomato leaf	211000	223000	217000	217000.00	6000.00
Oct-09	Crisphead lettuce root	10000	9800	9600	9800.00	200.00
Dec-09	Spinach stem	3400	3700	4200	3766.67	404.15
	Spinach	20100	21100	20500	20566.67	503.32
Jan-10	Cabbage soil	36000	32000	32000	33333.33	2309.40
	Cabbage root	8900	9100	8600	8866.67	251.66
	Crisphead lettuce root	205000	209000	215000	209666.67	5033.22
	Crisphead lettuce	300000	310000	350000	320000.00	26457.51
	Spinach stem	9000	9900	9300	9400.00	458.26
	Spinach	40000	41000	46000	42333.33	3214.55
Mar-10	Cabbage soil	32000	33000	33000	32666.67	577.35
	Cabbage root	10200	10600	10600	10466.67	230.94
	Crisphead lettuce root	254000	255000	258000	255666.67	2081.67
	Crisphead lettuce	267000	266000	263000	265333.33	2081.67
	Spinach	98000	97000	95000	96666.67	1527.53
Apr-10	Crisphead lettuce root	184000	181000	182000	182333.33	1527.53
	Crisphead lettuce	29400 29100	30000	29100	29400.00	424.26
	Jam tomato soil	5700	5600	5600	5633.33	57.74
	Jam tomato leaf	259000	261000	263000	261000.00	2000.00
May-10	Crisphead lettuce root	160000	162000	161000	161000.00	1000.00
	Crisphead lettuce	256000	291000	291000	279333.33	20207.26
	Jam tomato soil	6000	6300	6400	6233.33	208.17
	Jam tomato leaf	271000	272000	273000	272000.00	1000.00
	Spinach	102000	105000	103000	103333.33	1527.53
Jun-10	Crisphead lettuce root	136000	131000	139000	135333.33	4041.45
	Crisphead lettuce	231000	236000	236000	234333.33	2886.75
	Spinach stem	14500	15800	15200	15166.67	650.64
	Spinach	54000	57000	58000	56333.33	2081.67

SD = standard deviation; cfu/g = colony forming units per gram of sample

Table B22: Counts obtained for presumptive *L. monocytogenes* in different fresh produce collected from farm A1, over a one-year period.

Sampling period	**Samples**	***L. monocytogenes* counts (cfu/g)**				
		A	**B**	**C**	**Average**	**SD**
Jul-09	Cabbage soil	5500	5100	5300	5300.00	200.00
	Crisphead lettuce soil	6000	5400	5900	5766.67	321.46
	Crisphead lettuce	4200	3700	4200	4033.33	288.68
Aug-09	Broccoli soil	3000	3000	3000	3000.00	0.00
	Broccoli root	3400	3300	3300	3333.33	57.74
Dec-09	Spinach	3000	3400		3200.00	
Jan-10	Crisphead lettuce soil	4000	4900	5300	4733.33	665.83
	Crisphead lettuce root	9700	9600	9300	9533.33	208.17
	Crisphead lettuce	11100	11500	11200	11266.67	208.17
	Spinach	6900	6500	7200	6866.67	351.19
Apr-10	Crisphead lettuce soil	3000	3200	3000	3066.67	115.47
	Crisphead lettuce root	3500	3500	3400	3466.67	57.74
	Crisphead lettuce	8700	8600	8500	8600.00	100.00
	Jam tomato soil	3000	3200	3200	3133.33	115.47
	Jam tomato root	3100	3500	3300	3300.00	200.00
May-10	Crisphead lettuce	6700	6800	6800	6766.67	57.74
	Jam tomato soil	3000	3000	3000	3000.00	0.00
Jun-10	Crisphead lettuce root	5400	5900	5300	5533.33	321.46
	Spinach	5100	5400	5500	5333.33	208.17

SD = standard deviation; cfu/g = colony forming units per gram of sample

Table B23: Counts obtained for presumptive *Salmonella* spp. in different fresh produce collected from farm A1, over a one-year period.

Sampling period	**Samples**	***Salmonella* spp. counts (cfu/g)**				
		A	**B**	**C**	**Average**	**SD**
Jul-09	Broccoli soil	3100	3000	3300	3133.33	152.75
	Cabbage root	31000	33000	35000	33000.00	2000.00
	Crisphead lettuce root	6000	6500	6300	6266.67	251.66
	Jam tomato soil	4300	4200	4600	4366.67	208.17
Sep-09	Cabbage soil	4000	4600	4500	4366.67	321.46
Oct-09	Spinach root	9000	8600	8100	8566.67	450.92
Nov-09	Spinach stem	9600	9700	9100	9466.67	321.46
Dec-09	Spinach	3000	3400	3300	3233.33	208.17
Jan-10	Cabbage soil	10100	11200	10900	10733.33	568.62
	Cabbage root	3000	3500	3200	3233.33	251.66
	Crisphead lettuce root	3100	3200	3900	3400.00	435.89
	Crisphead lettuce	6500	6900	6900	6766.67	230.94
	Spinach	36000	36000	35000	35666.67	577.35
Feb-10	Crisphead lettuce soil	3000	3200	3200	3133.33	115.47
	Spinach	15000	15100	15700	15266.67	378.59
Mar-10	Cabbage soil	13200	13300	13100	13200.00	100.00
	Cabbage root	5500	5100	5200	5266.67	208.17
	Cabbage	3200	3300	3300	3266.67	57.74
	Crisphead lettuce root	6700	6600	6900	6733.33	152.75
	Crisphead lettuce	3300	3500	3800	3533.33	251.66
	Spinach	22900	22100	22500	22500.00	400.00
Apr-10	Crisphead lettuce	4500	4700	4800	4666.67	152.75
	Spinach	13500	13200	13600	13433.33	208.17
May-10	Crisphead lettuce	4900	5000	5300	5066.67	208.17
	Jam tomato soil	3000	3400	3500	3300.00	264.58
	Spinach	25000	25600	26100	25566.67	550.76
Jun-10	Crisphead lettuce root	6200	6300	6200	6233.33	57.74
	Crisphead lettuce	6900	6700	6400	6666.67	251.66
	Spinach	80000	83000	85000	82666.67	2516.61

SD = standard deviation; cfu/g = colony forming units per gram of sample

Table B24: Counts obtained for presumptive *Shigella* spp. in different fresh produce collected from farm A1, over a one-year period.

Sampling period	Samples	*Shigella* spp. counts (cfu/g)				
		A	B	C	Average	SD
Jul-09	Broccoli soil	88000	85000	90000	87666.67	2516.61
	Broccoli root	32000	37000	31000	33333.33	3214.55
	Broccoli stem	1580000	1550000	1540000	1556666.67	20816.66
	Broccoli leaf	24600 30000	24900	24300	25950.00	2711.09
	Broccoli	2690000	2700000	2730000	2706666.67	20816.66
	Cabbage soil	900000	880000	900000	893333.33	11547.01
	Cabbage root	330000	370000	330000	343333.33	23094.01
	Cabbage	420000	450000	410000	426666.67	20816.66
	Crisphead lettuce soil	145000	148000	154000	149000.00	4582.58
	Crisphead lettuce root	6000	6500	6200	6233.33	251.66
	Crisphead lettuce	390000	370000	360000	373333.33	15275.25
	Jam tomato soil	650000	630000	630000	636666.67	11547.01
	Jam tomato root	1360000	1330000	1350000	1346666.67	15275.25
	Jam tomato stem	1670000	1640000	1610000	1640000.00	30000.00
	Jam tomato leaf	6800	6900	6200	6633.33	378.59
	Jam tomato	104000	99000	102000	101666.67	2516.61
Aug-09	Broccoli soil	24000 30000	24100 33000	24500	27120.00	4140.89
	Broccoli root	241000	242000	245000	242666.67	2081.67
	Broccoli leaf	31000	32000	38000	33666.67	3785.94
	Broccoli	30000	31000	32000	31000.00	1000.00
	Cabbage soil	60000	61000	63000	61333.33	1527.53
	Cabbage root	250000	245000	251000	248666.67	3214.55
	Cabbage	3000	3000	3500	3166.67	288.68
	Crisphead lettuce soil	17000	18900	18100	18000.00	953.94
	Crisphead lettuce root	20100	20000	19800	19966.67	152.75
	Crisphead lettuce	6000	6200	6100	6100.00	100.00
	Jam tomato soil	40000	39000	38000	39000.00	1000.00
	Jam tomato root	81000	82000	85000	82666.67	2081.67
	Jam tomato stem	3500	3900	3100	3500.00	400.00
	Jam tomato leaf	4200	4800	4700	4566.67	321.46
	Jam tomato	3000	3100	3100	3066.67	57.74
Sep-09	Cabbage soil	40000	41000	41000	40666.67	577.35
	Cabbage root	310000	330000	340000	326666.67	15275.25
	Cabbage	300000	330000	320000	316666.67	15275.25
	Crisphead lettuce soil	19800	19600	19500	19633.33	152.75
	Crisphead lettuce root	25100	25300	26400	25600.00	700.00
	Crisphead lettuce	7600	7600	7300	7500.00	173.21
	Jam tomato soil	130000	132000	141000	134333.33	5859.47
	Jam tomato root	29800 30000	29500	29100	29550.00	391.58
	Jam tomato stem	29600	28500	28100	28733.33	776.75
	Jam tomato leaf	30000	31000	31000	30666.67	577.35
Oct-09	Broccoli soil	410000	490000	460000	453333.33	40414.52
	Broccoli root	59000	51000	52000	54000.00	4358.90
	Cabbage soil	22000	21200	21500	21566.67	404.15
	Cabbage	4700	4500	3900	4366.67	416.33
	Crisphead lettuce root	80000	89000	71000	80000.00	9000.00
	Spinach soil	13000	12800	12700	12833.33	152.75
	Spinach root	400000	360000	340000	366666.67	30550.50
	Spinach stem	3300	3100	3000	3133.33	152.75

Table B24/ Cont.

Sampling period	Samples	*Shigella* spp. counts (cfu/g)				
		A	**B**	**C**	**Average**	**SD**
	Spinach	64000	61000	60000	61666.67	2081.67
Nov-09	Spinach	4100	3000	3600	3566.67	550.76
Dec-09	Spinach	8300	9200	9300	8933.33	550.76
Jan-10	Cabbage soil	40000	46000	37000	41000.00	4582.58
	Cabbage root	35000	39000	38000	37333.33	2081.67
	Cabbage	4500	4500	4900	4633.33	230.94
	Crisphead lettuce root	20000	20500	19900	20133.33	321.46
	Crisphead lettuce	860000	840000	900000	866666.67	30550.50
	Spinach soil	15000	15100	15600	15233.33	321.46
	Spinach root	310000	350000	350000	336666.67	23094.01
	Spinach	209000	212000	211000	210666.67	1527.53
Feb-10	Crisphead lettuce soil	30000	35000	34000	33000.00	2645.75
	Crisphead lettuce root	40000	45000	41000	42000.00	2645.75
	Crisphead lettuce	29800	29500	29900	29733.33	208.17
	Spinach	8400	8500	8500	8466.67	57.74
Mar-10	Cabbage soil	60000	61000	63000	61333.33	1527.53
	Cabbage root	56000	51000	52000	53000.00	2645.75
	Cabbage	7100	7500	7500	7366.67	230.94
	Crisphead lettuce soil	6100	6200	6800	6366.67	378.59
	Crisphead lettuce root	92000	90000	91000	91000.00	1000.00
	Crisphead lettuce	9800	9200	9600	9533.33	305.51
	Spinach	25100	25400	25700	25400.00	300.00
Apr-10	Crisphead lettuce root	18300	18200	18600	18366.67	208.17
	Crisphead lettuce	650000	680000	680000	670000.00	17320.51
	Jam tomato soil	174000	173000	171000	172666.67	1527.53
	Jam tomato root	31000	32000	32000	31666.67	577.35
	Jam tomato stem	28400	28500	28100	28333.33	208.17
	Jam tomato leaf	65000	65000	65000	65000.00	0.00
	Spinach	7000	7100	7400	7166.67	208.17
May-10	Crisphead lettuce root	16000	16200	16500	16233.33	251.66
	Crisphead lettuce	530000	530000	540000	533333.33	5773.50
	Jam tomato soil	170000	172000	171000	171000.00	1000.00
	Jam tomato root	30000	33000	34000	32333.33	2081.67
	Jam tomato stem	27600	27600	27600	27600.00	0.00
	Jam tomato leaf	59000	60000	63000	60666.67	2081.67
	Spinach	22300	22700	22600	22533.33	208.17
Jun-10	Crisphead lettuce root	13000	13300	13500	13266.67	251.66
	Crisphead lettuce	298000	294000	293000	295000.00	2645.75
	Spinach soil	11000	11100	11800	11300.00	435.89
	Spinach root	201000	204000	193000	199333.33	5686.24
	Spinach	132000	137000	137000	135333.33	2886.75

SD = standard deviation; cfu/g = colony forming units per gram of sample

Table B25: Counts obtained for presumptive *Campylobacter* spp. in different fresh produce collected from farm A2, over a one-year period.

Sampling period	Samples	***Campylobacter* spp. counts (cfu/g)**				
		A	**B**	**C**	**Average**	**SD**
Jul-09	Broccoli soil	275000	262000	271000	269333.33	6658.33
	Broccoli root	264000	264000	270000	266000.00	3464.10
	Broccoli stem	294000	287000	290000	290333.33	3511.88
	Broccoli leaf	340000	360000	370000	356666.67	15275.25
	Cabbage soil	200000	208000	204000	204000.00	4000.00
	Cabbage root	1000000	980000	950000	976666.67	25166.11
	Cabbage	15200	16000	15600	15600.00	400.00
	Crisphead lettuce soil	300000	350000	320000	323333.33	25166.11
	Crisphead lettuce root	209000	210000	206000	208333.33	2081.67
	Crisphead lettuce	680000	650000	710000	680000.00	30000.00
	Spinach soil	44000	47000	43000	44666.67	2081.67
	Spinach root	1520000	1540000	1530000	1530000.00	10000.00
	Spinach stem	2790000	2800000	2840000	2810000.00	26457.51
	Spinach	2530000	2590000	2530000	2550000.00	34641.02
Aug-09	Broccoli soil	65000	78000	72000	71666.67	6506.41
	Broccoli root	440000	410000	380000	410000.00	30000.00
	Broccoli stem	8000	8900	9300	8733.33	665.83
	Broccoli leaf	70000	70000	61000	67000.00	5196.15
	Broccoli	60000	60000	57000	59000.00	1732.05
	Cabbage soil	1920000	1930000	1850000	1900000.00	43588.99
	Cabbage root	210000	221000	223000	218000.00	7000.00
	Cabbage	64000	66000	63000	64333.33	1527.53
	Crisphead lettuce soil	350000	380000	450000	393333.33	51316.01
	Crisphead lettuce root	1200000	1230000	1220000	1216666.67	15275.25
	Crisphead lettuce	70000	70000	73000	71000.00	1732.05
	Cauliflower soil	620000	610000	600000	610000.00	10000.00
	Cauliflower root	350000	390000	370000	370000.00	20000.00
	Cauliflower leaf	610000	630000	640000	626666.67	15275.25
	Cauliflower	90000	91000	97000	92666.67	3785.94
Sep-09	Broccoli soil	64000	69000	70000	67666.67	3214.55
	Broccoli root	1160000	1190000	1230000	1193333.33	35118.85
	Broccoli stem	20100	21200	20300	20533.33	585.95
	Broccoli leaf	251000	252000	257000	253333.33	3214.55
	Broccoli	21100	22600	21900	21866.67	750.56
	Cabbage soil	176000	181000	173000	176666.67	4041.45
	Cabbage root	1100000	1310000	1360000	1256666.67	137961.35
	Cabbage	1810000	1780000	1800000	1796666.67	15275.25
	Crisphead lettuce soil	92000	95000	105000	97333.33	6806.86
	Crisphead lettuce root	2010000	2150000	2320000	2160000.00	155241.75
	Crisphead lettuce	70000	76000	69000	71666.67	3785.94
Oct-09	Cabbage soil	70000	72000	76000	72666.67	3055.05
	Cabbage root	150000	151000	154000	151666.67	2081.67
	Cabbage	80000	86000	81000	82333.33	3214.55
	Crisphead lettuce soil	11000	10900	10700	10866.67	152.75
	Crisphead lettuce root	103000	110000	102000	105000.00	4358.90
	Crisphead lettuce	700000	690000	680000	690000.00	10000.00
	Spinach soil	45000	46000	49000	46666.67	2081.67
	Spinach root	150000	151000	156000	152333.33	3214.55
	Spinach stem	31000	33000	30000	31333.33	1527.53
	Spinach	12000	12300	12000	12100.00	173.21
Nov-09	Crisphead lettuce soil	760000	690000	660000	703333.33	51316.01
	Crisphead lettuce root	400000	420000	430000	416666.67	15275.25

Table B25/ Cont.

Sampling period	Samples	*Campylobacter* spp. counts (cfu/g)				
		A	B	C	Average	SD
	Crisphead lettuce	4000	4000	4200	4066.67	115.47
Dec-09	Crisphead lettuce soil	850000	890000	870000	870000.00	20000.00
	Crisphead lettuce root	400000	420000	430000	416666.67	15275.25
	Crisphead lettuce	45000	48000	50000	47666.67	2516.61
Jan-10	Broccoli soil	90000	91000	93000	91333.33	1527.53
	Broccoli root	900000	880000	870000	883333.33	15275.25
	Broccoli stem	19800	20000	20500	20100.00	360.56
	Broccoli leaf	251000	253000	256000	253333.33	2516.61
	Broccoli	19900	19700	19600	19733.33	152.75
	Crisphead lettuce soil	800000	850000	830000	826666.67	25166.11
	Crisphead lettuce root	610000	630000	690000	643333.33	41633.32
	Crisphead lettuce	29800 31000	28900 30000	27900	29520.00	1173.46
Apr-10	Crisphead lettuce soil	870000	880000	850000	866666.67	15275.25
	Crisphead lettuce root	470000	480000	470000	473333.33	5773.50
	Crisphead lettuce	4800	4900	5000	4900.00	100.00
May-10	Cabbage soil	111000	119000	116000	115333.33	4041.45
	Cabbage root	310000	300000	300000	303333.33	5773.50
	Cabbage	11700	11800	11000	11500.00	435.89
	Crisphead lettuce soil	930000	980000	980000	963333.33	28867.51
	Crisphead lettuce root	850000	880000	870000	866666.67	15275.25
	Crisphead lettuce	38000	37000	40000	38333.33	1527.53
Jun-10	Broccoli soil	123000	128000	129000	126666.67	3214.55
	Broccoli root	1540000	1560000	1590000	1563333.33	25166.11
	Broccoli stem	24500	24700	24800	24666.67	152.75
	Broccoli leaf	293000	295000	291000	293000.00	2000.00
	Broccoli	24300	24700	24000	24333.33	351.19
	Cabbage soil	239000	230000	236000	235000.00	4582.58
	Cabbage root	1340000	1390000	1360000	1363333.33	25166.11
	Cabbage	20100	20300	20400	20266.67	152.75

SD = standard deviation; cfu/g = colony forming units per gram of sample

Table B26: Counts obtained for presumptive coliforms in different fresh produce collected from farm A2, over a one-year period.

Sampling period	Samples	Coliforms (cfu/g)				
		A	B	C	Average	SD
Aug-09	Broccoli soil	4200	4200	4500	4300.00	173.21
	Broccoli root	103000	111000	112000	108666.67	4932.88
	Broccoli stem	8900	9000	9200	9033.33	152.75
	Broccoli leaf	9100	9000	9300	9133.33	152.75
	Broccoli	10000	10200	11400	10533.33	757.19
	Cabbage root	49000	49000	41000	46333.33	4618.80
	Cabbage	5000	5100	5500	5200.00	264.58
	Crisphead lettuce soil	4400	4500	4500	4466.67	57.74
	Crisphead lettuce root	1230000	1210000	1200000	1213333.33	15275.25
	Crisphead lettuce	29300	29500	28900	29233.33	305.51
	Cauliflower soil	7700	6900	7500	7366.67	416.33
	Cauliflower root	39000	40000	39000	39333.33	577.35
	Cauliflower leaf	370000	350000	380000	366666.67	15275.25
	Cauliflower	1100000	1120000	1110000	1110000.00	10000.00
Sep-09	Broccoli soil	8200	8300	8300	8266.67	57.74
	Broccoli root	1210000	1230000	1110000	1183333.33	64291.01
	Broccoli stem	30000	33000	30000	31000.00	1732.05
	Broccoli leaf	1980000	2030000	2040000	2016666.67	32145.50
	Broccoli	37000	32000	36000	35000.00	2645.75
	Cabbage soil	38000	34000	39000	37000.00	2645.75
	Cabbage root	176000	170000	172000	172666.67	3055.05
	Cabbage	80000	83000	82000	81666.67	1527.53
	Crisphead lettuce soil	850000	890000	860000	866666.67	20816.66
	Crisphead lettuce root	2200000	2230000	2160000	2196666.67	35118.85
	Crisphead lettuce	38000	35000	31000	34666.67	3511.88
Oct-09	Cabbage soil	5800	6000	6100	5966.67	152.75
	Cabbage root	13000	12800	12400	12733.33	305.51
	Crisphead lettuce soil	360000	320000	310000	330000.00	26457.51
	Crisphead lettuce root	990000	900000	920000	936666.67	47258.16
	Crisphead lettuce	860000	800000	790000	816666.67	37859.39
	Spinach soil	40000	41000	36000	39000.00	2645.75
	Spinach root	160000	161000	154000	158333.33	3785.94
	Spinach stem	4000	3900	3700	3866.67	152.75
	Spinach	7200	6900	7300	7133.33	208.17
Nov-09	Crisphead lettuce soil	490000	460000	450000	466666.67	20816.66
	Crisphead lettuce root	300000	310000		305000.00	
	Crisphead lettuce	91000	96000	87000	91333.33	4509.25
Dec-09	Crisphead lettuce soil	30000	38000	32000	33333.33	4163.33
	Crisphead lettuce root	9000	10800	11500	10433.33	1289.70
	Crisphead lettuce	350000	330000	410000	363333.33	41633.32
Jan-10	Broccoli soil	6500	4800	6300	5866.67	929.16
	Broccoli root	680000	660000	690000	676666.67	15275.25
	Broccoli leaf	2670000	2600000	2610000	2626666.67	37859.39
	Broccoli	48000	50000	51000	49666.67	1527.53
	Crisphead lettuce soil	28700 31000	28000	28200	28975.00	1381.73
	Crisphead lettuce root	12000	12100	12400	12166.67	208.17
	Crisphead lettuce	490000	450000	500000	480000.00	26457.51
Apr-10	Crisphead lettuce soil	520000	560000	500000	526666.67	30550.50
	Crisphead lettuce root	300000	350000	300000	316666.67	28867.51
	Crisphead lettuce	105000	104000	107000	105333.33	1527.53
May-10	Cabbage soil	3000	3100	3100	3066.67	57.74

Table B26/ Cont.

Sampling period	Samples	Coliforms (cfu/g)				
		A	B	C	Average	SD
Jun-10	Cabbage root	11000	11900	11800.0000	11566.67	493.29
	Crisphead lettuce soil	25100	25800	25700	25533.33	378.59
	Crisphead lettuce root	10200	10300	10300	10266.67	57.74
	Crisphead lettuce	156000	159000	157000	157333.33	1527.53
	Broccoli soil	3000	3200	3400	3200.00	200.00
	Broccoli root	82000	84000	83000	83000.00	1000.00
	Broccoli leaf	4500	4500	4800	4600.00	173.21
	Broccoli	5400	5800	5800	5666.67	230.94

SD = standard deviation; cfu/g = colony forming units per gram of sample

Table B27: Counts obtained for presumptive *E. coli* in different fresh produce collected from farm A2, over a one-year period.

Sampling period	Samples	***E. coli* counts (cfu/g)**				
		A	**B**	**C**	**Average**	**SD**
Sep-09	Crisphead lettuce root	17500	18000	18300	17933.33	404.15
Oct-09	Cabbage	43000	40000	41000	41333.33	1527.53
	Crisphead lettuce soil	4700	4000	4300	4333.33	351.19
Dec-09	Crisphead lettuce	3800	3600	3500	3633.33	152.75
Jan-10	Broccoli root	3900	3000	3000	3300.00	519.62
	Broccoli leaf	3200	3300	3700	3400.00	264.58
	Crisphead lettuce soil	71000	76000	79000	75333.33	4041.45
	Crisphead lettuce root	19700	18200	18600	18833.33	776.75
	Crisphead lettuce	900000	960000	910000	923333.33	32145.50
Apr-10	Crisphead lettuce soil	3000	3000	3200	3066.67	115.47
May-10	Crisphead lettuce soil	30000	32000	33000	31666.67	1527.53
	Crisphead lettuce	300000	380000	310000	330000.00	43588.99

SD = standard deviation; cfu/g = colony forming units per gram of sample

Table B28: Counts obtained for presumptive *L. monocytogenes* in different fresh produce collected from farm A2, over a one-year period.

Sampling period	Samples	***L. monocytogenes* counts (cfu/g)**				
		A	**B**	**C**	**Average**	**SD**
Jul-09	Broccoli soil	4000	3500	3400	3633.33	321.46
	Broccoli root	4000	4100	3800	3966.67	152.75
	Cabbage soil	4400	4100	3900	4133.33	251.66
Aug-09	Broccoli soil	3400	3000	3300	3233.33	208.17
Sep-09	Broccoli soil	3100	3200	3200	3166.67	57.74
	Cabbage soil	3400	3900	3500	3600.00	264.58
	Cabbage root	4200	4500	4000	4233.33	251.66
Oct-09	Spinach soil	3000	3100		3050.00	
Dec-09	Crisphead lettuce	3400	3000	3100	3166.67	208.17
Jan-10	Broccoli soil	3500	3600	3900	3666.67	208.17
	Broccoli leaf	3600	3000	3100	3233.33	321.46
	Crisphead lettuce	3600	3900	4200	3900.00	300.00
Apr-10	Crisphead lettuce	3000	3100	3000	3033.33	57.74
May-10	Crisphead lettuce	3000	3000	3000	3000.00	0.00
Jun-10	Broccoli soil	4500	4900	4100	4500.00	400.00
	Broccoli root	5400	5500	5200	5366.67	152.75
	Cabbage soil	4100	4200	4100	4133.33	57.74

SD = standard deviation; cfu/g = colony forming units per gram of sample

Table B29: Counts obtained for presumptive *Salmonella* spp. in different fresh produce collected from farm A2, over a one-year period.

Sampling period	**Samples**	***Salmonella* spp. counts (cfu/g)**				
		A	**B**	**C**	**Average**	**SD**
Jul-09	Cabbage root	70000	68000	65000	67666.67	2516.61
	Crisphead lettuce root	5400	5300	5100	5266.67	152.75
	Spinach root	31000	35000	32000	32666.67	2081.67
Sep-09	Cabbage soil	5100	5400	4300	4933.33	568.62
	Cabbage	14100	14500	14200	14266.67	208.17
Nov-09	Crisphead lettuce soil	30000	31000	36000	32333.33	3214.55
	Crisphead lettuce root	10000	9700	9600	9766.67	208.17
Dec-09	Crisphead lettuce root	3100	3600	3100	3266.67	288.68
	Crisphead lettuce	3500	3400		3450.00	
Jan-10	Broccoli soil	3000	3200	3500	3233.33	251.66
	Broccoli leaf	4500	4900	4600	4666.67	208.17
	Crisphead lettuce root	3000	3200	3100	3100.00	100.00
	Crisphead lettuce	25100	25400	26000	25500.00	458.26
Apr-10	Crisphead lettuce soil	64000	62000	62000	62666.67	1154.70
	Crisphead lettuce root	13400	13700	13400	13500.00	173.21
May-10	Cabbage soil	3500	3600	3800	3633.33	152.75
	Crisphead lettuce	20000	20100	21200	20433.33	665.83
Jun-10	Cabbage soil	3500	3300	3900	3566.67	305.51
	Cabbage root	78000	74000	76000	76000.00	2000.00

SD = standard deviation; cfu/g = colony forming units per gram of sample

Table B30: Counts obtained for presumptive *Shigella* spp. in different fresh produce collected from farm A2, over a one-year period.

Sampling period	Samples	*Shigella* spp. counts (cfu/g)				
		A	B	C	Average	SD
Jul-09	Broccoli soil	50000	52000	48000	50000.00	2000.00
	Broccoli stem	64000	65000	67000	65333.33	1527.53
	Cabbage soil	128000	130000	126000	128000.00	2000.00
	Cabbage root	31000 28400	33000	30000	30600.00	1925.27
	Cabbage	39000	31000	35000	35000.00	4000.00
	Crisphead lettuce soil	15400	15800	15700	15633.33	208.17
	Crisphead lettuce root	7300	6800	7000	7033.33	251.66
	Crisphead lettuce	9300	9200	8800	9100.00	264.58
	Spinach soil	173000	172000	175000	173333.33	1527.53
	Spinach root	75000	76000	79000	76666.67	2081.67
	Spinach stem	4500	4300	4200	4333.33	152.75
	Spinach	5100	5500	5200	5266.67	208.17
Aug-09	Broccoli soil	26000 33000	26100 31000	26600	28540.00	3244.69
	Broccoli root	70000	71000	70000	70333.33	577.35
	Broccoli	3000	3100	3000	3033.33	57.74
	Cabbage soil	8200	8300	8000	8166.67	152.75
	Cabbage root	33000	31000	30000	31333.33	1527.53
	Cabbage	101000	100000	99000	100000.00	1000.00
	Crisphead lettuce soil	5000	5100	5500	5200.00	264.58
	Crisphead lettuce root	100000	101000	111000	104000.00	6082.76
	Crisphead lettuce	80000	83000	85000	82666.67	2516.61
	Cauliflower soil	80000	81000	90000	83666.67	5507.57
	Cauliflower root	300000	310000	320000	310000.00	10000.00
	Cauliflower	300000	310000	330000	313333.33	15275.25
Sep-09	Broccoli soil	10500	10300	10600	10466.67	152.75
	Broccoli root	500000	510000	530000	513333.33	15275.25
	Broccoli leaf	1010000	1060000	990000	1020000.00	36055.51
	Broccoli	10000	10300	9800	10033.33	251.66
	Cabbage soil	30000	31000	33000	31333.33	1527.53
	Cabbage root	3000	3300	3900	3400.00	458.26
	Cabbage	28500	28000	29300	28600.00	655.74
	Crisphead lettuce soil	60000	65000	64000	63000.00	2645.75
	Crisphead lettuce root	1000000	980000	1030000	1003333.33	25166.11
	Crisphead lettuce	65000	61000	68000	64666.67	3511.88
Oct-09	Cabbage soil	5600	5100	5800	5500.00	360.56
	Cabbage root	8400	8000	8200	8200.00	200.00
	Crisphead lettuce soil	10500	10900	9800	10400.00	556.78
	Crisphead lettuce root	5300	6100	6200	5866.67	493.29
	Crisphead lettuce	33000	31000	36000	33333.33	2516.61
	Spinach soil	3500	3600	3900	3666.67	208.17
	Spinach root	30000	36000	31000	32333.33	3214.55
Nov-09	Crisphead lettuce soil	400000	410000	350000	386666.67	32145.50
	Crisphead lettuce root	140000	137000	132000	136333.33	4041.45
Dec-09	Crisphead lettuce soil	300000	310000	300000	303333.33	5773.50
	Crisphead lettuce root	90000	95000	86000	90333.33	4509.25
	Crisphead lettuce	3000	3200	3000	3100.00	
Jan-10	Broccoli leaf	1900000	1920000	1980000	1933333.33	41633.32
	Broccoli	1500000	1510000	1530000	1513333.33	15275.25
	Crisphead lettuce soil	350000	390000	420000	386666.67	35118.85
	Crisphead lettuce root	80000	85000	83000	82666.67	2516.61

Table B30/ Cont.

Sampling period	**Samples**	***Shigella* spp. counts (cfu/g)**				
		A	**B**	**C**	**Average**	**SD**
Apr-10	Crisphead lettuce	209000	211000	214000	211333.33	2516.61
	Crisphead lettuce soil	287000	288000	281000	285333.33	3785.94
	Crisphead lettuce root	87000	80000	85000	84000.00	3605.55
May-10	Crisphead lettuce	3100	3000	3000	3033.33	57.74
	Cabbage soil	13100	13000	12900	13000.00	100.00
	Cabbage root	5100	5000	4900	5000.00	100.00
Jun-10	Crisphead lettuce soil	201000	205000	208000	204666.67	3511.88
	Crisphead lettuce root	70000	71000	73000	71333.33	1527.53
	Crisphead lettuce	3000	3000	3000	3000.00	0.00
	Broccoli soil	9200	9400	9500	9366.67	152.75
	Broccoli root	300000	300000	340000	313333.33	23094.01
	Broccoli leaf	320000	380000	390000	363333.33	37859.39
	Broccoli	5400	5500	5600	5500.00	100.00
	Cabbage soil	87000	87000	89000	87666.67	1154.70
	Cabbage root	25600	25900	24900	25466.67	513.16
	Cabbage	30000	29400	30000	29800.00	346.41

SD = standard deviation; cfu/g = colony forming units per gram of sample

Table B31: Counts obtained for presumptive *Campylobacter* spp. in different fresh produce collected from farm B, over a one-year period.

Sampling period	Samples	*Campylobacter* spp. counts (cfu/g)				
		A	B	C	Average	SD
Jul-09	Broccoli soil	660000	670000	700000	676666.67	20816.66
	Broccoli root	1500000	1520000	1530000	1516666.67	15275.25
	Red cabbage soil	30000	31000	32000	31000.00	1000.00
	Red cabbage root	500000	520000	480000	500000.00	20000.00
	Red cabbage	16100	15600	15700	15800.00	264.58
	Crisphead lettuce soil	480000	500000	470000	483333.33	15275.25
	Crisphead lettuce root	64000	65000	66000	65000.00	1000.00
	Crisphead lettuce	3000	3100	3300	3133.33	152.75
	Spinach soil	125000	120000	123000	122666.67	2516.61
	Spinach root	1380000	1400000	1410000	1396666.67	15275.25
	Spinach stem	220000	210000	200000	210000.00	10000.00
	Spinach	6500	7000	7100	6866.67	321.46
Aug-09	Broccoli soil	2290000	2210000	2210000	2236666.67	46188.02
	Broccoli root	84000	85000	84000	84333.33	577.35
	Broccoli stem	32000	33000	35000	33333.33	1527.53
	Broccoli leaf	1260000	1160000	1100000	1173333.33	80829.04
	Broccoli	2370000	2210000	2380000	2320000.00	95393.92
	Red cabbage soil	90000	88000	73000	83666.67	9291.57
	Red cabbage root	8900	9000	9000	8966.67	57.74
	Red cabbage	5200	5000	5600	5266.67	305.51
	Crisphead lettuce soil	29800	29900	29500	29733.33	208.17
	Crisphead lettuce root	28100	28200	28100	28133.33	57.74
	Crisphead lettuce	1430000	1410000	1430000	1423333.33	11547.01
	Spinach soil	1000000	1030000	1010000	1013333.33	15275.25
	Spinach root	2170000	2050000	2010000	2076666.67	83266.64
	Spinach stem	54000	51000	53000	52666.67	1527.53
	Spinach	2000000	2110000	2050000	2053333.33	55075.71
Sep-09	Broccoli soil	30000	38000	31000	33000.00	4358.90
	Broccoli root	190000	192000	190000	190666.67	1154.70
	Broccoli stem	5800	4900	5000	5233.33	493.29
	Broccoli leaf	17000	17300	17200	17166.67	152.75
	Broccoli	34000	31000	30000	31666.67	2081.67
	Chinese cabbage soil	11000	11200	11900	11366.67	472.58
	Chinese cabbage root	95000	94000	103000	97333.33	4932.88
	Chinese cabbage	4500	4800	3900	4400.00	458.26
	Red cabbage soil	30000	31000	31000	30666.67	577.35
	Red cabbage root	90000	87000	81000	86000.00	4582.58
	Red cabbage	5000	5100	5300	5133.33	152.75
	Spinach soil	4500	4100	4200	4266.67	208.17
	Spinach root	45000	46000	41000	44000.00	2645.75
	Spinach stem	31000	33000	38000	34000.00	3605.55
	Spinach	6200	6200	6100	6166.67	57.74
Oct-09	Red cabbage soil	150000	146000	141000	145666.67	4509.25
	Red cabbage root	120000	126000	135000	127000.00	7549.83
	Red cabbage	12000	13200	13500	12900.00	793.73
	Crisphead lettuce soil	400000	460000	450000	436666.67	32145.50
	Crisphead lettuce root	2510000	2520000	2560000	2530000.00	26457.51
	Crisphead lettuce	800000	690000	930000	806666.67	120138.81
	Parsley soil	310000	320000		315000.00	
	Parsley root	35000	31000	30000	32000.00	2645.75
	Parsley stem	6000	5300	5100	5466.67	472.58
	Parsley	43000	42000	46000	43666.67	2081.67

Table B31/ Cont.

Sampling period	Samples	*Campylobacter* spp. counts (cfu/g)				
		A	B	C	Average	SD
	Spinach soil	141000	146000	159000	148666.67	9291.57
	Spinach root	4600	5900	4300	4933.33	850.49
	Spinach	68000	70000	76000	71333.33	4163.33
Nov-09	Cabbage soil	14000	14600	14100	14233.33	321.46
	Cabbage root	40000	43000	41000	41333.33	1527.53
	Cabbage	48000	46000	46000	46666.67	1154.70
	Parsley soil	60000	69000	63000	64000.00	4582.58
	Parsley root	700000	680000	670000	683333.33	15275.25
	Parsley stem	50000	58000	51000	53000.00	4358.90
	Parsley	7000	6800	6400	6733.33	305.51
	Spinach soil	600000	710000	700000	670000.00	60827.63
	Spinach root	1110000	1180000	1140000	1143333.33	35118.85
	Spinach stem	60000	58000	61000	59666.67	1527.53
	Spinach	65000	61000	60000	62000.00	2645.75
Dec-09	Cabbage soil	300000	310000	350000	320000.00	26457.51
	Cabbage root	31000	32000	30000	31000.00	1000.00
	Cabbage	11000	11200	11700	11300.00	360.56
	Crisphead lettuce soil	500000	510000	530000	513333.33	15275.25
	Crisphead lettuce root	390000	350000	360000	366666.67	20816.66
	Crisphead lettuce	80000	78000	79000	79000.00	1000.00
	Spinach soil	100000	98000	95000	97666.67	2516.61
	Spinach root	35000	38000	43000	38666.67	4041.45
	Spinach stem	9700	9900	10500	10033.33	416.33
	Spinach	350000	360000	390000	366666.67	20816.66
Jan-10	Cabbage soil	205000	209000	204000	206000.00	2645.75
	Cabbage root	120000	118000	124000	120666.67	3055.05
	Cabbage	21200	21900	22400	21833.33	602.77
	Spinach soil	590000	550000	510000	550000.00	40000.00
	Spinach root	1250000	1200000	1270000	1240000.00	36055.51
	Spinach stem	31000	36000	34000	33666.67	2516.61
	Spinach	205000	207000	210000	207333.33	2516.61
Feb-10	Bell pepper soil	29800	29900	28900	29533.33	550.76
	Bell pepper root	23500	23300	23500	23433.33	115.47
	Bell pepper leaf	27600	27800	27900	27766.67	152.75
	Bell pepper	11800	12500	12400	12233.33	378.59
Mar-10	Bell pepper soil	32000	33000	33000	32666.67	577.35
	Bell pepper root	24300	24300	24700	24433.33	230.94
	Bell pepper stem	5400	5500	5300	5400.00	100.00
	Bell pepper leaf	32000	36000	38000	35333.33	3055.05
	Bell pepper	15400	15500	15800	15566.67	208.17
	Broccoli soil	37000	38000	37000	37333.33	577.35
	Broccoli root	221000	234000	225000	226666.67	6658.33
	Broccoli stem	7900	8500	8400	8266.67	321.46
	Broccoli leaf	24300	23400	24100	23933.33	472.58
	Broccoli	43000	40000	38000	40333.33	2516.61
Apr-10	Bell pepper soil	30000	32000	32000	31333.33	1154.70
	Bell pepper root	22000	22100	22100	22066.67	57.74
	Bell pepper stem	3200	3500	3200	3300.00	173.21
	Bell pepper leaf	56000	56000	54000	55333.33	1154.70
	Bell pepper	23800	23700	23600	23700.00	100.00
	Crisphead lettuce soil	450000	460000	460000	456666.67	5773.50
	Crisphead lettuce root	380000	380000	380000	380000.00	0.00
	Crisphead lettuce	64000	65000	63000	64000.00	1000.00
May-10	Bell pepper soil	30000	29600	31000	30200.00	721.11

Table B31/ Cont.

Sampling period	**Samples**	***Campylobacter* spp. counts (cfu/g)**				
		A	**B**	**C**	**Average**	**SD**
Jun-10	Bell pepper root	22100	22300	22500	22300.00	200.00
	Bell pepper stem	3400	3400	3400	3400.00	0.00
	Bell pepper leaf	40000	36000	35000	37000.00	2645.75
	Bell pepper	13400	13500	13200	13366.67	152.75
	Crisphead lettuce soil	400000	410000	370000	393333.33	20816.66
	Crisphead lettuce root	310000	320000	300000	310000.00	10000.00
	Crisphead lettuce	61000	57000	56000	58000.00	2645.75
	Cabbage soil	287000	287000	284000	286000.00	1732.05
	Cabbage root	181000	189000	187000	185666.67	4163.33
	Cabbage	38000	39000	45000	40666.67	3785.94
	Crisphead lettuce soil	680000	690000	670000	680000.00	10000.00
	Crisphead lettuce root	890000	920000	970000	926666.67	40414.52
	Crisphead lettuce	108000	109000	106000	107666.67	1527.53

SD = standard deviation; cfu/g = colony forming units per gram of sample

Table B32: Counts obtained for presumptive coliforms in different fresh produce collected from farm B, over a one-year period.

Sampling period	Samples	Coliforms (cfu/g)				
		A	B	C	Average	SD
Jul-09	Red cabbage soil	3600	3600	3000	3400.00	346.41
	Red cabbage root	1450000	1470000	1500000	1473333.33	25166.11
	Red cabbage	11500	11400	11200	11366.67	152.75
	Crisphead lettuce soil	3600	3000	3100	3233.33	321.46
	Crisphead lettuce root	9000	9300	9700	9333.33	351.19
	Spinach soil	13500	13600	13700	13600.00	100.00
	Spinach root	13800	14000	14100	13966.67	152.75
	Spinach	7100	6900	6800	6933.33	152.75
Aug-09	Broccoli soil	2810000	2740000	2690000	2746666.67	60277.14
	Broccoli root	64000	64000	62000	63333.33	1154.70
	Broccoli stem	24400	23100	24100	23866.67	680.69
	Broccoli leaf	1900000	1920000	1810000	1876666.67	58594.65
	Broccoli	510000	490000	480000	493333.33	15275.25
	Red cabbage root	8600	8800	9100	8833.33	251.66
	Crisphead lettuce soil	2950000	2970000	2990000	2970000.00	20000.00
	Crisphead lettuce root	2900000	2950000	2890000	2913333.33	32145.50
	Crisphead lettuce	1270000	1300000	1320000	1296666.67	25166.11
	Spinach soil	2300000	2390000	2350000	2346666.67	45092.50
	Spinach root	2590000	2590000	2580000	2586666.67	5773.50
	Spinach stem	68000	58000	69000	65000.00	6082.76
	Spinach	2410000	2310000	2320000	2346666.67	55075.71
Sep-09	Broccoli soil	4300	5800	5900	5333.33	896.29
	Broccoli root	35000	39000	36000	36666.67	2081.67
	Broccoli stem	5000	5100	5700	5266.67	378.59
	Broccoli	6000	6100	6100	6066.67	57.74
	Chinese cabbage soil	4600	4600	4700	4633.33	57.74
	Chinese cabbage root	28000 30000	28100 33000	29300	29680.00	2036.42
	Chinese cabbage	6000	6200	6200	6133.33	115.47
	Red cabbage root	15000	14100	14200	14433.33	493.29
	Red cabbage	10000	10300	10100	10133.33	152.75
	Spinach root	4000	3900	4000	3966.67	57.74
	Spinach stem	18700	16900	16900	17500.00	1039.23
Oct-09	Red cabbage root	86000	89000	87000	87333.33	1527.53
	Red cabbage	18000	19200	20500	19233.33	1250.33
	Crisphead lettuce soil	120000	117000	113000	116666.67	3511.88
	Crisphead lettuce root	2010000	2080000	2050000	2046666.67	35118.85
	Crisphead lettuce	230000	227000	239000	232000.00	6245.00
	Parsley root	89000	85000	89000	87666.67	2309.40
	Parsley stem	99000	107000	100000	102000.00	4358.90
	Parsley	10200	10900	10500	10533.33	351.19
	Spinach root	630000	650000	620000	633333.33	15275.25
	Spinach	67000	65000	61000	64333.33	3055.05
Nov-09	Cabbage soil	102000	105000	118000	108333.33	8504.90
	Cabbage root	50000	48000	41000	46333.33	4725.82
	Cabbage	44000	49000	36000	43000.00	6557.44
	Parsley soil	7800	8300	8200	8100.00	264.58
	Parsley root	1410000	1490000	1300000	1400000.00	95393.92
	Parsley stem	7200	7100	7600	7300.00	264.58
	Parsley	19700	20100	20600	20133.33	450.92
	Spinach soil	770000	790000	740000	766666.67	25166.11
	Spinach root	2200000	2230000	2180000	2203333.33	25166.11

Table B32/ Cont.

Sampling period	Samples	Coliforms (cfu/g)				
		A	B	C	Average	SD
	Spinach stem	141000	136000	140000	139000.00	2645.75
	Spinach	21000	21100	20100	20733.33	550.76
Dec-09	Cabbage soil	5100	5000	4900	5000.00	100.00
	Cabbage	55000	59000	49000	54333.33	5033.22
	Crisphead lettuce soil	41000	40000	39000	40000.00	1000.00
	Crisphead lettuce root	17900	18100	18600	18200.00	360.56
	Spinach soil	7500	7900	8100	7833.33	305.51
	Spinach root	38000	36000	39000	37666.67	1527.53
	Spinach stem	3000	3900	3600	3500.00	458.26
	Spinach	760000	670000	690000	706666.67	47258.16
Jan-10	Cabbage soil	60000	61000	69000	63333.33	4932.88
	Cabbage root	69000	73000	70000	70666.67	2081.67
	Cabbage	80000	83000	79000	80666.67	2081.67
	Spinach soil	1100000	1110000	1130000	1113333.33	15275.25
	Spinach root	2450000	2410000	2490000	2450000.00	40000.00
	Spinach stem	170000	169000	172000	170333.33	1527.53
	Spinach	205000	210000	208000	207666.67	2516.61
Feb-10	Bell pepper soil	29800	29500	29600	29633.33	152.75
	Bell pepper root	30000	31000	31000	30666.67	577.35
	Bell pepper leaf	35000	36000	34000	35000.00	1000.00
	Bell pepper	6500	6600	6400	6500.00	100.00
Mar-10	Bell pepper soil	45000	43000	49000	45666.67	3055.05
	Bell pepper root	119000	111000	113000	114333.33	4163.33
	Bell pepper leaf	72000	72000	75000	73000.00	1732.05
	Bell pepper	8700	7600	8500	8266.67	585.95
	Broccoli soil	6500	6600	6300	6466.67	152.75
	Broccoli root	76000	72000	72000	73333.33	2309.40
	Broccoli leaf	3200	3300	3900	3466.67	378.59
	Broccoli	7500	7500	7100	7366.67	230.94
Apr-10	Bell pepper soil	30000	33000	33000	32000.00	1732.05
	Bell pepper root	130000	131000	130000	130333.33	577.35
	Bell pepper stem	3000	3100	3000	3033.33	57.74
	Bell pepper leaf	65000	65000	65000	65000.00	0.00
	Bell pepper	9700	9800	9500	9666.67	152.75
	Crisphead lettuce soil	30000	30000	31000	30333.33	577.35
	Crisphead lettuce root	16400	16100	16000	16166.67	208.17
May-10	Bell pepper soil	33000	32000	31000	32000.00	1000.00
	Bell pepper root	12300	12400	12300	12333.33	57.74
	Bell pepper leaf	70000	68000	68000	68666.67	1154.70
	Bell pepper	6000	6100	5700	5933.33	208.17
	Crisphead lettuce soil	32000	32000	34000	32666.67	1154.70
	Crisphead lettuce root	15300	15500	15800	15533.33	251.66
Jun-10	Cabbage soil	30000	34000	37000	33666.67	3511.88
	Cabbage root	48000	47000	47000	47333.33	577.35
	Cabbage	48000	48000	48000	48000.00	0.00
	Crisphead lettuce soil	4900	5000	5300	5066.67	208.17
	Crisphead lettuce root	12700	12900	12800	12800.00	100.00
	Crisphead lettuce	67000	67000	62000	65333.33	2886.75

SD = standard deviation; cfu/g = colony forming units per gram of sample

Table B33: Counts obtained for presumptive *E. coli* in different fresh produce collected from farm B, over a one-year period.

Sampling period	Samples	*E. coli* counts (cfu/g)				
		A	B	C	Average	SD
Oct-09	Parsley	43000	42000	46000	43666.67	2081.67
	Spinach	80000	81000	76000	79000.00	2645.75
Nov-09	Spinach stem	3100	3000	3600	3233.33	321.46
	Spinach	7000	6600	7900	7166.67	665.83
Jan-10	Spinach root	5500	5100	5700	5433.33	305.51
	Spinach stem	3000	3100	3000	3033.33	57.74
	Spinach	12500	12200	11700	12133.33	404.15
Mar-10	Bell pepper	3200	3300	3100	3200.00	100.00
Apr-10	Bell pepper leaf	3000	3300	3200	3166.67	152.75
	Bell pepper	5100	5400	5300	5266.67	152.75
Jun-10	Crisphead lettuce	3000	3500	3800	3433.33	404.15

SD = standard deviation; cfu/g = colony forming units per gram of sample

Table B34: Counts obtained for presumptive *Salmonella* spp. in different fresh produce collected from farm B, over a one-year period.

Sampling period	Samples	***Salmonella* spp. counts (cfu/g)**				
		A	B	C	Average	SD
Dec-09	Crisphead lettuce soil	6100	6300	6700	6366.67	305.51
Jan-10	Spinach	3500	3900	3600	3666.67	208.17
Feb-10	Bell pepper leaf	3000	3500	3300	3266.67	251.66
Mar-10	Bell pepper leaf	3600	3800	3800	3733.33	115.47
	Bell pepper	3100	3100	3200	3133.33	57.74
	Broccoli leaf	3000	3200	3200	3133.33	115.47
Apr-10	Bell pepper leaf	3100	3200	3400	3233.33	152.75
	Bell pepper	3300	3300	3500	3366.67	115.47
	Crisphead lettuce soil	5100	5500	5500	5366.67	230.94
May-10	Bell pepper leaf	3700	3900	4000	3866.67	152.75
	Bell pepper	3400	3300	3400	3366.67	57.74
	Crisphead lettuce	3500	3200	3300	3333.33	152.75
Jun-10	Crisphead lettuce	5200	5700	5200	5366.67	288.68

SD = standard deviation; cfu/g = colony forming units per gram of sample

Table B35: Counts obtained for presumptive *L. monocytogenes* in different fresh produce collected from farm B, over a one-year period.

Sampling period	**Samples**	***L. monocytogenes* counts (cfu/g)**				
		A	**B**	**C**	**Average**	**SD**
Jul-09	Broccoli soil	8800	9000	9100	8966.67	152.75
	Broccoli root	5000	4700	4600	4766.67	208.17
	Red cabbage soil	3000	3100	3000	3033.33	57.74
	Red cabbage	3300	3800	3000	3366.67	404.15
	Crisphead lettuce soil	13400	13600	13700	13566.67	152.75
	Crisphead lettuce root	6000	5900	6000	5966.67	57.74
Aug-09	Crisphead lettuce soil	3000	3100	3500	3200.00	264.58
	Crisphead lettuce root	5500	4800	3900	4733.33	802.08
Sep-09	Broccoli soil	3000	3000	3500	3166.67	288.68
	Broccoli root	3000	3100		3050.00	
	Red cabbage soil	3000	3100	3300	3133.33	152.75
Nov-09	Cabbage soil	3200	3000	3100	3100.00	100.00
	Cabbage root	3600	4000	4200	3933.33	305.51
Dec-09	Spinach root	3000	3100	3500	3200.00	264.58
Jan-10	Cabbage soil	3000	3100	3100	3066.67	57.74
	Cabbage root	4000	4500	5100	4533.33	550.76
	Spinach	3000	3100	3300	3133.33	152.75
Mar-10	Bell pepper	3500	3500	3700	3566.67	115.47
	Broccoli soil	4100	4000	4000	4033.33	57.74
	Broccoli root	4300	4500	4700	4500.00	200.00
	Broccoli leaf	3200	3200	3400	3266.67	115.47
Apr-10	Bell pepper	4500	4700	4700	4633.33	115.47
May-10	Bell pepper	3900	3600	3700	3733.33	152.75
Jun-10	Cabbage soil	6000	6400	6400	6266.67	230.94
	Cabbage root	9000	9300	9400	9233.33	208.17
	Crisphead lettuce soil	12000	12600	12800	12466.67	416.33
	Crisphead lettuce root	4300	4600	4700	4533.33	208.17
	Crisphead lettuce	4200	4300	4300	4266.67	57.74

SD = standard deviation; cfu/g = colony forming units per gram of sample

Table B36: Counts obtained for presumptive *Shigella* spp. in different fresh produce collected from farm B, over a one-year period.

Sampling period	Samples	*Shigella* spp. counts (cfu/g)				
		A	B	C	Average	SD
Jul-09	Broccoli soil	198000	201000	199000	199333.33	1527.53
	Broccoli root	2510000	2540000	2500000	2516666.67	20816.66
	Broccoli stem	198000	200000	196000	198000.00	2000.00
	Broccoli leaf	246000	244000	250000	246666.67	3055.05
	Red cabbage soil	154000	156000	155000	155000.00	1000.00
	Red cabbage root	1400000	1390000	1420000	1403333.33	15275.25
	Red cabbage	82000	85000	83000	83333.33	1527.53
	Crisphead lettuce soil	370000	400000	410000	393333.33	20816.66
	Crisphead lettuce root	2400000	2380000	2350000	2376666.67	25166.11
	Crisphead lettuce	15000	14900	14700	14866.67	152.75
	Spinach soil	120000	122000	117000	119666.67	2516.61
	Spinach root	1290000	1300000	1320000	1303333.33	15275.25
	Spinach	9600	9400	9700	9566.67	152.75
Aug-09	Broccoli soil	29900 33000	34000	31000	31975.00	1862.57
	Broccoli root	152000	156000	161000	156333.33	4509.25
	Broccoli leaf	6200	5800	5500	5833.33	351.19
	Broccoli	3000	3100	3300	3133.33	152.75
	Red cabbage soil	3100	3300	3900	3433.33	416.33
	Red cabbage root	3500	3700	4000	3733.33	251.66
	Red cabbage	3000	3200	3900	3366.67	472.58
	Crisphead lettuce soil	3500	3900	3100	3500.00	400.00
	Crisphead lettuce root	41000	42000	49000	44000.00	4358.90
	Crisphead lettuce	21200	20100	23100	21466.67	1517.67
	Spinach soil	25000	26100	27100	26066.67	1050.40
	Spinach root	264000	255000	260000	259666.67	4509.25
	Spinach	13800	14000	15100	14300.00	700.00
Sep-09	Broccoli root	298000 300000	275000 310000	298000	296200.00	12853.02
	Broccoli stem	3500	3100	3200	3266.67	208.17
	Broccoli	7000	7500	7800	7433.33	404.15
	Red cabbage root	75000	69000	72000	72000.00	3000.00
	Chinese cabbage root	161000	164000	168000	164333.33	3511.88
	Chinese cabbage	3500	3700	3600	3600.00	100.00
	Spinach root	3500	3600	5100	4066.67	896.29
	Spinach	4300	4000	4900	4400.00	458.26
Oct-09	Red cabbage root	26000 31000	25100 35000	26300	28680.00	4212.72
	Crisphead lettuce root	250000	231000	223000	234666.67	13868.43
	Crisphead lettuce	80000	83000	78000	80333.33	2516.61
	Parsley root	11000	11300	12600	11633.33	850.49
	Parsley	48000	49000	55000	50666.67	3785.94
	Spinach root	720000	760000	740000	740000.00	20000.00
	Spinach	11000	10900	10100	10666.67	493.29
Nov-09	Cabbage soil	700000	680000	660000	680000.00	20000.00
	Cabbage root	113000	116000	121000	116666.67	4041.45
	Cabbage	9000	8900	7600	8500.00	781.02
	Parsley soil	4200	4000	3700	3966.67	251.66
	Parsley root	320000	360000		340000.00	
	Spinach soil	296000	281000	283000	286666.67	8144.53
	Spinach root	600000	690000	510000	600000.00	90000.00
	Spinach stem	5000	4800	4300	4700.00	360.56

Table B36/ Cont.

Sampling period	Samples	*Shigella* spp. counts (cfu/g)				
		A	B	C	Average	SD
	Spinach	3100	3600		3350.00	
Dec-09	Cabbage soil	4000	4200	4300	4166.67	152.75
	Cabbage root	3000	3100	3300	3133.33	152.75
	Cabbage	42000	41000	46000	43000.00	2645.75
	Crisphead lettuce soil	10100	10300	11000	10466.67	472.58
	Crisphead lettuce root	29300	28700	29600	29200.00	458.26
	Spinach soil	3500	3600	3800	3633.33	152.75
	Spinach root	15100	15600	14800	15166.67	404.15
	Spinach stem	3000	3100	3600	3233.33	321.46
	Spinach	35000	31000	30000	32000.00	2645.75
Jan-10	Cabbage soil	500000	510000	530000	513333.33	15275.25
	Cabbage root	191000	198000	196000	195000.00	3605.55
	Cabbage	9800	11000	10600	10466.67	611.01
	Spinach soil	250000	254000	253000	252333.33	2081.67
	Spinach root	510000	540000	540000	530000.00	17320.51
	Spinach stem	5100	5500	5900	5500.00	400.00
	Spinach	150000	151000	154000	151666.67	2081.67
Feb-10	Bell pepper soil	154000	155000	156000	155000.00	1000.00
	Bell pepper root	28700	28900	29600	29066.67	472.58
	Bell pepper leaf	242000	241000	243000	242000.00	1000.00
	Bell pepper	8900	9100	9200	9066.67	152.75
Mar-10	Bell pepper soil	135000	137000	132000	134666.67	2516.61
	Bell pepper root	69000	75000	67000	70333.33	4163.33
	Bell pepper leaf	259000	267000	267000	264333.33	4618.80
	Bell pepper	17400	17300	17700	17466.67	208.17
	Broccoli soil	6000	6400	6700	6366.67	351.19
	Broccoli root	360000	320000	370000	350000.00	26457.51
	Broccoli leaf	9200	9400	9400	9333.33	115.47
	Broccoli	11300	11100	12100	11500.00	529.15
Apr-10	Bell pepper soil	131000	129000	128000	129333.33	1527.53
	Bell pepper root	51000	55000	51000	52333.33	2309.40
	Bell pepper leaf	240000	241000	242000	241000.00	1000.00
	Bell pepper	25400	25600	25500	25500.00	100.00
	Crisphead lettuce soil	9400	9500	9500	9466.67	57.74
	Crisphead lettuce root	26800	26900	26900	26866.67	57.74
May-10	Bell pepper soil	120000	124000	122000	122000.00	2000.00
	Bell pepper root	72000	77000	78000	75666.67	3214.55
	Bell pepper leaf	271000	272000	272000	271666.67	577.35
	Bell pepper	18500	18500	18500	18500.00	0.00
	Crisphead lettuce soil	9000	9300	9400	9233.33	208.17
	Crisphead lettuce root	25100	25500	25700	25433.33	305.51
Jun-10	Cabbage soil	300000	310000	320000	310000.00	10000.00
	Cabbage root	152000	155000	155000	154000.00	1732.05
	Cabbage	8300	8400	8900	8533.33	321.46
	Crisphead lettuce soil	293000	292000	291000	292000.00	1000.00
	Crisphead lettuce root	2100000	2100000	2030000	2076666.67	40414.52
	Crisphead lettuce	12100	12300	12600	12333.33	251.66

SD = standard deviation; cfu/g = colony forming units per gram of sample

Table B37: Counts obtained for presumptive *Campylobacter* spp. in different fresh produce collected from farm C, over a one-year period.

Sampling period	Samples	*Campylobacter* **spp. counts (cfu/g)**				
		A	**B**	**C**	**Average**	**SD**
Jul-09	Cabbage soil	51000	53000	48000	50666.67	2516.61
	Cabbage root	146000	148000	151000	148333.33	2516.61
	Cabbage	6000	6200	6500	6233.33	251.66
Aug-09	Cabbage soil	64000	65000	68000	65666.67	2081.67
	Cabbage root	1450000	1380000	1890000	1426666.67	40414.52
	Cabbage	1970000	1950000	1890000	1936666.67	41633.32
Sep-09	Cabbage soil	3000	3000	3200	3066.67	115.47
	Cabbage root	32000	36000	33000	33666.67	2081.67
	Cabbage	3500	3100	3200	3266.67	208.17
Jan-10	Jam tomato soil	3100	3100	3500	3233.33	230.94
	Jam tomato root	15100	15900	15500	15500.00	400.00
	Jam tomato leaf	3000	3000	3200	3066.67	115.47
Feb-10	Jam tomato root	9800	9600	9600	9666.67	115.47
	Jam tomato leaf	3500	3900	3500	3633.33	230.94
	Jam tomato	5400	5500	5800	5566.67	208.17

SD = standard deviation; cfu/g = colony forming units per gram of sample

Table B38: Counts obtained for presumptive coliforms in different fresh produce collected from farm C, over a one-year period.

Sampling period	Samples	**Coliforms (cfu/g)**				
		A	**B**	**C**	**Average**	**SD**
Jul-09	Cabbage soil	5800	6500	6700	6333.33	472.58
	Cabbage root	13000	13800	13600	13466.67	416.33
	Cabbage	68000	62000	66000	65333.33	3055.05
Aug-09	Cabbage soil	1890000	1710000	1790000	1796666.67	90185.00
	Cabbage root	690000	710000	720000	706666.67	15275.25
	Cabbage	410000	430000	440000	426666.67	15275.25
Sep-09	Cabbage soil	31000	31000	32000	31333.33	577.35
	Cabbage root	3100	3100	3500	3233.33	230.94
Jan-10	Jam tomato soil	2510000	2600000	2530000	2546666.67	47258.16
	Jam tomato root	1610000	1640000	1600000	1616666.67	20816.66
	Jam tomato stem	4900	4000	4100	4333.33	493.29
	Jam tomato leaf	40000	33000	32000	35000.00	4358.90
Feb-10	Jam tomato soil	2300000	2340000	2360000	2333333.33	30550.50
	Jam tomato root	300000	300000	340000	313333.33	23094.01
	Jam tomato leaf	183000	188000	186000	185666.67	2516.61

SD = standard deviation; cfu/g = colony forming units per gram of sample

Table B39: Counts obtained for presumptive *E. coli* (a) and *Salmonella* spp. (b) in different fresh produce collected from farm C, over a one-year period.

	Sampling period	Samples	**Microbial counts *(*cfu/g)**				
			A	**B**	**C**	**Average**	**SD**
(a)	Jul-09	Cabbage soil	6700	5900	6300	6300.00	400.00
(b)	Feb-10	Jam tomato leaf	3400	3400	3100	3300.00	173.21

SD = standard deviation; cfu/g = colony forming units per gram of sample

Table B40: Counts obtained for presumptive *L. monocytogenes* in different fresh produce collected from farm C, over a one-year period.

Sampling period	Samples	*L. monocytogenes* counts (cfu/g)				
		A	B	C	Average	SD
Jul-09	Cabbage root	3000	3100	3300	3133.33	152.75
Aug-09	Cabbage soil	7100	7300	7400	7266.67	152.75
	Cabbage root	6900	6800	6000	6566.67	493.29
Jan-10	Jam tomato soil	6100	6000	6500	6200.00	264.58
	Jam tomato root	7000	7200	7900	7366.67	472.58
Feb-10	Jam tomato soil	6000	6400	6500	6300.00	264.58
	Jam tomato root	5000	5200	5600	5266.67	305.51
	Jam tomato leaf	3500	3700	3500	3566.67	115.47

SD = standard deviation; cfu/g = colony forming units per gram of sample

Table B41: Counts obtained for presumptive *Shigella* spp. in different fresh produce collected from farm C, over a one-year period.

Sampling period	Samples	*Shigella* spp. counts (cfu/g)				
		A	B	C	Average	SD
Jul-09	Cabbage soil	4400	3700	3900	4000.00	360.56
	Cabbage root	720000	800000	830000	783333.33	56862.41
	Cabbage	2950000	2980000	2890000	2940000.00	45825.76
Aug-09	Cabbage soil	19800	20000	19100	19633.33	472.58
	Cabbage root	4500	4100	4000	4200.00	264.58
Sep-09	Cabbage soil	64000	63000	60000	62333.33	2081.67
	Cabbage root	30000	35000	37000	34000.00	3605.55
	Cabbage	8500	9000	9200	8900.00	360.56
Jan-10	Jam tomato soil	65000	68000	72000	68333.33	3511.88
	Jam tomato root	51000	55000	49000	51666.67	3055.05
	Jam tomato stem	3300	3800	4500	3866.67	602.77
	Jam tomato leaf	9000	9500	9300	9266.67	251.66
Feb-10	Jam tomato soil	198000	199000	196000	197666.67	1527.53
	Jam tomato root	300000	320000	320000	313333.33	11547.01
	Jam tomato stem	3400	3300	3000	3233.33	208.17
	Jam tomato leaf	18700	18800	19300	18933.33	321.46
	Jam tomato	6000	6500	6800	6433.33	404.15

SD = standard deviation; cfu/g = colony forming units per gram of sample

Table B42: Concentrations of heavy metals (mg/L) in irrigation water collected from farm A1, over a one-year period [a].

Heavy metal	Sampling period	Heavy metal concentrations (mg/L)				
		A	B	C	Average	SD
As	Jul-09	0.014	0.015	0.014	0.014	0.001
	Aug-09	0.004	0.004	0.004	0.004	0.000
	Sep-09	0.004	0.001	0.004	0.003	0.002
	Nov-09	0.005	0.002	0.004	0.004	0.002
	Dec-09	0.009	0.008	0.009	0.009	0.001
	Mar-10	N	0.009	0.006	0.008	
	Apr-10	0.015	0.008	0.007	0.010	0.004
	May-10	0.001	0.012	N	0.007	
Cd	Jul-09	0.015	0.015	0.015	0.015	0.000
	Sep-09	0.004	0.001	0.004	0.003	0.002
	Oct-09	0.003	0.003	0.003	0.003	0.000
	Nov-09	0.006	0.006	0.007	0.006	0.001
	Apr-10	0.001	0.001	0.001	0.001	0.000
	May-10	0.002	0.004	N	0.003	
Hg	Jul-09	0.006	0.006	0.007	0.006	0.001
	Aug-09	0.016	0.015	0.013	0.015	0.002
	Sep-09	0.007	0.007	0.001	0.005	0.003
	Oct-09	0.016	0.014	0.013	0.014	0.002
	Nov-09	0.006	0.009	0.008	0.008	0.002
	Feb-10	0.016	0.016	0.016	0.016	0.000
	Mar-10	0.001	0.005	0.006	0.004	0.003
	Apr-10	0.025	0.025	0.016	0.022	0.005
	Jun-10	0.046	0.047	0.077	0.057	0.018
Pb	Aug-09	0.019	0.018	0.018	0.018	0.001
	Sep-09	0.007	0.007	0.007	0.007	0.000
	Oct-09	0.026	0.027	0.027	0.027	0.001
	Nov-09	0.015	0.014	0.015	0.015	0.001
	Dec-09	0.007	0.002	0.007	0.005	0.003
	Jan-10	0.017	0.017	0.02	0.018	0.002
	Feb-10	0.009	0.009	0.009	0.009	0.000
	Apr-10	0.014	0.006	0.003	0.008	0.006
	May-10	0.004	0.006	0.006	0.005	0.001
	Jun-10	0.008	0.013	N	0.011	

N = heavy metal was not detected; SD = standard deviation; [a]detection limits for heavy metals (mg/L) were as follows: As (0.0053), Cd (0.0025), Hg (0.001), Pb (0.0042)

Table B43: Concentrations of heavy metals (mg/L) in irrigation water collected from farm A2, over a one-year period [a].

Heavy metal	Sampling period	Heavy metal concentrations (mg/L)				
		A	B	C	Average	SD
As	Jul-09	0.018	0.017	0.016	0.017	0.001
	Aug-09	0.004	0.004	0.003	0.004	0.001
	Sep-09	0.009	0.009	0.004	0.007	0.003
	Nov-09	0.006	0.013	N	0.010	
	Dec-09	0.005	0.008	0.012	0.008	0.004
	Mar-10	0.004	0.006	N	0.005	
	Apr-10	0.009	N	0.019	0.014	
	May-10	N	N	0.001	0.001	
Cd	Jul-09	0.018	0.017	0.016	0.017	0.001
	Sep-09	0.009	0.009	0.004	0.007	0.003
	Oct-09	0.003	0.003	0.003	0.003	0.000
	Nov-09	0.007	0.006	0.006	0.006	0.001
	Apr-10	0.001	0.001	0.001	0.001	0.000
Hg	Jul-09	0.007	0.006	0.007	0.007	0.001
	Aug-09	0.017	0.012	0.013	0.014	0.003
	Sep-09	0.007	0.007	0.009	0.008	0.001
	Oct-09	0.013	0.016	0.014	0.014	0.002
	Nov-09	0.005	0.008	0.008	0.007	0.002
	Feb-10	0.016	0.016	0.015	0.016	0.001
	Mar-10	0.008	0.008	0.018	0.011	0.006
	Apr-10	0.014	0.01	0.014	0.013	0.002
	Jun-10	0.024	0.03	0.047	0.034	0.012
Pb	Aug-09	0.02	0.016	0.019	0.018	0.002
	Sep-09	0.003	0.006	0.001	0.003	0.003
	Oct-09	0.023	0.025	0.027	0.025	0.002
	Nov-09	0.016	0.016	0.015	0.016	0.001
	Dec-09	0.001	0.002	0.003	0.002	0.001
	Jan-10	0.017	0.018	0.016	0.017	0.001
	Feb-10	0.006	0.004	0.012	0.007	0.004
	Apr-10	0.004	0.006	0.004	0.005	0.001

N = heavy metal was not detected; SD = standard deviation; [a]detection limits for heavy metals (mg/L) were as follows: As (0.0053), Cd (0.0025), Hg (0.001), Pb (0.0042)

Table B44: Concentrations of heavy metals (mg/L) in irrigation water collected from farm B, over a one-year period [a].

Heavy metal	Sampling period	Heavy metal concentrations (mg/L)				
		A	**B**	**C**	**Average**	**SD**
As	Jul-09	0.028	0.028	0.028	0.028	0.000
	Aug-09	0.001	0.001	0.002	0.001	0.001
	Sep-09	0.003	0.002	0.002	0.002	0.001
	Nov-09	0.002	0.007	0.007	0.005	0.003
	Dec-09	N	N	0.007	0.007	
	Jan-10	0.023	0.019	N	0.021	
	Feb-10	N	0.005	N	0.005	
	Mar-10	0.003	N	0.008	0.006	
	Apr-10	0.011	0.009	0.013	0.011	0.002
Cd	Jul-09	0.026	0.027	0.027	0.027	0.001
	Aug-09	0.003	0.003	0.002	0.003	0.001
	Sep-09	0.003	0.002	0.002	0.002	0.001
	Oct-09	0.003	0.003	0.004	0.003	0.001
	Nov-09	0.009	0.007	0.007	0.008	0.001
	Apr-10	0.001	0.001	0.001	0.001	0.000
Hg	Jul-09	0.033	0.034	0.033	0.033	0.001
	Aug-09	0.02	0.018	0.023	0.020	0.003
	Sep-09	0.043	0.021	0.019	0.028	0.013
	Oct-09	0.014	0.013	0.02	0.016	0.004
	Nov-09	0.011	0.013	0.016	0.013	0.003
	Feb-10	0.016	0.017	0.017	0.017	0.001
	Mar-10	0.01	0.005	0.006	0.007	0.003
	Apr-10	0.017	0.016	0.013	0.015	0.002
	Jun-10	0.042	0.036	0.036	0.038	0.003
Pb	Jul-09	0.037	0.037	0.038	0.037	0.001
	Aug-09	N	N	0.001	0.001	
	Sep-09	0.018	0.022	0.018	0.019	0.002
	Oct-09	0.025	0.025	0.025	0.025	0.000
	Nov-09	0.018	0.014	0.015	0.016	0.002
	Dec-09	0.008	0.003	N	0.006	
	Jan-10	0.015	0.017	0.017	0.016	0.001
	Feb-10	0.007	0.004	0.005	0.005	0.002
	Apr-10	0.006	0.005	0.003	0.005	0.002
	May-10	0.005	0.006	0.004	0.005	0.001

N = heavy metal was not detected; SD = standard deviation; [a]detection limits for heavy metals (mg/L) were as follows: As (0.0053), Cd (0.0025), Hg (0.001), Pb (0.0042)

Table B45: Concentrations of heavy metals (mg/L) in irrigation water collected from farm C, over a one-year period [a].

Heavy metals	**Sampling period**	**Heavy metal concentrations (mg/L)**				
		A	**B**	**C**	**Average**	**SD**
As	Jul-09	0.011	0.009	0.009	0.010	0.001
	Aug-09	0.002	0.001	0.001	0.001	0.001
	Nov-09	0.011	0.017	0.007	0.012	0.005
	Dec-09	0.008	0.005	0.005	0.006	0.002
	Jan-10	0.019	0.009	N	0.014	
	Feb-10	0.025	N	N	0.025	
	Mar-10	0.008	0.006	0.001	0.005	0.004
	Apr-10	0.01	0.01	0.015	0.012	0.003
Cd	Jul-09	0.016	0.013	0.013	0.014	0.002
	Aug-09	0.002	0.003	0.003	0.003	0.001
	Oct-09	0.004	0.004	0.004	0.004	0.000
	Nov-09	0.007	0.007	0.007	0.007	0.000
	Jan-10	0.001	0.001	N	0.001	
	Feb-10	0.003	N	N	0.003	
	Apr-10	N	0.001	N	0.001	
	May-10	N	N	0.001	0.001	
Hg	Jul-09	0.058	0.037	0.034	0.043	0.013
	Aug-09	0.027	0.026	0.024	0.026	0.002
	Sep-09	0.021	0.023	0.023	0.022	0.001
	Oct-09	0.014	0.015	0.013	0.014	0.001
	Nov-09	0.016	0.016	0.013	0.015	0.002
	Feb-10	0.018	0.015	0.017	0.017	0.002
	Mar-10	0.002	0.004	0.013	0.006	0.006
	Apr-10	0.016	0.017	0.02	0.018	0.002
	Jun-10	0.044	0.043	0.033	0.040	0.006
Pb	Jul-09	0.047	0.037	0.035	0.040	0.006
	Sep-09	0.012	0.015	0.017	0.015	0.003
	Oct-09	0.028	0.026	0.023	0.026	0.003
	Nov-09	0.018	0.017	0.016	0.017	0.001
	Dec-09	0.007	0.007	0.007	0.007	0.000
	Jan-10	0.021	0.021	0.017	0.020	0.002
	Feb-10	0.020	0.006	0.007	0.011	0.008
	Apr-10	0.007	0.005	0.003	0.005	0.002
	May-10	0.003	0.003	0.005	0.004	0.001

N = heavy metal was not detected; SD = standard deviation; [a]detection limits for heavy metals (mg/L) were as follows: As (0.0053), Cd (0.0025), Hg (0.001), Pb (0.0042)

Table B46: Concentrations of As (mg/L) in different fresh produce collected from farm A1, over a one-year period [a].

Sampling period	Samples	Concentration of As (mg/L)				
		A	B	C	Average	SD
Jul-09	Broccoli soil	0.016	0.016	0.015	0.016	0.001
	Broccoli root	0.020	0.022	0.019	0.020	0.002
	Broccoli stem	0.016	0.027	0.016	0.020	0.006
	Broccoli leaf	0.018	0.020	0.019	0.019	0.001
	Broccoli	0.014	0.015	0.015	0.015	0.001
	Cabbage soil	0.016	0.017	0.015	0.016	0.001
	Cabbage root	0.018	0.016	0.015	0.016	0.002
	Cabbage	0.016	0.017	0.016	0.016	0.001
	Crisphead lettuce soil	0.017	0.017	0.017	0.017	0.000
	Crisphead lettuce root	0.015	0.015	0.015	0.015	0.000
	Crisphead lettuce	0.068	0.015	0.018	0.034	0.030
	Jam tomato soil	0.015	0.015	0.017	0.016	0.001
	Jam tomato root	0.024	0.060	0.017	0.034	0.023
	Jam tomato stem	0.027	0.028	0.024	0.026	0.002
	Jam tomato leaf	0.016	0.018	0.016	0.017	0.001
	Jam tomato	0.015	0.015	0.018	0.016	0.002
Aug-09	Broccoli soil	0.009	0.009	0.009	0.009	0.000
	Broccoli root	0.004	0.004	0.004	0.004	0.000
	Broccoli stem	0.005	0.005	0.004	0.005	0.001
	Broccoli leaf	0.004	0.004	0.004	0.004	0.000
	Broccoli	0.004	0.004	0.004	0.004	0.000
	Cabbage soil	0.009	0.010	0.009	0.009	0.001
	Cabbage root	0.003	0.003	0.003	0.003	0.000
	Cabbage	0.005	0.001	0.003	0.003	0.002
	Crisphead lettuce soil	0.008	0.007	0.029	0.015	0.012
	Crisphead lettuce root	0.004	0.004	0.005	0.004	0.001
	Crisphead lettuce	0.004	0.003	0.004	0.004	0.001
	Jam tomato soil	0.007	0.005	0.008	0.007	0.002
	Jam tomato root	0.004	0.005	0.005	0.005	0.001
	Jam tomato stem	0.003	0.004	0.005	0.004	0.001
	Jam tomato leaf	0.013	0.005	0.007	0.008	0.004
	Jam tomato	0.005	0.004	0.004	0.004	0.001
Sep-09	Cabbage soil	0.008	0.012	0.020	0.013	0.006
	Cabbage root	0.002	N	N	0.002	
	Crisphead lettuce soil	0.028	0.006	0.007	0.014	0.012
	Crisphead lettuce	N	0.004	N	0.004	
	Jam tomato soil	0.017	0.013	0.011	0.014	0.003
	Jam tomato	N	N	0.001	0.001	
Oct-09	Broccoli root	N	0.005	N	0.005	
	Crisphead lettuce	N	N	0.005	0.005	
Nov-09	Spinach soil	0.005	N	0.001	0.003	
	Spinach root	0.011	N	0.014	0.013	
	Spinach stem	0.004	0.003	0.008	0.005	0.003
	Spinach	0.005	N	N	0.005	
Dec-09	Spinach stem	N	0.008	0.001	0.005	
	Spinach	N	N	0.006	0.006	
Jan-10	Cabbage root	0.017	0.001	0.049	0.022	0.024
	Cabbage	0.012	N	0.019	0.016	
	Crisphead lettuce root	0.005	0.013	N	0.009	
	Crisphead lettuce	0.011	0.013	0.042	0.022	0.017
	Spinach stem	0.009	0.019	0.033	0.020	0.012
	Spinach	0.006	0.011	0.017	0.011	0.006
Feb-10	Crisphead lettuce root	0.002	0.003	N	0.003	

Table B46/ Cont.

Sampling period	Samples	Concentration of As (mg/L)				
		A	B	C	Average	SD
	Crisphead lettuce	N	0.007	N	0.007	
	Spinach root	0.001	0.010	0.008	0.006	0.005
	Spinach stem	0.003	0.004	0.009	0.005	0.003
	Spinach	0.002	0.010	0.009	0.007	0.004
Mar-10	Cabbage soil	0.006	N	N	0.006	
	Cabbage root	0.008	0.007	0.003	0.006	0.003
	Crisphead lettuce soil	0.008	N	0.003	0.006	
	Crisphead lettuce root	0.010	0.014	0.014	0.013	0.002
	Crisphead lettuce	0.009	0.003	0.004	0.005	0.003
	Spinach soil	0.004	0.001	N	0.003	
	Spinach root	N	N	0.005	0.005	
	Spinach stem	0.007	0.005	0.007	0.006	0.001
	Spinach	0.007	0.007	0.006	0.007	0.001
Apr-10	Crisphead lettuce soil	0.016	0.016	0.015	0.016	0.001
	Crisphead lettuce root	0.008	0.018	0.018	0.015	0.006
	Crisphead lettuce	0.024	0.032	0.028	0.028	0.004
	Jam tomato soil	0.018	0.014	N	0.016	
	Jam tomato root	0.024	0.022	0.017	0.021	0.004
	Jam tomato stem	0.023	0.010	0.009	0.014	0.008
	Jam tomato leaf	0.010	0.007	0.011	0.009	0.002
	Jam tomato	0.040	0.013	0.019	0.024	0.014
	Spinach soil	0.006	0.001	0.002	0.003	0.003
	Spinach root	0.006	0.006	0.010	0.007	0.002
	Spinach	0.009	0.003	0.009	0.007	0.003
May-10	Crisphead lettuce root	0.006	0.013	0.019	0.013	0.007
	Crisphead lettuce	0.018	0.015	0.023	0.019	0.004
	Jam tomato soil	N	0.002	0.004	0.003	
	Jam tomato root	0.016	0.007	0.015	0.013	0.005
	Jam tomato stem	0.015	0.006	0.010	0.010	0.005
	Jam tomato leaf	0.016	0.017	0.019	0.017	0.002
	Jam tomato	N	0.006	0.009	0.008	
	Spinach soil	N	0.004	N	0.004	
	Spinach root	0.022	0.022	0.028	0.024	0.003
	Spinach stem	0.016	0.008	0.015	0.013	0.004
	Spinach	0.013	0.021	0.023	0.019	0.005
Jun-10	Crisphead lettuce root	0.016	0.019	0.020	0.018	0.002
	Crisphead lettuce	0.023	0.027	0.026	0.025	0.002
	Spinach soil	0.009	0.017	0.015	0.014	0.004
	Spinach root	0.087	0.086	0.082	0.085	0.003
	Spinach stem	0.010	0.008	0.006	0.008	0.002
	Spinach	0.024	0.023	0.028	0.025	0.003

N = heavy metal was not detected; SD = standard deviation; [a]detection limits for heavy metals (mg/L) were as follows: As (0.0053), Cd (0.0025), Hg (0.001), Pb (0.0042)

Table B47: Concentrations of Cd (mg/L) in different fresh produce collected from farm A1, over a one-year period ^{a}.

Sampling period	Samples	Concentration of Cd (mg/L)				
		A	B	C	Average	SD
Jul-09	Broccoli soil	0.017	0.017	0.016	0.017	0.001
	Broccoli root	0.013	0.012	0.014	0.013	0.001
	Broccoli stem	0.015	0.028	0.017	0.020	0.007
	Broccoli leaf	0.013	0.012	0.014	0.013	0.001
	Broccoli	0.015	0.014	0.015	0.015	0.001
	Cabbage soil	0.017	0.018	0.015	0.017	0.002
	Cabbage root	0.013	0.014	0.015	0.014	0.001
	Cabbage	0.014	0.014	0.014	0.014	0.000
	Crisphead lettuce soil	0.017	0.018	0.017	0.017	0.001
	Crisphead lettuce root	0.015	0.015	0.015	0.015	0.000
	Crisphead lettuce	0.071	0.016	0.016	0.034	0.032
	Jam tomato soil	0.016	0.016	0.018	0.017	0.001
	Jam tomato root	0.013	0.067	0.014	0.031	0.031
	Jam tomato stem	0.017	0.008	0.011	0.012	0.005
	Jam tomato leaf	0.015	0.015	0.015	0.015	0.000
	Jam tomato	0.016	0.016	0.017	0.016	0.001
Aug-09	Broccoli soil	0.006	0.005	0.007	0.006	0.001
	Broccoli root	0.001	0.001	0.002	0.001	0.001
	Cabbage soil	0.003	0.004	0.003	0.003	0.001
	Crisphead lettuce soil	0.005	0.004	0.024	0.011	0.011
	Crisphead lettuce root	0.001	0.001	0.002	0.001	0.001
	Jam tomato soil	0.002	N	0.004	0.003	
	Jam tomato root	0.001	0.003	0.002	0.002	0.001
	Jam tomato stem	0.000	0.000	0.002	0.001	0.001
	Jam tomato leaf	0.011	0.002	0.004	0.006	0.005
Sep-09	Cabbage soil	0.008	0.012	0.020	0.013	0.006
	Cabbage root	0.002	N	N	0.002	
	Crisphead lettuce soil	0.028	0.006	0.007	0.014	0.012
	Crisphead lettuce	N	0.004	N	0.004	
	Jam tomato soil	0.017	0.013	0.011	0.014	0.003
	Jam tomato	N	N	0.001	0.001	
Oct-09	Broccoli soil	0.005	0.006	0.005	0.005	0.001
	Broccoli root	0.004	0.004	0.004	0.004	0.000
	Broccoli stem	0.004	0.004	0.004	0.004	0.000
	Broccoli leaf	0.004	0.004	0.004	0.004	0.000
	Broccoli	0.007	0.006	0.005	0.006	0.001
	Cabbage soil	0.006	0.006	0.006	0.006	0.000
	Cabbage root	0.004	0.004	0.004	0.004	0.000
	Cabbage	0.004	0.004	0.004	0.004	0.000
	Crisphead lettuce soil	0.008	0.006	0.008	0.007	0.001
	Crisphead lettuce root	0.004	0.005	0.005	0.005	0.001
	Crisphead lettuce	0.004	0.005	0.004	0.004	0.001
	Spinach soil	0.006	0.006	0.005	0.006	0.001
	Spinach root	0.004	0.004	0.004	0.004	0.000
	Spinach stem	0.004	0.004	0.004	0.004	0.000
	Spinach	0.004	0.003	0.004	0.004	0.001
Nov-09	Spinach soil	0.007	0.008	0.008	0.008	0.001
	Spinach root	0.008	0.008	0.007	0.008	0.001
	Spinach stem	0.007	0.007	0.007	0.007	0.000
	Spinach	0.007	0.007	0.007	0.007	0.000
Jan-10	Cabbage soil	0.002	0.001	0.001	0.001	0.001
	Cabbage root	0.001	0.002	0.001	0.001	0.001
	Cabbage	0.001	0.001	0.001	0.001	0.000

Table B47/ Cont.

Sampling period	Samples	Concentration of Cd (mg/L)				
		A	B	C	Average	SD
	Crisphead lettuce soil	0.002	0.002	0.002	0.002	0.000
	Crisphead lettuce root	0.001	0.002	N	0.002	
	Crisphead lettuce	0.005	0.005	0.005	0.005	0.000
	Spinach soil	0.005	0.006	0.006	0.006	0.001
	Spinach root	0.002	0.001	0.002	0.002	0.001
	Spinach stem	0.003	0.002	0.002	0.002	0.001
	Spinach	0.005	0.005	0.004	0.005	0.001
Feb-10	Crisphead lettuce soil	0.003	0.001	0.002	0.002	0.001
	Spinach stem	N	0.014	N	0.014	
Mar-10	Cabbage soil	0.001	0.001	0.001	0.001	0.000
	Crisphead lettuce soil	0.002	0.002	0.002	0.002	0.000
	Spinach soil	0.002	0.004	0.002	0.003	0.001
	Spinach root	0.001	0.002	N	0.002	
	Spinach stem	0.002	0.001	0.001	0.001	0.001
	Spinach	0.001	0.001	0.001	0.001	0.000
Apr-10	Crisphead lettuce soil	0.001	0.001	0.001	0.001	0.000
	Crisphead lettuce root	0.001	0.001	0.001	0.001	0.000
	Crisphead lettuce	0.006	0.006	0.008	0.007	0.001
	Jam tomato root	0.002	0.003	0.004	0.003	0.001
	Jam tomato stem	0.002	0.002	0.001	0.002	0.001
	Jam tomato leaf	0.002	0.002	0.004	0.003	0.001
	Jam tomato	0.003	0.003	0.005	0.004	0.001
	Spinach soil	0.005	0.004	0.002	0.004	0.002
	Spinach root	0.002	0.002	0.004	0.003	0.001
	Spinach	0.001	0.003	0.002	0.002	0.001
May-10	Crisphead lettuce soil	0.011	0.008	0.008	0.009	0.002
	Crisphead lettuce	N	N	0.001	0.001	
	Jam tomato soil	0.006	0.006	0.006	0.006	0.000
	Jam tomato root	0.004	0.001	N	0.003	
	Jam tomato stem	0.001	N	N	0.001	
	Jam tomato leaf	0.002	N	0.002	0.002	
	Jam tomato	0.001	N	N	0.001	
	Spinach soil	0.006	0.002	0.012	0.007	0.005
	Spinach root	N	N	0.001	0.001	
	Spinach stem	0.001	N	N	0.001	
	Spinach	0.002	N	0.005	0.004	
Jun-10	Crisphead lettuce	0.004	0.003	0.001	0.003	0.002
	Spinach soil	0.009	0.009	0.009	0.009	0.000
	Spinach root	0.005	0.004	0.005	0.005	0.001
	Spinach stem	0.001	0.001	0.001	0.001	0.000
	Spinach	0.003	0.003	0.004	0.003	0.001

N = heavy metal was not detected; SD = standard deviation; [a]detection limits for heavy metals (mg/L) were as follows: As (0.0053), Cd (0.0025), Hg (0.001), Pb (0.0042)

Table B48: Concentrations of Hg (mg/L) in different fresh produce collected from farm A1, over a one-year period [a].

Sampling period	Samples	Concentration of Hg (mg/L)				
		A	B	C	Average	SD
Jul-09	Broccoli soil	0.005	0.006	0.006	0.006	0.001
	Broccoli stem	0.004	0.006	0.006	0.005	0.001
	Broccoli	0.006	0.004	0.006	0.005	0.001
	Cabbage soil	0.006	0.007	0.006	0.006	0.001
	Cabbage root	0.001	0.005	0.006	0.004	0.003
	Cabbage	0.004	0.002	0.003	0.003	0.001
	Crisphead lettuce soil	0.005	0.005	0.006	0.005	0.001
	Crisphead lettuce root	0.005	0.005	0.006	0.005	0.001
	Crisphead lettuce	0.005	0.006	0.004	0.005	0.001
	Jam tomato soil	0.008	0.009	0.009	0.009	0.001
	Jam tomato root	N	0.004	0.005	0.005	
	Jam tomato leaf	0.013	0.008	0.008	0.010	0.003
	Jam tomato	0.04	0.022	0.016	0.026	0.012
Aug-09	Broccoli soil	0.017	0.014	0.015	0.015	0.002
	Broccoli root	0.014	0.024	0.018	0.019	0.005
	Broccoli stem	0.019	0.08	0.018	0.039	0.036
	Broccoli leaf	0.019	0.027	0.018	0.021	0.005
	Broccoli	0.019	0.064	0.018	0.034	0.026
	Cabbage soil	0.018	0.015	0.012	0.015	0.003
	Cabbage root	0.018	0.019	0.02	0.019	0.001
	Cabbage	0.005	0.015	0.021	0.014	0.008
	Crisphead lettuce soil	0.011	0.013	0.013	0.012	0.001
	Crisphead lettuce root	0.018	0.021	0.031	0.023	0.007
	Crisphead lettuce	0.017	0.015	0.027	0.020	0.006
	Jam tomato soil	0.012	0.008	0.013	0.011	0.003
	Jam tomato root	0.172	0.032	0.034	0.079	0.080
	Jam tomato stem	0.033	0.022	0.024	0.026	0.006
	Jam tomato leaf	0.06	0.023	0.117	0.067	0.047
	Jam tomato	0.02	0.033	0.017	0.023	0.009
Sep-09	Cabbage soil	0.006	0.007	0.008	0.007	0.001
	Cabbage root	0.027	0.026	0.029	0.027	0.002
	Cabbage	0.023	0.024	0.025	0.024	0.001
	Crisphead lettuce soil	0.02	0.008	0.008	0.012	0.007
	Crisphead lettuce root	0.025	0.023	0.024	0.024	0.001
	Crisphead lettuce	0.023	0.022	0.022	0.022	0.001
	Jam tomato soil	0.006	0.002	0.006	0.005	0.002
	Jam tomato root	0.024	0.025	0.023	0.024	0.001
	Jam tomato stem	0.029	0.027	0.031	0.029	0.002
	Jam tomato leaf	0.023	0.024	0.027	0.025	0.002
	Jam tomato	0.024	0.023	0.022	0.023	0.001
Oct-09	Broccoli soil	0.02	0.017	0.017	0.018	0.002
	Broccoli root	0.022	0.032	0.034	0.029	0.006
	Broccoli stem	0.037	0.068	0.022	0.042	0.023
	Broccoli leaf	0.091	0.036	0.029	0.052	0.034
	Broccoli	0.013	0.011	0.011	0.012	0.001
	Cabbage soil	0.013	0.012	0.013	0.013	0.001
	Cabbage root	0.038	0.08	0.175	0.098	0.070
	Cabbage	0.104	0.217	0.052	0.124	0.084
	Crisphead lettuce soil	0.017	0.017	0.018	0.017	0.001
	Crisphead lettuce root	0.02	0.108	0.102	0.077	0.049
	Crisphead lettuce	0.015	0.02	0.019	0.018	0.003
	Spinach soil	0.008	0.009	0.01	0.009	0.001
	Spinach root	0.034	0.035	0.03	0.033	0.003

Table B48/ Cont.

Sampling period	Samples	Concentration of Hg (mg/L)				
		A	B	C	Average	SD
	Spinach stem	0.092	0.043	0.071	0.069	0.025
	Spinach	0.017	0.019	0.017	0.018	0.001
Nov-09	Spinach soil	0.007	0.007	0.006	0.007	0.001
	Spinach root	0.011	0.011	0.012	0.011	0.001
	Spinach stem	0.01	0.009	0.011	0.010	0.001
	Spinach	0.016	0.007	0.006	0.010	0.006
Jan-10	Crisphead lettuce root	N	0.005	N	0.005	
	Crisphead lettuce	0.019	0.026	0.013	0.019	0.007
	Spinach stem	0.003	0.003	N	0.003	
	Spinach	0.011	0.009	0.011	0.010	0.001
Feb-10	Crisphead lettuce soil	0.017	0.016	0.016	0.016	0.001
	Crisphead lettuce root	0.018	0.019	0.028	0.022	0.006
	Crisphead lettuce	0.03	0.017	0.017	0.021	0.008
	Spinach soil	0.016	0.018	0.016	0.017	0.001
	Spinach root	0.018	0.017	0.019	0.018	0.001
	Spinach stem	0.018	0.02	0.018	0.019	0.001
	Spinach	0.018	0.017	0.019	0.018	0.001
Mar-10	Cabbage soil	0.001	N	N	0.001	
	Cabbage root	0.003	0.001	0.002	0.002	0.001
	Crisphead lettuce root	0.002	N	N	0.002	
	Crisphead lettuce	0.001	0.002	0.002	0.002	0.001
	Spinach root	0.003	0.004	0.002	0.003	0.001
	Spinach stem	0.005	0.002	0.005	0.004	0.002
	Spinach	0.003	0.003	0.002	0.003	0.001
Apr-10	Crisphead lettuce soil	0.021	0.02	0.02	0.020	0.001
	Crisphead lettuce root	0.02	0.019	0.02	0.020	0.001
	Crisphead lettuce	0.026	0.031	0.03	0.029	0.003
	Jam tomato soil	0.019	0.023	0.021	0.021	0.002
	Jam tomato root	0.032	0.032	0.033	0.032	0.001
	Jam tomato stem	0.034	0.04	0.034	0.036	0.003
	Jam tomato leaf	0.028	0.026	0.026	0.027	0.001
	Jam tomato	0.04	0.037	0.037	0.038	0.002
	Spinach soil	0.02	0.019	0.021	0.020	0.001
	Spinach root	0.018	0.019	0.024	0.020	0.003
	Spinach	0.027	0.026	0.024	0.026	0.002
Jun-10	Crisphead lettuce soil	0.057	0.053	0.048	0.053	0.005

N = heavy metal was not detected; SD = standard deviation; [a]detection limits for heavy metals (mg/L) were as follows: As (0.0053), Cd (0.0025), Hg (0.001), Pb (0.0042)

Table B49: Concentrations of Pb (mg/L) in different fresh produce collected from farm A1, over a one-year period ^{a}.

Sampling period	Samples	Concentration of Pb (mg/L)				
		A	B	C	Average	SD
Jul-09	Broccoli soil	0.157	0.157	0.082	0.132	0.043
	Cabbage soil	0.109	0.127	0.024	0.087	0.055
	Cabbage root	N	N	0.003	0.003	
	Crisphead lettuce soil	0.139	0.142	0.181	0.154	0.023
	Jam tomato soil	0.250	0.266	0.160	0.225	0.057
	Jam tomato	0.017	0.018	0.001	0.012	0.010
Aug-09	Broccoli soil	0.124	0.139	0.123	0.129	0.009
	Broccoli root	0.025	0.026	0.021	0.024	0.003
	Broccoli stem	0.018	0.039	0.028	0.028	0.011
	Broccoli leaf	0.025	0.024	0.021	0.023	0.002
	Broccoli	0.024	0.033	0.031	0.029	0.005
	Cabbage soil	0.200	0.206	0.206	0.204	0.003
	Cabbage root	0.034	0.032	0.019	0.028	0.008
	Cabbage	0.021	0.032	0.010	0.021	0.011
	Crisphead lettuce soil	0.137	0.115	0.138	0.130	0.013
	Crisphead lettuce root	0.024	0.023	0.032	0.026	0.005
	Crisphead lettuce	0.019	0.014	0.015	0.016	0.003
	Jam tomato soil	0.117	0.095	0.205	0.139	0.058
	Jam tomato root	0.049	0.032	0.032	0.038	0.010
	Jam tomato stem	0.024	0.025	0.023	0.024	0.001
	Jam tomato leaf	0.045	0.035	0.050	0.043	0.008
	Jam tomato	0.048	0.027	0.020	0.032	0.015
Sep-09	Cabbage soil	0.131	0.137	0.160	0.143	0.015
	Cabbage root	0.029	0.049	0.069	0.049	0.020
	Cabbage	0.016	0.012	0.014	0.014	0.002
	Crisphead lettuce soil	0.094	0.145	0.121	0.120	0.026
	Crisphead lettuce root	0.023	0.027	0.024	0.025	0.002
	Crisphead lettuce	0.014	0.012	0.014	0.013	0.001
	Jam tomato soil	0.100	0.127	0.122	0.116	0.014
	Jam tomato root	0.024	0.021	0.020	0.022	0.002
	Jam tomato stem	0.017	0.016	0.027	0.020	0.006
	Jam tomato leaf	0.019	0.021	0.019	0.020	0.001
	Jam tomato	0.017	0.018	0.015	0.017	0.002
Oct-09	Broccoli soil	0.025	0.031	0.032	0.029	0.004
	Broccoli root	0.048	0.043	0.066	0.052	0.012
	Broccoli stem	0.033	0.027	0.026	0.029	0.004
	Broccoli leaf	0.024	0.026	0.024	0.025	0.001
	Broccoli	0.166	0.188	0.162	0.172	0.014
	Cabbage soil	0.089	0.096	0.104	0.096	0.008
	Cabbage root	0.090	0.061	0.068	0.073	0.015
	Cabbage	0.029	0.028	0.029	0.029	0.001
	Crisphead lettuce soil	0.029	0.029	0.075	0.044	0.027
	Crisphead lettuce root	0.043	0.120	0.087	0.083	0.039
	Crisphead lettuce	0.026	0.026	0.023	0.025	0.002
	Spinach soil	0.295	0.170	0.137	0.201	0.083
	Spinach root	0.034	0.036	0.053	0.041	0.010
	Spinach stem	0.031	0.028	0.028	0.029	0.002
	Spinach	0.027	0.025	0.027	0.026	0.001
Nov-09	Spinach soil	0.076	0.107	0.102	0.095	0.017
	Spinach root	0.067	0.062	0.029	0.053	0.021
	Spinach stem	0.017	0.021	0.017	0.018	0.002
	Spinach	0.017	0.023	0.020	0.020	0.003
Dec-09	Spinach soil	0.068	0.100	0.075	0.081	0.017

Table B49/ Cont.

Sampling period	Samples	Concentration of Pb (mg/L)				
		A	B	C	Average	SD
	Spinach stem	0.008	0.013	0.008	0.010	0.003
	Spinach	0.011	0.017	0.004	0.011	0.007
Jan-10	Cabbage soil	0.078	0.072	0.070	0.073	0.004
	Cabbage root	0.052	0.020	0.022	0.031	0.018
	Cabbage	0.016	0.018	0.018	0.017	0.001
	Crisphead lettuce soil	0.058	0.055	0.060	0.058	0.003
	Crisphead lettuce root	0.024	0.019	N	0.022	
	Crisphead lettuce	0.025	0.028	0.025	0.026	0.002
	Spinach soil	0.056	0.066	0.073	0.065	0.009
	Spinach root	0.053	0.026	0.055	0.045	0.016
	Spinach stem	0.023	0.018	0.018	0.020	0.003
	Spinach	0.026	0.024	0.024	0.025	0.001
Feb-10	Crisphead lettuce soil	0.082	0.083	0.111	0.092	0.016
	Crisphead lettuce root	0.008	0.012	0.013	0.011	0.003
	Crisphead lettuce	0.008	0.010	0.009	0.009	0.001
	Spinach soil	0.080	0.090	0.095	0.088	0.008
	Spinach root	0.015	0.004	0.010	0.010	0.006
	Spinach stem	0.009	0.031	0.005	0.015	0.014
	Spinach	0.010	0.021	0.018	0.016	0.006
Mar-10	Cabbage soil	0.057	0.062	0.074	0.064	0.009
	Cabbage root	0.002	0.013	0.010	0.008	0.006
	Crisphead lettuce soil	0.042	0.057	0.035	0.045	0.011
	Crisphead lettuce root	N	0.006	N	0.006	
	Spinach soil	0.041	0.101	0.030	0.057	0.038
	Spinach root	0.011	0.014	0.003	0.009	0.006
	Spinach stem	0.025	N	N	0.025	
Apr-10	Crisphead lettuce soil	N	0.012	0.010	0.011	
	Crisphead lettuce root	0.038	0.030	0.036	0.035	0.004
	Crisphead lettuce	0.035	0.023	0.044	0.034	0.011
	Jam tomato soil	N	0.003	0.004	0.004	
	Jam tomato root	0.021	0.015	0.021	0.019	0.003
	Jam tomato stem	0.008	0.007	0.005	0.007	0.002
	Jam tomato leaf	0.016	0.039	0.014	0.023	0.014
	Jam tomato	0.014	0.013	0.021	0.016	0.004
	Spinach soil	0.012	0.012	0.009	0.011	0.002
	Spinach root	0.018	0.011	0.033	0.021	0.011
	Spinach	0.022	0.029	0.013	0.021	0.008
May-10	Crisphead lettuce soil	0.181	0.120	0.159	0.153	0.031
	Crisphead lettuce root	0.002	0.005	0.001	0.003	0.002
	Crisphead lettuce	0.008	0.004	0.005	0.006	0.002
	Jam tomato soil	0.115	0.087	0.091	0.098	0.015
	Jam tomato root	0.039	0.043	0.046	0.043	0.004
	Jam tomato stem	0.013	0.009	0.011	0.011	0.002
	Jam tomato leaf	0.017	0.012	0.020	0.016	0.004
	Jam tomato	0.005	0.007	0.006	0.006	0.001
	Spinach soil	0.126	0.127	0.182	0.145	0.032
	Spinach root	0.030	0.035	0.031	0.032	0.003
	Spinach stem	0.006	0.004	0.002	0.004	0.002
	Spinach	0.044	0.048	0.043	0.045	0.003
Jun-10	Crisphead lettuce soil	0.161	0.162	0.167	0.163	0.003
	Crisphead lettuce root	0.010	0.015	0.017	0.014	0.004
	Crisphead lettuce	0.015	0.017	0.018	0.017	0.002
	Spinach soil	0.154	0.157	0.148	0.153	0.005
	Spinach root	0.056	0.064	0.063	0.061	0.004

Table B49/ Cont.

Sampling period	**Samples**	**Concentration of Pb (mg/L)**				
		A	**B**	**C**	**Average**	**SD**
	Spinach stem	0.007	0.007	0.007	0.007	0.000
	Spinach	0.065	0.067	0.062	0.065	0.003

N = heavy metal was not detected; SD = standard deviation; [a]detection limits for heavy metals (mg/L) were as follows: As (0.0053), Cd (0.0025), Hg (0.001), Pb (0.0042)

Table B50: Concentrations of As (mg/L) in different fresh produce collected from farm A2, over a one-year period [a].

Sampling period	Samples	Concentration of As (mg/L)				
		A	B	C	Average	SD
Jul-09	Broccoli soil	0.015	0.014	0.015	0.015	0.001
	Broccoli root	0.015	0.015	0.014	0.015	0.001
	Broccoli stem	0.015	0.015	0.015	0.015	0.000
	Broccoli leaf	0.015	0.016	0.017	0.016	0.001
	Cabbage soil	0.016	0.015	0.015	0.015	0.001
	Cabbage root	0.022	0.026	0.023	0.024	0.002
	Cabbage	0.017	0.016	0.017	0.017	0.001
	Crisphead lettuce soil	0.017	0.015	0.017	0.016	0.001
	Crisphead lettuce root	0.016	0.016	0.016	0.016	0.000
	Crisphead lettuce	0.020	0.021	0.019	0.020	0.001
	Spinach soil	0.015	0.016	0.015	0.015	0.001
	Spinach root	0.016	0.015	0.017	0.016	0.001
	Spinach stem	0.016	0.015	0.017	0.016	0.001
	Spinach	0.018	0.017	0.015	0.017	0.002
Aug-09	Broccoli soil	0.016	0.022	0.015	0.018	0.004
	Broccoli root	0.004	0.004	0.004	0.004	0.000
	Broccoli stem	0.008	0.009	0.005	0.007	0.002
	Broccoli leaf	0.006	0.004	0.004	0.005	0.001
	Broccoli	0.008	0.004	0.011	0.008	0.004
	Cabbage soil	0.044	0.013	0.015	0.024	0.017
	Cabbage root	0.005	0.004	0.004	0.004	0.001
	Cabbage	0.018	0.003	0.015	0.012	0.008
	Cauliflower soil	0.010	0.009	0.009	0.009	0.001
	Cauliflower root	0.006	0.005	0.005	0.005	0.001
	Cauliflower leaf	0.004	0.003	0.003	0.003	0.001
	Cauliflower	0.004	0.005	0.007	0.005	0.002
	Crisphead lettuce soil	0.007	0.010	0.007	0.008	0.002
	Crisphead lettuce root	0.005	0.004	0.004	0.004	0.001
	Crisphead lettuce	0.004	0.004	0.003	0.004	0.001
Sep-09	Broccoli soil	0.014	0.010	0.010	0.011	0.002
	Broccoli	N	0.002	N	0.002	
	Cabbage soil	0.010	0.084	0.009	0.034	0.043
	Crisphead lettuce soil	0.014	0.012	0.010	0.012	0.002
Oct-09	Crisphead lettuce root	N	0.002	N	0.002	
	Crisphead lettuce	0.001	N	N	0.001	
	Spinach stem	N	0.006	0.007	0.007	
Nov-09	Crisphead lettuce soil	0.010	0.010	0.007	0.009	0.002
	Crisphead lettuce root	0.001	0.009	0.001	0.004	0.005
Dec-09	Crisphead lettuce soil	0.002	N	N	0.002	
	Crisphead lettuce root	N	N	0.004	0.004	
Jan-10	Broccoli soil	0.088	0.027	0.130	0.082	0.052
	Broccoli root	0.021	0.014	0.018	0.018	0.004
	Broccoli stem	0.162	0.013	N	0.088	
	Broccoli leaf	0.008	0.004	0.057	0.023	0.030
	Broccoli	N	0.007	0.005	0.006	
	Crisphead lettuce soil	0.021	0.030	0.025	0.025	0.005
	Crisphead lettuce root	0.001	0.101	0.092	0.065	0.055
	Crisphead lettuce	0.041	0.002	0.006	0.016	0.021
Apr-10	Crisphead lettuce soil	0.022	0.007	0.024	0.018	0.009
	Crisphead lettuce root	0.009	0.004	0.008	0.007	0.003
	Crisphead lettuce	0.009	0.005	0.012	0.009	0.004
May-10	Cabbage soil	0.009	0.015	0.012	0.012	0.003
	Cabbage root	0.017	0.011	0.012	0.013	0.003

Table B50/ Cont.

Sampling period	Samples	Concentration of As (mg/L)				
		A	B	C	Average	SD
Jun-10	Cabbage	0.020	0.014	0.008	0.014	0.006
	Crisphead lettuce soil	0.017	0.018	0.018	0.018	0.001
	Crisphead lettuce root	0.017	0.019	0.021	0.019	0.002
	Crisphead lettuce	0.016	0.010	0.021	0.016	0.006
	Broccoli soil	0.023	0.027	0.027	0.026	0.002
	Broccoli root	0.028	0.028	0.028	0.028	0.000
	Broccoli stem	0.024	0.025	0.021	0.023	0.002
	Broccoli leaf	0.012	0.012	0.012	0.012	0.000
	Broccoli	0.009	0.009	0.009	0.009	0.000
	Cabbage soil	0.012	0.013	0.012	0.012	0.001
	Cabbage root	0.023	0.023	0.031	0.026	0.005
	Cabbage	0.018	0.016	0.024	0.019	0.004

N = heavy metal was not detected; SD = standard deviation; [a]detection limits for heavy metals (mg/L) were as follows: As (0.0053), Cd (0.0025), Hg (0.001), Pb (0.0042)

Table B51: Concentrations of Cd (mg/L) in different fresh produce collected from farm A2, over a one-year period [a].

Sampling period	Samples	Concentration of Cd (mg/L)				
		A	B	C	Average	SD
Jul-09	Broccoli soil	0.016	0.015	0.016	0.016	0.001
	Broccoli root	0.015	0.015	0.015	0.015	0.000
	Broccoli stem	0.014	0.015	0.015	0.015	0.001
	Broccoli leaf	0.015	0.015	0.015	0.015	0.000
	Cabbage soil	0.015	0.016	0.016	0.016	0.001
	Cabbage root	0.015	0.013	0.015	0.014	0.001
	Cabbage	0.017	0.016	0.017	0.017	0.001
	Crisphead lettuce soil	0.018	0.016	0.017	0.017	0.001
	Crisphead lettuce root	0.014	0.014	0.014	0.014	0.000
	Crisphead lettuce	0.020	0.020	0.018	0.019	0.001
	Spinach soil	0.016	0.017	0.016	0.016	0.001
	Spinach root	0.015	0.015	0.015	0.015	0.000
	Spinach stem	0.014	0.015	0.014	0.014	0.001
	Spinach	0.018	0.017	0.016	0.017	0.001
Aug-09	Broccoli soil	0.012	0.017	0.010	0.013	0.004
	Broccoli stem	0.003	0.005	N	0.004	
	Broccoli leaf	0.004	0.002	0.001	0.002	0.002
	Broccoli	0.003	N	0.006	0.005	
	Cabbage soil	0.040	0.009	0.012	0.020	0.017
	Cabbage	0.011	N	0.010	0.011	
	Cauliflower soil	0.007	0.007	0.006	0.007	0.001
	Cauliflower root	0.002	N	0.001	0.002	
	Cauliflower	0.002	0.002	0.004	0.003	0.001
	Crisphead lettuce soil	0.005	0.008	0.005	0.006	0.002
	Crisphead lettuce	0.001	N	N	0.001	
Sep-09	Broccoli soil	0.014	0.010	0.010	0.011	0.002
	Cabbage soil	0.010	0.084	0.009	0.034	0.043
	Crisphead lettuce soil	0.014	0.012	0.010	0.012	0.002
Oct-09	Cabbage soil	0.006	0.009	0.006	0.007	0.002
	Cabbage root	0.005	0.004	0.004	0.004	0.001
	Cabbage	0.004	0.004	0.004	0.004	0.000
	Crisphead lettuce soil	0.010	0.007	0.007	0.008	0.002
	Crisphead lettuce root	0.004	0.004	0.004	0.004	0.000
	Crisphead lettuce	0.004	0.004	0.005	0.004	0.001
	Spinach soil	0.005	0.005	0.006	0.005	0.001
	Spinach root	0.005	0.004	0.005	0.005	0.001
	Spinach stem	0.005	0.004	0.004	0.004	0.001
	Spinach	0.004	0.004	0.004	0.004	0.000
Nov-09	Crisphead lettuce soil	0.007	0.007	0.007	0.007	0.000
	Crisphead lettuce root	0.007	0.007	0.008	0.007	0.001
	Crisphead lettuce	0.008	0.008	0.008	0.008	0.000
Jan-10	Broccoli soil	0.006	0.006	0.007	0.006	0.001
	Broccoli root	0.009	0.002	0.002	0.004	0.004
	Broccoli stem	0.006	0.006	0.006	0.006	0.000
	Broccoli leaf	0.006	0.006	0.006	0.006	0.000
	Broccoli	0.002	0.001	0.002	0.002	0.001
	Crisphead lettuce soil	0.004	0.004	0.005	0.004	0.001
	Crisphead lettuce root	0.005	0.005	0.006	0.005	0.001
	Crisphead lettuce	0.005	0.004	0.004	0.004	0.001
Apr-10	Crisphead lettuce soil	0.001	0.001	0.001	0.001	0.000
	Crisphead lettuce root	0.001	0.001	0.001	0.001	0.000
	Crisphead lettuce	0.001	0.001	0.001	0.001	0.000
May-10	Cabbage soil	0.008	0.006	0.006	0.007	0.001

Table B51/ Cont.

Sampling period	Samples	Concentration of Cd (mg/L)				
		A	B	C	Average	SD
Jun-10	Cabbage root	0.005	N	N	0.005	
	Crisphead lettuce soil	0.007	0.006	0.006	0.006	0.001
	Crisphead lettuce root	0.003	0.001	0.002	0.002	0.001
	Broccoli soil	0.006	0.006	0.006	0.006	0.000
	Broccoli root	0.016	0.018	0.017	0.017	0.001
	Broccoli stem	0.001	N	0.001	0.001	
	Broccoli leaf	0.012	0.013	0.009	0.011	0.002
	Broccoli	0.007	0.007	0.005	0.006	0.001
	Cabbage soil	0.009	0.006	0.008	0.008	0.002
	Cabbage root	0.005	0.005	0.009	0.006	0.002
	Cabbage	0.001	0.001	0.001	0.001	0.000

N = heavy metal was not detected; SD = standard deviation; [a]detection limits for heavy metals (mg/L) were as follows: As (0.0053), Cd (0.0025), Hg (0.001), Pb (0.0042)

Table B52: Concentrations of Hg (mg/L) in different fresh produce collected from farm A2, over a one-year period [a].

Sampling period	Samples	Concentration of Hg (mg/L)				
		A	B	C	Average	SD
Jul-09	Broccoli soil	0.006	0.006	0.007	0.006	0.001
	Broccoli root	0.006	0.006	0.007	0.006	0.001
	Broccoli stem	0.005	0.006	0.006	0.006	0.001
	Broccoli leaf	0.006	0.005	0.003	0.005	0.002
	Cabbage soil	0.006	0.007	0.006	0.006	0.001
	Cabbage	0.008	0.007	0.007	0.007	0.001
	Crisphead lettuce soil	0.007	0.007	0.007	0.007	0.000
	Crisphead lettuce root	0.003	0.003	0.005	0.004	0.001
	Crisphead lettuce	0.004	0.004	0.005	0.004	0.001
	Spinach soil	0.006	0.005	0.006	0.006	0.001
	Spinach root	0.004	0.006	0.003	0.004	0.002
	Spinach stem	0.004	0.005	0.003	0.004	0.001
	Spinach	0.005	0.006	0.006	0.006	0.001
Aug-09	Broccoli soil	0.026	0.019	0.015	0.020	0.006
	Broccoli root	0.051	0.018	0.016	0.028	0.020
	Broccoli stem	0.029	0.032	0.018	0.026	0.007
	Broccoli leaf	0.020	0.021	0.040	0.027	0.011
	Broccoli	0.020	0.020	0.036	0.025	0.009
	Cabbage soil	0.019	0.019	0.018	0.019	0.001
	Cabbage root	0.026	0.019	0.021	0.022	0.004
	Cabbage	0.034	0.020	0.020	0.025	0.008
	Cauliflower soil	0.013	0.012	0.016	0.014	0.002
	Cauliflower root	0.054	0.028	0.026	0.036	0.016
	Cauliflower leaf	0.111	0.023	0.017	0.050	0.053
	Cauliflower	0.021	0.019	0.028	0.023	0.005
	Crisphead lettuce soil	0.021	0.014	0.017	0.017	0.004
	Crisphead lettuce root	0.017	0.053	0.019	0.030	0.020
	Crisphead lettuce	0.019	0.016	0.032	0.022	0.009
Sep-09	Broccoli soil	0.008	0.006	0.006	0.007	0.001
	Broccoli root	0.024	0.022	0.024	0.023	0.001
	Broccoli stem	0.024	0.024	0.024	0.024	0.000
	Broccoli leaf	0.025	0.021	0.023	0.023	0.002
	Broccoli	0.023	0.024	0.026	0.024	0.002
	Cabbage soil	0.008	0.008	0.007	0.008	0.001
	Cabbage root	0.024	0.023	0.023	0.023	0.001
	Crisphead lettuce soil	0.009	0.006	0.006	0.007	0.002
	Crisphead lettuce root	0.022	0.025	0.024	0.024	0.002
	Crisphead lettuce	0.022	0.026	0.028	0.025	0.003
Oct-09	Cabbage soil	0.013	0.013	0.013	0.013	0.000
	Cabbage root	0.022	0.017	0.015	0.018	0.004
	Cabbage	0.017	0.016	0.018	0.017	0.001
	Crisphead lettuce soil	0.034	0.017	0.017	0.023	0.010
	Crisphead lettuce root	0.015	0.016	0.014	0.015	0.001
	Crisphead lettuce	0.017	0.017	0.018	0.017	0.001
	Spinach soil	0.016	0.015	0.013	0.015	0.002
	Spinach root	0.022	0.017	0.018	0.019	0.003
	Spinach stem	0.016	0.016	0.016	0.016	0.000
	Spinach	0.015	0.013	0.013	0.014	0.001
Nov-09	Crisphead lettuce soil	0.009	0.010	0.010	0.010	0.001
	Crisphead lettuce root	0.009	0.012	0.008	0.010	0.002
	Crisphead lettuce	0.021	0.014	0.019	0.018	0.004
Dec-09	Crisphead lettuce	0.009	N	N	0.009	
Jan-10	Broccoli soil	0.008	0.011	0.004	0.008	0.004

Table B52/ Cont.

Sampling period	Samples	Concentration of Hg (mg/L)				
		A	B	C	Average	SD
	Broccoli root	0.002	N	0.001	0.002	
	Broccoli stem	0.005	0.008	0.010	0.008	0.003
	Broccoli leaf	0.009	0.009	0.010	0.009	0.001
	Crisphead lettuce soil	0.005	0.004	0.006	0.005	0.001
	Crisphead lettuce root	0.010	0.007	0.009	0.009	0.002
	Crisphead lettuce	0.002	0.006	0.005	0.004	0.002
Apr-10	Crisphead lettuce soil	0.019	0.025	0.024	0.023	0.003
	Crisphead lettuce root	0.033	0.027	0.026	0.029	0.004
	Crisphead lettuce	0.029	0.025	0.029	0.028	0.002

N = heavy metal was not detected; SD = standard deviation; [a]detection limits for heavy metals (mg/L) were as follows: As (0.0053), Cd (0.0025), Hg (0.001), Pb (0.0042)

Table B53: Concentrations of Pb (mg/L) in different fresh produce collected from farm A2, over a one-year period [a].

Sampling period	Samples	Concentration of Pb (mg/L)				
		A	B	C	Average	SD
Jul-09	Broccoli soil	0.031	0.020	0.058	0.036	0.020
	Broccoli leaf	N	0.032	N	0.032	
	Cabbage soil	0.021	0.024	0.010	0.018	0.007
	Crisphead lettuce soil	0.045	0.035	0.069	0.050	0.017
	Spinach soil	0.059	0.073	0.051	0.061	0.011
Aug-09	Broccoli soil	0.261	0.217	0.358	0.279	0.072
	Broccoli root	0.019	0.020	0.016	0.018	0.002
	Broccoli stem	0.034	0.028	0.028	0.030	0.003
	Broccoli leaf	0.026	0.023	0.022	0.024	0.002
	Broccoli	0.021	0.018	0.032	0.024	0.007
	Cabbage soil	0.139	0.132	0.108	0.126	0.016
	Cabbage root	0.030	0.020	0.019	0.023	0.006
	Cabbage	0.029	0.019	0.025	0.024	0.005
	Cauliflower soil	0.146	0.164	0.123	0.144	0.021
	Cauliflower root	0.036	0.024	0.032	0.031	0.006
	Cauliflower leaf	0.017	0.012	0.015	0.015	0.003
	Cauliflower	0.030	0.021	0.027	0.026	0.005
	Crisphead lettuce soil	0.114	0.112	0.108	0.111	0.003
	Crisphead lettuce root	0.026	0.035	0.017	0.026	0.009
	Crisphead lettuce	0.020	0.016	0.020	0.019	0.002
Sep-09	Broccoli soil	0.127	0.092	0.109	0.109	0.018
	Broccoli root	0.048	0.074	0.051	0.058	0.014
	Broccoli stem	0.013	0.014	0.011	0.013	0.002
	Broccoli leaf	0.015	0.019	0.025	0.020	0.005
	Broccoli	0.015	0.015	0.013	0.014	0.001
	Cabbage soil	0.051	0.063	0.090	0.068	0.020
	Cabbage root	0.019	0.015	0.042	0.025	0.015
	Crisphead lettuce soil	0.107	0.109	0.077	0.098	0.018
	Crisphead lettuce root	0.027	0.027	0.033	0.029	0.003
	Crisphead lettuce	0.022	0.018	0.019	0.020	0.002
Oct-09	Cabbage soil	0.096	0.109	0.096	0.100	0.008
	Cabbage root	0.099	0.063	0.066	0.076	0.020
	Cabbage	0.032	0.025	0.041	0.033	0.008
	Crisphead lettuce soil	0.080	0.072	0.078	0.077	0.004
	Crisphead lettuce root	0.059	0.052	0.052	0.054	0.004
	Crisphead lettuce	0.029	0.029	0.075	0.044	0.027
	Spinach soil	0.088	0.084	0.089	0.087	0.003
	Spinach root	0.060	0.047	0.061	0.056	0.008
	Spinach stem	0.030	0.027	0.027	0.028	0.002
	Spinach	0.028	0.030	0.028	0.029	0.001
Nov-09	Crisphead lettuce soil	0.056	0.034	0.036	0.042	0.012
	Crisphead lettuce root	0.055	0.047	0.033	0.045	0.011
	Crisphead lettuce	0.027	0.027	0.029	0.028	0.001
Dec-09	Crisphead lettuce soil	0.049	0.031	0.022	0.034	0.014
	Crisphead lettuce root	0.058	0.046	0.048	0.051	0.006
	Crisphead lettuce	0.021	0.021	0.022	0.021	0.001
Jan-10	Broccoli soil	0.051	0.053	0.063	0.056	0.006
	Broccoli root	0.054	0.026	0.027	0.036	0.016
	Broccoli stem	0.022	0.022	0.023	0.022	0.001
	Broccoli leaf	0.024	0.026	0.028	0.026	0.002
	Broccoli	0.021	0.026	0.021	0.023	0.003
	Crisphead lettuce soil	0.047	0.049	0.053	0.050	0.003
	Crisphead lettuce root	0.021	0.031	0.026	0.026	0.005

Table B53/ Cont.

Sampling period	Samples	Concentration of Pb (mg/L)				
		A	B	C	Average	SD
	Crisphead lettuce	0.025	0.030	0.023	0.026	0.004
Apr-10	Crisphead lettuce soil	0.004	0.012	0.011	0.009	0.004
	Crisphead lettuce root	0.025	0.009	0.014	0.016	0.008
	Crisphead lettuce	0.011	0.008	0.011	0.010	0.002
May-10	Cabbage soil	0.035	0.048	0.033	0.039	0.008
	Cabbage root	0.046	0.040	0.055	0.047	0.008
	Cabbage	0.004	0.005	0.008	0.006	0.002
	Crisphead lettuce soil	0.063	0.040	0.053	0.052	0.012
	Crisphead lettuce root	0.026	0.013	0.028	0.022	0.008
	Crisphead lettuce	0.007	0.011	0.006	0.008	0.003
Jun-10	Broccoli soil	0.075	0.075	0.073	0.074	0.001
	Broccoli root	0.036	0.038	0.042	0.039	0.003
	Broccoli stem	0.009	0.009	0.013	0.010	0.002
	Broccoli leaf	0.010	0.012	0.013	0.012	0.002
	Broccoli	0.008	0.007	0.008	0.008	0.001
	Cabbage soil	0.045	0.053	0.055	0.051	0.005
	Cabbage root	0.050	0.056	0.054	0.053	0.003
	Cabbage	0.007	0.006	0.007	0.007	0.001

N = heavy metal was not detected; SD = standard deviation; [a]detection limits for heavy metals (mg/L) were as follows: As (0.0053), Cd (0.0025), Hg (0.001), Pb (0.0042)

Table B54: Concentrations of As (mg/L) in different fresh produce collected from farm B, over a one-year period [a].

Sampling period	Samples	Concentration of As (mg/L)				
		A	B	C	Average	SD
Jul-09	Broccoli soil	0.032	0.036	0.032	0.033	0.002
	Broccoli root	0.027	0.028	0.028	0.028	0.001
	Broccoli stem	0.027	0.027	0.028	0.027	0.001
	Broccoli leaf	0.028	0.027	0.027	0.027	0.001
	Broccoli	0.031	0.028	0.028	0.029	0.002
	Red Cabbage soil	0.042	0.040	0.041	0.041	0.001
	Red Cabbage root	0.028	0.029	0.028	0.028	0.001
	Red Cabbage	0.028	0.029	0.028	0.028	0.001
	Crisphead lettuce soil	0.032	0.031	0.032	0.032	0.001
	Crisphead lettuce root	0.029	0.028	0.028	0.028	0.001
	Crisphead lettuce	0.028	0.028	0.028	0.028	0.000
	Spinach soil	0.031	0.030	0.030	0.030	0.001
	Spinach root	0.028	0.028	0.028	0.028	0.000
	Spinach stem	0.029	0.028	0.028	0.028	0.001
	Spinach	0.029	0.028	0.027	0.028	0.001
Aug-09	Broccoli soil	0.053	0.025	0.021	0.033	0.017
	Broccoli root	0.007	0.006	0.007	0.007	0.001
	Broccoli stem	0.003	0.004	0.003	0.003	0.001
	Broccoli leaf	0.002	0.003	0.014	0.006	0.007
	Broccoli	0.003	0.004	0.001	0.003	0.002
	Red Cabbage soil	0.043	0.026	0.017	0.029	0.013
	Red Cabbage root	0.006	0.005	0.005	0.005	0.001
	Red Cabbage	0.001	0.001	0.001	0.001	0.000
	Crisphead lettuce soil	0.026	0.024	0.029	0.026	0.003
	Crisphead lettuce root	0.006	0.007	0.006	0.006	0.001
	Crisphead lettuce	0.004	0.002	0.005	0.004	0.002
	Spinach soil	0.023	0.025	0.022	0.023	0.002
	Spinach root	0.001	0.001	N	0.001	
	Spinach stem	0.001	N	0.001	0.001	
	Spinach	0.002	0.001	0.001	0.001	0.001
Sep-09	Broccoli soil	0.030	0.032	0.035	0.032	0.003
	Broccoli	0.019	0.017	0.018	0.018	0.001
	Chinese cabbage	0.015	0.012	0.016	0.014	0.002
	Red Cabbage	0.015	0.016	0.015	0.015	0.001
	Spinach stem	0.023	0.008	0.005	0.012	0.010
Oct-09	Red Cabbage	0.001	N	N	0.001	
	Crisphead lettuce soil	0.012	N	N	0.012	
	Crisphead lettuce root	N	0.004	N	0.004	
	Parsley root	0.003	0.023	0.008	0.011	0.010
	Spinach root	N	0.003	N	0.003	
Nov-09	Cabbage soil	0.010	0.029	0.022	0.020	0.010
	Cabbage root	0.018	0.011	0.018	0.016	0.004
	Cabbage	0.018	0.016	0.001	0.012	0.009
	Parsley soil	0.010	0.016	0.017	0.014	0.004
	Parsley stem	0.010	0.011	0.014	0.012	0.002
	Parsley root	N	0.007	0.050	0.029	
	Spinach soil	0.010	0.013	0.002	0.008	0.006
	Spinach root	0.010	0.015	0.015	0.013	0.003
	Spinach stem	N	0.005	0.011	0.008	0.004
	Spinach	0.001	0.004	N	0.003	
Dec-09	Cabbage soil	0.007	0.016	N	0.012	
	Cabbage root	0.070	0.003	0.015	0.029	0.036
	Cabbage	0.004	0.014	0.006	0.008	0.005
	Crisphead lettuce soil	0.001	0.001	0.002	0.001	0.001

Table B54/ Cont.

Sampling period	Samples	Concentration of As (mg/L)				
		A	B	C	Average	SD
	Crisphead lettuce root	0.010	0.012	0.010	0.011	0.001
	Crisphead lettuce	0.015	0.005	0.007	0.009	0.005
	Spinach soil	0.009	N	0.009	0.009	
	Spinach root	0.007	0.010	0.010	0.009	0.002
	Spinach stem	0.003	N	0.003	0.003	
	Spinach	N	N	0.003	0.003	
Jan-10	Cabbage soil	0.056	0.010	0.056	0.041	0.027
	Cabbage root	N	N	0.011	0.011	
	Cabbage	0.118	0.036	0.013	0.056	0.055
	Spinach soil	0.017	0.016	0.017	0.017	0.001
	Spinach root	0.017	0.017	0.019	0.018	0.001
	Spinach stem	0.010	0.012	0.026	0.016	0.009
	Spinach	0.009	0.011	0.019	0.013	0.005
Feb-10	Bell pepper root	N	0.011	0.013	0.012	
	Bell pepper stem	N	0.003	0.001	0.002	
	Bell pepper leaf	0.005	N	0.008	0.007	
	Bell pepper	0.009	N	0.014	0.012	
Mar-10	Broccoli soil	0.012	0.012	N	0.012	
	Broccoli root	0.007	0.031	0.010	0.016	0.013
	Broccoli stem	0.003	0.004	N	0.004	
	Broccoli leaf	0.005	0.001	0.006	0.004	0.003
	Broccoli	0.010	0.010	0.013	0.011	0.002
	Bell pepper soil	N	0.010	N	0.010	
	Bell pepper root	0.015	N	0.002	0.009	
	Bell pepper stem	0.014	0.001	0.010	0.008	0.007
	Bell pepper leaf	0.002	0.005	0.007	0.005	0.003
	Bell pepper	0.007	0.002	0.009	0.006	0.004
Apr-10	Crisphead lettuce soil	0.011	0.001	0.016	0.009	0.008
	Crisphead lettuce root	0.010	0.017	0.012	0.013	0.004
	Crisphead lettuce	0.015	0.002	0.019	0.012	0.009
	Bell pepper soil	0.018	0.012	0.018	0.016	0.003
	Bell pepper root	0.012	0.014	0.008	0.011	0.003
	Bell pepper stem	0.014	0.013	0.012	0.013	0.001
	Bell pepper leaf	0.014	0.014	0.009	0.012	0.003
	Bell pepper	0.023	0.021	0.015	0.020	0.004
	Spinach soil	0.005	0.004	0.002	0.004	0.002
	Spinach root	0.005	0.005	0.005	0.005	0.000
	Spinach	0.009	0.003	0.009	0.007	0.003
May-10	Crisphead lettuce soil	0.003	0.002	0.002	0.002	0.001
	Crisphead lettuce root	0.017	0.027	0.033	0.026	0.008
	Crisphead lettuce	0.008	0.014	0.014	0.012	0.003
	Bell Pepper soil	0.021	0.024	0.016	0.020	0.004
	Bell Pepper root	0.014	0.022	0.026	0.021	0.006
	Bell Pepper stem	0.017	0.010	0.022	0.016	0.006
	Bell Pepper leaf	0.030	0.020	0.020	0.023	0.006
	Bell Pepper	0.023	0.013	0.018	0.018	0.005
Jun-10	Cabbage soil	0.017	0.020	0.022	0.020	0.003
	Cabbage root	0.023	0.028	0.026	0.026	0.003
	Cabbage	0.010	0.011	0.017	0.013	0.004
	Crisphead lettuce soil	0.026	0.028	0.028	0.027	0.001
	Crisphead lettuce root	0.015	0.017	0.015	0.016	0.001
	Crisphead lettuce	0.014	0.025	0.016	0.018	0.006

N = heavy metal was not detected; SD = standard deviation; [a]detection limits for heavy metals (mg/L) were as follows: As (0.0053), Cd (0.0025), Hg (0.001), Pb (0.0042)

Table B55: Concentrations of Cd (mg/L) in different fresh produce collected from farm B, over a one-year period [a].

Sampling period	Samples	Concentration of Cd (mg/L)				
		A	B	C	Average	SD
Jul-09	Broccoli soil	0.033	0.038	0.033	0.035	0.003
	Broccoli root	0.027	0.027	0.027	0.027	0.000
	Broccoli stem	0.027	0.027	0.027	0.027	0.000
	Broccoli leaf	0.027	0.027	0.027	0.027	0.000
	Broccoli	0.027	0.027	0.027	0.027	0.000
	Red Cabbage soil	0.042	0.040	0.042	0.041	0.001
	Red Cabbage root	0.027	0.027	0.027	0.027	0.000
	Red Cabbage	0.027	0.026	0.026	0.026	0.001
	Crisphead lettuce soil	0.033	0.032	0.032	0.032	0.001
	Crisphead lettuce root	0.026	0.027	0.027	0.027	0.001
	Crisphead lettuce	0.027	0.027	0.027	0.027	0.000
	Spinach soil	0.030	0.030	0.029	0.030	0.001
	Spinach root	0.027	0.027	0.027	0.027	0.000
	Spinach stem	0.027	0.027	0.027	0.027	0.000
	Spinach	0.027	0.027	0.027	0.027	0.000
Aug-09	Broccoli soil	0.057	0.028	0.024	0.036	0.018
	Broccoli root	0.010	0.010	0.010	0.010	0.000
	Broccoli stem	0.005	0.004	0.003	0.004	0.001
	Broccoli leaf	0.004	0.003	0.016	0.008	0.007
	Broccoli	0.004	0.005	0.003	0.004	0.001
	Red Cabbage soil	0.051	0.032	0.021	0.035	0.015
	Red Cabbage root	0.010	0.010	0.011	0.010	0.001
	Red Cabbage	0.003	0.003	0.003	0.003	0.000
	Crisphead lettuce soil	0.032	0.030	0.036	0.033	0.003
	Crisphead lettuce root	0.010	0.010	0.010	0.010	0.000
	Crisphead lettuce	0.004	0.003	0.005	0.004	0.001
	Spinach soil	0.026	0.027	0.025	0.026	0.001
	Spinach root	0.003	0.003	0.003	0.003	0.000
	Spinach stem	0.003	0.003	0.002	0.003	0.001
	Spinach	0.004	0.002	0.003	0.003	0.001
Sep-09	Broccoli soil	0.030	0.032	0.035	0.032	0.003
	Broccoli	0.019	0.017	0.018	0.018	0.001
	Chinese cabbage	0.015	0.012	0.016	0.014	0.002
	Red Cabbage	0.015	0.016	0.015	0.015	0.001
	Spinach stem	0.023	0.008	0.005	0.012	0.010
Oct-09	Red Cabbage soil	0.007	0.001	0.007	0.005	0.003
	Red Cabbage root	0.006	0.009	0.010	0.008	0.002
	Red Cabbage	0.005	0.005	0.006	0.005	0.001
	Crisphead lettuce soil	0.007	0.005	0.009	0.007	0.002
	Crisphead lettuce root	0.006	0.005	0.005	0.005	0.001
	Crisphead lettuce	0.005	0.007	0.005	0.006	0.001
	Parsley soil	0.017	0.018	0.013	0.016	0.003
	Parsley stem	0.013	0.007	0.006	0.009	0.004
	Parsley root	0.007	0.006	0.005	0.006	0.001
	Parsley	0.005	0.005	0.007	0.006	0.001
	Spinach soil	0.011	0.014	0.006	0.010	0.004
	Spinach root	0.006	0.006	0.005	0.006	0.001
	Spinach	0.006	0.005	0.006	0.006	0.001
Nov-09	Cabbage soil	0.009	0.012	0.014	0.012	0.003
	Cabbage root	0.007	0.007	0.008	0.007	0.001
	Cabbage	0.007	0.007	0.007	0.007	0.000
	Parsley soil	0.022	0.027	0.017	0.022	0.005
	Parsley stem	0.008	0.007	0.007	0.007	0.001

Table B55/ Cont.

Sampling period	Samples	Concentration of Cd (mg/L)				
		A	B	C	Average	SD
	Parsley root	0.007	0.008	0.008	0.008	0.001
	Parsley	0.008	0.009	0.008	0.008	0.001
	Spinach soil	0.010	0.009	0.033	0.017	0.014
	Spinach root	0.007	0.007	0.008	0.007	0.001
	Spinach stem	0.007	0.007	0.007	0.007	0.000
	Spinach	0.007	0.007	0.007	0.007	0.000
Dec-09	Cabbage soil	N	0.001	0.002	0.002	
	Crisphead lettuce soil	0.009	0.005	0.007	0.007	0.002
Jan-10	Cabbage soil	0.006	0.006	0.006	0.006	0.000
	Cabbage root	0.001	0.003	0.001	0.002	0.001
	Cabbage	0.006	0.004	0.004	0.005	0.001
	Spinach soil	0.005	0.005	0.005	0.005	0.000
	Spinach root	0.002	0.001	0.002	0.002	0.001
	Spinach stem	0.004	0.004	0.005	0.004	0.001
	Spinach	0.005	0.005	0.005	0.005	0.000
Feb-10	Bell pepper soil	0.024	0.032	0.023	0.026	0.005
	Bell pepper leaf	0.008	N	N	0.008	
Mar-10	Broccoli soil	0.016	0.019	0.017	0.017	0.002
	Broccoli root	0.001	N	0.001	0.001	
	Broccoli stem	0.001	0.001	0.001	0.001	0.000
	Broccoli leaf	0.010	0.012	0.011	0.011	0.001
	Bell pepper soil	0.023	0.018	0.022	0.021	0.003
	Bell pepper root	N	0.001	0.002	0.002	
	Bell pepper stem	0.001	0.001	N	0.001	
	Bell pepper leaf	0.002	0.001	N	0.002	
	Bell pepper	0.001	0.001	0.002	0.001	0.001
Apr-10	Crisphead lettuce soil	0.004	0.003	0.004	0.004	0.001
	Crisphead lettuce root	0.001	0.002	0.001	0.001	0.001
	Crisphead lettuce	0.001	0.001	0.001	0.001	0.000
	Bell pepper soil	0.001	0.001	0.001	0.001	0.000
	Bell pepper root	0.002	0.002	0.002	0.002	0.000
	Bell pepper stem	0.001	0.002	0.002	0.002	0.001
	Bell pepper leaf	0.001	0.001	0.001	0.001	0.000
	Bell pepper	0.004	0.005	0.007	0.005	0.002
	Spinach soil	0.002	0.002	0.002	0.002	0.000
	Spinach root	0.002	0.002	0.002	0.002	0.000
	Spinach	0.001	0.003	0.002	0.002	0.001
May-10	Crisphead lettuce soil	0.019	0.026	0.017	0.021	0.005
	Crisphead lettuce root	0.002	N	0.002	0.002	
	Crisphead lettuce	0.002	0.001	0.005	0.003	0.002
	Bell Pepper soil	0.028	0.018	0.018	0.021	0.006
	Bell Pepper root	0.003	0.002	0.003	0.003	0.001
	Bell Pepper stem	0.001	N	0.003	0.002	
	Bell Pepper leaf	0.003	0.006	0.001	0.003	0.003
	Bell Pepper	0.004	0.003	0.002	0.003	0.001
Jun-10	Cabbage soil	0.018	0.017	0.017	0.017	0.001
	Cabbage root	0.003	0.003	0.003	0.003	0.000
	Cabbage	0.006	0.006	0.006	0.006	0.000
	Crisphead lettuce soil	0.032	0.034	0.034	0.033	0.001
	Crisphead lettuce root	0.005	0.005	0.004	0.005	0.001
	Crisphead lettuce	N	0.001	N	0.001	

N = heavy metal was not detected; SD = standard deviation; [a]detection limits for heavy metals (mg/L) were as follows: As (0.0053), Cd (0.0025), Hg (0.001), Pb (0.0042)

Table B56: Concentrations of Hg (mg/L) in different fresh produce collected from farm B, over a one-year period [a].

Sampling period	Samples	Concentration of Hg (mg/L)				
		A	B	C	Average	SD
Jul-09	Broccoli soil	0.033	0.033	0.034	0.033	0.001
	Broccoli root	0.034	0.034	0.033	0.034	0.001
	Broccoli stem	0.034	0.034	0.034	0.034	0.000
	Broccoli leaf	0.034	0.034	0.034	0.034	0.000
	Broccoli	0.035	0.034	0.034	0.034	0.001
	Red Cabbage soil	0.034	0.034	0.033	0.034	0.001
	Red Cabbage root	0.034	0.032	0.033	0.033	0.001
	Red Cabbage	0.033	0.032	0.032	0.032	0.001
	Crisphead lettuce soil	0.034	0.034	0.034	0.034	0.000
	Crisphead lettuce root	0.032	0.033	0.032	0.032	0.001
	Crisphead lettuce	0.034	0.034	0.033	0.034	0.001
	Spinach soil	0.034	0.034	0.033	0.034	0.001
	Spinach root	0.034	0.032	0.033	0.033	0.001
	Spinach stem	0.033	0.033	0.034	0.033	0.001
	Spinach	0.033	0.034	0.034	0.034	0.001
Aug-09	Broccoli soil	N	0.075	0.051	0.063	
	Broccoli root	0.033	0.034	0.034	0.034	0.001
	Broccoli stem	0.044	0.042	0.036	0.041	0.004
	Broccoli leaf	0.031	0.032	0.032	0.032	0.001
	Broccoli	0.035	0.033	0.031	0.033	0.002
	Red Cabbage soil	0.027	0.026	0.023	0.025	0.002
	Red Cabbage root	0.038	0.033	0.035	0.035	0.003
	Red Cabbage	0.027	0.025	0.025	0.026	0.001
	Crisphead lettuce soil	0.026	0.023	0.024	0.024	0.002
	Crisphead lettuce root	0.035	0.035	0.036	0.035	0.001
	Crisphead lettuce	0.027	0.027	0.025	0.026	0.001
	Spinach soil	0.031	0.029	0.032	0.031	0.002
	Spinach root	0.031	0.027	0.029	0.029	0.002
	Spinach stem	0.029	0.029	0.047	0.035	0.010
	Spinach	0.029	0.03	0.032	0.030	0.002
Sep-09	Broccoli soil	0.006	0.006	0.004	0.005	0.001
	Broccoli	0.004	0.004	0.005	0.004	0.001
	Chinese cabbage	0.004	0.006	0.004	0.005	0.001
	Red Cabbage	0.001	0.003	0.005	0.003	0.002
	Spinach root	0.021	N	N	0.021	
	Spinach stem	0.017	0.01	0.007	0.011	0.005
	Spinach	0.028	0.024	0.027	0.026	0.002
Oct-09	Red Cabbage soil	0.010	0.009	0.011	0.010	0.001
	Red Cabbage root	0.022	0.014	0.019	0.018	0.004
	Red Cabbage	0.017	0.017	0.021	0.018	0.002
	Crisphead lettuce soil	0.011	0.009	0.007	0.009	0.002
	Crisphead lettuce root	0.015	0.016	0.014	0.015	0.001
	Crisphead lettuce	0.016	0.016	0.019	0.017	0.002
	Parsley soil	0.013	0.01	0.01	0.011	0.002
	Parsley stem	0.018	0.014	0.015	0.016	0.002
	Parsley root	0.017	0.017	0.015	0.016	0.001
	Parsley	0.025	0.022	0.021	0.023	0.002
	Spinach soil	0.043	0.016	0.014	0.024	0.016
	Spinach root	0.017	0.016	0.016	0.016	0.001
	Spinach	0.016	0.02	0.017	0.018	0.002
Nov-09	Cabbage soil	0.009	0.008	0.012	0.010	0.002
	Cabbage root	0.009	0.01	0.01	0.010	0.001
	Cabbage	0.018	0.018	0.021	0.019	0.002

Table B56/ Cont.

Sampling period	Samples	Concentration of Hg (mg/L)				
		A	B	C	Average	SD
	Parsley soil	0.009	0.007	0.01	0.009	0.002
	Parsley stem	0.017	0.016	0.014	0.016	0.002
	Parsley root	0.011	0.012	0.011	0.011	0.001
	Parsley	0.027	0.043	0.028	0.033	0.009
	Spinach soil	0.011	0.009	0.009	0.010	0.001
	Spinach root	0.016	0.019	0.015	0.017	0.002
	Spinach stem	0.017	0.022	0.022	0.020	0.003
	Spinach	0.009	0.011	0.013	0.011	0.002
Dec-09	Cabbage root	0.014	0.024	0.025	0.021	0.006
	Crisphead lettuce root	N	0.053	0.039	0.046	
	Crisphead lettuce	0.038	0.036	0.055	0.043	0.010
	Spinach stem	N	0.01	0.004	0.007	
Jan-10	Cabbage soil	0.008	0.007	0.006	0.007	0.001
	Cabbage	0.006	0.006	0.007	0.006	0.001
	Spinach soil	0.009	0.01	0.009	0.009	0.001
	Spinach stem	0.009	0.008	0.009	0.009	0.001
	Spinach	0.08	0.028	0.021	0.043	0.032
Feb-10	Bell pepper soil	0.017	0.018	0.016	0.017	0.001
	Bell pepper root	0.034	0.018	0.018	0.023	0.009
	Bell pepper stem	0.017	0.017	0.018	0.017	0.001
	Bell pepper leaf	0.018	0.018	0.019	0.018	0.001
	Bell pepper	0.018	0.017	0.023	0.019	0.003
Mar-10	Broccoli soil	0.001	N	N	0.001	
	Broccoli root	0.004	N	0.007	0.006	
	Broccoli stem	0.005	0.004	0.004	0.004	0.001
	Broccoli leaf	0.006	0.008	0.006	0.007	0.001
	Broccoli	N	0.001	0.002	0.002	
	Bell pepper root	0.001	0.001	0.004	0.002	0.002
	Bell pepper stem	0.026	0.009	0.007	0.014	0.010
	Bell pepper leaf	N	N	0.001	0.001	
	Bell pepper	0.004	0.005	0.005	0.005	0.001
Apr-10	Crisphead lettuce soil	0.024	0.023	0.023	0.023	0.001
	Crisphead lettuce root	0.026	0.028	0.028	0.027	0.001
	Crisphead lettuce	0.025	0.026	0.028	0.026	0.002
	Bell pepper soil	0.021	0.023	0.02	0.021	0.002
	Bell pepper root	0.021	0.021	0.02	0.021	0.001
	Bell pepper stem	0.029	0.035	0.024	0.029	0.006
	Bell pepper leaf	0.022	0.025	0.021	0.023	0.002
	Bell pepper	0.047	0.044	0.042	0.044	0.003
	Spinach soil	0.015	0.014	0.013	0.014	0.001
	Spinach root	0.016	0.016	0.016	0.016	0.000
	Spinach	0.027	0.026	0.024	0.026	0.002
Jun-10	Crisphead lettuce	0.051	0.05	0.049	0.050	0.001

N = heavy metal was not detected; SD = standard deviation; ^{a}detection limits for heavy metals (mg/L) were as follows: As (0.0053), Cd (0.0025), Hg (0.001), Pb (0.0042)

Table B57: Concentrations of Pb (mg/L) in different fresh produce collected from farm B, over a one-year period ^{a}.

Sampling period	Samples	Concentration of Pb (mg/L)				
		A	B	C	Average	SD
Jul-09	Broccoli soil	0.168	0.214	0.195	0.192	0.023
	Broccoli root	0.038	0.038	0.045	0.040	0.004
	Broccoli stem	0.040	0.040	0.039	0.040	0.001
	Broccoli leaf	0.050	0.041	0.039	0.043	0.006
	Broccoli	0.032	0.039	0.039	0.037	0.004
	Red Cabbage soil	0.148	0.132	0.117	0.132	0.016
	Red Cabbage root	0.039	0.043	0.040	0.041	0.002
	Red Cabbage	0.049	0.035	0.034	0.039	0.008
	Crisphead lettuce soil	0.163	0.170	0.148	0.160	0.011
	Crisphead lettuce root	0.052	0.036	0.042	0.043	0.008
	Crisphead lettuce	0.038	0.039	0.037	0.038	0.001
	Spinach soil	0.083	0.078	0.072	0.078	0.006
	Spinach root	0.035	0.036	0.038	0.036	0.002
	Spinach stem	0.048	0.043	0.039	0.043	0.005
	Spinach	0.053	0.040	0.037	0.043	0.009
Aug-09	Broccoli soil	0.912	0.304	0.362	0.526	0.336
	Broccoli root	0.049	0.038	0.047	0.045	0.006
	Broccoli stem	0.006	0.002	0.005	0.004	0.002
	Broccoli leaf	N	0.001	0.006	0.004	
	Broccoli	0.004	0.001	N	0.003	
	Red Cabbage soil	0.221	0.021	0.163	0.135	0.103
	Red Cabbage root	0.045	0.031	0.031	0.036	0.008
	Red Cabbage	0.003	0.002	N	0.003	
	Crisphead lettuce soil	0.213	0.204	0.221	0.213	0.009
	Crisphead lettuce root	0.038	0.048	0.038	0.041	0.006
	Crisphead lettuce	0.010	0.001	0.019	0.010	0.009
	Spinach soil	0.162	0.178	0.176	0.172	0.009
	Spinach root	0.028	N	N	0.028	
	Spinach stem	N	N	0.004	0.004	
	Spinach	0.007	0.004	0.001	0.004	0.003
Sep-09	Broccoli soil	0.226	0.207	0.229	0.221	0.012
	Broccoli	0.359	0.351	0.370	0.360	0.010
	Chinese cabbage	0.283	0.199	0.325	0.269	0.064
	Red Cabbage	0.322	0.320	0.311	0.318	0.006
	Spinach root	0.013	N	N	0.013	
	Spinach stem	0.013	0.006	0.002	0.007	0.006
	Spinach	0.015	0.016	0.015	0.015	0.001
Oct-09	Red Cabbage soil	0.166	0.181	0.208	0.185	0.021
	Red Cabbage root	0.120	0.236	0.147	0.168	0.061
	Red Cabbage	0.059	0.026	0.027	0.037	0.019
	Crisphead lettuce soil	0.140	0.137	0.157	0.145	0.011
	Crisphead lettuce root	0.041	0.040	0.033	0.038	0.004
	Crisphead lettuce	0.026	0.029	0.026	0.027	0.002
	Parsley soil	0.259	0.259	0.145	0.221	0.066
	Parsley stem	0.037	0.026	0.029	0.031	0.006
	Parsley root	0.043	0.041	0.031	0.038	0.006
	Parsley	0.033	0.029	0.026	0.029	0.004

Table B57/ Cont.

Sampling period	Samples	Concentration of Pb (mg/L)				
		A	B	C	Average	SD
	Spinach soil	0.141	0.164	0.146	0.150	0.012
	Spinach root	0.053	0.053	0.034	0.047	0.011
	Spinach	0.029	0.025	0.031	0.028	0.003
Nov-09	Cabbage soil	0.036	0.073	0.101	0.070	0.033
	Cabbage root	0.027	0.022	0.042	0.030	0.010
	Cabbage	0.017	0.024	0.028	0.023	0.006
	Parsley soil	0.156	0.204	0.108	0.156	0.048
	Parsley stem	0.021	0.015	0.016	0.017	0.003
	Parsley root	0.035	0.051	0.030	0.039	0.011
	Parsley	0.051	0.057	0.097	0.068	0.025
	Spinach soil	0.066	0.067	0.074	0.069	0.004
	Spinach root	0.035	0.041	0.067	0.048	0.017
	Spinach stem	0.016	0.042	0.030	0.029	0.013
	Spinach	0.019	0.041	0.066	0.042	0.024
Dec-09	Cabbage soil	0.026	0.069	0.074	0.056	0.026
	Cabbage root	0.005	0.019	0.015	0.013	0.007
	Cabbage	0.008	0.016	0.020	0.015	0.006
	Crisphead lettuce soil	0.184	0.156	0.200	0.180	0.022
	Crisphead lettuce root	0.012	0.007	0.005	0.008	0.004
	Crisphead lettuce	0.003	0.010	0.006	0.006	0.004
	Spinach soil	0.040	0.047	0.003	0.030	0.024
	Spinach root	0.024	0.026	0.027	0.026	0.002
	Spinach stem	0.012	0.048	0.021	0.027	0.019
	Spinach	0.007	0.035	0.055	0.032	0.024
Jan-10	Cabbage soil	0.048	0.042	0.039	0.043	0.005
	Cabbage root	0.040	0.030	0.057	0.042	0.014
	Cabbage	0.027	0.020	0.018	0.022	0.005
	Spinach soil	0.028	0.035	0.031	0.031	0.004
	Spinach root	0.021	0.023	0.039	0.028	0.010
	Spinach stem	0.016	0.020	0.026	0.021	0.005
	Spinach	0.028	0.024	0.024	0.025	0.002
Feb-10	Bell pepper soil	0.160	0.156	0.148	0.155	0.006
	Bell pepper root	0.022	0.004	0.006	0.011	0.010
	Bell pepper stem	0.005	0.009	0.009	0.008	0.002
	Bell pepper leaf	0.011	0.010	0.009	0.010	0.001
	Bell pepper	0.007	0.016	0.005	0.009	0.006
Mar-10	Broccoli soil	0.087	0.108	0.106	0.100	0.012
	Broccoli root	0.034	0.019	0.057	0.037	0.019
	Broccoli stem	N	0.006	N	0.006	
	Broccoli leaf	0.051	0.061	0.054	0.055	0.005
	Broccoli	N	0.001	N	0.001	
	Bell pepper soil	0.101	0.080	0.094	0.092	0.011
	Bell pepper root	0.001	0.002	0.026	0.010	0.014
	Bell pepper stem	0.008	0.002	N	0.005	
	Bell pepper leaf	N	N	N	N	
	Bell pepper	0.001	0.002	0.002	0.002	0.001
Apr-10	Crisphead lettuce soil	0.009	0.010	0.012	0.010	0.002

Table B57/ Cont.

Sampling period	**Samples**	**Concentration of Pb (mg/L)**				
		A	**B**	**C**	**Average**	**SD**
	Crisphead lettuce root	0.023	0.054	0.021	0.033	0.019
	Crisphead lettuce	0.039	0.038	0.023	0.033	0.009
	Bell pepper soil	0.006	0.003	0.007	0.005	0.002
	Bell pepper root	0.026	0.043	0.040	0.036	0.009
	Bell pepper stem	0.008	0.021	0.019	0.016	0.007
	Bell pepper leaf	0.007	0.005	0.006	0.006	0.001
	Bell pepper	0.018	0.017	0.021	0.019	0.002
	Spinach soil	0.008	0.009	0.008	0.008	0.001
	Spinach root	0.015	0.014	0.014	0.014	0.001
	Spinach	0.022	0.029	0.013	0.021	0.008
May-10	Crisphead lettuce soil	0.154	0.159	0.158	0.157	0.003
	Crisphead lettuce root	0.018	0.011	0.021	0.017	0.005
	Crisphead lettuce	0.018	0.009	0.016	0.014	0.005
	Bell Pepper soil	0.132	0.114	0.100	0.115	0.016
	Bell Pepper root	0.035	0.033	0.039	0.036	0.003
	Bell Pepper stem	0.016	0.009	0.017	0.014	0.004
	Bell Pepper leaf	0.025	0.039	0.024	0.029	0.008
	Bell Pepper	0.024	0.021	0.032	0.026	0.006
Jun-10	Cabbage soil	0.166	0.165	0.168	0.166	0.002
	Cabbage root	0.027	0.032	0.035	0.031	0.004
	Cabbage	0.036	0.036	0.039	0.037	0.002
	Crisphead lettuce soil	0.143	0.135	0.143	0.140	0.005
	Crisphead lettuce root	0.045	0.047	0.041	0.044	0.003
	Crisphead lettuce	0.025	0.029	N	0.027	

N = heavy metal was not detected; SD = standard deviation; ^{a}detection limits for heavy metals (mg/L) were as follows: As (0.0053), Cd (0.0025), Hg (0.001), Pb (0.0042)

Table B58: Concentrations of As (mg/L) in different fresh produce collected from farm C, over a one-year period[a].

Sampling period	Samples	Concentration of As (mg/L)				
		A	B	C	Average	SD
Jul-09	Cabbage soil	0.011	0.01	0.01	0.010	0.001
	Cabbage root	0.006	0.006	0.006	0.006	0.000
Aug-09	Cabbage soil	0.073	0.015	0.015	0.034	0.033
	Cabbage root	0.001	0.002	0.001	0.001	0.001
	Cabbage	0.001	0.001	0.002	0.001	0.001
Sep-09	Cabbage	0.007	0.01	0.012	0.010	0.003
Jan-10	Jam tomato root	0.012	0.018	0.01	0.013	0.004
	Jam tomato stem	0.055	0.142	0.101	0.099	0.044
	Jam tomato leaf	0.003	0.009	0.009	0.007	0.003
Feb-10	Jam tomato soil	N	0.015	0.001	0.008	
	Jam tomato root	N	N	0.012	0.012	
	Jam tomato stem	N	0.005	0.003	0.004	
	Jam tomato leaf	0.01	0.01	0.011	0.010	0.001
	Jam tomato	0.006	0.015	N	0.011	

N = heavy metal was not detected; SD = standard deviation; [a]detection limits for heavy metals (mg/L) were as follows: As (0.0053), Cd (0.0025), Hg (0.001), Pb (0.0042)

Table B59: Concentrations of Cd (mg/L) in different fresh produce collected from farm C, over a one-year period[a].

Sampling period	Samples	Concentration of Cd (mg/L)				
		A	B	C	Average	SD
Jul-09	Cabbage soil	0.015	0.014	0.014	0.014	0.001
	Cabbage root	0.011	0.012	0.011	0.011	0.001
	Cabbage	0.011	0.011	0.012	0.011	0.001
Aug-09	Cabbage soil	0.083	0.018	0.018	0.040	0.038
	Cabbage root	0.003	0.002	0.002	0.002	0.001
	Cabbage	0.003	0.004	0.002	0.003	0.001
Sep-09	Cabbage	0.007	0.01	0.012	0.010	0.003
Jan-10	Jam tomato soil	0.005	0.006	0.006	0.006	0.001
	Jam tomato root	0.002	0.002	0.002	0.002	0.000
	Jam tomato stem	0.005	0.006	0.006	0.006	0.001
	Jam tomato leaf	0.002	0.002	0.002	0.002	0.000
Feb-10	Jam tomato soil	0.012	0.01	0.006	0.009	0.003
	Jam tomato	0.009	0.009	0.009	0.009	0.000

N = heavy metal was not detected; SD = standard deviation; [a]detection limits for heavy metals (mg/L) were as follows: As (0.0053), Cd (0.0025), Hg (0.001), Pb (0.0042)

Table B60: Concentrations of Hg (mg/L) in different fresh produce collected from farm C, over a one-year period[a].

Sampling period	Samples	Concentration of Hg (mg/L)				
		A	B	C	Average	SD
Jul-09	Cabbage soil	0.036	0.037	0.035	0.036	0.001
	Cabbage root	0.034	0.033	0.032	0.033	0.001
	Cabbage	0.033	0.034	0.034	0.034	0.001
Aug-09	Cabbage soil	0.025	0.026	0.026	0.026	0.001
	Cabbage root	0.027	0.029	0.024	0.027	0.003
	Cabbage	0.027	0.026	0.025	0.026	0.001
Sep-09	Cabbage soil	0.02	0.022	0.024	0.022	0.002
	Cabbage root	0.024	0.025	0.022	0.024	0.002
	Cabbage	0.023	0.021	0.018	0.021	0.003
Jan-10	Jam tomato root	0.016	0.012	0.018	0.015	0.003
	Jam tomato stem	0.012	0.014	0.01	0.012	0.002
	Jam tomato leaf	N	0.001	0.002	0.002	
Feb-10	Jam tomato soil	0.021	0.018	0.017	0.019	0.002
	Jam tomato root	0.018	0.018	0.017	0.018	0.001
	Jam tomato stem	0.019	0.018	0.034	0.024	0.009
	Jam tomato leaf	0.019	0.018	0.018	0.018	0.001
	Jam tomato	0.017	0.016	0.016	0.016	0.001

N = heavy metal was not detected; SD = standard deviation; [a]detection limits for heavy metals (mg/L) were as follows: As (0.0053), Cd (0.0025), Hg (0.001), Pb (0.0042)

Table B61: Concentrations of Pb (mg/L) in different fresh produce collected from farm C, over a one-year period[a].

Sampling period	Samples	Concentration of Pb (mg/L)				
		A	B	C	Average	SD
Jul-09	Cabbage soil	0.143	0.121	0.133	0.132	0.011
	Cabbage root	0.034	0.037	0.035	0.035	0.002
	Cabbage	0.045	0.031	0.046	0.041	0.008
Aug-09	Cabbage soil	0.085	0.063	0.063	0.070	0.013
	Cabbage root	0.019	0.015	0.015	0.016	0.002
Sep-09	Cabbage soil	0.012	0.016	0.017	0.015	0.003
	Cabbage root	0.019	0.037	0.019	0.025	0.010
	Cabbage	0.295	0.237	0.166	0.233	0.065
Jan-10	Jam tomato soil	0.071	0.076	0.066	0.071	0.005
	Jam tomato root	0.027	0.023	0.027	0.026	0.002
	Jam tomato stem	0.025	0.029	0.032	0.029	0.004
	Jam tomato leaf	0.022	0.019	0.018	0.020	0.002
Feb-10	Jam tomato soil	0.137	0.108	0.088	0.111	0.025
	Jam tomato root	0.070	0.050	0.033	0.051	0.019
	Jam tomato stem	0.007	0.003	0.012	0.007	0.005
	Jam tomato leaf	0.006	0.004	0.003	0.004	0.002
	Jam tomato	0.093	0.101	0.098	0.097	0.004

N = heavy metal was not detected; SD = standard deviation; [a]detection limits for heavy metals (mg/L) were as follows: As (0.0053), Cd (0.0025), Hg (0.001), Pb (0.0042)

Table B62: The effect of *P. aeruginosa*, at varying concentrations, on *L. monocytogenes*.

	Diameter of zone of inhibition (mm)				
	10^4	**10^5**	**10^6**	**10^7**	**10^8**
	N	N	N	11	14
	N	N	N	11	14
	N	N	N	12	14
	N	N	N	12	14
	N	N	N	12	14
	N	N	N	11	14
Average				11.50	14.00
SD				0.55	0.00

N = no inhibition was observed; SD = standard deviation

Table B63: The effect of temperature on the inhibitory activity of *P. aeruginosa* against *L. monocytogenes*.

	Diameter of zone of inhibition (mm)				
Temperature (°C)	**10**	**15**	**20**	**25**	**30**
	9	10	11	11	12
	9	10	10.5	11	12
	10	10	11	12	11
	10	11	11	12	12
	10	11	11	12	12
	10	11	11	12	12
Average	9.67	10.50	10.92	11.67	11.83
SD	0.52	0.55	0.20	0.52	0.41

SD = standard deviation

Table B64: The effect of pH on the inhibitory activity of *P. aeruginosa* against *L. monocytogenes*.

	Diameter of zone of inhibition (mm)		
pH	**5**	**7**	**9**
	10	12	11
	9	12	11
	9	12	11
	9	12	11
	9	11.5	11
	9	11.5	11
Average	9.17	11.83	11.00
SD	0.41	0.26	0.00

SD = standard deviation

Table B65: The presence of Total Heterotrophic bacteria (THB) and *P. aeruginosa* populations in non-autoclaved soil at week 0, before spiking, in the greenhouse experiment.

Microbial population	**cfu/g**	**log cfu/g**	**Average**	**SD**
THB	9.4×10^8	8.97	8.97	0.01
	9.3×10^8	8.97		
	9.2×10^8	8.96		
P. aeruginosa	6.3×10^5	5.80	5.81	0.02
	6.5×10^5	5.81		
	6.7×10^5	5.83		

SD = standard deviation; cfu/g = colony forming units per gram of sample

Table B66: The presence of Total Heterotrophic bacteria (THB), *L. monocytogenes* and *P. aeruginosa* populations in non-autoclaved soil and at week 1, in the greenhouse experiment.

Microbial population	cfu/g	log cfu/g	Average	SD
THB	1.34×10^{16}	16.13	16.12	0.01
	1.37×10^{16}	16.12		
	1.37×10^{16}	16.12		
L. monocytogenes	28.6×10^{3}	4.46	4.46	0.00
	28.7×10^{3}	4.46		
	28.5×10^{3}	4.46		
P. aeruginosa	5.1×10^{10}	10.71	10.72	0.01
	5.3×10^{10}	10.72		
	5.4×10^{10}	10.73		

SD = standard deviation; cfu/g = colony forming units per gram of sample

Table B67: The presence of Total Heterotrophic bacteria (THB), *L. monocytogenes* and *P. aeruginosa* populations in non-autoclaved soil at week 2, in the greenhouse experiment.

Microbial population	cfu/g	log cfu/g	Average	SD
THB	2.30×10^{12}	12.36	12.36	0.01
	2.30×10^{12}	12.36		
	2.35×10^{12}	12.37		
L. monocytogenes	3600	3.56	3.54	0.02
	3500	3.54		
	3400	3.53		
P. aeruginosa	4.5×10^{7}	7.65	7.65	0.03
	4.8×10^{7}	7.68		
	4.3×10^{7}	7.63		

SD = standard deviation; cfu/g = colony forming units per gram of sample

Table B68: The presence of Total Heterotrophic bacteria (THB) and *P. aeruginosa* populations in non-autoclaved soil and at week 3, in the greenhouse experiment.

Microbial population	cfu/g	log cfu/g	Average	SD
THB	3.5×10^{9}	9.54	9.56	0.03
	3.9×10^{9}	9.59		
	3.6×10^{9}	9.56		
P. aeruginosa	5.3×10^{5}	5.72	5.74	0.02
	5.6×10^{5}	5.75		
	5.6×10^{5}	5.75		

SD = standard deviation; cfu/g = colony forming units per gram of sample

Table B69: The presence of Total Heterotrophic bacteria (THB) and *P. aeruginosa* populations in non-autoclaved soil at week 4, in the greenhouse experiment.

Microbial population	cfu/g	log cfu/g	Average	SD
THB	15.6×10^7	8.19	8.20	0.01
	15.9×10^7	8.2		
	16.1×10^7	8.21		
P. aeruginosa	10.2×10^4	5.01	5.01	0.00
	10.2×10^4	5.01		
	10.2×10^7	5.01		

SD = standard deviation; cfu/g = colony forming units per gram of sample

Table B70: The presence of *L. monocytogenes* and *P. aeruginosa* populations in autoclaved soil at week 1, of the greenhouse experiment.

Microbial population	cfu/g	log cfu/g	Average	SD
L. monocytogenes	3×10^5	5.48	5.48	0.01
	3.1×10^5	5.49		
	3×10^5	5.48		
P. aeruginosa	3×10^6	6.48	6.50	0.03
	3.4×10^6	6.53		
	3×10^6	6.48		

SD = standard deviation; cfu/g = colony forming units per gram of sample

Table B71: The presence of *L. monocytogenes* and *P. aeruginosa* populations in autoclaved soil and the presence of *L. monocytogenes* in the lettuce, itself, at week 2, of the greenhouse experiment.

Sample	Microbial population	cfu/g	log cfu/g	Average	SD
Lettuce soil	*L. monocytogenes*	185000	5.27	5.27	0.01
		183000	5.26		
		186000	5.27		
Lettuce	*L. monocytogenes*	25400	4.41	4.41	0.00
		25700	4.41		
		25400	4.41		
Lettuce soil	*P. aeruginosa*	450000	5.65	5.66	0.02
		450000	5.65		
		490000	5.69		

SD = standard deviation; cfu/g = colony forming units per gram of sample

Table B72: The presence of *L. monocytogenes* and *P. aeruginosa* populations in autoclaved soil and the presence of *L. monocytogenes* in the lettuce, itself, at week 3 of the greenhouse experiment.

Sample	Microbial population	cfu/g	log cfu/g	Average	SD
Lettuce soil	*L. monocytogenes*	29800	4.47	4.48	0.02
		29700	4.47		
		29300	4.47		
		32000	4.51		
Lettuce	*L. monocytogenes*	27600	4.44	4.44	0.00
		27800	4.44		
		27700	4.44		
Lettuce soil	*P. aeruginosa*	203000	5.28	5.29	0.01
		204000	5.28		
		204000	5.30		

SD = standard deviation; cfu/g = colony forming units per gram of sample

Table B73: The presence of *L. monocytogenes* and *P. aeruginosa* populations in autoclaved soil and the presence of *L. monocytogenes* in the lettuce, itself, at week 4 of the greenhouse experiment.

Sample	Microbial population	cfu/g	log cfu/g	Average	SD
Lettuce soil	*L. monocytogenes*	18700	4.27	4.27	0.01
		18600	4.27		
		18300	4.26		
Lettuce	*L. monocytogenes*	29800	4.47	4.47	0.01
		29100	4.46		
		29600	4.47		
Lettuce soil	*P. aeruginosa*	15100	4.18	4.19	0.01
		15500	4.19		
		15400	4.19		

SD = standard deviation; cfu/g = colony forming units per gram of sample

Table B74: The presence of *L. monocytogenes* population in autoclaved soil and in the lettuce, itself, at week 1 of the greenhouse experiment.

Samples	cfu/g	log cfu/g	Average	SD
Lettuce soil	31000	4.49	4.51	0.02
	34000	4.53		
	32000	4.51		
Lettuce	3400	3.53	3.55	0.02
	3700	3.57		
	3500	3.54		

SD = standard deviation; cfu/g = colony forming units per gram of sample

Table B75: The presence of *L. monocytogenes* in autoclaved soil and in the lettuce, itself, at week 2 of the greenhouse experiment.

Samples	cfu/g	log cfu/g	Average	SD
Lettuce soil	2130000	6.33	6.34	0.01
	2160000	6.34		
	2180000	6.34		
Lettuce	222000	5.35	5.35	0.00
	225000	5.35		
	225000	5.35		

SD = standard deviation; cfu/g = colony forming units per gram of sample

Table B76: The presence of *L. monocytogenes* in autoclaved soil and in the lettuce, itself, at week 3 of the greenhouse experiment.

Samples	cfu/g	log cfu/g	Average	SD
Lettuce soil	1560000	6.19	6.19	0.00
	1560000	6.19		
	1560000	6.19		
Lettuce	274000	5.44	5.44	0.00
	273000	5.44		
	273000	5.44		

SD = standard deviation; cfu/g = colony forming units per gram of sample

Table B77: The presence of *L. monocytogenes* in autoclaved soil and in the lettuce, itself, at week 4 of the greenhouse experiment.

Samples	cfu/g	log cfu/g	Average	SD
Lettuce soil	12100	4.08	4.09	0.01
	12300	4.09		
	12400	4.09		
Lettuce	530000	5.72	5.72	0.01
	530000	5.72		
	510000	5.71		

SD = standard deviation; cfu/g = colony forming units per gram of sample

Table B78: The presence of different presumptive pathogens in the control (unwashed) and in the fresh produce washed using different treatment methods, before refrigeration (day 0).

Presumptive microbial pathogens	Treatment	Samples	cfu/g	log cfu/g	Average	SD
Campylobacter spp.	control	Broccoli	24500	4.39	4.39	0.00
			24400	4.39		
			24900	4.40		
		Cabbage	20000	4.30	4.31	0.01
			20400	4.31		
			20900	4.32		
		Crisphead lettuce	27400	4.44	4.43	0.00
			27100	4.43		
			26900	4.43		
		Spinach	28000	4.45	4.45	0.00
			28400	4.45		
			28500	4.45		
	i	Broccoli	20100	4.30	4.31	0.01
			20600	4.31		
			20300	4.31		
		Cabbage	19000	4.28	4.28	0.00
			19200	4.28		
			19100	4.28		
		Crisphead lettuce	26100	4.42	4.42	0.00
			26100	4.42		
			26400	4.42		
		Spinach	28600	4.46	4.45	0.00
			28300	4.45		
			28000	4.45		
	ii	Broccoli	18300	4.26	4.26	0.00
			18200	4.26		
			18600	4.27		
		Cabbage	17200	4.24	4.24	0.00
			17300	4.24		
			17500	4.24		
		Crisphead lettuce	23100	4.36	4.37	0.00
			23500	4.37		
			23400	4.37		
		Spinach	24300	4.39	4.39	0.00
			24500	4.39		
			24800	4.39		
	iii	Broccoli	25500	4.41	4.41	0.00
			25500	4.41		
			25500	4.41		
		Cabbage	20000	4.30	4.29	0.01
			19700	4.29		
			19400	4.29		
		Crisphead lettuce	26500	4.42	4.42	0.00
			26400	4.42		
			26400	4.42		
		Spinach	27400	4.44	4.44	0.00
			27100	4.43		
			27500	4.44		
	v	Broccoli	24300	4.39	4.38	0.00
			24300	4.39		
			24100	4.38		
		Cabbage	20100	4.30	4.29	0.01

Table B78/ Cont.

Presumptive microbial pathogens	Treatment	Samples	cfu/g	log cfu/g	Average	SD
			19300	4.29		
			19400	4.29		
		Crisphead lettuce	25300	4.40	4.40	0.01
			25100	4.40		
			25800	4.41		
		Spinach	25100	4.40	4.41	0.01
			26100	4.42		
			26100	4.42		
Coliforms	control	Broccoli	5000	3.70	3.70	0.02
			4900	3.69		
			5300	3.72		
		Crisphead lettuce	39000	4.59	4.57	0.02
			35000	4.54		
			37000	4.57		
		Spinach	45000	4.65	4.66	0.01
			45000	4.65		
			46000	4.66		
	i	Broccoli	3000	3.48	3.50	0.02
			3200	3.51		
			3200	3.51		
		Crisphead lettuce	29800	4.47	4.47	0.00
			29400	4.47		
			29300	4.47		
		Spinach	29400	4.47	4.47	0.01
			29800	4.47		
			29100	4.46		
	ii	Crisphead lettuce	28300	4.45	4.45	0.01
			27900	4.45		
			27500	4.44		
		Spinach	27400	4.44	4.44	0.00
			27600	4.44		
			27800	4.44		
	iii	Spinach	3500	3.54	3.51	0.03
			3100	3.49		
			3200	3.51		
	vi	Spinach	3200	3.51	3.51	0.00
			3200	3.51		
			3200	3.51		
E. coli	control	Crisphead lettuce	23300	4.37	4.37	0.01
			23800	4.38		
			23100	4.36		
		Spinach	28300	4.45	4.45	0.00
			28400	4.45		
			28400	4.45		
	i	Crisphead lettuce	18600	4.27	4.27	0.01
			18900	4.28		
			18200	4.26		
		Spinach	27300	4.44	4.44	0.01
			27800	4.44		
			27100	4.43		
	ii	Crisphead lettuce	16200	4.21	4.21	0.00
			16300	4.21		
			16100	4.21		

Table B78/ Cont.

Presumptive microbial pathogens	Treatment	Samples	cfu/g	log cfu/g	Average	SD
		Spinach	25400	4.40	4.41	0.01
			25800	4.41		
			25100	4.40		
	iii	Spinach	3000	3.48	3.54	0.06
			3500	3.54		
			3900	3.59		
L .monocytogenes	control	Spinach	3500	3.54	3.59	0.04
			4100	3.61		
			4200	3.62		
	i	Spinach	4100	3.61	3.58	0.04
			3400	3.53		
			4000	3.60		
	ii	Spinach	3300	3.52	3.51	0.01
			3200	3.51		
			3200	3.51		
	iii	Spinach	3500	3.54	3.55	0.01
			3700	3.57		
			3500	3.54		
	v	Spinach	3000	3.48	3.49	0.01
			3100	3.49		
			3100	3.49		
	vi	Spinach	3000	3.48	3.48	
			3000	3.48		
Salmonella spp.	control	Crisphead lettuce	7500	3.88	3.90	0.03
			8000	3.90		
			8500	3.93		
		Spinach	65000	4.81	4.84	0.02
			70000	4.85		
			71000	4.85		
	i	Crisphead lettuce	6500	3.81	3.79	0.02
			6000	3.78		
			6100	3.79		
		Spinach	35000	4.54	4.53	0.01
			34000	4.53		
			33000	4.52		
	ii	Crisphead lettuce	4200	3.62	3.63	0.02
			4500	3.65		
			4200	3.62		
		Spinach	28500	4.45	4.47	0.04
			34000	4.53		
			28500	4.45		
			28300	4.45		
	iii	Crisphead lettuce	3000	3.48	3.50	0.02
			3200	3.51		
			3300	3.52		
	v	Spinach	3000	3.48	3.49	0.01
			3100	3.49		
			3100	3.49		
Shigella spp.	control	Broccoli	5400	3.73	3.75	0.02
			5500	3.74		
			5900	3.77		
		Cabbage	31000	4.49	4.53	0.04
			38000	4.58		

Table B78/ Cont.

Presumptive microbial pathogens	Treatment	Samples	cfu/g	log cfu/g	Average	SD
			34000	4.53		
		Crisphead lettuce	35000	4.54	4.58	0.03
			39000	4.59		
			39000	4.59		
		Spinach	54000	4.73	4.75	0.02
			56000	4.75		
			59000	4.77		
	i	Cabbage	29100	4.46	4.47	0.00
			29300	4.47		
			29200	4.47		
		Crisphead lettuce	15100	4.18	4.19	0.01
			15700	4.20		
			15700	4.20		
		Spinach	29300	4.47	4.47	0.00
			29100	4.46		
			29300	4.47		
	ii	Cabbage	26400	4.42	4.42	0.00
			26400	4.42		
			26900	4.43		
		Crisphead lettuce	13200	4.12	4.12	0.01
			13600	4.13		
			13100	4.12		
		Spinach	26500	4.42	4.42	0.00
			26200	4.42		
			26400	4.42		
	iii	Spinach	3500	3.54	3.53	0.02
			3400	3.53		
			3200	3.51		
	v	Spinach	3200	3.51	3.53	0.02
			3400	3.53		
			3500	3.54		
	vi	Crisphead lettuce	4300	3.63	3.62	0.01
			4100	3.61		
			4200	3.62		
		Spinach	3100	3.49	3.52	0.02
			3400	3.53		
			3400	3.53		
THB	control	Broccoli	350000	5.54	5.53	0.02
			320000	5.51		
			350000	5.54		
		Cabbage	890000	5.95	5.96	0.01
			910000	5.96		
			930000	5.97		
		Crisphead lettuce	8400000	6.92	6.94	0.01
			8700000	6.94		
			8900000	6.95		
		Spinach	1020000000	9.01	9.02	0.01
			1090000000	9.04		
			1050000000	9.02		
	i	Broccoli	36000	4.56	4.54	0.02
			35000	4.54		
			33000	4.52		
		Cabbage	91000	4.96	4.96	0.00

Table B78/ Cont.

Presumptive microbial pathogens	Treatment	Samples	cfu/g	log cfu/g	Average	SD
			92000	4.96		
			93000	4.97		
		Crisphead lettuce	760000	5.88	5.90	0.02
			780000	5.89		
			840000	5.92		
		Spinach	1060000000	9.03	9.02	0.00
			1040000000	9.02		
			1040000000	9.02		
	ii	Broccoli	201000	5.30	5.31	0.00
			204000	5.31		
			201000	5.30		
		Cabbage	287000	5.46	5.45	0.00
			283000	5.45		
			281000	5.45		
		Crisphead lettuce	2710000	6.43	6.44	0.01
			2670000	6.43		
			2840000	6.45		
		Spinach	245000000	8.39	8.38	0.01
			243000000	8.39		
			239000000	8.38		
	iii	Broccoli	3500	3.54	3.54	0.01
			3600	3.56		
			3400	3.53		
		Cabbage	4000	3.60	3.62	0.02
			4300	3.63		
			4200	3.62		
		Crisphead lettuce	5300	3.72	3.74	0.02
			5500	3.74		
			5700	3.76		
		Spinach	6300	3.80	3.82	0.02
			6700	3.83		
			6900	3.84		
	v	Broccoli	3700	3.57	3.59	0.02
			3900	3.59		
			4100	3.61		
		Cabbage	4500	3.65	3.63	0.03
			4300	3.63		
			4000	3.60		
		Crisphead lettuce	5600	3.75	3.75	0.02
			5400	3.73		
			5800	3.76		
		Spinach	6100	3.79	3.80	0.02
			6500	3.81		
			6500	3.81		
	vi	Broccoli	5300	3.72	3.74	0.02
			5700	3.76		
			5400	3.73		
		Cabbage	6700	3.83	3.82	0.01
			6500	3.81		
			6800	3.83		
		Crisphead lettuce	7600	3.88	3.88	0.01
			7800	3.89		
			7500	3.88		

Table B78/ Cont.

Presumptive microbial pathogens	Treatment	Samples	cfu/g	log cfu/g	Average	SD
		Spinach	8200	3.91	3.89	0.04
			6900	3.84		
			8100	3.91		

i = tap water, ii = NaCl, iii = chlorine, iv = blanching, v = H_2O_2, vi = UV; SD = standard deviation; cfu/g = colony forming units per gram of sample; THB = total heterotrophic bacteria

Table B79: The presence of different presumptive pathogens in the control (unwashed) and in the fresh produce washed using different treatment methods, after refrigeration (day 6).

Presumptive microbial pathogens	Treatment	Samples	cfu/g	log cfu/g	Average	SD
Campylobacter spp.	Control	Broccoli	67000	4.83	4.83	0.00
			68000	4.83		
			67000	4.83		
		Cabbage	32000	4.51	4.51	0.01
			33000	4.52		
			33000	4.52		
		Crisphead lettuce	76000	4.88	4.88	0.01
			78000	4.89		
			75000	4.88		
		Spinach	320000	5.51	5.52	0.01
			330000	5.52		
			340000	5.53		
	i	Broccoli	29800	4.47	4.46	0.01
			28700	4.46		
			28700	4.46		
		Cabbage	178000	5.25	5.25	0.00
			176000	5.25		
			176000	5.25		
		Crisphead lettuce	275000	5.44	5.44	0.00
			274000	5.44		
			278000	5.44		
		Spinach	2350000	6.37	6.38	0.01
			2390000	6.38		
			2430000	6.39		
	ii	Broccoli	21400	4.33	4.33	0.00
			21600	4.33		
			21700	4.34		
		Cabbage	21800	4.34	4.34	0.00
			21600	4.33		
			21700	4.34		
		Crisphead lettuce	27600	4.44	4.44	0.00
			27900	4.45		
			27600	4.44		
		Spinach	32000	4.51	4.55	0.04
			37000	4.57		
			37000	4.57		
	iii	Broccoli	21000	4.32	4.32	0.00
			21000	4.32		
			21000	4.32		
		Cabbage	21800	4.34	4.34	0.00
			21900	4.34		
			22000	4.34		
		Crisphead lettuce	28600	4.46	4.46	0.00
			28500	4.45		
			28900	4.46		
		Spinach	29400	4.47	4.47	0.00
			29400	4.47		
			29100	4.46		
	v	Broccoli	20100	4.30	4.31	0.00
			20500	4.31		
			20500	4.31		
		Cabbage	22100	4.34	4.34	0.00

Table B79/ Cont.

Presumptive microbial pathogens	Treatment	Samples	cfu/g	log cfu/g	Average	SD
Coliforms		Crisphead lettuce	22100	4.34	4.45	0.00
			22100	4.34		
			28600	4.46		
		Spinach	28400	4.45	4.52	0.01
			28300	4.45		
			32000	4.51		
	control	Broccoli	33000	4.52	4.75	0.02
			34000	4.53		
			53000	4.72		
		Crisphead lettuce	57000	4.76	5.25	0.01
			58000	4.76		
			179000	5.25		
		Spinach	176000	5.25	5.51	0.01
			174000	5.24		
			320000	5.51		
	i	Broccoli	330000	5.52	5.24	0.01
			310000	5.49		
			173000	5.24		
		Cabbage	178000	5.25	3.53	0.03
			175000	5.24		
			3200	3.51		
		Crisphead lettuce	3300	3.52	5.63	0.02
			3600	3.56		
			430000	5.63		
		Spinach	440000	5.64	5.74	0.00
			410000	5.61		
			540000	5.73		
	ii	Crisphead lettuce	550000	5.74	4.45	0.01
			540000	5.73		
			28300	4.45		
		Spinach	27900	4.45	4.44	0.00
			27500	4.44		
			27400	4.44		
	iii	Broccoli	27600	4.44	3.51	0.01
			27800	4.44		
			3200	3.51		
		Spinach	3300	3.52	5.73	0.03
			3200	3.51		
			530000	5.72		
	iv	Broccoli	520000	5.72	3.50	0.02
			580000	5.76		
			3000	3.48		
	v	Broccoli	3300	3.52	3.74	0.00
			3200	3.51		
			5400	3.73		
		Crisphead lettuce	5500	3.74	3.83	0.01
			5500	3.74		
			6500	3.81		
		Spinach	6800	3.83	5.02	0.01
			6900	3.84		
			101000	5.00		
			105000	5.02		
			106000	5.03		

Table B79/ Cont.

Presumptive microbial pathogens	Treatment	Samples	cfu/g	log cfu/g	Average	SD
	vi	Broccoli	3400	3.53	3.52	0.03
			3500	3.54		
			3100	3.49		
		Crisphead lettuce	3000	3.48	3.49	0.01
			3100	3.49		
			3200	3.51		
		Spinach	3900	3.59	3.58	0.01
			3800	3.58		
			3700	3.57		
E. coli	control	Cabbage	5400	3.73	3.74	0.01
			5500	3.74		
			5600	3.75		
		Crisphead lettuce	34000	4.53	4.56	0.03
			38000	4.58		
			37000	4.57		
		Spinach	36000	4.56	4.55	0.01
			36000	4.56		
			34000	4.53		
	i	Cabbage	54000	4.73	4.75	0.02
			56000	4.75		
			58000	4.76		
		Crisphead lettuce	59000	4.77	4.71	0.06
			46000	4.66		
			49000	4.69		
		Spinach	32000	4.51	4.52	0.01
			33000	4.52		
			34000	4.53		
	ii	Cabbage	75000	4.88	4.88	0.01
			78000	4.89		
			76000	4.88		
		Crisphead lettuce	65000	4.81	4.82	0.01
			67000	4.83		
			64000	4.81		
		Spinach	30000	4.48	4.50	0.02
			33000	4.52		
			33000	4.52		
	iii	Cabbage	32000	4.51	4.50	0.01
			31000	4.49		
			31000	4.49		
		Crisphead lettuce	27600	4.44	4.46	0.03
			27500	4.44		
			31000	4.49		
		Spinach	31000	4.49	4.50	0.02
			31000	4.49		
			34000	4.53		
	v	Cabbage	32000	4.51	4.50	0.02
			33000	4.52		
			30000	4.48		
		Crisphead lettuce	54000	4.73	4.75	0.01
			56000	4.75		
			57000	4.76		
		Spinach	31000	4.49	4.50	0.01
			32000	4.51		

Table B79/ Cont.

Presumptive microbial pathogens	**Treatment**	**Samples**	**cfu/g**	**log cfu/g**	**Average**	**SD**
			31000	4.49		
L. monocytogenes	control	Spinach	4100	3.61	3.62	0.01
			4300	3.63		
			4200	3.62		
	i	Spinach	4300	3.63	3.65	0.01
			4600	3.66		
			4500	3.65		
	ii	Spinach	3700	3.57	3.58	0.01
			3800	3.58		
			3800	3.58		
	iii	Broccoli	3200	3.51	3.51	0.01
			3100	3.49		
			3300	3.52		
		Cabbage	3200	3.51	3.51	0.03
			3000	3.48		
			3500	3.54		
		Crisphead lettuce	3200	3.51	3.51	0.03
			3000	3.48		
			3500	3.54		
		Spinach	4200	3.62	3.64	0.02
			4200	3.62		
			4600	3.66		
	iv	Cabbage	3100	3.49	3.53	0.05
			3200	3.51		
			3800	3.58		
		Crisphead lettuce	6200	3.79	3.79	0.01
			6100	3.79		
			6300	3.80		
		Spinach	5400	3.73	3.74	0.01
			5400	3.73		
			5700	3.76		
	v	Spinach	3400	3.53	3.54	0.01
			3500	3.54		
			3600	3.56		
	vi	Spinach	3400	3.53	3.51	0.02
			3200	3.51		
			3200	3.51		
Salmonella spp.	control	Crisphead lettuce	7200	3.86	3.85	0.01
			6900	3.84		
			6900	3.84		
		Spinach	62000	4.79	4.79	0.00
			62000	4.79		
			62000	4.79		
	i	Crisphead lettuce	6300	3.80	3.79	0.00
			6200	3.79		
			6200	3.79		
		Spinach	45000	4.65	4.66	0.02
			44000	4.64		
			48000	4.68		
	ii	Crisphead lettuce	5400	3.73	3.72	0.01
			5200	3.72		
			5100	3.71		
		Spinach	31000	4.49	4.49	0.01

Table B79/ Cont.

Presumptive microbial pathogens	Treatment	Samples	cfu/g	log cfu/g	Average	SD
			32000	4.51		
			30000	4.48		
	iii	Spinach	21000	4.32	4.34	0.01
			22100	4.34		
			22300	4.35		
	iv	Cabbage	3200	3.51	3.50	0.01
			3100	3.49		
			3200	3.51		
		Crisphead lettuce	5800	3.76	3.76	0.00
			5800	3.76		
			5800	3.76		
		Spinach	8900	3.95	3.97	0.02
			9700	3.99		
			9100	3.96		
	v	Spinach	3000	3.48	3.49	0.01
			3200	3.51		
			3100	3.49		
Shigella spp.	control	Broccoli	25300	4.40	4.40	0.00
			25100	4.40		
			25600	4.41		
		Cabbage	287000	5.46	5.46	0.01
			288000	5.46		
			281000	5.45		
		Crisphead lettuce	273000	5.44	5.44	0.00
			275000	5.44		
			271000	5.43		
		Spinach	106000	5.03	5.03	0.01
			109000	5.04		
			105000	5.02		
	i	Broccoli	32000	4.51	4.50	0.01
			32000	4.51		
			31000	4.49		
		Cabbage	35000	4.54	4.50	0.04
			31000	4.49		
			30000	4.48		
		Crisphead lettuce	34000	4.53	4.51	0.02
			33000	4.52		
			31000	4.49		
		Spinach	29700	4.47	4.49	0.01
			31000	4.49		
			31000	4.49		
	ii	Broccoli	27800	4.44	4.44	0.00
			27600	4.44		
			28100	4.45		
		Cabbage	27100	4.43	4.43	0.00
			27200	4.43		
			27100	4.43		
		Crisphead lettuce	24500	4.39	4.40	0.01
			25300	4.40		
			25100	4.40		
		Spinach	29300	4.47	4.43	0.03
			26100	4.42		
			26300	4.42		

Table B79/ Cont.

Presumptive microbial pathogens	**Treatment**	**Samples**	**cfu/g**	**log cfu/g**	**Average**	**SD**
	iii	Broccoli	3400	3.53	3.51	0.02
			3200	3.51		
			3100	3.49		
		Cabbage	13400	4.13	4.13	0.00
			13300	4.12		
			13400	4.13		
		Crisphead lettuce	22400	4.35	4.35	0.00
			22100	4.34		
			22100	4.34		
		Spinach	20100	4.30	4.31	0.00
			20300	4.31		
			20400	4.31		
	iv	Crisphead lettuce	31000	4.49	4.51	0.02
			33000	4.52		
			34000	4.53		
		Spinach	30000	4.48	4.48	0.00
			30000	4.48		
			30000	4.48		
	v	Cabbage	3200	3.51	3.53	0.02
			3500	3.54		
			3400	3.53		
		Crisphead lettuce	13100	4.12	4.12	0.01
			13400	4.13		
			13300	4.12		
		Spinach	14500	4.16	4.17	0.02
			14300	4.16		
			15700	4.20		
	vi	Broccoli	3000	3.48	3.48	0.01
			3100	3.49		
			3000	3.48		
THB	control	Broccoli	14400000	7.16	7.16	0.01
			14500000	7.16		
			14900000	7.17		
		Cabbage	256000000	8.41	8.41	0.00
			255000000	8.41		
			259000000	8.41		
		Crisphead lettuce	520000000	8.72	8.73	0.01
			530000000	8.72		
			550000000	8.74		
		Spinach	2240000000	9.35	9.36	0.01
			2260000000	9.35		
			2310000000	9.36		
	i	Broccoli	30000000	7.48	7.49	0.01
			32000000	7.51		
			31000000	7.49		
		Cabbage	350000000	8.54	8.54	0.01
			330000000	8.52		
			350000000	8.54		
		Crisphead lettuce	1020000000	9.01	9.01	0.01
			1050000000	9.02		
			1030000000	9.01		
		Spinach	2750000000	9.44	9.44	0.00
			2780000000	9.44		

Table B79/ Cont.

Presumptive microbial pathogens	Treatment	Samples	cfu/g	log cfu/g	Average	SD
			2750000000	9.44		
	ii	Broccoli	580000	5.76	5.76	0.01
			590000	5.77		
			560000	5.75		
		Cabbage	10400000	7.02	7.02	0.01
			10200000	7.01		
			10500000	7.02		
		Crisphead lettuce	8500000	6.93	6.93	0.02
			8900000	6.95		
			8300000	6.92		
		Spinach	5600000	6.75	6.74	0.00
			5500000	6.74		
			5500000	6.74		
	iii	Broccoli	25300000	7.40	7.41	0.00
			25400000	7.40		
			25800000	7.41		
		Cabbage	350000000	8.54	8.54	0.01
			330000000	8.52		
			350000000	8.54		
		Crisphead lettuce	1020000000	9.01	9.01	0.01
			1050000000	9.02		
			1030000000	9.01		
		Spinach	2750000000	9.44	9.44	0.00
			2780000000	9.44		
			2750000000	9.44		
	iv	Broccoli	4200	3.62	3.59	0.03
			3800	3.58		
			3700	3.57		
		Cabbage	31000	4.49	4.53	0.03
			35000	4.54		
			36000	4.56		
		Crisphead lettuce	2930000	6.47	6.46	0.01
			2870000	6.46		
			2780000	6.44		
		Spinach	2540000	6.40	6.41	0.00
			2590000	6.41		
			2580000	6.41		
	v	Broccoli	117000	5.07	5.07	0.01
			119000	5.08		
			116000	5.06		
		Cabbage	134000	5.13	5.14	0.02
			139000	5.14		
			146000	5.16		
		Crisphead lettuce	1210000	6.08	6.09	0.01
			1240000	6.09		
			1270000	6.10		
		Spinach	7000000	6.85	6.85	0.01
			7200000	6.86		
			7200000	6.86		
	vi	Broccoli	257000	5.41	5.41	0.00
			257000	5.41		
			259000	5.41		
		Cabbage	73000	4.86	4.87	0.01

Table B79/ Cont.

Presumptive microbial pathogens	Treatment	Samples	cfu/g	log cfu/g	Average	SD
		Crisphead lettuce	76000	4.88	4.90	0.02
			72000	4.86		
			82000	4.91		
			79000	4.90		
		Spinach	76000	4.88	4.81	0.03
			65000	4.81		
			68000	4.83		
			59000	4.77		

i = tap water, ii = NaCl, iii = chlorine, iv = blanching, v = H_2O_2, vi = UV; SD = standard deviation; cfu/g = colony forming units per gram of sample; THB = total heterotrophic bacteria

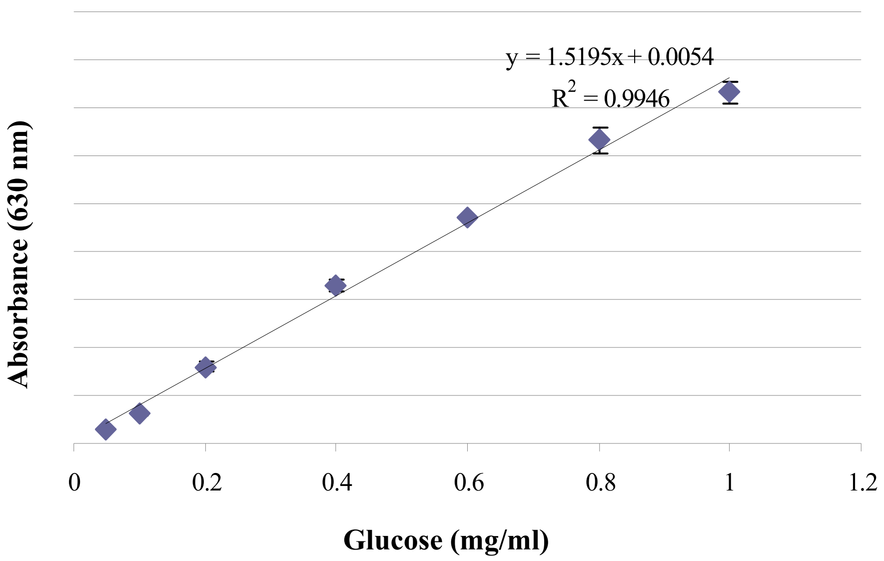

Figure B1: Standard curve for the estimation of total carbohydrate (TC) content by the Anthrone method.

Table B80: The concentrations (mg/g) of total carbohydrates (TC) obtained though the Anthrone method at day 0 from four different fresh produce, which were unwashed and subjected to the tap water treatment method.

			Total carbohydrates (mg/g)									
			Unwashed control					**Tap water**				
Day	**Samples**					**Average**	**SD**				**Average**	**SD**
0			0.14	0.14	0.14			0.14	0.12	0.15		
	Broccoli	con	0.09	0.09	0.09			0.09	0.08	0.09		
		calc	87.56	86.90	88.22	87.56	0.66	88.87	76.37	92.17	85.80	8.34
			0.13	0.14	0.14			0.14	0.13	0.13		
	Cabbage	con	0.08	0.09	0.09			0.09	0.08	0.08		
		calc	82.95	88.22	86.90	86.02	2.74	86.24	82.95	84.92	84.70	1.66
			0.10	0.12	0.13			0.12	0.13	0.12		
	Lettuce	con	0.06	0.08	0.08			0.08	0.08	0.08		
		calc	63.20	76.37	81.63	73.73	9.49	76.37	80.32	77.68	78.12	2.01
			0.13	0.11	0.13			0.13	0.13	0.12		
	Spinach	con	0.08	0.07	0.08			0.08	0.08	0.08		
		calc	79.00	65.83	79.66	74.83	7.80	79.00	80.32	76.37	78.56	2.01
6			0.10	0.11	0.11			0.10	0.10	0.11		
	Broccoli	con	0.06	0.07	0.07			0.06	0.07	0.07		
		calc	64.52	65.83	68.47	66.27	2.01	63.86	65.17	66.49	65.17	1.32
			0.10	0.09	0.10			0.10	0.10	0.10		
	Cabbage	con	0.06	0.06	0.06			0.06	0.06	0.07		
		calc	61.88	57.93	60.57	60.13	2.01	63.86	64.52	65.17	64.52	0.66
			0.08	0.08	0.07			0.08	0.08	0.08		
	Lettuce	con	0.05	0.05	0.05			0.05	0.05	0.05		
		calc	46.74	46.08	45.42	46.08	0.66	50.03	46.74	47.40	48.06	1.74
			0.08	0.08	0.09			0.09	0.08	0.08		
	Spinach	con	0.05	0.05	0.05			0.05	0.05	0.05		
		calc	50.69	50.03	53.98	51.57	2.12	54.64	51.35	52.01	52.67	1.74

con = concentration of TC as shown by the standard curve; calc = calculation of the concentration of TC using the concentrations obtained from the standard curve; SD = standard deviation

Table B81: The concentrations (mg/g) of total carbohydrates (TC) obtained though the Anthrone method at day 0 from four different fresh produce, which were subjected to the NaCl and chlorine treatment methods.

			Total carbohydrate (mg/g)									
					NaCl					Chlorine		
Day	Samples					Average	SD				Average	SD
0			0.14	0.14	0.14			0.15	0.14	0.13		
	Broccoli	con	0.09	0.09	0.09			0.09	0.09	0.08		
		calc	85.58	88.22	86.24	86.68	1.37	92.82	89.53	82.29	88.22	5.39
			0.13	0.13	0.13			0.11	0.14	0.14		
	Cabbage	con	0.08	0.08	0.08			0.07	0.09	0.09		
		calc	84.27	82.95	82.29	83.17	1.01	68.47	87.56	85.58	80.54	10.50
			0.13	0.12	0.13			0.12	0.13	0.13		
	Lettuce	con	0.08	0.08	0.08			0.07	0.08	0.08		
		calc	81.63	78.34	79.66	79.88	1.66	73.73	82.29	79.66	78.56	4.38
			0.13	0.12	0.12			0.13	0.12	0.12		
	Spinach	con	0.08	0.08	0.08			0.08	0.08	0.08		
		calc	80.97	78.34	77.68	79.00	1.74	81.63	78.34	77.68	79.22	2.12
6			0.11	0.12	0.12			0.12	0.11	0.12		
	Broccoli	con	0.07	0.08	0.08			0.08	0.07	0.07		
		calc	69.78	75.71	77.68	74.39	4.11	75.05	71.10	73.07	73.07	1.97
			0.10	0.11	0.11			0.11	0.11	0.11		
	Cabbage	con	0.07	0.07	0.07			0.07	0.07	0.07		
		calc	65.17	67.15	68.47	66.93	1.66	65.83	65.83	67.15	66.27	0.76
			0.08	0.07	0.07			0.09	0.08	0.07		
	Lettuce	con	0.05	0.04	0.04			0.05	0.05	0.05		
		calc	48.72	43.45	42.13	44.77	3.48	53.98	46.08	45.42	48.50	4.76
			0.08	0.09	0.08			0.09	0.08	0.09		
	Spinach	con	0.05	0.05	0.05			0.06	0.05	0.05		
		calc	51.35	52.67	50.69	51.57	1.01	57.93	50.69	52.67	53.76	3.74

con = concentration of TC as shown by the standard curve; calc = calculation of the concentration of TC using the concentrations obtained from the standard curve; SD = standard deviation

Table B82: The concentrations (mg/g) of total carbohydrates (TC) obtained though the Anthrone method at day 0 from four different fresh produce, which were subjected to the blanching and hydrogen peroxide treatment methods.

			Total carbohydrate (mg/g)									
			Blanching					Hydrogen peroxide				
Day	**Samples**					**Average**	**SD**				**Average**	**SD**
0			0.13	0.11	0.13			0.14	0.14	0.14		
	Broccoli	**con**	0.08	0.07	0.08			0.09	0.09	0.09		
		calc	82.95	65.83	84.92	77.90	10.50	86.24	86.24	87.56	86.68	0.76
			0.14	0.13	0.13			0.13	0.14	0.13		
	Cabbage	**con**	0.09	0.08	0.08			0.08	0.09	0.08		
		calc	88.22	82.95	82.29	84.49	3.25	82.29	86.90	82.95	84.05	2.49
			0.13	0.13	0.12			0.13	0.13	0.11		
	Lettuce	**con**	0.08	0.08	0.08			0.08	0.08	0.07		
		calc	82.95	80.32	77.68	80.32	2.63	81.63	81.63	71.76	78.34	5.70
			0.12	0.13	0.13			0.13	0.13	0.12		
	Spinach	**con**	0.08	0.08	0.08			0.08	0.08	0.08		
		calc	75.71	80.32	79.00	78.34	2.37	80.32	79.66	76.37	78.78	2.12
6			0.12	0.12	0.12			0.11	0.11	0.12		
	Broccoli	**con**	0.08	0.07	0.08			0.07	0.07	0.07		
		calc	76.37	74.39	76.37	75.71	1.14	71.10	71.76	73.07	71.98	1.01
			0.09	0.08	0.08			0.10	0.10	0.11		
	Cabbage	**con**	0.05	0.05	0.05			0.07	0.06	0.07		
		calc	52.67	50.03	50.03	50.91	1.52	65.17	64.52	65.83	65.17	0.66
			0.05	0.05	0.05			0.08	0.08	0.07		
	Lettuce	**con**	0.03	0.03	0.03			0.05	0.05	0.04		
		calc	30.28	31.60	26.33	29.41	2.74	52.01	50.03	42.79	48.28	4.85
			0.06	0.06	0.06			0.09	0.09	0.08		
	Spinach	**con**	0.04	0.04	0.04			0.05	0.05	0.05		
		calc	36.87	38.18	38.84	37.96	1.01	53.32	53.32	51.35	52.67	1.14

con = concentration of TC as shown by the standard curve; calc = calculation of the concentration of TC using the concentrations obtained from the standard curve; SD = standard deviation

Table B83: The concentrations (mg/g) of total carbohydrates (TC) obtained though the Anthrone method at day 0 from four different fresh produce, which were subjected to Ultra-Violet light.

			Total carbohydrate (mg/g)				
Day	**Samples**					**Average**	**SD**
0			0.13	0.14	0.13		
	Broccoli	con	0.08	0.09	0.08		
		calc	82.95	87.56	83.61	84.70	2.49
			0.14	0.13	0.14		
	Cabbage	con	0.09	0.08	0.09		
		calc	88.22	80.32	85.58	84.70	4.02
			0.12	0.12	0.12		
	Lettuce	con	0.08	0.08	0.08		
		calc	76.37	77.68	78.34	77.46	1.01
			0.12	0.12	0.12		
	Spinach	con	0.08	0.08	0.08		
		calc	77.68	75.71	77.02	76.80	1.01
6			0.12	0.12	0.13		
	Broccoli	con	0.08	0.08	0.08		
		calc	76.37	77.68	79.66	77.90	1.66
			0.12	0.12	0.12		
	Cabbage	con	0.07	0.07	0.08		
		calc	72.42	74.39	75.05	73.95	1.37
			0.08	0.08	0.08		
	Lettuce	con	0.05	0.05	0.05		
		calc	50.03	52.01	50.69	50.91	1.01
			0.09	0.09	0.08		
	Spinach	con	0.05	0.05	0.05		
		calc	52.67	53.98	51.35	52.67	1.32

con = concentration of TC as shown by the standard curve; calc = calculation of the concentration of TC using the concentrations obtained from the standard curve; SD = standard deviation

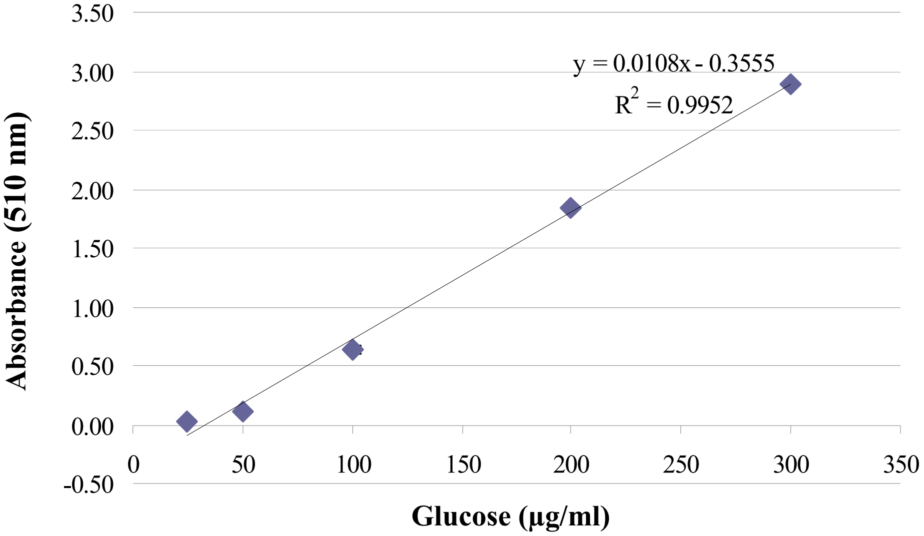

Figure B2: Standard curve used for the determination of reducing sugar content (DNS method) of the samples.

Table B84: The absorbance and concentrations (µg/g) of reducing sugars obtained though the DNS method at day 0 and 6 from four different fresh produce, which were unwashed and subjected to tap water treatment.

			Reducing sugars (µg/g)									
			unwashed control					**tap water**				
Samples		**Day**				**Average**	**SD**				**Average**	**SD**
Broccoli	absorbance	0	0.154	0.157	0.156			0.143	0.157	0.153		
	concentration		471.76	474.54	473.61	473.30	1.41	461.57	474.54	470.83	468.98	6.68
	absorbance	6	0.104	0.104	0.107			0.104	0.1	0.093		
	concentration		425.46	425.46	428.24	426.39	1.60	425.46	421.76	415.28	420.83	5.16
Cabbage	absorbance	0	0.135	0.135	0.132			0.132	0.139	0.141		
	concentration		454.17	454.17	451.39	453.24	1.60	451.39	457.87	459.72	456.33	4.38
	absorbance	6	0.117	0.118	0.118			0.117	0.119	0.114		
	concentration		437.50	438.43	438.43	438.12	0.53	437.50	439.35	434.72	437.19	2.33
Lettuce	absorbance	0	0.128	0.127	0.126			0.127	0.121	0.122		
	concentration		447.69	446.76	445.83	446.76	0.93	446.76	441.20	442.13	443.36	2.98
	absorbance	6	0.117	0.117	0.114			0.116	0.113	0.121		
	concentration		437.50	437.50	434.72	436.57	1.60	436.57	433.80	441.20	437.19	3.74
Spinach	absorbance	0	0.134	0.136	0.138			0.137	0.135	0.132		
	concentration		453.24	455.09	456.94	455.09	1.85	456.02	454.17	451.39	453.86	2.33
	absorbance	6	0.119	0.124	0.12			0.115	0.117	0.119		
	concentration		439.35	443.98	440.28	441.20	2.45	435.65	437.50	439.35	437.50	1.85

SD = standard deviation

Table B85: The absorbance and concentrations (μg/g) of reducing sugars obtained though the DNS method at day 0 and 6 from four different fresh produce, which were to NaCl and chlorine treatment.

			Reducing sugars (μg/g)									
			NaCl					Chlorine				
Samples		**Day**				**Average**	**SD**				**Average**	**SD**
Broccoli	absorbance	0	0.151	0.153	0.159			0.157	0.151	0.148		
	concentration		468.98	470.83	476.39	472.07	3.85	474.54	468.98	466.20	469.91	4.24
	absorbance	6	0.104	0.104	0.106			0.104	0.106	0.105		
	concentration		425.46	425.46	427.31	426.08	1.07	425.46	427.31	426.39	426.39	0.93
Cabbage	absorbance	0	0.135	0.141	0.138			0.138	0.134	0.136		
	concentration		454.17	459.72	456.94	456.94	2.78	456.94	453.24	455.09	455.09	1.85
	absorbance	6	0.121	0.126	0.117			0.122	0.121	0.12		
	concentration		441.20	445.83	437.50	441.51	4.18	442.13	441.20	440.28	441.20	0.93
Lettuce	absorbance	0	0.129	0.137	0.124			0.126	0.128	0.121		
	concentration		448.61	456.02	443.98	449.54	6.07	445.83	447.69	441.20	444.91	3.34
	absorbance	6	0.109	0.114	0.116			0.117	0.114	0.113		
	concentration		430.09	434.72	436.57	433.80	3.34	437.50	434.72	433.80	435.34	1.93
Spinach	absorbance	0	0.134	0.135	0.138			0.139	0.137	0.141		
	concentration		453.24	454.17	456.94	454.78	1.93	457.87	456.02	459.72	457.87	1.85
	absorbance	6	0.124	0.116	0.118			0.123	0.124	0.124		
	concentration		443.98	436.57	438.43	439.66	3.85	443.06	443.98	443.98	443.67	0.53

SD = standard deviation

Table B86: The absorbance and concentrations (µg/g) of reducing sugars obtained though the DNS method at day 0 and 6 from four different fresh produce, which were to blanching and hydrogen peroxide treatment.

			Reducing sugars (µg/g)									
			Blanching					Hydrogen peroxide				
Samples		**Days**				**Average**	**SD**				**Average**	**SD**
Broccoli	absorbance	0	0.142	0.157	0.159			0.161	0.162	0.148		
	concentration		460.65	474.54	476.39	470.52	8.60	478.24	479.17	466.20	474.54	7.23
	absorbance	6	0.123	0.124	0.127			0.115	0.118	0.117		
	concentration		443.06	443.98	446.76	444.60	1.93	435.65	438.43	437.50	437.19	1.41
Cabbage	absorbance	0	0.131	0.139	0.137			0.134	0.138	0.139		
	concentration		450.46	457.87	456.02	454.78	3.85	453.24	456.94	457.87	456.02	2.45
	absorbance	6	0.083	0.082	0.081			0.117	0.118	0.113		
	concentration		406.02	405.09	404.17	405.09	0.93	437.50	438.43	433.80	436.57	2.45
Lettuce	absorbance	0	0.12	0.12	0.127			0.124	0.128	0.129		
	concentration		440.28	440.28	446.76	442.44	3.74	443.98	447.69	448.61	446.76	2.45
	absorbance	6	0.115	0.113	0.113			0.076	0.063	0.06		
	concentration		435.65	433.80	433.80	434.41	1.07	399.54	387.50	384.72	390.59	7.87
Spinach	absorbance	0	0.134	0.137	0.134			0.138	0.134	0.138		
	concentration		453.24	456.02	453.24	454.17	1.60	456.94	453.24	456.94	455.71	2.14
	absorbance	6	0.051	0.056	0.061			0.116	0.118	0.114		
	concentration		376.39	381.02	385.65	381.02	4.63	436.57	438.43	434.72	436.57	1.85

SD = standard deviation

Table B87: The absorbance and concentrations (µg/g) of reducing sugars obtained though the DNS method at day 0 and 6 from four different fresh produce, which were subjected to Ultra-Violet treatment.

			Reducing sugars (µg/g)				
Samples		**Day**				**Average**	**SD**
Broccoli	absorbance	0	0.157	0.153	0.154		
	concentration		474.54	470.83	471.76	472.38	1.93
	absorbance	6	0.127	0.123	0.121		
	concentration		446.76	443.06	441.20	443.67	2.83
Cabbage	absorbance	0	0.147	0.135	0.138		
	concentration		465.28	454.17	456.94	458.80	5.78
	absorbance	6	0.123	0.121	0.125		
	concentration		443.06	441.20	444.91	443.06	1.85
Lettuce	absorbance	0	0.124	0.126	0.127		
	concentration		443.98	445.83	446.76	445.52	1.41
	absorbance	6	0.116	0.114	0.114		
	concentration		436.57	434.72	434.72	435.34	1.07
Spinach	absorbance	0	0.137	0.135	0.144		
	concentration		456.02	454.17	462.50	457.56	4.38
	absorbance	6	0.121	0.123	0.121		
	concentration		441.20	443.06	441.20	441.82	1.07

SD = standard deviation

Table B88: The absorbance and total chlorophyll, chlorophyll *a* and chlorophyll *b* (µg/g) contents detected in the control and tap water treated fresh produce at day 0 and 6.

			Wash methods									
			Control (unwashed)					**Tap water**				
Day	**Samples**					**Average**	**SD**				**Average**	**SD**
0		A_{645}	0.042	0.044	0.044			0.041	0.041	0.039		
		A_{663}	0.12	0.122	0.121			0.128	0.124	0.127		
	Broccoli	Chlorophyll	181.08	186.72	185.92	184.58	3.05	185.48	182.27	180.63	182.79	2.46
		Chlorophyll *a*	141.10	143.10	141.83	142.01	1.01	151.53	146.45	150.80	149.59	2.75
		Chlorophyll *b*	40.02	43.66	44.13	42.61	2.25	33.99	35.86	29.87	33.24	3.06
		A_{645}	0.02	0.024	0.021			0.022	0.027	0.021		
		A_{663}	0.09	0.091	0.092			0.088	0.091	0.09		
	Cabbage	Chlorophyll	112.58	121.46	116.20	116.75	4.47	115.02	127.52	114.60	119.05	7.34
		Chlorophyll *a*	108.92	109.11	111.19	109.74	1.26	105.84	108.31	108.65	107.60	1.53
		Chlorophyll *b*	3.68	12.37	5.03	7.03	4.68	9.20	19.24	5.97	11.47	6.92
		A_{645}	0.075	0.074	0.071			0.075	0.076	0.079		
		A_{663}	0.245	0.243	0.245			0.235	0.234	0.233		
	Lettuce	Chlorophyll	347.99	344.37	339.91	344.09	4.05	339.97	341.19	346.45	342.53	3.44
		Chlorophyll *a*	290.98	288.70	292.05	290.58	1.71	278.28	276.74	274.66	276.56	1.81
		Chlorophyll *b*	57.09	55.74	47.93	53.59	4.94	61.77	64.53	71.87	66.05	5.22
		A_{645}	0.201	0.208	0.21			0.206	0.201	0.207		
		A_{663}	0.805	0.81	0.801			0.794	0.801	0.804		
	Spinach	Chlorophyll	1051.63	1069.78	1066.60	1062.67	9.69	1052.91	1048.42	1062.95	1054.76	7.44
		Chlorophyll *a*	968.28	972.75	960.78	967.27	6.05	952.97	963.20	965.40	960.52	6.63
		Chlorophyll *b*	83.55	97.24	106.03	95.61	11.33	100.15	85.42	97.76	94.44	7.90
6		A_{645}	0.025	0.028	0.029			0.026	0.024	0.025		
		A_{663}	0.1	0.108	0.107			0.102	0.1	0.104		
	Broccoli	Chlorophyll	130.70	143.18	144.39	139.42	7.58	134.32	128.68	133.91	132.30	3.15
		Chlorophyll *a*	120.28	129.63	128.09	126.00	5.02	122.55	120.54	125.36	122.82	2.42
		Chlorophyll *b*	10.45	13.58	16.33	13.45	2.94	11.80	8.16	8.58	9.51	1.99
		A_{645}	0.018	0.019	0.018			0.019	0.018	0.019		
		A_{663}	0.085	0.086	0.082			0.083	0.085	0.086		
	Cabbage	Chlorophyll	104.53	107.35	102.12	104.67	2.62	104.95	104.53	107.35	105.61	1.52
		Chlorophyll *a*	103.11	104.11	99.30	102.17	2.54	100.30	103.11	104.11	102.51	1.98
		Chlorophyll *b*	1.44	3.26	2.84	2.52	0.95	4.67	1.44	3.26	3.12	1.62
		A_{645}	0.055	0.056	0.056			0.051	0.05	0.052		
		A_{663}	0.239	0.238	0.235			0.238	0.239	0.233		

Table B88/ Cont.

		Wash methods									
		Control (unwashed)					Tap water				
Day	**Samples**				**Average**	**SD**				**Average**	**SD**
Lettuce	Chlorophyll	302.78	304.00	301.59	302.79	1.20	293.90	292.68	291.91	292.83	1.00
	Chlorophyll *a*	288.74	287.20	283.39	286.44	2.75	288.54	290.08	281.92	286.85	4.33
	Chlorophyll *b*	14.10	16.86	18.26	16.40	2.12	5.41	2.65	10.04	6.03	3.73
	A_{645}	0.185	0.182	0.185			0.188	0.181	0.178		
	A_{663}	0.806	0.805	0.801			0.8	0.801	0.802		
Spinach	Chlorophyll	1020.11	1013.25	1016.10	1016.49	3.45	1021.36	1008.02	1002.76	1010.72	9.59
	Chlorophyll *a*	973.86	973.39	967.51	971.58	3.54	965.43	968.58	970.66	968.22	2.63
	Chlorophyll *b*	46.44	40.04	48.78	45.09	4.53	56.12	39.62	32.28	42.68	12.21

SD = standard deviation; A = absorbance

Table B89: The absorbance and total chlorophyll, chlorophyll *a* and chlorophyll *b* (µg/g) contents detected in the NaCl and chlorine treated fresh produce at day 0 and 6.

			Wash methods									
				NaCl					**Chlorine**			
Day	**Samples**					**Average**	**SD**				**Average**	**SD**
0	Broccoli	A_{645}	0.038	0.042	0.042			0.047	0.041	0.045		
		A_{663}	0.121	0.12	0.128			0.119	0.116	0.127		
		Chlorophyll	173.80	181.08	187.50	180.79	6.85	190.38	175.85	192.75	186.33	9.15
		Chlorophyll *a*	143.45	141.10	151.26	145.27	5.32	138.49	136.29	149.19	141.32	6.90
		Chlorophyll *b*	30.39	40.02	36.28	35.56	4.85	51.94	39.60	43.61	45.05	6.29
	Cabbage	A_{645}	0.02	0.024	0.026			0.024	0.022	0.025		
		A_{663}	0.09	0.094	0.098			0.091	0.087	0.091		
		Chlorophyll	112.58	123.87	131.12	122.52	9.34	121.46	114.21	123.48	119.72	4.87
		Chlorophyll *a*	108.92	112.92	117.47	113.10	4.28	109.11	104.57	108.85	107.51	2.55
		Chlorophyll *b*	3.68	10.97	13.68	9.44	5.17	12.37	9.66	14.66	12.23	2.50
	Lettuce	A_{645}	0.071	0.076	0.075			0.072	0.071	0.072		
		A_{663}	0.231	0.234	0.237			0.238	0.23	0.231		
		Chlorophyll	328.68	341.19	341.57	337.15	7.33	336.32	327.88	330.70	331.63	4.29
		Chlorophyll *a*	274.27	276.74	280.82	277.27	3.31	282.89	273.00	274.00	276.63	5.44
		Chlorophyll *b*	54.48	64.53	60.83	59.95	5.08	53.50	54.95	56.77	55.07	1.64
	Spinach	A_{645}	0.209	0.204	0.211			0.204	0.201	0.201		
		A_{663}	0.809	0.8	0.8			0.801	0.811	0.807		
		Chlorophyll	1071.00	1053.68	1067.82	1064.17	9.22	1054.48	1056.44	1053.23	1054.72	1.62
		Chlorophyll *a*	971.21	961.12	959.24	963.86	6.44	962.39	975.90	970.82	969.71	6.82
		Chlorophyll *b*	100.00	92.76	108.79	100.52	8.03	92.29	80.74	82.61	85.22	6.20
6	Broccoli	A_{645}	0.025	0.029	0.028			0.027	0.028	0.027		
		A_{663}	0.108	0.1	0.101			0.102	0.103	0.102		
		Chlorophyll	137.12	138.78	137.56	137.82	0.86	136.34	139.17	136.34	137.28	1.63
		Chlorophyll *a*	130.44	119.20	120.74	123.46	6.09	122.28	123.28	122.28	122.61	0.58
		Chlorophyll *b*	6.71	19.61	16.85	14.39	6.80	14.09	15.92	14.09	14.70	1.05
	Cabbage	A_{645}	0.017	0.017	0.018			0.021	0.021	0.02		
		A_{663}	0.081	0.079	0.08			0.084	0.086	0.086		
		Chlorophyll	99.30	97.70	100.52	99.17	1.42	109.79	111.39	109.37	110.18	1.07
		Chlorophyll *a*	98.30	95.76	96.76	96.94	1.28	101.03	103.57	103.84	102.81	1.55
		Chlorophyll *b*	1.02	1.96	3.78	2.25	1.40	8.78	7.84	5.55	7.39	1.66
		A_{645}	0.053	0.05	0.052			0.052	0.053	0.052		

Table B89/ Cont.

			Wash methods									
			NaCl					Chlorine				
Day	**Samples**					**Average**	**SD**				**Average**	**SD**
	Lettuce	A_{663}	0.231	0.238	0.235			0.236	0.234	0.237		
		Chlorophyll	292.32	291.88	293.51	292.57	0.84	294.31	294.73	295.11	294.72	0.40
		Chlorophyll *a*	279.11	288.81	284.46	284.13	4.86	285.73	282.92	287.00	285.22	2.09
		Chlorophyll *b*	13.26	3.12	9.10	8.49	5.10	8.63	11.86	8.16	9.55	2.01
	Spinach	A_{645}	0.185	0.184	0.181			0.18	0.182	0.186		
		A_{663}	0.798	0.804	0.806			0.802	0.802	0.8		
		Chlorophyll	1013.70	1016.49	1012.03	1014.07	2.25	1006.80	1010.84	1017.32	1011.66	5.30
		Chlorophyll *a*	963.70	971.58	974.93	970.07	5.77	970.12	969.58	965.97	968.56	2.26
		Chlorophyll *b*	50.19	45.09	37.28	44.19	6.50	36.86	41.44	51.54	43.28	7.51

SD = standard deviation; A = absorbance

Table B90: The absorbance and total chlorophyll, chlorophyll *a* and chlorophyll *b* (μg/g) contents detected in the blanching and hydrogen peroxide treated fresh produce at day 0 and 6.

			Wash methods									
			Blanching					Hydrogen peroxide				
Day	Samples					Average	SD				Average	SD
0		A_{645}	0.046	0.048	0.047			0.044	0.049	0.041		
		A_{663}	0.12	0.123	0.124			0.121	0.122	0.128		
	Broccoli	Chlorophyll	189.16	195.61	194.39	193.05	3.42	185.92	196.82	185.48	189.41	6.43
		Chlorophyll *a*	140.03	143.30	144.84	142.72	2.46	141.83	141.76	151.53	145.04	5.62
		Chlorophyll *b*	49.18	52.36	49.60	50.38	1.73	44.13	55.11	33.99	44.41	10.57
		A_{645}	0.02	0.022	0.023			0.023	0.024	0.021		
		A_{663}	0.09	0.093	0.095			0.091	0.094	0.091		
	Cabbage	Chlorophyll	112.58	119.03	122.65	118.09	5.10	119.44	123.87	115.40	119.57	4.23
		Chlorophyll *a*	108.92	112.19	114.46	111.86	2.79	109.38	112.92	109.92	110.74	1.91
		Chlorophyll *b*	3.68	6.86	8.21	6.25	2.33	10.08	10.97	5.50	8.85	2.93
		A_{645}	0.072	0.07	0.071			0.074	0.077	0.072		
		A_{663}	0.231	0.236	0.231			0.238	0.235	0.237		
	Lettuce	Chlorophyll	330.70	330.67	328.68	330.02	1.16	340.36	344.01	335.51	339.96	4.26
		Chlorophyll *a*	274.00	280.89	274.27	276.39	3.90	282.35	277.74	281.62	280.57	2.48
		Chlorophyll *b*	56.77	49.85	54.48	53.70	3.53	58.08	66.35	53.96	59.46	6.31
		A_{645}	0.201	0.2	0.205			0.2	0.204	0.2		
		A_{663}	0.802	0.804	0.809			0.805	0.805	0.801		
	Spinach	Chlorophyll	1049.22	1048.81	1062.92	1053.65	8.03	1049.61	1057.69	1046.40	1051.23	5.82
		Chlorophyll *a*	964.47	967.28	972.29	968.01	3.96	968.55	967.47	963.47	966.50	2.68
		Chlorophyll *b*	84.95	81.73	90.84	85.84	4.62	81.26	90.42	83.13	84.94	4.84
6		A_{645}	0.029	0.027	0.026			0.029	0.027	0.028		
		A_{663}	0.104	0.103	0.104			0.101	0.105	0.104		
	Broccoli	Chlorophyll	141.99	137.15	135.93	138.35	3.21	139.58	138.75	139.97	139.43	0.62
		Chlorophyll *a*	124.28	123.55	125.09	124.30	0.77	120.47	126.09	124.55	123.70	2.90
		Chlorophyll *b*	17.74	13.63	10.87	14.08	3.46	19.14	12.69	15.45	15.76	3.24
		A_{645}	0.024	0.026	0.028			0.018	0.019	0.019		
		A_{663}	0.092	0.091	0.091			0.084	0.081	0.085		
	Cabbage	Chlorophyll	122.26	125.50	129.54	125.77	3.65	103.73	103.34	106.55	104.54	1.75
		Chlorophyll *a*	110.38	108.58	108.04	109.00	1.23	101.84	97.76	102.84	100.81	2.69
		Chlorophyll *b*	11.90	16.95	21.53	16.80	4.82	1.91	5.60	3.73	3.75	1.85

Table B90/ Cont.

			Wash methods									
			Blanching					Hydrogen peroxide				
Day	**Samples**					**Average**	**SD**				**Average**	**SD**
	Lettuce	A_{645}	0.056	0.058	0.059			0.053	0.052	0.055		
		A_{663}	0.238	0.24	0.246			0.234	0.235	0.238		
		Chlorophyll	304.00	309.64	316.47	310.04	6.25	294.73	293.51	301.98	296.74	4.58
		Chlorophyll *a*	287.20	289.20	296.55	290.98	4.92	282.92	284.46	287.47	284.95	2.31
		Chlorophyll *b*	16.86	20.50	19.98	19.11	1.97	11.86	9.10	14.57	11.84	2.73
	Spinach	A_{645}	0.187	0.196	0.193			0.185	0.183	0.179		
		A_{663}	0.802	0.804	0.805			0.801	0.809	0.802		
		Chlorophyll	1020.94	1040.73	1035.47	1032.38	10.25	1016.10	1018.48	1004.78	1013.12	7.32
		Chlorophyll *a*	968.24	968.36	970.43	969.01	1.23	967.51	978.20	970.39	972.03	5.54
		Chlorophyll *b*	52.89	72.57	65.23	63.56	9.94	48.78	40.46	34.57	41.27	7.14

SD = standard deviation; A = absorbance

Table B91: The absorbance and total chlorophyll, chlorophyll *a* and chlorophyll *b* (µg/g) contents detected in the UV treated fresh produce at day 0 and 6.

Day	Samples					Average	SD
0	Broccoli	A_{645}	0.045	0.044	0.048		
		A_{663}	0.122	0.124	0.127		
		Chlorophyll	188.74	188.33	198.81	191.96	5.94
		Chlorophyll *a*	142.84	145.64	148.38	145.62	2.77
		Chlorophyll *b*	45.95	42.73	50.48	46.39	3.90
	Cabbage	A_{645}	0.025	0.024	0.024		
		A_{663}	0.094	0.094	0.091		
		Chlorophyll	125.89	123.87	121.46	123.74	2.22
		Chlorophyll *a*	112.66	112.92	109.11	111.56	2.13
		Chlorophyll *b*	13.26	10.97	12.37	12.20	1.15
	Lettuce	A_{645}	0.079	0.077	0.078		
		A_{663}	0.235	0.239	0.234		
		Chlorophyll	348.05	347.22	345.23	346.83	1.45
		Chlorophyll *a*	277.20	282.82	276.20	278.74	3.57
		Chlorophyll *b*	70.93	64.48	69.11	68.17	3.33
	Spinach	A_{645}	0.211	0.21	0.207		
		A_{663}	0.809	0.801	0.81		
		Chlorophyll	1075.04	1066.60	1067.76	1069.80	4.57
		Chlorophyll *a*	970.67	960.78	973.02	968.16	6.49
		Chlorophyll *b*	104.58	106.03	94.95	101.85	6.02
6	Broccoli	A_{645}	0.033	0.031	0.032		
		A_{663}	0.111	0.107	0.105		
		Chlorophyll	155.68	148.43	148.85	150.99	4.07
		Chlorophyll *a*	132.09	127.55	124.74	128.13	3.71
		Chlorophyll *b*	23.62	20.91	24.14	22.89	1.73
	Cabbage	A_{645}	0.022	0.02	0.021		
		A_{663}	0.086	0.085	0.085		
		Chlorophyll	113.41	108.57	110.59	110.86	2.43
		Chlorophyll *a*	103.30	102.57	102.30	102.72	0.52
		Chlorophyll *b*	10.13	6.02	8.31	8.15	2.06
	Lettuce	A_{645}	0.054	0.053	0.052		
		A_{663}	0.234	0.237	0.234		
		Chlorophyll	296.75	297.13	292.71	295.53	2.45
		Chlorophyll *a*	282.65	286.73	283.19	284.19	2.22
		Chlorophyll *b*	14.15	10.45	9.57	11.39	2.43
	Spinach	A_{645}	0.192	0.193	0.193		
		A_{663}	0.809	0.808	0.807		
		Chlorophyll	1036.66	1037.88	1037.07	1037.20	0.62
		Chlorophyll *a*	975.78	974.24	972.97	974.33	1.41
		Chlorophyll *b*	61.07	63.83	64.29	63.06	1.74

SD = standard deviation; A = absorbance

Table B92: Sensory evaluation performed by 8 (A-H) random people before (day 0) and after refrigeration (day 6) on the tap water and NaCl treated broccoli samples, compared to a control.

Characteristics of the product	Washing method																			
	Tap water										NaCl									
Day 0	Visual quality																			
	A	**B**	**C**	**D**	**E**	**F**	**G**	**H**	**Average**	**SD**	**A**	**B**	**C**	**D**	**E**	**F**	**G**	**H**	**Average**	**SD**
Gloss	5	5	5	5	5	5	5	5	5.00	0.00	5	5	5	5	5	5	5	5	5.00	0.00
Freshness	5	5	10	10	10	10	10	10	8.75	2.31	5	10	10	10	5	10	10	10	8.75	2.31
Colour uniformity and intensity	5	5	5	5	5	5	5	5	5.00	0.00	5	5	5	5	0	5	5	5	4.38	1.77
	Additional characteristics																			
Colour	5	5	5	5	5	5	5	5	5.00	0.00	5	5	5	5	3	5	5	5	4.69	0.88
Texture	5	5	5	5	5	5	5	5	5.00	0.00	5	5	5	5	3	5	5	5	4.69	0.88
	Defects of the product																			
Off-odours	0	0	0	0	0	0	0	0	0.00	0.00	0	0	0	0	0	0	0	0	0.00	0.00
Decay	0	0	0	0	0	0	0	0	0.00	0.00	0	0	0	0	0	0	0	0	0.00	0.00
Tissue damage	0	0	0	0	0	2.5	0	0	0.31	0.88	0	0	0	0	0	0	0	0	0.00	0.00
Day 6	**Visual quality**																			
Gloss	5	5	5	5	5	5	5	5	5.00	0.00	5	5	5	5	5	5	5	5	5.00	0.00
Freshness	10	5	10	10	10	10	10	10	9.38	1.77	10	10	10	10	10	10	10	10	10.00	0.00
Colour uniformity and intensity	5	5	5	5	5	5	5	5	5.00	0.00	5	5	5	5	5	5	5	5	5.00	0.00
	Additional characteristics																			
Colour	5	5	5	5	5	5	5	5	5.00	0.00	5	5	5	5	5	5	5	5	5.00	0.00
Texture	5	5	5	5	5	5	5	5	5.00	0.00	5	5	5	5	5	5	5	5	5.00	0.00
	Defects of the product																			
Off-odours	0	0	0	0	0	0	0	0	0.00	0.00	0	0	0	0	0	0	0	0	0.00	0.00
Decay	0	0	0	0	0	0	0	0	0.00	0.00	0	0	0	0	0	0	0	0	0.00	0.00
Tissue damage	0	0	0	0	2.5	2.5	0	0	0.63	1.16	0	0	0	0	0	0	0	0	0.00	0.00

SD = standard deviation; Ave = Average

Table B93: Sensory evaluation performed by 8 (A-H) random people before (day 0) and after refrigeration (day 6) on the chlorine treated and blanched broccoli samples, compared to a control.

Characteristics of the product	Washing method																			
	Chlorine										Blanching									
Day 0								**Visual quality**												
	A	**B**	**C**	**D**	**E**	**F**	**G**	**H**	**Average**	**SD**	**A**	**B**	**C**	**D**	**E**	**F**	**G**	**H**	**Average**	**SD**
Gloss	5	5	5	5	5	5	5	5	5.00	0.00	10	10	10	10	10	10	10	10	10.00	0.00
Freshness	5	0	5	10	10	10	5	10	6.88	3.72	10	10	10	10	5	10	10	5	8.75	2.31
Colour uniformity and intensity	5	5	5	5	5	5	5	5	5.00	0.00	10	10	10	10	10	10	10	10	10.00	0.00
									Additional characteristics											
Colour	5	2.5	5	5	5	3	5	5	4.38	1.16	5	5	5	5	5	5	5	5	5.00	0.00
Texture	5	0	5	5	5	0	5	5	3.75	2.31	5	2.5	5	5	5	5	5	5	4.69	0.88
									Defects of the product											
Off-odours	0	0	3	3	3	5	3	3	2.19	1.60	0	0	0	0	3	3	5	0	1.25	1.89
Decay	0	0	3	0	0	0	3	0	0.63	1.16	0	0	0	0	0	0	0	0	0.00	0.00
Tissue damage	0	0	0	0	0	0	0	0	0.00	0.00	0	0	0	0	0	0	0	0	0.00	0.00
Day 6								**Visual quality**												
Gloss	5	5	5	5	5	5	5	5	5.00	0.00	10	10	10	10	10	10	10	10	10.00	0.00
Freshness	10	10	10	10	10	10	10	10	10.00	0.00	10	10	10	10	10	10	10	10	10.00	0.00
Colour uniformity and intensity	5	5	5	5	5	5	5	5	5.00	0.00	10	10	10	10	10	10	10	10	10.00	0.00
									Additional characteristics											
Colour	5	5	5	5	5	5	5	5	5.00	0.00	5	5	5	5	5	5	5	5	5.00	0.00
Texture	5	5	0	5	5	0	5	5	3.75	2.31	5	5	5	5	5	5	5	5	5.00	0.00
									Defects of the product											
Off-odours	0	0	0	3	3	0	5	3	1.56	1.86	0	0	0	0	0	0	0	0	0.00	0.00
Decay	0	0	0	3	0	0	0	0	0.31	0.88	0	0	0	0	0	0	0	0	0.00	0.00
Tissue damage	0	0	0	0	0	0	0	0	0.00	0.00	0	0	0	0	0	0	0	0	0.00	0.00

SD = standard deviation; Ave = Average

Table B94: Sensory evaluation performed by 8 (A-H) random people before (day 0) and after refrigeration (day 6) on the hydrogen peroxide and Ultra-violet (UV) light treated broccoli samples, compared to a control.

Characteristics of the product	**Treatment method**																			
	Hydrogen peroxide										**UV**									
	Day 0										**Visual quality**									
	A	**B**	**C**	**D**	**E**	**F**	**G**	**H**	**Average**	**SD**	**A**	**B**	**C**	**D**	**E**	**F**	**G**	**H**	**Average**	**SD**
Gloss	5	5	5	5	5	5	5	5	5.00	0.00	5	5	5	5	5	10	5	10	6.25	2.31
Freshness	10	10	10	10	10	10	10	10	10.00	0.00	5	5	10	10	10	10	10	10	8.75	2.31
Colour uniformity and intensity	5	5	5	5	5	5	5	5	5.00	0.00	5	5	5	5	5	10	5	10	6.25	2.31
Additional characteristics																				
Colour	5	2.5	5	5	5	5	5	5	4.69	0.88	5	2.5	5	5	5	5	5	5	4.69	0.88
Texture	5	2.5	5	3	5	3	5	3	3.75	1.34	5	2.5	5	5	5	5	5	5	4.69	0.88
Defects of the product																				
Off-odours	0	2.5	0	3	5	3	0	5	2.19	2.09	0	2.5	0	0	0	0	0	0	0.31	0.88
Decay	0	0	2.5	0	0	0	0	0	0.31	0.88	0	0	0	0	0	0	0	0	0.00	0.00
Tissue damage	0	0	2.5	0	0	0	0	0	0.31	0.88	0	0	0	0	0	0	0	0	0.00	0.00
Day 6	**Visual quality**																			
Gloss	5	5	5	5	5	5	5	5	5.00	0.00	5	10	10	5	5	5	5	10	6.88	2.59
Freshness	10	10	10	10	10	10	10	10	10.00	0.00	5	10	10	10	10	10	10	10	9.38	1.77
Colour uniformity and intensity	5	5	5	5	5	5	5	5	5.00	0.00	5	5	10	5	5	5	10	10	6.88	2.59
Additional characteristics																				
Colour	5	5	5	5	5	5	5	5	5.00	0.00	5	5	5	5	5	5	5	5	5.00	0.00
Texture	5	5	5	5	5	5	5	5	5.00	0.00	5	5	5	5	5	5	5	5	5.00	0.00
Defects of the product																				
Off-odours	0	0	0	3	0	0	0	0	0.31	0.88	0	0	0	0	0	0	0	0	0.00	0.00
Decay	0	5	0	0	0	0	0	0	0.63	1.77	0	0	0	0	0	0	0	0	0.00	0.00
Tissue damage	0	0	0	0	0	0	3	0	0.31	0.88	0	0	0	0	0	0	0	0	0.00	0.00

SD = standard deviation; Ave = Average

Table B95: Sensory evaluation performed by 8 (A-H) random people before (day 0) and after refrigeration (day 6) on the tap water and NaCl treated cabbage samples, compared to a control.

Characteristics of the product	Washing method																			
	Tap water										NaCl									
Day 0	**Visual quality**																			
	A	**B**	**C**	**D**	**E**	**F**	**G**	**H**	**Average**	**SD**	**A**	**B**	**C**	**D**	**E**	**F**	**G**	**H**	**Average**	**SD**
Gloss	5	10	5	5	5	5	5	5	5.63	1.77	5	5	5	5	5	5	5	5	5.00	0.00
Freshness	10	10	10	10	10	10	10	10	10.00	0.00	10	5	10	10	10	10	10	10	9.38	1.77
Colour uniformity and intensity	5	10	5	5	5	5	5	5	5.63	1.77	5	5	5	5	5	5	5	5	5.00	0.00
	Additional characteristics																			
Colour	5	5	5	5	5	5	5	5	5.00	0.00	5	5	5	5	5	5	5	5	5.00	0.00
Texture	5	5	5	5	5	5	5	5	5.00	0.00	5	5	5	5	5	5	5	5	5.00	0.00
	Defects of the product																			
Off-odours	0	0	0	0	0	0	0	0	0.00	0.00	0	0	0	0	0	0	0	0	0.00	0.00
Decay	0	0	0	0	0	0	0	0	0.00	0.00	0	0	0	0	0	0	0	0	0.00	0.00
Tissue damage	0	0	0	0	0	0	0	0	0.00	0.00	0	0	0	0	0	0	0	0	0.00	0.00
Day 6	**Visual quality**																			
Gloss	5	5	5	5	5	5	5	5	5.00	0.00	5	5	5	5	5	5	5	5	5.00	0.00
Freshness	10	10	10	10	10	10	10	10	10.00	0.00	10	10	10	10	10	10	10	10	10.00	0.00
Colour uniformity and intensity	5	5	5	5	5	5	5	5	5.00	0.00	5	5	5	5	5	5	5	5	5.00	0.00
	Additional characteristics																			
Colour	5	5	5	5	5	5	5	5	5.00	0.00	5	5	5	5	5	5	5	5	5.00	0.00
Texture	5	5	5	5	5	5	5	5	5.00	0.00	5	5	5	5	5	5	5	5	5.00	0.00
	Defects of the product																			
Off-odours	0	0	0	0	0	0	0	0	0.00	0.00	0	0	0	0	0	0	0	0	0.00	0.00
Decay	0	0	2.5	2.5	2.5	0	0	0	0.94	1.29	0	0	2.5	0	2.5	2.5	0	0	0.94	1.29
Tissue damage	0	0	0	0	0	0	0	0	0.00	0.00	0	0	0	0	0	0	0	0	0.00	0.00

SD = standard deviation; Ave = Average

Table B96: Sensory evaluation performed by 8 (A-H) random people before (day 0) and after refrigeration (day 6) on the chlorine treated and blanched cabbage samples, compared to a control.

Characteristics of the product	Washing method																			
	Chlorine										Blanching									
Day 0							**Visual quality**													
	A	**B**	**C**	**D**	**E**	**F**	**G**	**H**	**Average**	**SD**	**A**	**B**	**C**	**D**	**E**	**F**	**G**	**H**	**Average**	**SD**
Gloss	5	5	5	5	5	5	5	5	5.00	0.00	10	10	10	10	10	10	10	10	10.00	0.00
Freshness	10	5	10	10	10	10	10	10	9.38	1.77	10	5	5	10	5	10	10	10	8.13	2.59
Colour uniformity and intensity	5	5	5	5	5	5	5	5	5.00	0.00	10	10	10	10	10	10	10	10	10.00	0.00
									Additional characteristics											
Colour	5	2.5	5	5	5	5	5	5	4.69	0.88	5	5	5	5	5	5	5	5	5.00	0.00
Texture	5	2.5	5	5	5	5	5	5	4.69	0.88	2.5	2.5	2.5	2.5	0	2.5	0	0	1.56	1.29
									Defects of the product											
Off-odours	0	0	2.5	5	5	0	2.5	0	1.88	2.22	2.5	0	0	2.5	0	2.5	2.5	0	1.25	1.34
Decay	0	0	0	0	0	0	0	0	0.00	0.00	0	0	0	0	0	0	0	0	0.00	0.00
Tissue damage	0	0	0	0	0	0	0	0	0.00	0.00	0	0	0	0	0	0	0	0	0.00	0.00
Day 6							**Visual quality**													
Gloss	5	5	5	5	5	5	5	5	5.00	0.00	10	10	10	10	10	10	10	10	10.00	0.00
Freshness	10	5	10	5	5	10	10	10	8.13	2.59	10	0	0	0	0	5	5	0	2.50	3.78
Colour uniformity and intensity	5	5	5	5	5	5	5	5	5.00	0.00	10	10	10	10	10	10	10	10	10.00	0.00
									Additional characteristics											
Colour	5	5	5	5	5	5	5	5	5.00	0.00	5	5	5	5	5	5	5	5	5.00	0.00
Texture	5	5	5	5	5	5	5	5	5.00	0.00	0	2.5	0	2.5	2.5	0	0	0	0.94	1.29
									Defects of the product											
Off-odours	0	0	0	5	0	0	2.5	5	1.56	2.29	2.5	5	5	2.5	0	2.5	2.5	0	2.50	1.89
Decay	0	0	0	0	0	0	0	0	0.00	0.00	0	0	2.5	5	0	5	0	2.5	1.88	2.22
Tissue damage	0	0	0	0	0	0	0	0	0.00	0.00	0	0	0	0	0	0	0	0	0.00	0.00

SD = standard deviation; Ave = Average

Table B97: Sensory evaluation performed by 8 (A-H) random people before (day 0) and after refrigeration (day 6) on the hydrogen peroxide and ultra-violet (UV) light treated cabbage samples, compared to a control.

Characteristics of the product	Treatment method																			
	Hydrogen peroxide										UV									
Day 0	**Visual quality**																			
	A	**B**	**C**	**D**	**E**	**F**	**G**	**H**	**Average**	**SD**	**A**	**B**	**C**	**D**	**E**	**F**	**G**	**H**	**Average**	**SD**
Gloss	5	10	5	5	5	5	5	5	5.63	1.77	5	5	5	5	5	5	5	5	5.00	0.00
Freshness	10	5	10	5	5	5	5	10	6.88	2.59	10	10	10	10	10	10	10	10	10.00	0.00
Colour uniformity and intensity	2.5	2.5	5	0	2.5	2.5	2.5	2.5	2.50	1.34	5	5	5	5	5	5	5	5	5.00	0.00
	Additional characteristics																			
Colour	0	2.5	0	0	2.5	2.5	0	0	0.94	1.29	5	5	5	5	5	5	5	5	5.00	0.00
Texture	5	0	2.5	5	2.5	0	5	2.5	2.81	2.09	5	5	5	5	5	5	5	5	5.00	0.00
	Defects of the product																			
Off-odours	0	0	2.5	5	2.5	0	5	2.5	2.19	2.09	0	5	0	5	0	0	0	0	1.25	2.31
Decay	0	0	0	0	0	0	0	0	0.00	0.00	0	0	0	0	0	0	0	0	0.00	0.00
Tissue damage	0	0	0	0	0	0	0	0	0.00	0.00	0	0	0	0	0	0	0	0	0.00	0.00
Day 6	**Visual quality**																			
Gloss	5	5	5	5	5	5	5	5	5.00	0.00	5	5	5	5	5	5	5	5	5.00	0.00
Freshness	10	10	10	10	10	10	10	10	10.00	0.00	10	10	10	10	10	10	10	10	10.00	0.00
Colour uniformity and intensity	2.5	5	5	0	2.5	2.5	2.5	2.5	2.81	1.60	5	5	5	5	5	5	5	5	5.00	0.00
	Additional characteristics																			
Colour	2.5	0	5	2.5	2.5	2.5	5	0	2.50	1.89	5	5	5	5	5	5	5	5	5.00	0.00
Texture	5	5	5	5	5	5	5	5	5.00	0.00	5	5	5	5	5	5	5	5	5.00	0.00
	Defects of the product																			
Off-odours	0	0	0	0	0	0	2.5	0	0.31	0.88	0	0	0	2.5	0	0	0	0	0.31	0.88
Decay	0	0	0	0	0	0	0	0	0.00	0.00	0	0	0	0	0	0	0	0	0.00	0.00
Tissue damage	0	0	0	0	0	0	0	0	0.00	0.00	0	0	0	0	0	2.5	0	0	0.31	0.88

SD = standard deviation; Ave = Average

Table B98: Sensory evaluation performed by 8 (A-H) random people before (day 0) and after refrigeration (day 6) on the tap water and NaCl treated lettuce samples, compared to a control.

Characteristics of the product	Washing method																			
	Tap water										NaCl									
Day 0	**Visual quality**																			
	A	**B**	**C**	**D**	**E**	**F**	**G**	**H**	**Average**	**SD**	**A**	**B**	**C**	**D**	**E**	**F**	**G**	**H**	**Average**	**SD**
Gloss	5	10	10	10	5	10	5	10	8.13	2.59	5	5	5	5	5	5	5	5	5.00	0.00
Freshness	10	0	5	5	10	10	10	5	6.88	3.72	10	5	10	10	10	10	10	10	9.38	1.77
Colour uniformity and intensity	5	5	10	10	10	10	10	10	8.75	2.31	5	5	5	5	5	5	5	5	5.00	0.00
	Additional characteristics																			
Colour	5	2.5	5	5	5	5	5	5	4.69	0.88	5	5	5	5	5	5	5	5	5.00	0.00
Texture	5	2.5	5	5	5	5	5	5	4.69	0.88	5	5	5	5	5	5	5	5	5.00	0.00
	Defects of the product																			
Off-odours	0	0	0	0	0	0	0	0	0.00	0.00	0	0	0	0	0	0	0	0	0.00	0.00
Decay	0	0	0	0	0	0	0	0	0.00	0.00	0	0	0	0	0	0	0	0	0.00	0.00
Tissue damage	0	2.5	0	0	0	0	0	0	0.31	0.88	0	0	0	0	0	0	0	0	0.00	0.00
Day 6	**Visual quality**																			
Gloss	5	5	10	5	5	5	5	5	5.63	1.77	5	5	5	5	5	5	5	5	5.00	0.00
Freshness	10	10	10	10	10	10	10	10	10.00	0.00	10	10	10	10	10	10	10	10	10.00	0.00
Colour uniformity and intensity	5	5	10	5	5	5	5	5	5.63	1.77	5	5	5	5	5	5	5	5	5.00	0.00
	Additional characteristics																			
Colour	5	5	2.5	5	5	2.5	5	5	4.38	1.16	5	5	5	5	5	5	5	5	5.00	0.00
Texture	5	5	5	5	5	2.5	2.5	5	4.38	1.16	5	5	5	5	5	5	5	5	5.00	0.00
	Defects of the product																			
Off-odours	0	0	0	0	0	0	0	0	0.00	0.00	0	0	0	0	0	0	0	0	0.00	0.00
Decay	2.5	0	2.5	0	5	5	5	0	2.50	2.31	0	0	0	0	0	0	0	0	0.00	0.00
Tissue damage	0	0	0	0	0	0	0	0	0.00	0.00	0	0	0	0	0	0	0	0	0.00	0.00

SD = standard deviation; Ave = Average

Table B99: Sensory evaluation performed by 8 (A-H) random people before (day 0) and after refrigeration (day 6) on the chlorine treated and blanched lettuce samples, compared to a control.

Characteristics of the product	Washing method																			
	Chlorine										Blanching									
Day 0	**Visual quality**																			
	A	**B**	**C**	**D**	**E**	**F**	**G**	**H**	**Average**	**SD**	**A**	**B**	**C**	**D**	**E**	**F**	**G**	**H**	**Average**	**SD**
Gloss	5	5	5	5	5	5	5	5	5.00	0.00	0	0	0	5	0	0	0	0	0.63	1.77
Freshness	10	10	10	10	10	10	10	10	10.00	0.00	0	0	0	0	0	0	0	0	0.00	0.00
Colour uniformity and intensity	5	5	5	5	10	5	5	5	5.63	1.77	10	0	10	10	10	5	5	5	6.88	3.72
	Additional characteristics																			
Colour	5	2.5	5	5	5	5	5	2.5	4.38	1.16	5	0	5	5	5	5	5	5	4.38	1.77
Texture	5	5	5	5	5	5	5	5	5.00	0.00	0	0	2.5	0	2.5	2.5	0	0	0.94	1.29
	Defects of the product																			
Off-odours	0	0	0	2.5	0	0	2.5	2.5	0.94	1.29	0	0	2.5	5	5	5	0	0	2.19	2.48
Decay	0	0	0	0	0	0	0	0	0.00	0.00	0	2.5	0	0	0	0	0	0	0.31	0.88
Tissue damage	0	2.5	0	2.5	2.5	0	2.5	2.5	1.56	1.29	5	2.5	2.5	2.5	2.5	5	5	2.5	3.44	1.29
Day 6	**Visual quality**																			
Gloss	5	5	5	5	5	5	5	5	5.00	0.00	0	0	0	5	0	0	0	0	0.63	1.77
Freshness	10	10	10	10	10	10	10	10	10.00	0.00	0	0	0	0	0	0	0	0	0.00	0.00
Colour uniformity and intensity	5	5	5	5	5	5	5	5	5.00	5.00	10	5	5	5	5	5	5	5	5.63	1.77
	Additional characteristics																			
Colour	5	5	5	5	5	5	5	5	5.00	0.00	5	0	5	5	5	5	5	5	4.38	1.77
Texture	5	5	5	5	5	5	5	5	5.00	0.00	0	0	2.5	0	2.5	2.5	0	0	0.94	1.29
	Defects of the product																			
Off-odours	0	0	0	0	0	0	0	0	0.00	0.00	5	5	5	5	5	5	0	5	4.38	1.77
Decay	0	0	0	0	0	0	0	0	0.00	0.00	0	0	0	0	0	0	0	0	0.00	0.00
Tissue damage	0	0	0	2.5	2.5	2.5	2.5	2.5	1.56	1.29	5	2.5	5	5	5	5	5	5	4.69	0.88

SD = standard deviation; Ave = Average

Table B100: Sensory evaluation performed by 8 (A-H) random people before (day 0) and after refrigeration (day 6) on the hydrogen peroxide and ultra-violet (UV) light treated lettuce samples, compared to a control.

Characteristics of the product	Treatment method																			
	Hydrogen peroxide										UV									
Day 0	**Visual quality**																			
	A	**B**	**C**	**D**	**E**	**F**	**G**	**H**	**Average**	**SD**	**A**	**B**	**C**	**D**	**E**	**F**	**G**	**H**	**Average**	**SD**
Gloss	5	10	5	5	5	5	5	5	5.63	1.77	5	5	5	5	5	5	5	5	5.00	0.00
Freshness	10	5	10	5	5	5	5	10	6.88	2.59	10	10	10	10	10	10	10	10	10.00	0.00
Colour uniformity and intensity	2.5	2.5	5	0	2.5	2.5	2.5	2.5	2.50	1.34	5	5	5	5	5	5	5	5	5.00	0.00
	Additional characteristics																			
Colour	0	2.5	0	0	2.5	2.5	0	0	0.94	1.29	5	5	5	5	5	5	5	5	5.00	0.00
Texture	5	0	2.5	5	2.5	0	5	2.5	2.81	2.09	5	5	5	5	5	5	5	5	5.00	0.00
	Defects of the product																			
Off-odours	0	0	2.5	5	2.5	0	5	2.5	2.19	2.09	0	5	0	5	0	0	0	0	1.25	2.31
Decay	0	0	0	0	0	0	0	0	0.00	0.00	0	0	0	0	0	0	0	0	0.00	0.00
Tissue damage	0	0	0	0	0	0	0	0	0.00	0.00	0	0	0	0	0	0	0	0	0.00	0.00
Day 6	**Visual quality**																			
Gloss	5	5	5	5	5	5	5	5	5.00	0.00	5	5	5	5	5	5	5	5	5.00	0.00
Freshness	10	10	10	10	10	10	10	10	10.00	0.00	10	10	10	10	10	10	10	10	10.00	0.00
Colour uniformity and intensity	2.5	5	5	0	2.5	2.5	2.5	2.5	2.81	1.60	5	5	5	5	5	5	5	5	5.00	0.00
	Additional characteristics																			
Colour	2.5	0	5	2.5	2.5	2.5	5	0	2.50	1.89	5	5	5	5	5	5	5	5	5.00	0.00
Texture	5	5	5	5	5	5	5	5	5.00	0.00	5	5	5	5	5	5	5	5	5.00	0.00
	Defects of the product																			
Off-odours	0	0	0	0	0	0	2.5	0	0.31	0.88	0	0	0	2.5	0	0	0	0	0.31	0.88
Decay	0	0	0	0	0	0	0	0	0.00	0.00	0	0	0	0	0	0	0	0	0.00	0.00
Tissue damage	0	0	0	0	0	0	0	0	0.00	0.00	0	0	0	0	0	2.5	0	0	0.31	0.88

SD = standard deviation; Ave = Average

Table B101: Sensory evaluation performed by 8 (A-H) random people before (day 0) and after refrigeration (day 6) on the tap water and NaCl treated spinach samples, compared to a control.

	Washing method																			
Characteristics of the product	**Tap water**										**NaCl**									
Day 0	**Visual quality**																			
	A	**B**	**C**	**D**	**E**	**F**	**G**	**H**	**Average**	**SD**	**A**	**B**	**C**	**D**	**E**	**F**	**G**	**H**	**Average**	**SD**
Gloss	5	5	0	5	5	5	5	5	4.38	1.77	5	5	5	5	5	5	5	5	5.00	0.00
Freshness	10	10	0	10	10	5	10	10	8.13	3.72	10	5	5	5	10	10	10	5	7.50	2.67
Colour uniformity and intensity	5	5	5	5	5	5	5	5	5.00	0.00	5	5	5	5	5	5	5	5	5.00	0.00
	Additional characteristics																			
Colour	5	5	2.5	5	5	5	5	5	4.69	0.88	5	5	5	5	5	5	5	5	5.00	0.00
Texture	5	5	2.5	5	5	2.5	5	5	4.38	1.16	5	2.5	5	5	5	5	5	5	4.69	0.88
	Defects of the product																			
Off-odours	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0.00	0.00
Decay	0	0	0	0	0	0	0	0	0.00	0.00	0	0	0	0	0	0	0	0	0.00	0.00
Tissue damage	0	0	0	0	0	2.5	0	0	0.31	0.88	0	0	0	0	0	0	0	0	0.00	0.00
Day 6	**Visual quality**																			
Gloss	5	5	5	5	5	5	5	5	5.00	0.00	5	5	5	5	5	5	5	5	5.00	0.00
Freshness	10	10	10	10	10	5	5	5	8.13	2.59	10	10	10	5	10	5	5	5	7.50	2.67
Colour uniformity and intensity	5	5	5	5	5	5	5	5	5.00	0.00	5	5	5	5	5	5	5	5	5.00	0.00
	Additional characteristics																			
Colour	5	5	2.5	5	5	5	5	5	4.69	0.88	5	5	5	5	5	5	5	5	5.00	0.00
Texture	5	5	2.5	5	5	2.5	5	5	4.38	1.16	5	5	5	5	5	5	5	5	5.00	0.00
	Defects of the product																			
Off-odours	0	2.5	2.5	5	0	0	0	0	1.25	1.89	0	0	0	0	0	0	0	0	0.00	0.00
Decay	0	0	0	0	0	0	0	0	0.00	0.00	0	0	0	0	0	0	0	0	0.00	0.00
Tissue damage	0	0	0	0	0	2.5	0	0	0.31	0.88	0	0	0	0	0	0	0	0	0.00	0.00

SD = standard deviation; Ave = Average

Table B102: Sensory evaluation performed by 8 (A-H) random people before (day 0) and after refrigeration (day 6) on the chlorine treated and blanched spinach samples, compared to a control.

	Washing method																			
Characteristics of the product	**Chlorine**										**Blanching**									
Day 0	**Visual quality**																			
	A	**B**	**C**	**D**	**E**	**F**	**G**	**H**	**Average**	**SD**	**A**	**B**	**C**	**D**	**E**	**F**	**G**	**H**	**Average**	**SD**
Gloss	5	5	5	5	5	5	5	5	5.00	0.00	5	5	5	5	5	5	5	5	5.00	0.00
Freshness	10	5	10	10	10	10	10	10	9.38	1.77	10	10	10	10	10	10	5	10	9.38	1.77
Colour uniformity and intensity	5	5	5	5	5	5	5	5	5.00	0.00	5	5	5	5	5	5	5	5	5.00	0.00
	Additional characteristics																			
Colour	5	5	5	5	5	5	5	5	5.00	0.00	5	5	5	2.5	5	5	2.5	5	4.38	1.16
Texture	5	2.5	5	5	5	5	5	5	4.69	0.88	0	0	5	5	2.5	0	0	0	1.56	2.29
	Defects of the product																			
Off-odours	0	0	0	5	5	2.5	2.5	2.5	2.19	2.09	2.5	0	5	0	5	2.5	2.5	2.5	2.50	1.89
Decay	0	0	0	0	0	0	0	0	0.00	0.00	2.5	0	0	0	0	0	0	0	0.31	0.88
Tissue damage	0	0	0	0	0	0	0	0	0.00	0.00	5	0	2.5	5	2.5	2.5	2.5	2.5	2.81	1.60
Day 6	**Visual quality**																			
Gloss	5	5	5	5	5	5	5	5	5.00	0.00	5	5	5	5	5	5	5	5	5.00	0.00
Freshness	10	10	10	10	10	10	10	10	10.00	0.00	10	10	10	10	10	10	5	10	9.38	1.77
Colour uniformity and intensity	5	5	5	5	5	5	5	5	5.00	0.00	5	5	5	5	5	5	5	5	5.00	0.00
	Additional characteristics																			
Colour	5	5	5	5	5	5	5	5	5.00	0.00	5	5	5	5	5	5	5	5	5.00	0.00
Texture	5	5	5	5	5	5	5	5	5.00	0.00	0	0	0	0	0	0	0	0	0.00	0.00
	Defects of the product																			
Off-odours	0	0	0	0	0	0	0	0	0.00	0.00	2.5	5	5	5	5	5	2.5	5	4.38	1.16
Decay	0	0	0	0	0	0	0	0	0.00	0.00	2.5	0	0	0	0	0	0	0	0.31	0.88
Tissue damage	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.50	0.00	5	5	0	5	2.5	5	5	2.5	3.75	1.89

SD = standard deviation; Ave = Average

Table B103: Sensory evaluation performed by 8 (A-H) random people before (day 0) and after refrigeration (day 6) on the hydrogen peroxide and ultra-violet (UV) light treated spinach samples, compared to a control.

Characteristics of the product	Treatment method																			
	Hydrogen peroxide										UV									
Day 0	**Visual quality**																			
	A	**B**	**C**	**D**	**E**	**F**	**G**	**H**	**Average**	**SD**	**A**	**B**	**C**	**D**	**E**	**F**	**G**	**H**	**Average**	**SD**
Gloss	5	5	5	5	5	5	5	5	5.00	0.00	5	10	5	5	5	5	5	10	6.25	2.31
Freshness	0	5	0	10	10	10	10	10	6.88	4.58	10	10	5	10	5	10	10	10	8.75	2.31
Colour uniformity and intensity	5	5	5	5	5	5	5	5	5.00	0.00	5	5	5	5	5	5	5	5	5.00	0.00
	Additional characteristics																			
Colour	5	5	5	5	5	5	5	5	5.00	0.00	5	5	5	5	5	5	5	5	5.00	0.00
Texture	5	2.5	5	2.5	2.5	2.5	2.5	2.5	3.13	1.16	5	5	2.5	5	2.5	5	5	5	4.38	1.16
	Defects of the product																			
Off-odours	0	0	2.5	0	0	0	0	0	0.31	0.88	0	0	0	0	2.5	0	0	0	0.31	0.88
Decay	0	0	2.5	0	0	0	0	0	0.31	0.88	0	5	5	0	2.5	2.5	0	0	1.88	2.39
Tissue damage	0	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.19	0.88	0	0	5	0	2.5	0	0	0	0.94	1.86
Day 6	**Visual quality**																			
Gloss	5	5	5	5	5	5	5	5	5.00	0.00	5	5	5	5	5	5	5	5	5.00	0.00
Freshness	10	10	10	10	10	10	10	10	10.00	0.00	10	5	10	10	10	10	10	10	9.38	1.77
Colour uniformity and intensity	5	5	5	5	5	5	5	5	5.00	0.00	5	10	5	5	5	5	5	5	5.63	1.77
	Additional characteristics																			
Colour	5	5	5	5	5	5	5	5	5.00	0.00	5	5	5	5	5	5	5	5	5.00	0.00
Texture	5	5	5	5	5	5	5	5	5.00	0.00	5	5	5	5	5	5	5	5	5.00	0.00
	Defects of the product																			
Off-odours	0	0	0	0	0	0	0	0	0.00	0.00	0	0	0	0	0	0	0	0	0.00	0.00
Decay	0	0	0	0	0	0	0	0	0.00	0.00	0	0	0	0	0	0	0	0	0.00	0.00
Tissue damage	0	0	0	0	0	0	0	0	0.00	0.00	0	0	0	0	0	0	0	0	0.00	0.00

SD = standard deviation; Ave = Average