The frequency of insulin resistance and hyperlipidaemia in women with Polycystic Ovarian Syndrome (PCOS) attending Inkosi Albert Luthuli Central Hospital

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This dissertation is submitted to the University of KwaZulu Natal in fulfilment of the requirement for the degree of MMed.

Declaration

I, Nitasha Magan , hereby declare that the work on which this dissertation is based is original and is my own unaided work carried out by me, under the supervison of Professor J.S. Bagratee.

Signed: _____

Date : _____

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I would like to thank my parents and brother for their tolerance, support and encouragement.

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GLOSSARY

BMI	Body Mass Index
DHEAS	Dihydroepiandrosterone sulphate
FSH	Follicular Stimulating Hormone
GNRH	Gonadotropin Releasing Hormone
HDL	High density lipoprotein
IGF	Insulin Like Growth Factor
IALCH	Inkosi Albert Luthuli Central Hospital
LH	Lutenising Hormone
LDL	Low density lipoprotein
17ά OHP	17ά Hydroxyprogesterone
PCOS	Polycystic Ovarian Syndrome
PRL	Prolactin
SD	Standard Deviation
SHBG	Sex Hormone Binding Globulin
TSH	Thyroid Stimulating Hormone
T4	Thyroxin

ABSTRACT

BACKGROUND

Polycystic ovarian syndrome is one of the commonest endocrinopathies in women of reproductive age. The prevalence of the disease is estimated to be around 5 % in general population (Azziz, 2004). Literature on the prevalence of PCOS in Black women is limited (Knochenhauer, 1998). This syndrome is a diagnostic conundrum due to the phenotypic variability of these women. The PCOS woman also has a greater disposition for impaired glucose homeostasis as well as hyperlipidaemia.

OBJECTIVE

The hormonal and metabolic profiles of South African women with PCOS have not been described. Ethnic differences in the prevalence of PCOS have also not been well explored. Our study aims to describe and compare the phenotypic profile of African and Indian women with PCOS and to determine the frequency of insulin resistance and hyperlipidaemia in these women.

METHODS

A retrospective audit of all patients attending gynaecology endocrine and infertility clinics over the period June 2005 to June 2009 was carried out. The biochemical and clinical profiles were analysed and a comparative analysis between the two largest groups, Indian and Black women were done. All women that attended these clinics were subjected to a fasting lipogram and fasting serum glucose. An abnormal fasting serum glucose would have necessitated a full glucose tolerance test.

RESULTS

A total of 110 patients were analysed in this study. There were 87 Indian patients, 16 Black patients, 5 Coloured patients and 2 White patients. Eighty nine percent of PCOS women studied had an increased body mass index (>25). There was an increased LH:FSH in 66 (75.9%) of Indian women and 13 (81.3%) of Black women. Increased androgens were present in 26 (30.2%) in Indian women and 6 (37.5%) of Black women. An increase in fasting insulin was found in 48 (55.2%) of the Indian women and 5 (31.3%) of the Black women. Twenty five (29.1%) Indian women had an increase in fasting serum glucose compared to 1 (6.3%) in Black women. In the Indian population, 13(14.9%) were found to have Diabetes Mellitus, and 9 (10.3%) had an impaired glucose tolerance test. In the Black population only 1 patient had impaired glucose tolerance. There were no Black patients with Diabetes Mellitus. No Black women were found to have hyperlipidaemia, however 12 (14.3%) Indian women were affected. None of these differences between the races were statistically significant. The major limitation of the study was the sample size of Black women. This is an ongoing study, and aims to recruit more Black women. This will be able to adequately address the correct perspective regarding the metabolic and cardiovascular abnormalities in these women.

CONCLUSION

The prevalence of insulin resistance and hyperlipidaemia in local women with PCOS was 50.9%.and 11.3% respectively. Menstrual irregularities and infertility are the most frequent presenting complaints of women with PCOS. Features of hyperandrogenism are not common presenting complaints in South African women. There are no differences in the hormonal and clinical profile of South African Indian and Black women with PCOS, however, there is a trend toward Indian women having a greater prevalence of glucose abnormalities than Black women. We recommend further studies in the management of the metabolic abnormalities in local women with PCOS, in an attempt to develop a protocol to manage the metabolic complexities of PCOS.

1. **INTRODUCTION**

Polycystic ovarian syndrome (PCOS) is one of the commonest endocrinopathies in women of reproductive age. The prevalence of the disease is estimated to be around 5 % in the general population (Azziz et al, 2004).

The exact aetiology of PCOS remains a conundrum. A genetic association has been suggested, however, the gene or genes responsible are yet to be discovered (Kahsar-Miller et al, 2001). Other theories suggest androgen overexposure in the prenatal period (Xita et al, 2002). The exact mechanism of this excessive androgen exposure resulting in PCOS, as yet, remains undefined. A strong familial link has also been noted (Balen et al, 2004). An estimated fifty per cent of first degree relatives have PCOS suggesting a dominant pattern of inheritance.

Women with PCOS have a variety of clinical presentations. These range from increased weight gain, dysfunctional uterine bleeding, oligo/amenorrhea, acne, hirsuitism, acanthosis nigricans and infertility. Psychosocial implications emanating from negative self-image can manifest in depression, body dysmorphic disoder and eating disorders (Bishop et al, 2009).

The diagnosis of PCOS had previously been difficult due to the diverse phenotypes of the patient population. The American Society for Reproductive Medicine and the European Society of Human Reproduction and Embryology have since developed the Rotterdam Criteria (2003) to diagnose PCOS.

The diagnosis is made on the patient having two of the three criteria

i) Oligo or anovulation

ii) Hyperandrogenism – clinical or biochemical

iii) ultrasound evidence of PCO.

The characteristic hormonal marker of PCOS is hyperandrogenism. Ovarian hyperandrogenaemia occurs as a result of an increase in luteinising hormone secretion due to aberrant gonadotropic releasing hormone secretion. This hyperandrogenaemia is further perpetuated by insulin, due to insulin induced suppression of hepatic production of sex hormone binding globulin and insulin-like growth factor binding protein. Hyperinsulinaemia or insulin resistance is present in 20 – 60 per cent of women with PCOS (Azziz et al, 2003).

Insulin resistance and hyperandrogenism have several metabolic sequelae. Diabetes Mellitis and impaired glucose tolerance are present in 10 per cent and 35 per cent of women with PCOS respectively (Legro et al, 1999). Obesity, as defined as a body mass index (BMI) greater than 30 kg/m^2, is present in at least 50 per cent of women with PCOS (Martinez-Bermejo, 2007). The combination of hyperinsulinaemia, hyperandrogenaemia and obesity predisposes these women to cardiovascular disease and the Metabolic Syndrome. NHANES III survey showed the prevalence of the Metabolic syndrome in women in the general population to be 23.7% (Ford et al , 2002). The Metabolic Syndrome is prevalent in 43% of women with PCOS. (Apridonidze et al, 2004).

Treatment strategies for women with PCOS are symptomatic. Non pharmacological methods include weight loss, cosmetic depilation and lifestyle modifications, such as exercise. Pharmacological agents are used as an adjunct to non pharmacological methods. These are directed at reducing hyperinsulinaemia, hyperandrogenaemia and the relevant metabolic sequelae. Drugs commonly used are oral hypoglycaemics eg. Metformin and antiandrogens such as cyproterone acetate. Oral contraceptives are used to regulate menstruation as well as providing an anti androgenic effect.

PCOS has been shown to demonstrate phenotypic variability amongst different ethnic groups. The highest reported prevalence of PCOS has been the South Asian population that have immigrated to Britain (Wijeyaratne et al, 2002). These South Asian women with PCOS were found to have a greater degree of insulin resistance as compared to their Caucasian counterparts. In addition, these women also display features of PCOS at an earlier age.

Mexican American women have also been shown to have higher prevalence of insulin resistance than White, American women (Kauffman, 2002). There is also ethnic variation in the presenting complaints and quality of life in women with PCOS (Schmid,2004; Hashimoto,2003)

Ethnic differences in the prevalence of PCOS in Black women have not been well explored. Knochenhauer et al (1998) in their study of Black and White women of reproductive age found no significant difference in the prevalence of PCOS between both the race groups. This study, however, did not include the metabolic profile of these women. The severity of PCOS symptoms, demographics and hormonal

profiles of local South African women with PCOS has as yet not been described. Studies done locally have predominantly focussed on alternate treatment methods. In this study we aim to ascertain the phenotypic and biochemical profile of local women with PCOS. This study will outline the profile of local Black women and local Indian women with PCOS, and in addition assess the prevalence of hyperinsulinaemia and hyperlipidaemia amongst these women.

2.Background

Polycystic Ovarian Syndrome (PCOS) is an endocrine disorder that affects an estimated 5% of all women in the reproductive age group (Solomon,1999). Initially described in the early 1900's (Hart, 2004), the disorder has evolved into a multifaceted disease pathology that has proved over the years to be a diagnostic conundrum, due to its variety of symptoms. The Rotterdam Criteria as suggested at the ASRM/ESHRE consensus meeting, has thus far refined the diagnosis of PCOS (Rotterdam ESHRE/ASRM- Sponsored PCOS consensus workshop, 2003).

Women with PCOS may present with several symptoms such as: features of hyperandrogenism (alopecia, acne, hirsuitism), menstrual disturbances (oligo/amenorrhea or dysfunctional uterine bleeding), infertility and obesity (Michelmore et al, 1999). The serum biochemistry, in these women, may display abnormalities such as hyperandrogenaemia, an imbalance in the normal relationship of the gonadotrophic hormones (follicule stimulating hormone and lutenising hormone), hyperinsulinaemia, as well as a hyperlipidaemia (Michelmore et al, 1999).

PCOS has serious metabolic implications. Women with PCOS have a predisposition to non insulin dependant diabetes mellitus due to insulin resistance and resultant impaired glucose homeostasis (Aganovic, 1996). These women are at greater risk for developing cardiovascular complications and dyslipidaemias (Christian et al, 2003) and also have a greater predisposition to develop the Metabolic Syndrome (Apridonidze et al, 2004).

The variety of clinical presentations in PCOS, lends itself to several treatment modalities for these women. These include the oral contraceptive pill, insulin sensitizers, anti-androgens as well as non-pharmacological methods such as bariatric surgery, diets and lifestyle modifications (Escobar-Morreale , 2008). In women with PCOS, a comparison of lifestyle modification, to medical options such as clomiphene citrate and metformin has shown that lifestyle modifications are more effective in improving metabolic parameters, with a 20% success compared to 12 and 14 % for clomiphene citrate and metformin respectively (Karimzadeh, 2010).

2. 1 HISTORY OF PCOS

PCOS was first described by Doctors' Irving Stein and Michael Leventhal in the early 1930s. They reported their findings in 1935 in the American Journal of Obstetrics and Gynecology (Balen, 2003). They had described seven women that presented to them with amenorrhea and infertility. They determined that these women had enlarged ovaries with cysts. Surgical intervention was carried out in the belief that removal of these cysts would remove the obstructive effect of these cysts on these ovaries. Post operatively these women had resumption of their menses and five achieved conception. On a follow up report, in 1945, obesity and male type hair distribution were added to their growing list of women's symptoms.

It was not until the 1970's that further progress was made with the introduction of gonadrotropin releasing hormones and oral contraceptive pills in the management of PCOS. This was due to Solomon Berson and Rosalyn Yalow's 1967 development of radioimmunoassay (Rebar, 1976). The initial assays were done with insulin and later technology was extrapolated to measure hormonal levels of gonadotropin releasing hormones. The biochemical abnormalities of PCOS were then identified. Oral contraceptives were then utilized to combat these anomalies.

Hyperandrogenism, infertility and methods to overcome these problems were extensively researched in the 1980's. The clinical features of hyperandrogenism were expanded to include acne, skin tags and male pattern balding. Pharmacotherapy directed at these features included the use of prednisone, ketoconazole, cyproterone acetate, spironolactone, flutamide, and finasteride. Infertility treatments aimed at ovulation induction with the use of clomiphene citrate

as first line therapy. Clomiphene citrate is thought to inhibit the negative feedback mechanism and results in increased follicle stimulating hormone. Surgical intervention by way of ovarian surgery is recommended if ovulation induction is unsuccessful. The most common technique presently used is ovarian drilling. Ovarian surgery should be used in conjunction with clomiphene citrate if ovulation does not occur within 12 weeks of ovarian drilling (Bayram, 2004). Other techniques employed are the assisted reproduction techniques such as in vitro fertilization. The success rate of in vitro fertilization, in women with PCOS, ranges from 5% – 24% (Lintsen et al, 2010).

Pregnancy in women with PCOS is associated with an increased incidence of gestational diabetes, gestational hypertension and pre eclampsia (Homburg, 2006). This can be attributed to underlying insulin resistance and obesity. It is therefore recommended that these patients be counseled and achieve a BMI less than 30 prior to commencing fertility treatments (Balen et al, 2006).

The role of insulin resistance and the metabolic milieu of PCOS were explored during the 1990's and the first decade of the 2000's. Metabolic concerns in women with PCOS are confounded by obesity which was recognised as one of the initial signs of PCOS. This remains one of the major setbacks in the treatment of women with PCOS.

2.2 Aetiology

The aetiology of PCOS is largely unknown. Recent advances suggest that PCOS may have a genetic component (Franks et al, 1997). In addition, it is also postulated that exposure to androgens during intrauterine fetal development contributes to the phenotypic expression of PCOS (Xita et al, 2002).

Androgen exposure, in utero, has been documented to cause a diminished effect of the GnRH negative feedback mechanism, with resultant neural reprogramming (Sullivan et al, 2004). Experiments done in rats showed that the concentration of androgens during early development was central to the hypothalamus programmed release of gonadotophic hormones (Barraclough et al, 1961). These female rats later went on to develop anovulatory sterility and polycystic ovaries. Additional studies done in sheep, showed that androgen exposure in utero, resulted in a decreased sensitivity of the gonadotrophin –steroid hormone feedback mechanism (Padmanabhan et al, 1998). This decreased efficiency of the feedback mechanism resulted in there being an increased LH secretion coupled with aberrant ovarian follicular development. The neural impairment of this pathway was also postulated to be as a result of excessive in utero androgen exposure .

Studies done in Rhesus monkeys showed that females exposed, in utero, to androgen levels similar to their male counterparts, went on to develop, in later life, the clinical and biochemical features of PCOS that are exhibited in adult women (Eisner et al, 2002). These female monkeys also displayed menstrual irregularities and ovulatory dysfunction (Abbott et al, 1998). The menstrual and ovulatory

dysfunction was more severe in the female monkeys with increased body mass indices, suggesting a hyperinsulinaemic component.
Metabolic abnormalities described in these monkeys also included aberrant secretion and action of insulin – glucose homeostasis and an abnormal distribution of adipose tissue. This included an increase in visceral fat. These are similar characteristics experienced by adult women with PCOS.

Genetic association studies have identified suggested maternal androgens as a source of increased androgens that the fetus is exposed to. The fetus is normally protected from maternal androgens by placental aromatases and high amounts of sex hormone binding globulin. In pregnant women with PCOS the efficacy of this protective system is reduced by other factors such as hyperinsulinaemia (Sir-Petermann et al, 2002). Maternal hyperinsulinaemia also can lead to excessive placental HCG production resulting in fetal ovarian hyperplasia and resultant hyperandrogenism (Barbieri et al,1986).

Ovarian hyperandrogenism is characteristic of PCOS (Legro et al, 1998). The enzymatic abnormalities are an increased functional activity of cytochrome P450, 3-beta-hydroxysteroid dehydrogenase and intracellular protein kinases. These are essential for normal ovarian theca cell steroidogenesis. Several genes have been implicated in the pathogenesis of PCOS. Genes involved in serine phosphorylation that may have a contributory role in the development of PCOS and its sequelae are the genes regulating the cytochrome P450 C17 alpha enzyme, aldehyde dehydrogenase-6 and the CYP11 alpha and CYP17 genes, involved in theca cell

steroidogenesis. To date, however, no conclusive evidence has thus far demonstrated a direct genetic cause for PCOS (Escobar-Morreale, 2005).

Another postulated mechanism for the development of PCOS in hyperandrogenised females, is the activin-follistatin-inhibin pathway. Follistatin is a binding protein of activin. It neutralises the biological activity of activin. Activin and follistatin can be found in the pancreas, adrenal cortex and ovary. In the ovary, activin enhances the development of ovarian follicles, inhibits the production of ovarian androgens by the theca cells, increases pituitary FSH release and beta cell insulin secretion (Mather et al, 1997). Follistatin has the opposite effect. The inhibins are comprised of an alpha and beta subunit. They play a role in the in FSH suppression during the late follicular phase and luteal phase of the menstrual cycle. Anovulatory PCOS women have decreased levels of inhibin B and lack the pulsatile secretory pattern. Serum concentrations of inihibin A are also decreased in anovulatory women with PCOS (Segal, 2010). Animal studies have also shown that hyperandrogenised females expressed increased levels of follistatin and decreased levels of activin beta B mRNA (Norman et al, 2001). This is however thought to have a modest contribution to the later development of PCOS.

2.3 Pathogenesis

The pathogenesis of PCOS is multifactoral. It is a combination of neuroendocrine, ovarian and metabolic mechanisms that lead to an overall propagation of its varied presentations and sequelae.

Neuroendocrine mechanism

Hypothalamic - pituitary - ovarian dysfunction is a key factor in the pathophysiology of PCOS. There is an increased LH secretion from the pituitary. This is a heightened response to the GnRH negative feedback mechanism, in response to the concentration of the circulating steroid hormones. An increased circulating LH delivered to the ovaries, results in an increased stimulation of the theca cells, leading to an increased androgen production (Burger et al, 1985).

Ovarian Dysfunction

For normal ovarian steroidogenesis, it is essential that there is trophic regulation of steroid hormone secretion. This is dependent on various peptides, such as the Insulin- like Growth Factor (IGF) and Insulin-like growth factor binding protein, and is responsible for cytochrome P450 C17alpha gene expression. Women with PCOS have low serum levels of Insulin-like growth factor binding protein (Homburg , 1992). The resultant deficiency causes impairment in steroid hormone secretion with eventual excess of ovarian androgen production (Franks et al,1999).

Intraovarian androgen production is a normal physiological phenomenon that is essential for ovarian follicular development. An excess of androgens, leads to poor

follicular development and eventually follicular atresia. This results in anovulation, oligo/amenorrhea, dysfunctional uterine bleeding and infertility.

Insulin resistance

Some women with PCOS have hyperinsulinaemia. Insulin levels seemed to correlate with androgen levels, suggesting the probable involvement of insulin in the pathogenesis of PCOS (Burghen et al, 1980).

Peripheral insulin resistance increases pancreatic beta-cell secretion of insulin. In normal circumstances this relationship is constant to maintain euglycaemia. In women with PCOS, there is an abnormal relationship of this homeostasis, compared to weight matched controls (Dunaif et al, 1996). Evidence of pancreatic beta – cell dysfunction has been demonstrated by studies using oscillatory glucose infusions and analysing insulin responses in women with PCOS (Erhmann et al, 1995).

The mechanisms for insulin contributing to PCOS are several:

Insulin at the level of the pituitary increases serum LH concentration by two mechanisms. The first is an increase in the amount of LH secreted by the pituitary and the second is by increasing the frequency of LH pulses. Peripherally, insulin acts on the liver by increasing glucose uptake and storage. High levels of insulin suppress hepatic secretion of Insulin like Growth Factor Binding Protein (IGFBP) with resultant hyperinsulinaemia. Insulin also decreases the concentration of sex hormone binding globulin (SHBG) by inducing hepatic suppression of its production. This contributes to the increased free pool of circulating androgens.

A normal relationship of insulin to IGF is essential for normal steroidogenesis in the ovary. In PCOS, there is an imbalance of this relationship. This leads to inhibition of normal ovarian steroidogenesis. In addition, the excessive circulating LH then acts on the ovarian theca cells and causes them to undergo follicular atresia. This results in anovulation and an increase in intraovarian androgen and later hyperandrogenaemia.

The prevalence of abnormal glucose tolerance tests in these women is 45% (Legro et al, 2005). This insulin resistance and resultant abnormal glucose tolerance, including Type II Diabetes Mellitus, associated with PCOS, is independent of body weight (Barcellos et al, 2007).

Obesity and Hyperlipidaemia

Approximately fifty per cent of women with PCOS are obese (Gambineri, 2002). This obesity is predominantly central. Central obesity is due to an increase in visceral fat.

The adipose tissue in central obesity is more metabolically active and contributes toward hyperglycaemia, via the release of free fatty acids from adipocytes. This in turn leads to a decrease in glucose uptake by skeletal muscle and the liver, resulting in increased circulating glucose. Hyperinsulinaemia in obesity also occurs as a result of impaired hepatic insulin sensitivity and an increase in hepatic glucose production (Boden et al, 1997).

Women with PCOS are prone to hyperlipidaemia. The predominant dyslipidaemia is a higher serum triglyceride level, lower HDL level, a higher LDL/HDL ratio and an

increase in LDL levels. PCOS women therefore have a prolonged exposure to a higher level of circulating cholesterol than normal women and have an increased prevalence of atherosclerosis (Talbott et al, 2006; Christian et al, 2003). In addition, compared with their age related counterparts, women with PCOS have an increased risk of experiencing a myocardial infarction (Dahlgren et al, 1992).

2.4 DIAGNOSIS

The syndrome has a wide array of phenotypic variables, which, for some time has led to controversy regarding its diagnosis. Symptoms include oligo/amenorrhea, infertility, hirsuitism, acne and obesity.

The European Society for Human Reproduction and Embryology and the American Society for Reproductive Medicine (ESHRE / ASRM) proposed the following criteria for the diagnosis of PCOS (Rotterdam Criteria). These criteria includes two out of three of the following :

1) oligo/anovulation

2) clinical or biochemical markers of hyperandrogenism

3) ultrasonographic evidence of polycystic ovaries.

In addition the criteria excludes any medical disorder that causes oligo/amenorrhea and hyperandrogenism

Medical disorders that can present with similar symptoms are Congenital Adrenal Hyperplasia (CAH) as well as Cushing's syndrome. CAH can be excluded by biochemically determining the 17α OHP levels. Cushing's syndrome, however, requires a careful history taking, examination and biochemical testing. The features that suggest Cushing's syndrome are facial plethora, rounded facies, violaceous striae, thin skin, bruising and proximal muscle weakness. In addition, in younger patients they can present with growth arrest and primary amenorrhea. A dexamethasone suppression test can be performed for diagnosis.

2.4.1 Oligo/Anovulation

This is suggestive by a history of oligo/ amenorrhea, and confirmed by the absence of the appropriate biochemical hormonal response to ovulation. A raised LH to FSH ratio of greater than 1 is also suggestive of PCOS (Hsu et al, 2009).

2.4.2 Hyperandrogenism

Clinical markers of hyperandrogenism include the presence of acne and hirsuitism. Hirsuitism is classified using the Modified Ferrimen Gallwey Score. A score greater than 8 is suggestive of hyperandrogenism, however this is subject to ethnicity.

Biochemical markers are that of raised androgens. This includes raised serum testosterone, Dehydroepiandrosterone-sulfate (DHEAS), 17 ά Hydroxyprogesterone (17 ά OHP) and Androstenedione.

2.4.3 Ultrasound Features

These features are:

i) an increased ovarian volume (greater than 10 cm^3) – on transvaginal scan

ii) 8- 12 peripheral cysts follicles of between 2 -10mm size

iii) increased echogenicity of ovarian stroma

The above features should be demonstrated on an ultrasound scan done during ovarian inactivity, preferably during day 2-5 of the menstrual cycle. Unilateral features demonstrative of PCOS on ultrasound is acceptable for diagnosis.

2.5 **PCOS and the Metabolic Syndrome**

The National Cholesterol Education Program Adult Treatment Panel (NCEP ATP III) guidelines define the Metabolic Syndrome as the presence of three or more of the following criteria:

a) waist circumference in females ≥ 88cm
b) fasting serum glucose ≥110mg/dl (6.11mmol/l)
c) fasting serum triglycerides ≥ 150mg/dl (1.7mmol/l)
d) serum HDL cholesterol ≤ 50mg/dl (1.3mmol/l)
e) blood pressure ≥ 130/85 mmHg

Approximately 43% of women with PCOS have the Metabolic Syndrome (Apridonidze et al, 2004). This is in comparison with 6 and 15% in the 20-29 and 30-39 year age group, in women without PCOS, respectively (NHANES III). The Metabolic Syndrome as well as PCOS share a common thread of central obesity, hyperinsulinaemia and hyperlipidaemia. This triad of metabolic derangements play a pivotal role in the evolution of serious cardiovascular morbidity (Goode et al, 1998).

The most frequent abnormalities detected in women who have both the Metabolic Syndrome and PCOS are:

- low HDL in 68% of women
- raised body mass index in 67% of women
- raised blood pressure in 45 % of women
- hypertriglyceridaemia in 35% of women
- increased fasting plasma glucose in 4% of women (Wild et al,1985)

These abnormalities are also risk factors for cardiovascular disease and Diabetes Mellitus type 2 and are more prevalent in women with PCOS than a weight matched control (Legro et al, 2002). The exact mechanism of cardiovascular disease is not as yet clearly defined, however insulin resistance seems to be the main contributory factor. Studies done, using imaging and echocardiography, suggest that there might be both a structural and functional abnormality of the cardiovascular system in women with PCOS. These abnormalities include endothelial dysfunction and arterial stiffness (Cussons et al, 2006).

The metabolic dysfunction results in an increased cardiovascular morbidity in women with PCOS. Dahlgren et al (1992) studied the impact of cardiovascular disease on women with PCOS. The independent risk factors analysed were age, hypertension, diabetes mellitus, central obesity measured as increased waist to hip circumference ratio and serum triglyceride concentration. Women with PCOS were found to have an increased risk of a myocardial infarction, compared to age matched counterparts.

2.6 MANAGEMENT

Treatment of women with PCOS is individualised and based on women's symptoms and needs.

LIFESTYLE

Clinically, obese women with PCOS have an increased BMI and waist to hip ratio. An increased waist to hip ratio is a greater predictor of cardiovascular disease than BMI (Noble R, 2001). Lifestyle modification and weight loss is the cornerstone of therapy for these obese women with PCOS. Dietary modification and strict calorie restricted diet is essential to aid exercise to achieve weight reduction. Weight reduction, per se, improves all effects of PCOS. In addition activities such as cigarette smoking should be curtailed in an effort to reduce the cardiovascular effects of PCOS.

The use of statin therapy for women with PCOS has been experimental. To date there has been no evidence that the use of statins improve menstrual regularity, insulin resistance, hirsuitism or acne. Statins, however, have been shown to decrease lipid and testosterone levels (Raval et al, 2011).

Surgical intervention such as bariatric surgery is, of late, a treatment modality that is available for the morbidly obese (Escobar- Morreale, 2005). Further research into weight loss techniques is still ongoing.

REGULATION OF MENSES

Oilgo/ amenorrhea as well as dysfunctional uterine bleeding are amongst the key features of PCOS, and have a negative psycho-social implication for these women.

Non-pharmacological methods, incorporating lifestyle modifications and weight loss may benefit ovarian function and menstruation (Kiddy et al, 1992).

The combined oral contraceptive has been effective in regulating menses. However, in women with PCOS and the Metabolic Syndrome, judicious use of the oral contraceptive pill, is recommended due to the increased cardiovascular risk for both conditions.

Regular menstruation, as a result of improved cyclical ovarian function, has followed surgical interventions such as wedge resection and ovarian drilling.

INSULIN RESISTANCE

Insulin resistance is involved in the pathogenesis of PCOS. An essential strategy to reduce insulin resistance is weight loss and exercise.
Pharmacotherapy is used as an adjunct to these lifestyle modifications.

The use of insulin-sensitizing drugs such as metformin and thiazolidinediones have been suggested as one of pharmacological treatment strategies for women with PCOS. These drugs are used as they exhibit both a metabolic function and an endocrine/ovarian function (Katsiki et al, 2009).

Metformin is a biguanide and it reduces insulin resistance (Pirwany et al,1999). Insulin sensitising agents also increase circulating levels of SHBG thus lowering levels of androgens and this has been shown to improve follicular maturation, ovulation and eventual menstruation in PCOS women (Costello et al, 2003). The reduction in androgens also improves hirsuitism and acne.

HIRSUITISM and ACNE

Hirsuitism and acne occur as a result of hyperandrogenism. There are 2 modalities of treatment, the first being topical or mechanical methods and the latter addressing the hyperandrogenaemia.

Mechanical methods of managing hirsuitism involve shaving, application of wax, depilatory creams, electrolysis and laser photothermolysis. Electrolysis and laser photothermolysis are the most effective but these methods are not permanent (Clayton et al, 2005). Topical preparations for acne include retinoids, antibiotics and antibacterial creams.

Oral contraceptives have been used to increase circulating levels of SHBG, thus decreasing the levels of free androgens, however this mechanism has only shown to be successful in a limited number of women (Wiegratz et al, 2003). Metformin targeting insulin resistance, has demonstrated an even lower ability to target hirsuitism and acne in women with PCOS (Harborne et al, 2003). Spironolactone, an aldosterone antagonist, and a competitive antagonist for the testosterone receptor, reduces hirsuitism by up to fifty per cent (Crosby et al, 1991) when used alone, and up to seventy five per cent when used in combination with an oral contraceptive

(Erenus et al ,1996). Another anti - androgenic agent that has been used is cyproterone acetate. It is usually used in combination with an oral contraceptive pill.

INFERTILITY

Lifestyle modification and weight loss alone positively influence fertility (Moran et al, 2003).

The addition of pharmacological agents acts as an adjunct to lifestyle modification and weight loss. Ovulation induction with clomiphene citrate either alone or in conjunction with metformin has been used. Dosages used in PCOS have to be adjusted to body mass index for a greater efficacy (Shepard et al,1979).

The ideal therapy for infertility treatment of women with PCOS has to date been controversial. Various therapies have been considered viz. metformin, clomiphene citrate and a combination therapy of metformin and clomiphene citrate.

Metformin has not been found to increase fertility rates when used as an adjunct to assisted reproductive techniques (Tso et al, 2009). Metformin however, improved pregnancy outcomes of women with PCOS (Glueck et al, 2001). Clomiphene Citrate has been found to be more effective in non-obese women with PCOS, and may be recommended as the first line agent (Legro et al, 2001; Zain et al 2009). Meta-analysis of these therapies, however, has not been able to demonstrate superiority of any of these therapies over the other (Palomba et al 2009).

Ovarian drilling, either alone or in conjunction with an ovulation inducer has been shown to enhance fertility rates (David et al, 2008).

None of the different fertility treatment options have not demonstrated an increased rate of multiple pregnancy in women with PCOS (Ott et al, 2010).

3. Methods

Subjects

Case files of women attending the Gynae-Endocrinology and Gynae – Infertility outpatient clinics were retrieved from the IALCH medicom database.

Diagnosis of PCOS was made according to the Rotterdam Criteria as proposed by The European Society for Human Reproduction and Embryology and the American Society for Reproductive Medicine (ESHRE / ASRM). This diagnosis was made on two out of three criteria :

- oligo/anovulation
- Clinical or biochemical markers of hyperandrogenism
- ultrasonographic evidence of polycystic ovaries

In addition the diagnosis was made after the exclusion of other medical disorders that cause oligo/amenorrhea and hyperandrogenism

An audit of all women attending the Gynaecology Endocrine and Infertility clinics over the period June 2005 to June 2009 was carried out

Demographic data collected included age, race group and presenting complaint/s. Clinical data included signs of hirsuitism, acne, insulin resistance, blood pressure, height, weight, and waist and hip measurements. Body mass index and waist hip ratio were then calculated.

All women were subjected to serum biochemistry that included FSH, LH, Estradiol, Prolactin, TSH, T4, DHEAS, fasting Insulin, Androstenedione, 17 OHP, Testosterone.

All women underwent a screening glucose tolerance test. This included the ingestion of 75g of glucose and a serum glucose being measured 2 hours later. A value of greater than 6.0 mm/l was considered to be abnormal. Any woman with abnormal screening glucose tests were then subjected to a full oral glucose tolerance test.

The lipid profiles of these subjects were tested after a fast. This profile included the measurement of HDL, LDL, total cholesterol as well as triglyceride levels.

Transvaginal ultrasounds were performed on all women. All ultrasound examinations were performed by a qualified ultrasonographer, employed by the hospital. The diagnosis of polycystic ovaries on ultrasound was made in accordance with the American Society for Reproductive Medicine (ESHRE / ASRM) criteria for the ultrasonographic features of PCOS. These include:

i) an increased ovarian volume (greater than 10 cm^3) – transvaginal scan

ii) 8- 12 peripheral follicles/cysts

iii) increased echogenicity of ovarian stroma

Laboratory analysis

The hormonal profiles were analysed using the Advia Centaur. Trophic hormones such as TSH, FSH, LH, Prolactin, as well as Testosterone and Insulin were analysed using chemiluminescent immunoassay. DHEAS and Androstenedione were analysed via radio immunoassay – manual method. Glucose was analysed using the enzymatic method using hexokinase. Total Cholesterol and Trigylceride levels were analysed using end point enzymatic assay. HDL was analysed using 2 point kinetic enzymatic assay. The intra and interassay coefficients of variances for prolactin was 9.9% and 12% ; for FSH 15.3% and 9.2% ; for LH 28.5% and 11% ; for TSH 11.2% and 9.5% ; for free thyroxine 11.5% and 7.3% ; for testosterone 13.6% and 14.4%; for DHEAS 9.8% and 8.0%; for 17 α OHP 6.7% and 7.1%; for androstenedione 11.1 and 9.8%; for estradiol 12.1% and 8.2% respectively. The intra and interassay coefficients of variances for the metabolic profile of these patients were as follows: for insulin 10.2% and 7.9%; for glucose 6.9% and 3.4%; for triglycerides 5.6% and 2.9%; for cholesterol 5.9% and 1.2% and for HDL 8.4% and 9.9% respectively.

Statistical analysis was done with the aid of a statistician. The software package used was the SPSS system. Demographic and historic dichotomous variables were compared by x^2 analysis using the Pearson test as well as Fisher's exact test. Continuous variables were analysed using the Student's *t* test.

4. RESULTS

The case files of 480 women from the period June 2005 to June 2009 were analysed. Of these 480 files, 110 had the diagnosis of PCOS according to the Rotterdam Criteria (Rotterdam ESHRE/ASRM- Sponsored PCOS consensus workshop, 2003) and were included in this study. There were 87 Indian women, 16 Black women, 5 Coloured women and 2 White women with PCOS. The Coloured and White women were excluded from further analysis due to the small number of women. The Indian and Black women were analysed separately and compared to each other. A p-value of <0.05 was accepted as significant.

Demographic data is displayed in Table 1. The mean age of PCOS women attending IALCH was 27 years and no statistical differences between the ages of the Indian and Black population was found. Eighty nine per cent of women in this study had an increased body mass index (BMI >25). The majority of these women (69.7%) were classified obese (BMI >30), and 19.3 % were overweight (BMI 26-30). There was no significant difference in the body mass indices between the Indian and Black groups.

TABLE 1. Clinical profile (Mean and SD) of Indian and Black women with PCOS SD :

	INDIAN (n=67)		**BLACK** (n=16)		**p-value**
AGE (years)	27.80	(4.78)	27.00	(6.69)	0.54
HEIGHT (m)	1.58	(0.06)	1.59	(0.06)	0.47
WEIGHT (kg)	81.44	(19.24)	77.78	(19.41)	0.49
BMI (kg/m^2)	32.10	(7.51)	30.50	(6.46)	0.44

Standard deviation

Table 2 displays the frequency of the presenting complaints of women with PCOS. Women with PCOS presented with oligo/amenorrhea, dysfunctional uterine bleeding, acne, hirsuitism and infertility. Infertlity was defined as a failure to conceive after a year of trying, in the absence of contraception.

The most frequent presenting complaint, menstrual irregularities, was found in 55 women (50%). Menstrual irregularities were subdivided into oligo/amenorrhea and dysfunctional uterine bleeding. Infertility (44.5%) was the second commonest reason to warrant medical assistance. Hyperandrogenic symptoms were amongst the main complaints in 8.2% of these women.

Table 2. Presenting complaints of all* women with PCOS (*n=110)*

PRESENTING COMPLAINTS	***n***	***%***
OLIGO/AMENORRHEA	35	32
DYSFUNCTIONAL UTERINE BLEEDING	17	15.4
ACNE/HIRSUITISM	6	5.5
INFERTILITY	49	44.5
OLIGO/AMENORRHEA + ACNE/HIRSUITISM	3	2.8

All women = Black, Indian, Coloured and White

Table 3 shows the presenting complaints in Indian and Black women with PCOS. There were no statistical differences between Indian and Black women regarding oligo/amenorrhea, dysfunctional uterine bleeding, acne/ hirsuitism as well as infertility.

Table 3. Presenting complaints in 103 Indian and Black women with PCOS

	INDIAN *n=87*	**%**	**BLACK** *n=16*	**%**	**p-value**
OLIGO/AMENORRHEA	29	33.70%	4	25.00%	0.58
DYSFUNCTIONAL UTERINE BLEEDING	14	16.30%	1	6.30%	0.46
ACNE/HIRSUITISM	4	4.70%	1	6.30%	0.58
INFERTILITY	35	40.00%	10	62.50%	0.11

Table 4 shows the biochemical parameters of Indian and Black women with PCOS. The biochemical parameters analysed were thyroid profiles, estradiol, levels, androgen profiles as well as prolactin levels. Individual biochemical parameters amongst Indian and Black women with PCOS revealed no statistically significant differences between the groups.

Table 4. Hormone profile {mean and (SD)} of African and Indian women with PCOS

	INDIAN		**BLACK**		**p-value**
FSH (mIU/mL)	4.97	(2.0)	5.32	(2.44)	0.53
LH (mIU/mL**)**	8.94	(5.63)	9.98	(6.15)	0.51
PRL (mIU/ml)	347.1	(392.6)	356.00	(291.77)	0.93
TSH (mIU/L)	6.85	(34.64)	1.87	(0.98)	0.58
T4 (pmol/L)	14.92	(2.87)	15.84	(2.43)	0.39
ESTRADIOL (pmol/L)	276.85	(430.74)	372.20	(268.19)	0.45
TESTOSTERONE (nmol/L)	2.07	(1.25)	2.20	(1.38)	0.74
ANDROSTENEDIONE (pmol/L)	14.2	(7.3)	9.00	(3.81)	0.53
17ά OHP (mg/L)	4.71	(3.30)	5.32	(2.30)	0.61
DHEAS (nmol/L)	6.22	(4.01)	7.92	(6.43)	0.23

Normal values available in Appendix

Table 5 displays the biochemical profiles of women with PCOS. In our study population, we found an increased LH to FSH ratio in 78.2% of women. Assessment of the androgenic milieu included total serum testosterone, DHEAS, androstenedione and 17ά OHP. Hyperandrogenaemia was defined as an elevated serum testosterone and/or androstenedione and /or DHEAS. Thirty four percent of women studied were found to have raised androgen levels. Fasting hyperinsulinaemia (>25 uIU/L) was found in half the population of women studied (50.9%). An increased fasting serum glucose (>6 mmol/L) of 23.9 % and hyperlipidaemia of 11.3% was found in women with PCOS.

Table 5. Biochemical Profile of all women with PCOS *(n=110)*

	n	**%**
INCREASED LH:FSH	85	78.2
HYPERANDROGENAEMIA	37	34.3
FASTING HYPERINSULINAEMIA	55	50.9
FASTING HYPERGLYCAEMIA	26	23.9
FASTING HYPERLIPIDAEMIA	12	11.3

A comparative analysis of these parameters between Indian and Black women showed no statistical differences, as demonstrated in Table 6. There is, however, a trend toward there being more fasting hyperglycaemia amongst Indian women as compared to Black women with PCOS.

TABLE 6. Comparative Biochemical profiles of Indian and Black women with PCOS

	INDIAN *n=87*		**BLACK** *n=16*		**p-value**
RAISED LH:FSH	66	(75.9%)	13	(81.3%)	0.46
RAISED ANDROGENS	26	(30.2%)	6	(37.5%)	0.57
RAISED INSULIN	48	(55.2%)	5	(31.3%)	0.08
RAISED GLUCOSE	25	(29.1%)	1	(6.3%)	0.05
RAISED CHOLESTEROL	12	(14.3%)	0	(0%)	0.12

The prevalence of lipid abnormalities is shown in Table 7. Thirty women had lipid abnormalities. Raised triglycerides and low levels of HDL were found to be more frequent manifestations in our study population.

Table 7. Prevalence of Lipid abnormalities in all women with PCOS (n=110)

	n	***%***
RAISED TOTAL CHOLESTEROL (6.22mmol/L)	12	11
RAISED TRIGLYCERIDES (>2.26mmol/L)	23	21
RAISED TRIGLYCERIDES + CHOLESTEROL	2	1.8
LOW HDL (<1.6mmol/L)	10	9
RAISED LDL (>4.1mmol/L)	8	7

Comparative analysis of the lipid profiles between the Indian and Black women showed no statistical difference (Table 8).

Table 8. Comparative Lipid profile (Mean and SD) of all women with PCOS

	INDIAN		**BLACK**		**p-value**
TOTAL CHOLESTEROL	4.96	(1.07)	4.69	(0.75)	0.41
TRIGLYCERIDES	1.76	(1.05)	1.56	(0.74)	0.52
HDL	1.32	(0.23)	1.30	(0.26)	0.91
LDL	2.84	(0.92)	3.58	(3.44)	0.12

All values expressed in mmol/L

Table 9 shows women with an abnormal screening glucose tolerance test. In the Indian population, 13 (14.9%) were found to have Diabetes Mellitus, and 9 (10.3%) had an impaired glucose tolerance test. In the Black population only 1 patient had impaired glucose tolerance. There were no Black women with Diabetes Mellitus.

Table 9. Abnormal serum glucose in Indian and Black women with PCOS

	INDIAN *(n=87)*		**BLACK** *(n=16)*		**p-value**
IMPAIRED GLUCOSE TOLERANCE	9	(10.3%)	1	(6.3%)	0.44
DIABETES MELLITUS	13	(14.9%)		0	0.21

5. DISCUSSION

The objectives of this study were three-fold. The first objective was to establish the profile regarding the clinical presentation and biochemical parameters of women with PCOS attending IALCH. The second objective was to assess the frequency of hyperlipidaemia and insulin resistance in these women, and thirdly to compare these profiles in Indian and Black women with PCOS.

The most frequent presenting complaint was menstrual irregularity in 49.5% of women. Forty four point five percent of women presented with infertility and 8.2% presented with hyperandrogenism. The clinical profiles of our women did not differ from previous studies (Shi et al, 2007). The majority of our women were found to be obese (69.7%). This is higher than that found in other PCOS populations where the prevalence of obesity is between 30 –35% (Li et al, 2005; Li et al, 2007). Features of hyperandrogenism were present in 8.2% of women. The prevalence on insulin resistance in our study population was 50.9%. Twelve per cent of these women also had hyperlipidaemia.

The most frequent reasons for women with PCOS seeking medical help were related to menstrual irregularities and infertility (Table 2). The prevalence of these leading main complaints are similar to previous studies (Shi et al, 2007; Vutyavanich et al, 2007). It is interesting to note that only 5-6% of women presented with the primary complaint of hyperandrogenism as compared to other studies (Ozdemir, 2010; Li et al, 2007). The possible reason for this might be the social acceptance of excessive hair growth in the population studied or the widespread use of depilatory techniques.

None of the women with PCOS presented with symptoms of Diabetes Mellitus or symptoms of cardiovascular disease being their main complaints.
Studies reported by Erhmann et al (1999) and Legro et al (2005) showed the prevalence of glucose abnormalities in women with PCOS being 45 % and 38.6% respectively. The prevalence of type 2 Diabetes Mellitus in these studies was ranged from 7.5% to 10% and impaired glucose tolerance from 31.1% to 35%. In our study, the prevalence of Type 2 Diabetes Mellitus was 14.9% and impaired glucose tolerance was found in 10.3 % of the Indian women. Only 1 Black woman in our study had impaired glucose tolerance (Table 9). This is in keeping with previous studies which showed that the Indian population in Durban has a higher prevalence of Diabetes as compared to the Black population, the prevalence rates being 10% and 1.6% in Indians and Blacks respectively (Asmal et al, 1981).

Lipid abnormalities are closely associated with PCOS. The major serum lipid abnormalities described are a reduced HDL, increased triglycerides, elevated LDL and a higher LDL – HDL ratio (Wild et al, 1985). Women with PCOS who have an associated lipid abnormality have a greater predisposition to develop cardiovascular disease and morbidity (Christian et al, 2003). In local Indian women, the prevalence of hyperlipidaemia was 14.3%, in contrast to no Black woman in our study having hyperlipidaemia (Table 6). Despite the majority of the women having normal serum cholesterol, atherogenic abnormalities have been found in one-third of women with PCOS who have a normal lipid pattern (Berneis et al, 2009).

The presence of insulin resistance, glucose abnormalities, hyperlipidaemia and obesity precedes the development of the Metabolic Syndrome. The prevalence of the Metabolic Syndrome in women with PCOS is about 43% (Apridonidze et al,

2004). The most frequent abnormalities detected in women who have both the Metabolic Syndrome and PCOS are low HDL, raised body mass index, raised blood pressure, hypertriglyceridaemia and an increased fasting plasma glucose (Wild et al,1985). In our study population, an increased BMI >25 was found in 89% of women. Eighty eight per cent of Indian women had a BMI>25 compared to 81% in Black women. Low HDL was found in 66.7 % of women, raised triglycerides in 79% and only 23.9% of women had raised fasting glucose. This suggests that the Metabolic Syndrome may be present in a significant number of local Indian women with PCOS.

Ultrasound findings showed that the majority of the local population studied, had polycystic ovaries. Polycystic ovaries on ultrasound, either bilateral or unilateral were found in 83% of Indian women and 93.8% of Black women.
This has clinical significance as the absence of this radiological feature should not exclude the diagnosis of PCOS. Conversely, the presence of polycystic ovaries on ultrasound should not be used in isolation to make the diagnosis of PCOS. The prevalence of isolated polycystic ovaries on ultrasound can occur in 25% of normal women of reproductive age (Franks et al, 2004).

In terms of public health relevance, our findings suggest that South African women with PCOS, have similar presentations and profiles as women with PCOS globally. However, in Black women, the most common presenting complaint was infertility and not menstrual irregularities and hyperandrogenism (Table 3). In local Indian women, the presence of type 2 Diabetes Mellitus is greater than international rates (Vrbikova,

2009). This emphasizes the need for establishing the metabolic parameters in every patient with PCOS, and to maintain long term regular follow up of these women.

Our study has found that PCOS is an entity in Black women as well. It can be presumed, that the small number of Black women is a shortcoming of this study. However it must be noted that PCOS in Black women has been seldom described in the literature (Knochenhauer et al, 1998). This study highlights the low number of Black women seen and indirectly suggests a possible lower prevalence of PCOS in the Black population. This study gives insight into the main presenting complaint and biochemical variables in Black women with PCOS. A bigger sample size of Black women with PCOS may reveal other differences in Indian and Black women with PCOS.

6. RECOMMENDATIONS

Contemporary management of women with PCOS consists of varying investigations and treatment modalities. These investigations target women's symptoms to aid the diagnosis of PCOS. This approach and the management of a patient with PCOS in this manner can completely disregard the metabolic disarrangement experienced by these women.

Our results show that metabolic abnormalities in South African women with PCOS do exist irrespective of race. If left untreated, these abnormalities can progress to serious cardiovascular morbidity. It is essential to recognise these women as high risk and screen for metabolic disarrangements. Judicious follow up these for these women is recommended to identify and institute early necessary treatment if required.

There are as yet, no local treatment protocols for the management of women with PCOS. In addition, there are no clear guidelines to screen these women for metabolic disorders. Our study shows the prevalence metabolic sequelae in Black and Indian women with PCOS. We therefore recommend the development of a treatment protocol addressing the metabolic consequences of women with PCOS.

Appendix

Normal values used by the chemical pathology laboratory at IALCH.

FSH			
Follicular phase		2.5 -10.2	mIU/mL
Midcycle peak		3.4 -33.4	mIU/mL
Luteal phase		1.5 -1.9	mIU/mL
LH			
Follicular phase		1.9 -12.5	mIU/mL
Midcycle peak		8.7 -76.3	mIU/mL
Luteal phase		0.5 -16.9	mIU/mL
Prolactin		121 -619	mIU/ml
TSH		0.27 -4.2	mIU/L
T4		11.5 -22.7	mIU/L
Estradiol			
Follicular phase	D2	40 -253	pmol/L
	D10	231-606	pmol /L
Midcycle	D13	536-1930	pmol /L
Luteal phase	D16	121-551	pmol/L
	D20	250-719	pmol/L
	D26	132-448	pmol/L
Testosterone		0.5-2.6	nmol/L
Androstenedione		1-12.2	nmol/L

DHEAS	0.9-11.7	nmol/L
17 OHP	0.88 – 12.1	nmol/L
Fasting insulin	3-25	uIU/L
Fasting glucose	<7.8	mmol/L
Total Cholesterol	<6.22	mmol/L
Triglycerides	<2.26	mmol/L
HDL	>1.6	mmol/L
LDL	>4.1	mmol/L

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