International Journal of Basic and Applied Medical Sciences ISSN: 2277-2103 (Online)
An Open Access, Online International Journal Available at http://www.cibtech.org/jms.htm
2018 Vol. 8 (3) September-December, pp. 42-50/Geethanjali et al.
Research Article

A COMPARATIVE STUDY OF ANTI MULLERIAN HORMONE IN POLYCYSTIC OVARIAN SYNDROME (PCOS) AND NORMALLY OVULATING WOMEN

Geethanjali G[1], [*]Manisha Sharma[1], Harsh Vardhan Singh[2], P.K. Jain[3], Rajeev Ranjan[2], Sanjay Jain[4], Rajni Mittal[1]

[1]Department of Obst & Gynae, [2]Department of Biochemistry, [3]Department of Radiology, [4]Department of Microbiology, Hindu Rao Hospital & NDMC Medical College, NDMC, Delhi

[*]Author for Correspondence: drmanishasharma63@gmail.com

ABSTRACT

Polycystic ovary syndrome (PCOS) is a syndrome of ovarian dysfunction affecting reproductive age group women with the prevalence of 1 in 15 worldwide. It is characterized by anovulation, hyperandrogenism and polycystic ovarian morphology. Affected women are at risk of irregular cycle, psychological and behavioral disorders. The study was conducted to compare Anti mullerian Hormone (AMH) level in PCOS women and normally ovulating women to study its relationship of with clinical parameters, hormonal levels and ultrasonological morphology. It was a hospital observational study done by taking 40 PCOS and 40 normally ovulating women of similar age group. PCOS women were defined on the basis of Rotterdam ESHRE/ ASRM –Sponsored consensus with presence of at least two of the three criteria. All the women were evaluated for serum levels of AMH, LH (Luteinising hormone), FSH (Follicle stimulating hormone), Prolactin, TSH (Thyroid stimulating hormone), estradiol, SHBG (sex hormone binding globulin), DHEA-S (Dihydroepiandrostenedione – sulphate) and Testosterone on day 2 of menstrual cycle. Serum AMH was significantly high in PCOS women (p value 0.000) as compared to normally ovulating women and was positively correlated with ovarian volume (p value 0.000), antral follicle count (p value 0.000) and androgens (testosterone and DHEA-S with p value 0.000, 0.000 respectively). AMH was also positively correlated with LH and a negatively correlated with FSH and SHBG in PCOS women but the correlation was statistically not significant (p value 0. 405, 0.792 and 0.368 respectively). Raised serum AMH level was associated with increased antral follicle count and ovarian volume and it correlated well with clinical and biochemical hyperandrogenism. So it can replace ultrasonound which requires expert sonologists and multiple tests which are done for testing different androgens and thus cost effective.

Keywords: *Polycystic Ovary Syndrome, Anti Mullerian Hormone, Hyperandrogenism, Anovulation*

INTRODUCTION

Polycystic ovary syndrome (PCOS) is a syndrome of ovarian dysfunction, where affected women are at increased risk of psychological and behavioral disorder. It is the most common endocrine disorder with a worldwide prevalence of one in fifteen among women of reproductive age group (Norman *et al.*, 2007). Approximately 75% of anovulatory women of any cause have polycystic ovaries and 20–25% of women with normal ovulation demonstrate ultrasound findings typical of polycystic ovaries. The full blown syndrome of hyperandrogenism, chronic anovulation and polycystic ovaries affect 4–6% of women (Malhotra *et al.*, 2014).

The association of amenorrhea with bilateral polycystic ovaries and obesity was first described in 1935 by Stein and Leventhal. They described the syndrome in presence of amenorrhea, hirsutism and enlarged polycystic ovaries which is now recognized to be characteristic of extreme cases (Speroff and Fritz, 2011). In 1990 National Institutes of Health Conference on PCOS, recommended that diagnostic criteria should include evidence of ovulatory dysfunction with clinical and / or biochemical hyperandrogenism

with exclusion of other etiologies to diagnose polycystic ovary syndrome (Malhotra *et al.,* 2014), but it did not satisfy many because it omitted the ultrasound criteria.

In 2003, the European Society for Human Reproduction and Embryology (ESHRE) and the American Society for Reproductive Medicine (ASRM) - Sponsored PCOS consensus workshop group at Rotterdam, (ESHRE TR, 2004) proposed two of the following three criteria for diagnosis of Polycystic Ovary Syndrome.

1. Anovulation and/or Oligo - ovulation
2. Poly cystic ovarian morphology (≥12 follicle measuring 2-9 mm in diameter and /or volume >10cc in each ovary)
3. Hyperandrogenism (Clinical/Biochemical) with exclusion of other etiologies (congenital adrenal hyperplasia, androgen secreting tumors, Cushing's syndrome, Prolactinemia).

Anti-Mullerian hormone (AMH) also known as mullerian inhibiting substance, a glycoprotein and a member of transforming growth factor β family, is secreted from sertoli cells of testis in males during embryogenesis and causes regression of mullerian ducts and development of mesonephric duct. At the same time it is not secreted in the female embryo, thus allowing development of the female sexual organs from Mullerian ducts which do not regress. Bioactive AMH secreted by granulosa cells is first detected at 36 weeks of gestation in a female fetus and reaches maximum concentration at puberty, then begins to decrease in adulthood and finally disappears completely following menopause (Rajpert *et al.,* 1999), (La Marca *et al.,* 2009).

Follicle development in the ovaries comprises two distinct stages: initial recruitment and cyclical recruitment. The initial recruitment takes place by AMH in which it makes primordial follicles to mature but inhibits transition of follicles from primordial into maturation stages and thereby has an important role in regulating the number of follicles remaining in the primordial pool. It also has an inhibitory effect on follicular sensitivity to follicle stimulating hormone (FSH) and therefore has a role in the process of follicular selection. Cyclic recruitment is controlled by FSH in which there is growth of a cohort of small antral follicles, among which the dominant follicle (destined to ovulate) is subsequently selected (Speroff and Fritz, 2011). AMH is produced maximally from granulosa cells of primary follicles in pre antral stage (app 6 mm) and minimally by greater antral follicles. When follicle growth becomes FSH dependent, AMH expression diminishes and becomes undetectable in the follicle (La Marca *et al.,* 2009).

Although exact pathogenesis of PCOS is obscure, the distinctive feature is failure of follicular maturation, despite initial recruitment which results in anovulation and accumulation of preantral and small antral follicles which contribute significantly to the production of AMH. Thus high level of AMH is found in PCOS patients due to increase in number of antral follicles (La Marca *et al.,* 2009).

Till date it is difficult to diagnose PCOS due to its complex presentation. Clinical diagnosis of Hyperandrogenemia associated with PCOS by Ferriman – Gallway score is subjective and the biochemical diagnosis requires multiple tests for confirmation. Studies have shown that AMH correlated well with hyperandrogenemia and can replace multiple tests required for testing different androgens. Very few studies have been done on Indian population so the present study was undertaken in order to evaluate AMH in PCOS and normally ovulating women in our hospital and to study its correlation with other biochemical and hormonal parameters of PCOS.

MATERIALS AND METHODS

This was a hospital based observational study conducted at Gynae and Obstetrics department at Hindu Rao hospital, Delhi for a period of one year from June 2017 to May 2018. In this study 40 women with PCOS (Polycystic ovarian syndrome) and 40 normally ovulating women with age from 18-35 years were studied. PCOS women were defined on the basis of Rotterdam ESHRE/ ASRM –Sponsored consensus with presence of at least two of the three criteria.

Anovulation and/or Oligo ovulation was defined as presence of secondary amenorrhea or < 8 menstrual periods per year.

Hyperandrogenism (Clinical) was defined as presence of hirsuitism ie terminal (coarse) hairs in a male-like pattern on upper lip, chin, chest, upper back, upper abdomen, lower abdomen, upper arms, forearms, thighs, legs. Ferriman Gallwey scoring was used in the study with grading from 0 to 4 where 0 = no growth of terminal hair 4=extensive hair growth. Hirsuitism was defined with score > 8. Minor signs such as acne or seborrhea or excessive growth of coarse hairs of the lower forearms and lower legs alone were not included.[2]

Poly cystic ovarian morphology – Transvaginal or transabdominal sonography was done in follicular phase (day 3) for detection of the number of small follicles and ovarian volume which was calculated using the formula of ellipsoid (pi/6 multiplied by length, width and height). When at least 1 ovary had 12 or more follicles < 10 mm in diameter and volume >10mm[1], it was diagnosed as polycystic ovary. Presence of peripherally arranged follicles or thickened stroma was not included as a case of PCO. **Normally ovulating women** had regular ovulatory cycle (21-35days) with normal ultrasonic ovulatory morphology.

Women with pregnancy or using oral contraceptives or with ovarian cyst/endometrioma/surgery on ovaries or inadequate visualization of ovary were excluded. Women with H/O hyperprolactinemia, primary thyroid disorder, congenital adrenal hyperplasia, androgen secreting tumor, cushing's syndrome, rapidly progressing hirsutism or those who refused to participate in the study were also excluded.

After a detailed history, general physical examination and systemic examination was done. Anthropometric evaluation was done for all the women and body mass index (BMI) was calculated. USG was done by Samsung machine in close association with radiologist for antral follicle count and ovarian volume.

All the women were evaluated for serum levels of AMH, LH (Luteinising hormone), FSH, Prolactin, TSH, estradiol, SHBG (sex hormone binding globulin), DHEA-S (Dihydroepiandrostenedione – sulphate) and Testosterone. Thyroid function test and prolactin levels done on all patients but patients with thyroid disorder and hyperprolactinemia were excluded. All the hormone tests were done by electro-chemiluminescence immunoassay (ECLIA) technique using Elecsys immunoassay analyzer 2010 from the company Roche Diagnostics. The study was ethically and scientifically approved by the institution.

Data Analysis

Statistical analysis was done by using IBM SPSS version 25 and the obtained data was expressed as mean with standard deviation. The Independent sample Mann-Whitney U Test was used to assess the difference in Ovarian volume, LH, FSH, Prolactin, TSH, Serum estradiol, SHBG, DHEAS, AMH, Testosterone between PCOS women and normally ovulating women. The Spearman's rho was used to find correlation between AMH and other variables. P value of <0.05 was taken as statistically significant.

RESULTS

The study was conducted on 40 PCOS and 40 normally ovulating women. Out of 40 women with PCOS 37(92.5%) presented with oligomenorrhoea but without hyperandogenemia while 7.5% presented with oligomenorrhoea and hyperandogenemia.

Table 1: Distribution of women according to age, BMI and ultrasonological findings

	PCOS women (n=40)	Normally ovulating women (n=40)	P value[1]
Mean age (years)	23.28± 4.8	24.09± 4.4	0.112
BMI (kg/sq cm)	25.6 ± 3.8	23.78 ± 3.4	0.000
Mean follicle number	16.69±3.08	5.37±2.24	0.000
Mean volume (cu cm)	12.35±6.16	3.92±0.2	0.000

[1]*Independent sample Mann – Whitney U Test*

Table 2: Distribution of mean hormonal levels in PCOS and normally ovulating women

Mean of hormonal level	PCOS women (n=40)	Normally ovulating women (n=40)	P value[1]
AMH (ng/ml)	9.43±9.50	2.16±3.26	0.000
LH (mIU/ml)	15.28±26.6	6.77±3.22	0.000
FSH (value)	7.4±3.4	9.66±7.2	0.001
LH/FSH ratio	2.04±2.86	0.74±0.42	0.000
Serum estradiol (pg/ml)	58.88±162.4	66.33±177.32	0.927
Total Testosterone (ng/ml)	0.40033±0.66	0.16±0.24	0.000
SHBG (nmol/ml)	91.42±54.4	112.35±56.34	0.156
DHEA-S (nmol/l)	149.49±85.19	99.31±76.22	0.011
Free Androgen Index (total Te ×100/SHBG)	1.63±12	0.16±0.24	0.000

[1]*Independent sample Mann – Whitney U Test*

Table 1 shows the mean age, mean BMI, mean antral follicle count and mean ovarian volume in PCOS women and normally ovulating women. Mean level of anti Mullerian hormone, luteinizing hormone, total Testosterone, DHEA-S was high in PCOS women as compared to normally ovulating women as shown in table – 2. Mean LH/FSH ratio and free Androgen Index was also high in PCOS women while mean follicular stimulating hormone and sex hormone binding globulin was low in PCOS women.

Table 3: Relationships between the AMH plasma levels with clinical parameters and ultrasonic features in women having PCOS and normally ovulating women

Parameters	Coefficient of correlation in PCOS women	P value[1]	Coefficient of correlation in normally ovulating women	P value[1]
AMH, Age	0.278	0.086	0.025	0.885
AMH, BMI	0.255	0.117	-0.091	0.603
AMH, clinical hyperandrogenemia	0.347	0.031	-	-
AMH , Ovarian volume	0.525	0.001	0.241	0.164
AMH, Follicle number	0.615	0.000	-0.012	0.947

[1]*Spearman's rho*

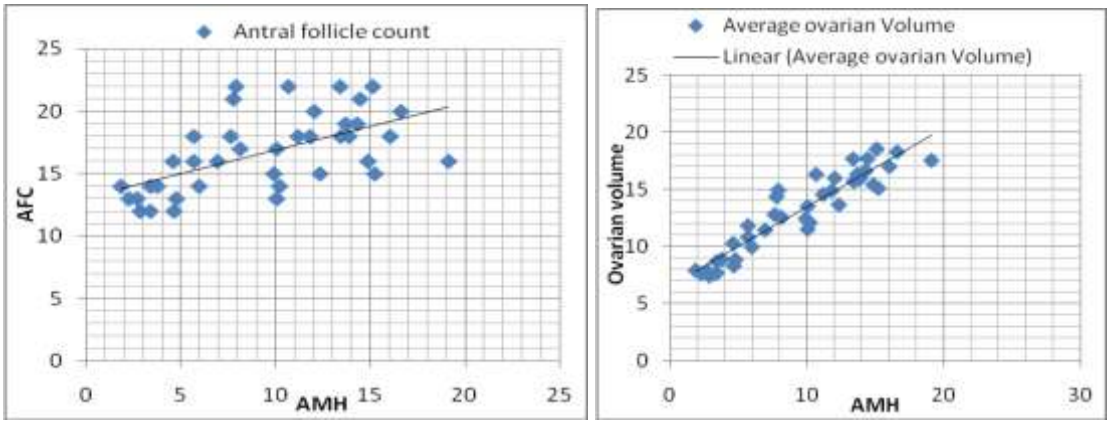

Figure 1: Correlation of serum AMH level with AFC and Ovarian volume in PCOS women

Table 3 shows the correlation of AMH with age, BMI, clinical hyperandrogenemia, ovarian volume and follicle number in PCOS and normal ovulating women. Table 4 shows the correlation of AMH with FSH, LH, androgens (testosterone, DHEA-S), SHBG and free androgen index in PCOS and normal ovulating women.

Table 4: Relationship between AMH plasma levels with various hormones in women having PCOS and normally ovulating women

Parameters	Coefficient of correlation in PCOS women	P value[1]	Coefficient of correlation in normally ovulating women	P value[1]
AMH, FSH	-0.044	0.792	0.232	0.180
AMH, LH	0.137	0.405	0.143	0.413
AMH, Testosterone	0.685	0.000	0.525	0.001
AMH, DHEAS	0.587	0.000	0.309	0.071
AMH, SHBG	-0.148	0.368	0.315	0.066
AMH, FAI	0.558	0.000	0.406	0.015

[1]Spearman's rho

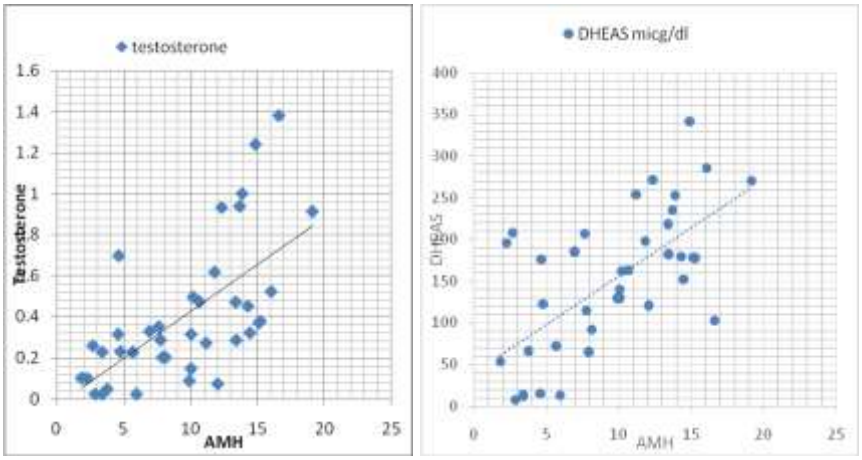

Figure 2: Correlation of AMH with testosterone and DHEA – S in PCOS women

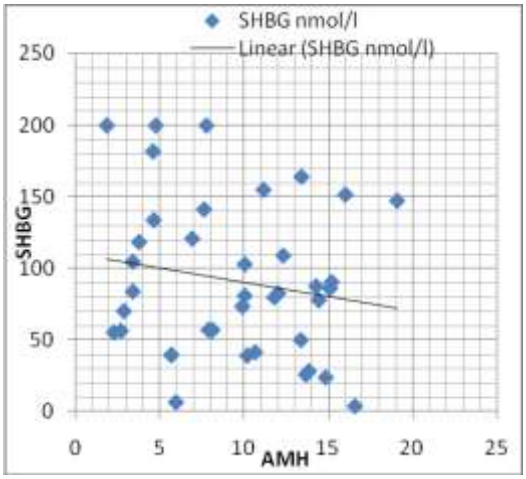

Figure 3: Correlation of AMH with SHBG in PCOS women

DISCUSSION

Polycystic ovarian syndrome is the most frequent endocrine disorder of women in the reproductive age group, diagnosis of which always remains a challenge to the gynecologists and endocrinologists. Various criteria have been proposed for the diagnosis of PCOS for past many years. These included clinical (irregular cycles, infertility, hirsutism, acne, features of hyperandrogenism etc.), hormonal (raised LH levels, LH/FSH ratio, serum testosterone, serum androstenedione, DHEA-S), and ultrasound features.

Rotterdam criteria included the ultrasound features and broadened the diagnostic criteria for PCOS. But it also received criticism as counting the follicle number per ovary (FNPO) may not be sufficiently reliable and with a threshold of 12 for the FNPO only 75% of PCOS patients were diagnosed (Pigny *et al.,* 2006). Also for studying ovarian morphology by ultrasound requires examination by radiologist with appropriate skills and is time consuming. At the same time a large proportion of adolescents will be diagnosed as PCOS who may have polycystic ovaries (10-48%) only and not have the syndrome (Roe and Dokras, 2011).

The reproductive women of similar age group were taken in the study as the mean age was 23.28±4.8 years in PCOS women and 24.08±4.4 years in normally ovulating women with no statistically significant difference (p value 0.112). Mean antral follicle count (16.69±3.08) and mean ovarian volume (12.35±6.16 cu cm) was higher among PCOS women as compared to normally ovulating women (5.37±2.24 follicles and 3.92±0.2 cu cm respectively) in our study and the difference was statistically significant (p value 0.000). Increased antral follicle count and ovarian volume in PCOS women as compared to normal ovulating women was found in various studies (Pigny *et al.,* 2003, Piouka *et al.,* 2009, Begawy *et al.,* 2010 and Elersten *et al.,* 2012 with p value 0.0001, 0.001, 0.001and 0.001 respectively). In PCOS there is intra ovarian hyperandrogenism which promotes early follicular growth with more follicles able to enter the growing cohort instead of a single follicle. This excessive number of growing follicles then becomes arrested at 6 to 9 mm size. Increased stromal volume which is seen in PCOS is the main cause of ovarian enlargement.

Mean serum AMH was 9.43±9.50 ng/ml in PCOS women and was 2.16±3.26 ng/ml in normally ovulating women and the difference was statistically significant (p value 0.000). With 95% CI for serum AMH level was between 6.49 to 12.37 mg/dl in PCOS women. Various authors have also found higher levels of AMH in their studies as shown in table – 5 (La Marca *et al.,* 2004, Begawy *et al.,* 2010, Parco *et al.,* 2011, Woo *et al.,* 2012 and Verma *et al.,* 2016). The increase in AMH level is due to increased synthesis and secretion of AMH by polycystic ovaries in PCOS women. Das *et al.,* 2008 found significantly higher levels of AMH levels in follicular fluid ($P < 0.0001$) in women with anovulatory PCOS compared with normal-ovulatory controls. They also found 60 times higher mean follicular fluid AMH levels than in the serum of PCOS patients and concluded that increased circulating concentrations of AMH are partly due to the increased production of AMH by individual follicles and not simply attributable to the increased number of small antral follicles and suggested an intrinsic abnormality in the ovarian follicles themselves in PCOS.

Table 5: Comparison of AMH level in PCOS women and normally ovulating women with other studies

Studies	AMH level in PCOS women	AMH level in normally ovulating women	P value
La Marca *et al.,* 2004	7.4±1.7 ng/ml	3.5±1.5 ng/ml	0.001
Begawy *et al.,* 2010	9.50 ± 5.11 ng/ml	3.53±1.95 ng/ml	0.001
Parco *et al.,* 2011	10.0±2.28 ng/ml	3.64±1.51 ng/ml	<0.05
Woo *et al.,* 2012	11.58±6.31 ng/ml	5.38±2.99 ng/ml	<0.001
Verma *et al.,* 2016	6.90±5.09	2.15±1.86	0.001
In our study, 2018	9.43±9.50 ng/ml	2.16±3.26 ng/ml	0.000

As shown in table 4, there was significantly higher LH level and lower level of FSH in PCOS women compare to normally ovulating women in our study (p value 0.000). Significantly elevated levels of LH was found by various authors in PCOS women in their studies (Pigny *et al.,* 2003, Piouku *et al.,* 2009, Begawy *et al.,* 2010, Homburg et al 2013 and Wiweko *et al.,* 2014 with p value 0.0001, 0.0001, 0.001, 0.001 and 0.001 respectively). They also found lower amount of FSH in PCOS women in their study. LH/FSH ratio was higher in PCOS women than in normally ovulating women and the difference was statistically significant (p value 0.000). Follicle stimulating hormone (FSH) is not increased, probably because of the synergistic negative feedback of chronically elevated oestrogen levels and normal follicular inhibin (Norman *et al.,* 2007). Begawy *et al.,* 2010 also found increased LH/FSH ratio in their study (p value 0.012). There was significantly raised Androgen (Testosterone, DHEAS, FAI) level in women with PCOS compared to normally ovulating women (p value 0.000, 0.011 and 0.000 respectively). The SHBG level was low in PCOS women compared to normally ovulating women but was statistically not significant (p value 0.156). Pigny *et al.,* 2003, Eldar-Geva *et al.,* 2005, Nardo *et al.,* 2009, Begawy et al 2010 and Woo *et al.,* 2012 also found significant higher level of androgens in PCOS group compared to normal controls in their study (p value 0.0001, 0.0001, <0.001, 0.001, and 0.001).
AMH was positively correlated with clinical hyperandrogenemia in PCOS women (p value 0.031). Hence raised serum AMH may be seen in women presenting with clinical hyperandrogenism
There was statistically significant positive correlation of AMH with antral follicle count and ovarian volume (p value 0.000 and 0.001 respectively) in PCOS women while its correlation was statistically not significant in normally ovulating women (p value 0.947). Pigny *et al.,* 2003, Nardo *et al.,* 2009 and Begawy *et al.,* 2010 also found statistically positive correlation between AMH with antral follicle count in PCOS women while Begawy et al 2010 and Koutlaki *et al.,* 2013 found statistically significant positive correlation between AMH and ovarian volume in PCOS women. Hence in PCOS women if follicle count increases AMH level also increases. The Hyperandrogenism may also contribute to increased number of antral follicle leading to increased AMH secretion (Begawy, 2010).
AMH correlated positively with LH and negatively with FSH in PCOS women but the correlation was not statistically significant (p value 0.405 and 0.792 respectively). Begawy *et al.,* 2010 and Woo *et al.,* 2012 also found positive correlation between AMH and LH and negative correlation between AMH and FSH. Raised LH may be responsible for increasing AMH by causing follicular arrest to some extent in PCOS women. Also multiple small follicles can suppress FSH levels in PCOS women and increase AMH level. There was significant positive correlation of AMH with testosterone, FAI and DHEA-S with p value 0.000, 0.000 and 0.000 respectively in women having PCOS and in normally ovulating women (p value 0.001, 0.015, 0.071 respectively) in our study. Nardo *et al.,* 2009, Begawy *et al.,* 2010 and Woo *et al.,* 2012 also found significant positive correlation of AMH with androgens in their study in PCOS women and support our study. Negative correlation of AMH with FSH may suggest that AMH reduces aromatization activity in PCOS women and contribute to androgens. Thus AMH is a biochemical marker which can replace multiple tests done for testing different androgens for the diagnosis of PCOS.

CONCLUSION

Increased antral follicle count and ovarian volume is commonly associated with raised serum AMH level, so serum AMH can replace ultrasonological examination which requires expert radiologist and time consuming. Hyperandrogenemia and related complications like acne, abnormal hair growth etc are common in PCOS women and as serum AMH was correlating well with clinical and biochemical hyperandrogenism, it can replace multiple tests which are done for testing different androgens and thus cost effective. We conclude that AMH, as a biochemical marker, can increase sensitivity of Rotterdam criteria for diagnosing PCOS, so it should be incorporated in the definition of the Rotterdam criteria for PCOS.

REFERENCES

Begawy AF, El-Mazny AN, Abou-Salem NA and El-Taweel NE (2010). Anti-Müllerian hormone in polycystic ovary syndrome and normo-ovulatory women: Correlation with clinical, hormonal and ultrasonographic parameters. *Middle East Fertility Society Journal* **15**(4) 253-8.

Das M, Gillott DJ, Saridogan E and Djahanbakhch O (2008). Anti-Mullerian hormone is increased in follicular fluid from unstimulated ovaries in women with polycystic ovary syndrome. *Human Reproduction* **23**(9) 2122-6.

Eilertsen TB, Vanky E and Carlsen SM (2012). Anti-Mullerian hormone in the diagnosis of polycystic ovary syndrome: can morphologic description be replaced? *Human reproduction* **27**(8) 2494-502.

Eldar-Geva T, Margalioth EJ, Gal M, Ben-Chetrit A, Algur N, Zylber-Haran E, Brooks B, Huerta M and Spitz IM (2005). Serum anti-Mullerian hormone levels during controlled ovarian hyperstimulation in women with polycystic ovaries with and without hyperandrogenism. *Human Reproduction* **20**(7) 1814-9.

ESHRE TR, ASRM-Sponsored PCOS Consensus Workshop Group (2004). Revised 2003 consensus on diagnostic criteria and long-term health risks related to polycystic ovary syndrome. *Fertility and sterility* **81**(1) 19-25.

Homburg R, Ray A, Bhide P, Gudi A, Shah A, Timms P and Grayson K (2013). The relationship of serum anti-Mullerian hormone with polycystic ovarian morphology and polycystic ovary syndrome: a prospective cohort study. *Human Reproduction* **28**(4) 1077-83.

Koutlaki N, Dimitraki M, Zervoudis S, Poiana C, Psillaki A, Nikas I, Liberis A, Badiu C and Liberis V (2013). The relationship between Anti-Müllerian hormone and other reproductive parameters in normal women and in women with polycystic ovary syndrome. *Journal of medicine and life* **6**(2) 146.

La Marca A, Broekmans FJ, Volpe A, Fauser BC and Macklon NS (2009). Anti-Müllerian hormone (AMH): what do we still need to know? *Human Reproduction* **24**(9) 2264-75.

La Marca A, Orvieto R, Giulini S, Jasonni VM, Volpe A and De Leo V (2004). Müllerian-inhibiting substance in women with polycystic ovary syndrome: relationship with hormonal and metabolic characteristics. *Fertility and Sterility* **82**(4) 970-2.

Malhotra N, Kumar P, Malhotra J, Malhotra BN and Mittal P (2014). Polycystic ovary syndrome. In: *Jeffcoate's Principles of Gynaecology*. 8th edition. New Delhi; (Jaypee Brothers Medical Publishers (P) LTD) 360-368.

Nardo LG, Gelbaya TA, Wilkinson H, Roberts SA, Yates A, Pemberton P and Laing I (2009). Circulating basal anti-Müllerian hormone levels as predictor of ovarian response in women undergoing ovarian stimulation for in vitro fertilization. *Fertility and sterility* **92**(5) 1586-93.

Norman RJ, Dewailly D, Legro RS and Hickey TE (2007). Polycystic ovary syndrome. *The Lancet* **370**(9588) 685-97.

Parco S, Novelli C, Vascotto F and Princi T (2011). Serum anti-Müllerian hormone as a predictive marker of polycystic ovarian syndrome. *International journal of general medicine* **4** 759.

Pigny P, Jonard S, Robert Y and Dewailly D (2006). Serum anti-Mullerian hormone as a surrogate for antral follicle count for definition of the polycystic ovary syndrome. *The Journal of Clinical Endocrinology & Metabolism* **91**(3) 941-5.

Pigny P, Merlen E, Robert Y, Cortet-Rudelli C, Decanter C, Jonard S and Dewailly D (2003). Elevated serum level of anti-mullerian hormone in patients with polycystic ovary syndrome: relationship to the ovarian follicle excess and to the follicular arrest. *The Journal of Clinical Endocrinology & Metabolism* **88**(12) 5957-62.

Piouka A, Farmakiotis D, Katsikis I, Macut D, Gerou S and Panidis D (2009). Anti-Mullerian hormone levels reflect severity of PCOS but are negatively influenced by obesity: relationship with

increased luteinizing hormone levels. *American Journal of Physiology-Endocrinology and Metabolism* **296**(2) E238-43.

Rajpert-De Meyts E, J rgensen , Gr m , M ller J, Cate R and Skakkeb k E (1999). Expression of anti-Mullerian hormone during normal and pathological gonadal development: association with differentiation of Sertoli and granulosa cells. *The Journal of Clinical Endocrinology & Metabolism* **84**(10) 3836-44.

Roe AH and Dokras A (2011). The diagnosis of polycystic ovary syndrome in adolescents. *Review in Obstetrics and Gynecology* **4**(2) 45.

Speroff L and Fritz MA (2011). Polycystic ovary syndrome. In: *Clinical Gynecologic Endocrinology and Infertility*. 8th Edition, Lippincott Williams & Wilkins 143-148 & 500-522.

Verma AK, Rajbhar S, Mishra J, Gupta M, Sharma M, Deshmukh G and Ali W (2016). Anti-mullerian hormone: a marker of ovarian reserve and its association with polycystic ovarian syndrome. *Journal of Clinical and Diagnostic Research*: *JCDR* **10**(12) QC10.

Wiweko B, Maidarti M, Priangga MD, Shafira N, Fernando D, Sumapraja K, Natadisastra M and Hestiantoro A (2014). Anti-mullerian hormone as a diagnostic and prognostic tool for PCOS patients. *Journal of assisted reproduction and genetics* **31**(10) 1311-6.

Woo HY, Kim KH, Rhee EJ, Park H and Lee MK (2012). Differences of the association of anti-Müllerian hormone with clinical or biochemical characteristics between women with and without polycystic ovary syndrome. *Endocrine journal* **59**(9) 781-90.