Antral Follicle Count and AMH Level for Ovarian Reserve Comparison

Authors
Dr Shrikant Adsul[1], Dr Rajendra Rajguru[2], Dr Neeta Adsul[3], Dr Asmita Patil[4]
[1]DNB FCPS DGO MBBS Gyneacology , Assistant Professor At Rajiv Gandhi Medical College Kalwa
[2]FCPS DGO MBBS, Gyneacology, Consultant At Shreenika Hospital Kalwa
[3]MD MEDICINE, IDCCM, Physician, Consultant At Shreenika Hospital Kalwa
[4]DNB DGO Gyneacology, Assistant Professor At Rajiv Gandhi Medical College Kalwa West

Abstract
Background: *The chronological age at which menopause happens is evocative that reproductive aging is flexible amongst females, and shows that age is not a lone factor indicating the females' reproductive potential. Various tests have been used to determine ovarian reserve, they are primarily used in Infertility clinics. Endocrine markers like anti-Mullerian hormone, estrogen and follicle stimulating hormone and inhibin B indirectly specify the increasing follicle count. The aim of the present study was to compare the antral follicle count and AMH level for ovarian reserve.*
Materials and Methods: *The present study was conducted in the department of gynecology for a period of 1 year. Plasma was separated and frozen at -70 degree Celsius. The demographic details of all the subjects was recorded in the forms. All the subjects were asked about age at menarche. The level of AMH was estimated in ng/ml. Student t test was used as a test of significance and probability value of less than 0.05 was considered as significant.*
Results: *The study involved 57 females with the mean age between 35.78 +/- 5.32 years. The median age of the subjects was 34 years with the range between 28-39 years. Between 20-25 years of age, the mean antral follicle count was 15.6+/-4.1. Between 26-30 years of age, the mean antral follicle count was 14.5+/- 4.9. The level of AMH amongst subjects who smoked was 1.8 (0.8–3.5) and those who didn't smoke was 2.2 (1.2–3.2). The level of AMH amongst subjects who consumed alcohol was 2.2 (1.0–3.3) and those who didn't was 1.7 (1.2–3.2).*
Conclusion: *From the above study, we can conclude that levels of AMH and AFC significantly correlate with reproductive age.*
Keywords: *Antral follicle, Infertility, Mullerian.*

Introduction

Reproductive aging in females is determined by ovarian aging and expressed as a decrease in both the quantity and quality of ovarian follicles.[1] Peak fertility age amongst the females is during the mid-20s, after which the fertility starts to decay until menopause[2,3]. The chronological age at which menopause happens is evocative that reproductive aging is flexible amongst females, and shows that age is not a lone factor indicating the females' reproductive potential.[4] The decline in follicle number was initially regarded biphasic with a sudden decline in number at 37 years of age[5], but subsequent studies suggested that there

is gradual loss over time.[6] Various tests have been used to determine ovarian reserve, they are primarily used in Infertility clinics. Endocrine markers like anti-Mullerian hormone, estrogen and follicle stimulating hormone and inhibin B indirectly specify the increasing follicle count[7], ultrasound can be used to directly estimatethe follicle count.[8] Females with trisomic pregnancy have shown early onset of menopause.[9]Various studies suggests that circulating anti-Mullerian hormone concentrations are highly linked with the number of antral follicles and are more strongly connected to ovarian reserve as compared to the clinical markers like follicle-stimulating hormone (FSH), inhibin B and estradiol.[10-12]It is also known as Mullerian inhibiting substance and it is a fellow of the transforming growth factor-b family.[13] It is seen in both the sexesat different phases of development and it is best remembered for its partin fetal sex differentiation.[14] The aim of the present study was to compare the antral follicle count and AMH level for ovarian reserve.

Materials and Methods

The present study was conducted in the department of gynecology for a period of 1 year. The study was approved by the institutional ethical board and all the subjects were informed about the study and a written consent was obtained from all in their vernacular language. The subjects with first stimulation of ovary treatment, both ovaries visible on ultrasound, no previous hormone replacement therapy, no autoimmune disease, no past ovarian surgery and no history of radiation therapy were included in the study.Blood samples were drawn and stored in lithium heparin tubes. Plasma was separated and frozen at -70 degree Celsius. The demographic details of all the subjects was recorded in the forms. All the subjects were asked about age at menarche. The level of AMH was estimated in ng/ml. The subjects were divided as smokers and non-smokers, alcoholics and non-alcoholics and were also divided per the gravidae. The figure of years meanwhile menarche was obtained by

subtracting age with age at menarche. Gravidity encompassed any spontaneous pregnancy by the woman irrespective of the outcome of the same. All the data was arranged in a tabulated form and analyzed using SPSS software. Student t test was used as a test of significance and probability value of less than 0.05 was considered as significant.

Results

The study involved 57 females with the mean age between 35.78 +/- 5.32 years.

Table 1 shows the characteristics of the study population. The median age of the subjects was 34 years with the range between 28-39 years. The median antral follicle count was 10 with the range of 8-16. The median age at menarche was 12 years with the range between 11-15 years. The median level of AMH was 1.9 ng/ml with the range between 1.2-3.3 ng/ml.

Table 2 shows the descriptive values of antral follicle count. Between 20-25 years of age, the mean antral follicle count was 15.6+/-4.1. Between 26-30 years of age, the mean antral follicle count was 14.5+/- 4.9. Between 31-35 years of age, the mean antral follicle count was 11.1+/-3.3. Between 36-40 years of age, the mean antral follicle count was 10.3+/- 3.6.

Table 3 shows the association between ovarian reserve markers and lifestyle. The level of AMH amongst subjects who smoked was 1.8 (0.8–3.5) and those who didn't smoke was 2.2 (1.2–3.2). The level of AMH amongst subjects who consumed alcohol was 2.2 (1.0–3.3) and those who didn't was 1.7 (1.2–3.2). There was no significant difference amongst the smokers and non-smokers. The AFC amongst the smokers and alcohol intake subjects was 9 (6–20) and 10 (8–16) respectively. The AFC count amongst primigravidae is 11 (6–13) and multigravidae is 10 (9–14). There was no significant difference in the AFC count amongst gravidae index.

Table 1: Characteristics of study population

Variable	Median	range
Age (years)	34	28-39
Antral follicle count	10	8-16
Age at menarche (years)	12	11-15
AMH (ng/ml)	1.9	1.2-3.3

Table 2: Antral follicle count descriptive values.

Age (years)	Mean+/- SD	Minimum-Maximum
20-25	15.6+/-4.1	5-25
26-30	14.5+/- 4.9	4-30
31-35	11.1+/-3.3	3-31
36-40	10.3+/- 3.6	2-33

Table 3: Association between ovarian reserve markers and lifestyle

Lifestyle factors		AMH		AFC	
		Median (range)	P value	Median (range)	P value
Smoking	Yes	1.8 (0.8–3.5)	>0.05	9 (6–20)	>0.05
	No	2.2 (1.2–3.2)		10 (7–14)	
Alcohol	Yes	2.2 (1.0–3.3)	>0.05	10 (8–16)	>0.05
	no	1.7 (1.2–3.2)		10 (6–17)	
Gravidae	0	2.2 (1.2–3.7)	>0.05	11 (6–13)	>0.05
	>/=1	1.5 (0.8–2.9)		10 (9–14)	

Discussion

Ovarian reserve tests play a critical role in the management of assisted reproductive techniques related to the estimation of poor[15] or elevated ovarian response[16], disputing the controlled ovarian increased stimulation decorum[17,18] and dosing of gonadotropin level[19,20] to estimate the optimal frequency of oocyte. They are also used as marker to define polycystic ovarian syndrome, but the optimal level and the type of marker that should be regarded is not exactly clear.[21,22] The expected age of menopause can now be predicted using the various ovarian reserve estimation tests, even in with wide interval of confidence.[23,24] Recently, ovarian reserve screening amongst the general population has been contraindicated under the ethical concerns pertaining to planning reproductive life of women.[25]Variety of hormonal and ultrasonic markers have been studied for ovarian reserve estimation amongst females with a poor response to ovulation induction during the anti-retroviral therapy. Markers like antral follicle estimation have found to be more valuable than markers previously studies like follicular stimulating hormone level and levels of follicular phase inhibin B.[26]In our study, between 20-25 years of age, the mean antral follicle count was 15.6+/-4.1. Between 26-30 years of age, the mean antral follicle count was 14.5+/- 4.9. Between 31-35 years of age, the mean antral follicle count was 11.1+/-3.3. Between 36-40 years of age, the mean antral follicle count was 10.3+/- 3.6. Levels of serum AMH indicate the size of the primordial follicles present in ovary indirectly and show limited cycle variation during the intra and inter cycle period. The ultrasonic estimation of antral follicle count during the early follicular phase is directly related to ovarian reserve[22] and its decline should be considered as the sign of ovarian aging. In our study, the level of AMH amongst subjects who smoked was 1.8 (0.8–3.5) and those who didn't smoke was 2.2 (1.2–3.2). The level of AMH amongst subjects who consumed alcohol was 2.2 (1.0–3.3) and those who didn't was 1.7 (1.2–3.2). There was no significant difference amongst the smokers and non-smokers. The AFC amongst the smokers and alcohol intake subjects was 9 (6–20) and 10 (8–16) respectively. The AFC count amongst primigravidae is 11 (6–13) and multigravidae is 10 (9–14). There was no significant difference in the AFC count amongst gravidae index. Initial studies[10,11] had shown that level of AMH correlate significantly with antral follicle count. In females, anti mullerian hormone is generally not detected before 36weeks of gestation[27] and is produced throughout the entire reproductive life.[10,28] AMH is produced by thegranulosa cells of the preantral and young antral follicles[29-31] and gradually decreases in the advancing stages of development of follicle until disappearing with the large-sized antral follicles.[32-34] This hormone is not found in follicles which show features of atresia.[35]

Conclusion

From the above study, we can conclude that levels of AMH and AFC significantly correlate with reproductive age. This study concluded that both AMH and AFC are equally efficacious in predicting the level of ovarian follicle reserve. However, smoking, alcohol intake and gravidae do no significantly alter the AFC and AMH level. With advancing age decline, has been observed in the antral follicle count.

References

1. te Velde ER, Scheffer GJ, Dorland M, Broekmans FJ, Fauser BC. Developmental and endocrine aspects of normal ovarian aging. Mol Cell Endocrinol 1998;145:67–73.

2. Menken J, Trussell J, Larsen U. Age and infertility. Science 1986; 233:1389–1394.

3. van Noord-Zaadstra BM, Looman CW, Alsbach H, Habbema JD, te Velde ER, Karbaat J. Delaying childbearing: effect of age on fecundity and outcome of pregnancy. BMJ 1991;302:1361–1365.

4. El-Toukhy T, Khalaf Y, Hart R, Taylor A, Braude P. Young age does not protect against the adverse effects of reduced ovarian reserve—an eight year study. Hum Reprod2002;17:1519–1524.

5. Faddy MJ, Gosden RG, Gougeon A, Richardson SJ, Nelson JF. Accelerated disappearance of ovarian follicles in mid-life: implications for forecasting menopause. Hum Reprod1992;7:1342–1346.

6. Hansen KR, Knowlton NS, Thyer AC, Charleston JS, Soules MR, Klein NA. A new model of reproductive aging: the decline in ovarian non-growing follicle number from birth to menopause. Hum Reprod 2008;23: 699–708.

7. Broekmans FJ, de Ziegler D, Howles CM, Gougeon A, Trew G, Olivennes F. The antral follicle count: practical reco-mmendations for better standardization. FertilSteril2010;94:1044–1051.

8. Hendriks DJ, Mol BW, Bancsi LF, Te Velde ER, Broekmans FJ. Antral follicle count in the prediction of poor ovarian response and pregnancy after in vitro fertilization: a meta-analysis and co-mparison with basal follicle-stimulating hormone level. FertilSteril 2005;83:291–301.

9. Kline J, Kinney A, Levin B, Warburton D. Trisomic pregnancy and earlier age at menopause. Am J Hum Genet 2000; 67:395–404.

10. de Vet A, Laven JS, de Jong FH, Themmen AP, Fauser BC. Anti-mu¨ llerian hormone a putative marker for ovarian aging. Fertil Steril2002;77:357–362.

11. Fanchin R, Schona¨uer LM, Righini C, Guibourdenche J, Frydman R, Taieb J. Serum anti-Mu¨ llerian hormone is more strongly related to ovaran follicular status than inhibin B, estradiol, FSH and LH on day 3. Hum Reprod 2003;18: 323–327.

12. Muttukrishna S, Suharjono H, McGarrigle H, Sathanandan M. Inhibin B and anti-Mu¨ llerian hormone: markers of ovarian response in IVF/ICSI patients? Br J Obstet Gynaecol 2004; 111:1248–1253.

13. Visser JA, Themmen AP. Anti-mu¨ llerian hormone and folliculogenesis. Mol Cell Endocrinol 2004;234:81–86.

14. Josso N, di Clemente N. TGF-b family members and gonadal development. Trends Endocrinol Metab 1999;10: 216–222.

15. Broer SL, van Disseldorp J, Broeze KA, et al. Added value of ovarian reserve testing on patient characteristics in the prediction of ovarian response and ongoing pregnancy: an individual patient data approach. Hum Reprod Update 2013;19:26–36.

16. Broer SL, Dolleman M, van Disseldorp J, et al. Prediction of an excessive response in in vitro fertilization from patient characteristics and ovarian reserve tests and comparison in subgroups: an individual patient data metaanalysis. FertilSteril2013;100:420–9 e7.

17. Nelson SM, Yates RW, Lyall H, et al. Anti-Mullerian hormone-based approach to controlled ovarian stimulation for assisted conception. Hum Reprod 2009;24:867–75.

18. Yates AP, Rustamov O, Roberts SA, et al. Anti-Mullerian hormone-tailored stimulation protocols improve outcomes whilst reducing adverse effects and costs of IVF. Hum Reprod2011;26:2353–62.

19. La Marca A, Grisendi V, Giulini S, et al. Individualization of the FSH starting dose in IVF/ICSI cycles using the antral follicle count. J Ovarian Res 2013;6:11.

20. La Marca A, Papaleo E, Grisendi V, Argento C, Giulini S, Volpe A. Development of a nomogram based on markers of ovarian reserve for the individualisation of the follicle-stimulating hormone starting dose in in vitro fertilisation cycles. BJOG 2012;119:1171–9.

21. Dewailly D, Lujan ME, Carmina E, et al. Definition and significance of polycystic ovarian morphology: a task force report from the Androgen Excess and Polycystic Ovary Syndrome Society. Hum Reprod Update 2014;20:334–52.

22. Iliodromiti S, Kelsey TW, Anderson RA, Nelson SM. Can anti-Mullerian hormone predict the diagnosis of polycystic ovary syndrome? A systematic review and meta-analysis of extracted data. J Clin Endocrinol Metab 2013;98:3332–40.

23. Broer SL, Eijkemans MJ, Scheffer GJ, et al. Anti-Mullerian hormone predicts menopause: a long-term follow-up study in normoovulatory women. J Clin Endocrinol Metab2011;96:2532–9.

24. Wellons MF, Bates GW, Schreiner PJ, Siscovick DS, Sternfeld B, Lewis CE. Antral follicle count predicts natural menopause in a population-based sample: the Coronary Artery Risk Development in Young Adults Women's Study. Menopause 2013;20:825–30.

25. Tremellen K, Savulescu J. Ovarian reserve screening: a scientific and ethical analysis. Hum Reprod2014;29:2606–14.

26. Toner JP, Philput CB, Jones GS, Muasher SJ. Basal follicle-stimulating hormone level is a better predictor of in vitro fertilization performance than age. FertilSteril1991;55:784–791.

27. Rajpert-De Meyts E, Jorgensen N, Graem N, Muller J,Cate RL, Skakkebaek NE. Expression of anti-Mu¨ llerianhormone during normal and pathological gonadal development:association with different-iation of Sertoli and granulosacells. J Clin Endocrinol Metab1999;84:3836–3844.

28. Lee MM, Donahoe PK, Hasegawa T, Silverman B, Crist GB,Best S, Hasegawa Y, Noto RA, Schoenfeld D, Mac Laughlin DT. Mullerian inhibiting substance in humans: normal levelsfrom infancy to adulthood. J Clin Endocrinol Metab 1996;81:571–576.

29. Durlinger AL, Visser JA, Themmen AP. Regulation of ovarianfunction: the role of anti-mu¨ llerian hormone. Reproduction 2002; 124:601–609.

30. Baarends WM, Uilenbroek JT, Kramer P, Hoogerbrugge JW,van Leeuwen EC, Themmen AP, Grootegoed JA. Anti-Mu¨ llerian hormone and anti-Mu¨ llerian hormone type IIreceptor messenger ribonucleic acid expression in rat ovariesduring postnatal development, the estrous cycle, and gonadotrophin-induced follicle growth. Endocrinology 1995;136:4951–4962.

31. Vigier B, Picard JY, Tran D, Legeai L, Josso N. Production ofanti-Mu¨ llerian hormone: another homology between Sertoliand granulosa cells. Endocrinology 1984;114:1315–1320.

32. Cook CL, Siow Y, Taylor S, Fallat ME. Serum mu¨ llerian inhibiting substance levels during normal menstrual cycles. Fertil Steril2000;73:859–861.

33. Weenen C, Laven JS, von Bergh AR, CranfieldM, Groome NP,Visser JA, Kramer P, Fauser BC, Themmen AP. Anti-Mü llerianhormone expression pattern in the human ovary: potentialimplications for initial and cyclic follicle recruitment. MolHumReprod2004;10:77–83.

34. Fauser BC, van Heusden AM. Manipulation of humanovarian function: physiological concepts and clinical consequences. Endocr Rev 1997;18:71–106.

35. Ueno S, Takahashi M, Manganaro TF, Ragin RC,Donahoe PK. Cellular localization of mü llerian inhibiting substancein the developing rat ovary. Endocrinology 1989;124:1000–1006.