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Esraa Ali Abd Alameer Saed
Babylon Health Directorate, Babylon, Iraq

Basil O Saleh
Department of Biochemistry, University of Baghdad, College of Medicine, Baghdad, Iraq

Corresponding Author:
Esraa Ali Abd Alameer Saed
Babylon Health Directorate, Babylon, Iraq

Association of serum AMH concentration with age & body mass index in a sample of apparently healthy Iraqi women of reproductive & preimenopausal age range

Esraa Ali Abd Alameer Saed and Basil O Saleh

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Abstract

Background: Anti-Müllerian hormone (AMH) is a glycoprotein secreted primarily by granulosa cells of preantral and early antral follicles, serving as a reliable biomarker for ovarian reserve. Serum AMH levels decline with age and become undetectable near menopause.

Aim: This study aimed to assess age-related changes in circulating AMH levels among healthy Iraqi women and to estimate the predicted menopausal age.

Methods: A cross-sectional study was conducted at the Department of Biochemistry, University of Baghdad, College of Medicine, from January to August 2022, under the supervision of the Iraqi Board of Medical Specializations. A total of 123 apparently healthy women aged 25-55 years with regular menstrual cycles were enrolled. Participants were categorized into six age groups (5-year intervals) and by body mass index (BMI) into normal weight, overweight, and obese. Serum AMH concentrations were measured using ELISA.

Results: The overall mean serum AMH level was 1.06±0.94 ng/mL. A significant decline in AMH was observed with increasing age (p<0.0001), with the highest levels in the 25-29.9-year group and the lowest in the 51-55-year group. No significant difference was found between the 45-49.9-year and 50-55-year groups (P=0.54). Overweight (0.91±0.30 ng/mL) and obese women (0.59±0.20 ng/mL) had significantly lower AMH than normal-weight women (1.59±0.77 ng/mL, p<0.01), with no significant difference between overweight and obese groups (P=0.74).

Conclusion: AMH levels decline progressively with age, with menopause estimated around 51±5 years in Iraqi women. Higher BMI is associated with lower AMH levels.

Keywords: Serum, AMH, age, body mass index, women, reproductive, preimenopausal

Introduction

The ovaries are paired female reproductive organs located within the ovarian fossae on either side of the uterus, covered by a modified peritoneum and connected to the uterus via the ovarian ligament. They are responsible for housing and releasing ova and for the production of sex hormones essential for reproduction and secondary sexual characteristics [1-3]. Ovarian size and function vary with age and hormonal status; they are largest during the reproductive years and undergo progressive atrophy after menopause [1-3]. The female reproductive system exhibits cyclical physiological changes, known as the menstrual cycle, which prepares the endometrium for implantation. The average cycle lasts 28 days, beginning at menarche (ages 10-16) and ending at menopause, around the age of 51 years [4-6]. The cycle is regulated by a hypothalamic-pituitary-ovarian axis, in which gonadotropin-releasing hormone (GnRH) stimulates follicle-stimulating hormone (FSH) and luteinizing hormone (LH) secretion, thereby regulating follicular growth, ovulation, and corpus luteum function through coordinated feedback mechanisms involving estrogen, progesterone, inhibin, and activin [7-10]. Folliculogenesis begins in fetal life, with a finite pool of primordial follicles established by birth, declining progressively until menopause [11, 12]. Follicle recruitment and selection are influenced by intraovarian growth factors, gonadotropins, and metabolic signals. Anti-Müllerian hormone (AMH), a dimeric glycoprotein belonging to the transforming growth factor-beta (TGF-β) superfamily, is secreted by granulosa cells of preantral and small antral follicles [13, 14]. In females, AMH production starts in late fetal life, peaks in early adulthood, and declines steadily with age until becoming undetectable at menopause [15, 16].

AMH plays a regulatory role in folliculogenesis by inhibiting the initial recruitment of primordial follicles and modulating follicular sensitivity to FSH [17, 18]. Serum AMH concentration correlates with the number of small growing follicles and serves as a reliable, cycle-independent biomarker of ovarian reserve [19, 20]. Its clinical applications include predicting ovarian response in assisted reproduction, estimating reproductive lifespan, and assessing gonadotoxicity risk after chemotherapy [21, 22]. Several factors can influence AMH levels, including ovarian surgery [23], polycystic ovary syndrome (PCOS) [24], use of oral contraceptives [25], and obesity, which has been associated with significantly lower AMH levels [26]. Given its stability across the menstrual cycle and its strong association with ovarian reserve, AMH measurement offers a valuable tool for assessing reproductive aging and predicting the timing of menopause. However, data on AMH dynamics and menopausal age prediction among healthy Iraqi women remain scarce. Aim of the study: This study was designed to estimate the age-dependent decline in circulating AMH levels in a sample of apparently healthy Iraqi women and to predict their menopausal age.

Methods

This cross-sectional control study was conducted at the Department of Biochemistry, College of Medicine, University of Baghdad, in collaboration with the Iraqi Board for Medical Specializations, from January to August 2022. A total of 123 apparently healthy Iraqi women aged 25-55 years, from Babil and Baghdad, with regular menstrual cycles, were recruited from the general population. Participants were stratified into six age groups at five-year intervals (25-29.9, 30-34.9, 35-39.9, 40-44.9, 45-49.9, and 50-55 years). Based on body mass index (BMI), calculated as weight (kg)/height² (m²) according to WHO criteria [66], they were categorized as normal weight (18.5-24.9 kg/m²), overweight (25.0-29.9 kg/m²), or obese (≥30 kg/m²). Ethical approval was obtained from the Department of Biochemistry, Iraqi Board for Medical Specializations, and the Ministry of Health and Environment. Informed oral consent was secured from all participants. Exclusion criteria included chronic systemic diseases (e.g., liver or renal failure, diabetes mellitus, ischemic heart disease), gynecological disorders (e.g., PCOS, hyperprolactinemia, ovarian/uterine malignancies), use of hormonal or fertility medications, and history of *in vitro* fertilization. Data collection included demographic details, menstrual history, medical and drug history, and previous gynecological interventions. Height and weight were measured, and BMI was calculated. Sample collection and storage: Three milliliters of venous blood were drawn under aseptic conditions into sterile gel tubes, allowed to clot at room temperature for 20 minutes, and centrifuged at 2500 rpm for 10 minutes. Serum was separated and stored at −20 °C until analysis. Laboratory analysis: Serum AMH levels were measured using a human AMH ELISA kit (Biotin double antibody sandwich method), following the manufacturer's instructions. The assay employed monoclonal anti-AMH antibodies, biotinylated detection antibodies, and streptavidin-HRP for signal generation, with colorimetric detection at 450 nm. Statistical analysis: Data were analyzed using SPSS version 25.0. Descriptive statistics included frequency, percentage, mean, and standard deviation. ANOVA was used to compare means between groups. Pearson's correlation coefficient (r) assessed relationships between variables, interpreted as weak (r<0.3), moderate (0.3-0.5), or strong (>0.5). Statistical significance was set at $p<0.05$

Results

Table (1) shows the mean (±SD) values of age, BMI, and serum AMH levels of the entire included 123 apparently healthy Iraqi women. The mean (±SD) value of age was 40.02±8.68 year and that of BMI was 26.56±3.42 Kg/m^2. The mean (±SD) value of serum AMH levels of all enrolled women was 1.06±0.94 ng/ml and ranges from 0.02 ng/ml to 3.32 ng/ml.

Table 1: Mean ± SD value and range of age, BMI, and serum AMH levels of entire women group

Parameter	Entire group of women (N=123)	Minimum	Maximum
Age (year)	40.02±8.68	25.90	55.00
BMI (Kg/m2)	26.56±3.42	20.07	41.62
AMH (ng/ml)	1.06±0.94	0.02	3.32

Participants were aged 25-55 years, with distribution as follows: 25-29.9 years (16.3%), 30-34.9 years (18.7%), 35-39.9 years (16.3%), 40-44.9 years (16.3%), 45-49.9 years (15.4%), and 50-55 years (17.1%). BMI categories showed 27.6% normal weight, 58.5% overweight, and 13.8% obese. Marital status revealed 74.8% married and 25.2% single. As in Table 2.

Table 2: Distribution of participants by age group, BMI category, and marital status

Variable	Category	Number (%)	Total (%)
Age group	25-29.9 yrs.	20 (16.3%)	
Age group	30-34.9 yrs.	23 (18.7%)	
Age group	35-39.9 yrs.	20 (16.3%)	
Age group	40-44.9 yrs.	20 (16.3%)	
Age group	45-49.9 yrs.	19 (15.4%)	
Age group	50-55 yrs.	21 (13.6%)	100%
BMI group	Normal BMI	34 (28%)	
BMI group	Overweight	72 (58%)	
BMI group	Obese	17 (14%)	100%
Marital status	Married	92 (74.8%)	
Marital status	Single	31 (25.2%)	100%

The findings showed that the mean AMH levels significantly decreased as women's ages increased. The mean (±SD) values for the age groups of 25-29 years, 30-34 years, 35-39 years, 40-44 years, 45-49 years, and 50-55 years were 2.46±0.32 ng/ml, 1.93±0.28 ng/ml, 1.16±0.33 ng/ml, 0.49±0.19 ng/ml, 0.49±0.19 ng/ml, 0.15±0.08 ng/ml, and 0.02±0.03 ng/ml, respectively. However, as shown, there was no discernible difference in serum AMH levels between the 45-49 and 50-55 year subgroups (p-value 0.54) as in Table 3.

Table 3: Mean (±SD) values and range of AMHs level among subgroups of age and BMI

Age subgroups (Year)	Number	AMH (ng/ml)	Minimum	Maximum	P-Value
25-29.9 yrs.	20	2.46±0.32	1.79	3.32	< 0.0001*
30-34.9 yrs.	23	1.93±0.28	1.42	2.38	
35-39.9 yrs.	20	1.16±0.33	0.70	1.63	
40-44.9 yrs.	20	0.49±0.19	0.25	0.88	
45-49.9 yrs.	19	0.15±0.08	0.02	0.30	
50-55 yrs.	21	0.02±0.03	0.02	0.10	
Total	123	1.06±0.94	0.02	3.32	
Age subgroup / BMI	**Number**	**AMH (ng/ml)**	**Minimum**	**Maximum**	**P-Value**
Normal BMI	34	1.59±0.77	0.09	2.6	< 0.0001
Overweight	72	0.91±0.30	0.02	3.31	
Obese	17	0.59±0.20	0.02	3.12	
Total	123	1.06±0.94	0.02	3.32	

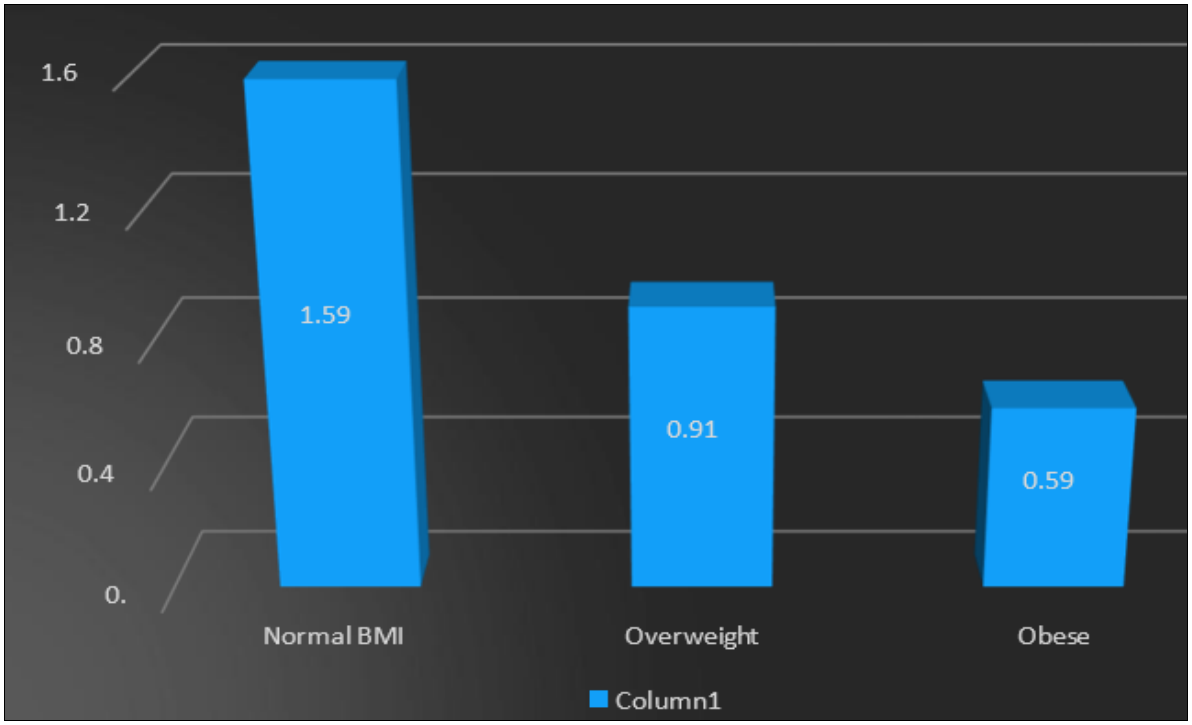

Fig 1: Mean of AMH in ng/ml among participant BMI groups

Table 4: Number and percentages of women according to menses & mean and range of age, AMH & BMI

Status of menstrual cycle	Number	Percentage	Mean ± SD of age & age range (years)	Mean ± SD AMH CONc. & range (ng/ml)	Mean ± SD of BMI Kg/m^2.
Regular menses	97	78.9%	36.76±6.6 (25.9-46.4)	1.33±o.87 (0.1-3.32)	26.2±3.7
Irregular menses	11	8.9%	50±1.6 (45.8-52)	0.07±0.03 0.4-0.1	26.9±1.8
Menopause	15	12.2%	53.5±1.7 48.5-55	Undetectable to 0.02	28.1±1.6
Total	123	100%	40.02±8.68, 25.9-55	1.06±0.94	26.56±3.42
P-Value			<0.0001	<0.0001	0.12

*p-value< 0.05

Pearson correlation coefficient was used to evaluate correlation between two numerical variables. There was significant negative strong correlation between age values and AMH concentrations (r=-0.93, p-value < 0.0001) as well as positive moderate correlation between age values and BMI (r=0.32, p<0.32). In addition, there was significant negative moderate correlation between AMH concentration and BMI (r=-0.34, p<0.0001), as in Table 2, 3.

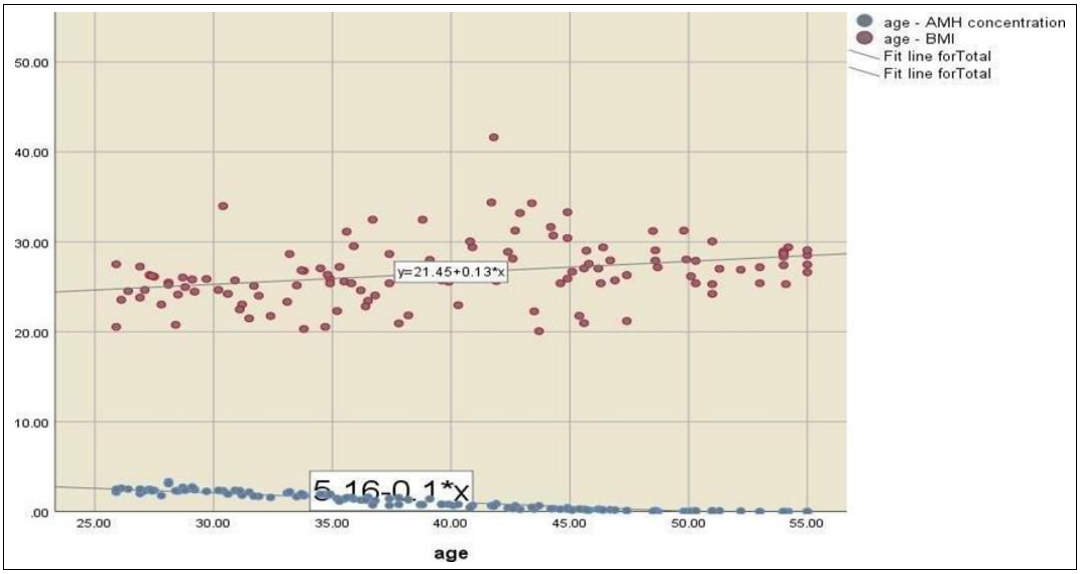

Fig 2: Scattered dot diagram of correlation among AMH, BMI and age

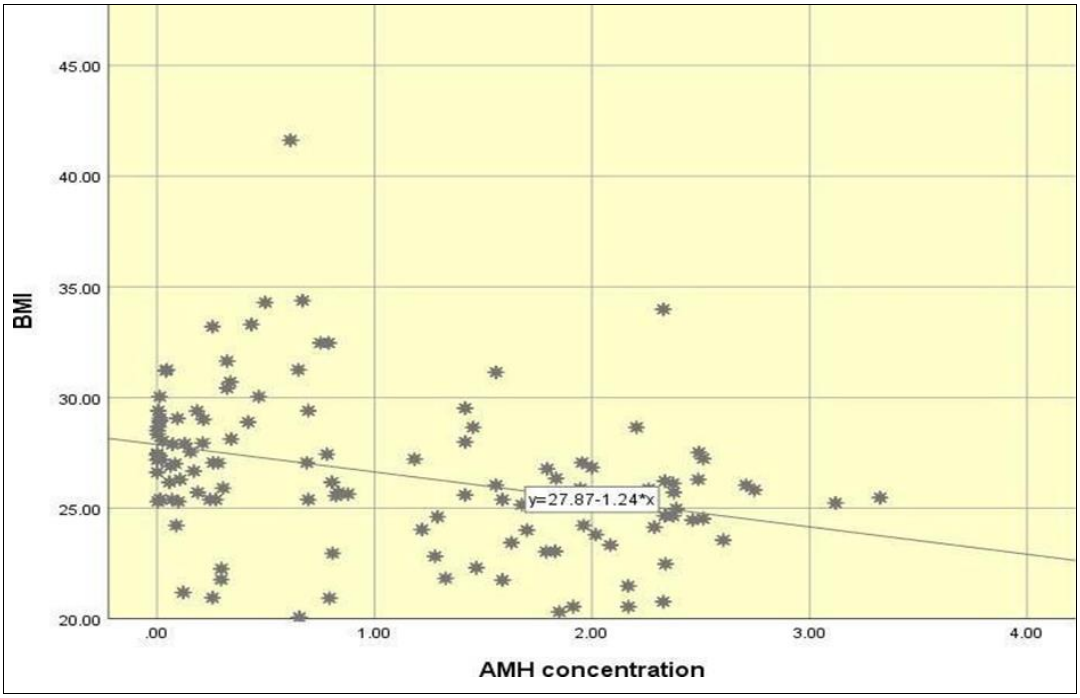

Fig 3: Scattered dot diagram of correlation among AMH

Discussion

The present study demonstrated a significant age-dependent decline in serum anti-Müllerian hormone (AMH) levels among healthy Iraqi women aged 25-55 years, with the highest levels observed in the 25-29.9-year group and the lowest in women above 45 years. This finding aligns with the established physiological role of AMH as a marker of functional ovarian reserve, reflecting the quantity of small antral and preantral follicles that progressively decrease with age until becoming undetectable near menopause [27]. The mean serum AMH concentration for all participants (1.06±0.94 ng/mL) was lower than that reported in a large Arabian Peninsula cohort (2.59±2.9 ng/mL) involving younger women with a broader reproductive age range (19-50 years) [28]. The difference may be attributed to the exclusion of women younger than 25 years in our study, as AMH peaks around 24-25 years and subsequently declines [29]. A Lebanese study (N=1190, age 17-54 years) also recorded a higher mean AMH (2.47±2.29 ng/mL), with the peak seen in those aged 17-20 years (6.71±2.91 ng/mL), further supporting the influence of age structure on mean values [30]. Similarly, Kotlyar and Seifer [31] demonstrated a consistent negative correlation between AMH and age across ethnicities, highlighting age as the dominant determinant of AMH variation. Egyptian data from PLOS One [32] confirmed a strong inverse relationship between AMH and age in 998 healthy women aged 15-49 years, corroborating our findings. The absence of a significant difference between the 45-49. 9-year and 50-55-year groups in our study likely reflects the advanced depletion of the follicular pool by this stage. In terms of BMI, we found that overweight and obese women had significantly lower mean AMH levels than normal-weight participants, consistent with previous research showing that increased adiposity may suppress AMH production by antral follicles [33]. Bernardi *et al*. [34] reported a similar inverse relationship in African-American women, with progressively lower AMH in those overweight or obese at age 18. The mechanism remains

unclear but may involve altered gonadotropin dynamics, insulin resistance, or changes in follicular microenvironment. However, conflicting evidence exists. Harlow SD [7] found no significant correlation between BMI and AMH in women under 45 years, suggesting that the BMI-AMH relationship may vary by population characteristics or methodological differences. Overall, our findings support the use of AMH as an age-sensitive marker of ovarian reserve and suggest that obesity is associated with reduced AMH levels in Iraqi women. These results have implications for fertility counseling, early reproductive planning, and predicting menopausal timing in this population.

Conclusion

This first study on healthy Iraqi women found a progressive age-related decline in serum AMH, with menopause expected around 50±5 years. The highest AMH levels occurred at ages 25-30, and the lowest at 50-55 years. Higher BMI was significantly associated with lower AMH levels.

Conflict of Interest

Not available

Financial Support

Not available

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