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Perceived stress, psychological distress and serum anti-Müllerian hormone levels among infertile and fertile women in North-central Nigeria

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Abstract

Background: Previous studies reported that self-reported stress, stressful life events, and psychological distress influence ovarian ageing and response. However, there are limited, yet conflicting findings on the effect of stress and psychological distress on serum anti-Müllerian hormone (AMH) as a biomarker of ovarian reserve, response, and ageing. This case-control study aimed to determine if stress and psychological distress levels were associated with serum anti-Müllerian hormone levels among 81 infertile and 109 fertile women of reproductive age attending the gynaecology and immunization clinics of a tertiary hospital in North-central Nigeria, respectively. Stress and psychological distress were measured using the Perceived Stress scale-10 and Kessler Psychological Distress Scale-10, respectively. The serum concentration of AMH was determined using enzyme-linked immunosorbent assays.

Results: There was a significantly higher stress ($p = 0.001$) and psychological distress ($p = 0.005$) levels among infertile women; however, there was no difference in serum anti-Müllerian hormone levels between the two groups ($p = 0.409$). There was no significant correlation found between perceived stress and serum anti-Müllerian hormone in both infertile ($r = 0.041$, $p = 0.719$) and fertile ($r = -0.090$, $p = 0.353$) women. There was also no correlation between psychological distress and serum anti-Müllerian hormone in the infertile ($r = -0.020$, $p = 0.860$) and fertile ($r = -0.049$, $p = 0.636$) groups. Controlling for age and body mass index in multivariate linear regression; stress and psychological distress were not significantly associated with serum anti-Müllerian hormone ($B = -0.005$; $p = 0.370$ and $B = -0.001$; $p = 0.811$).

Conclusion: Self-reported stress and psychological distress are not associated with serum anti-Müllerian hormone levels in infertile and fertile women of reproductive age. Also, ovarian ageing was accelerated in infertile women when compared to fertile women; however, this does not appear to be related to stress or psychological distress. More research is needed to understand factors that may contribute to this accelerated decline.

Keywords: Infertility, Anti-Müllerian hormone, Stress, Psychological distress, Fertility

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Background

Infertility is a stressful condition for the couples involved [1]. Studies have shown that the state of being infertile and the medical interventions that follow cause social, biological, emotional, and economic distress in both men and women affected—with women bearing more of the burden [1–3]. These have also been shown to affect the overall outcome of fertility interventions [4–6].

The word stress and distress are often used interchangeably; although they are related concepts, they are different psychological occurrences. Stress is a state of physiological and psychological imbalance associated with an external (environmental or psychological) or internal conditions (illness or medical procedure) which are found to be taxing or threatening or exceed the coping capacity of an individual due to insufficient environmental or personal resources [7]. The stress process involves three components: (1) the stressor, (2) the perception of stress, and (3) the affective, behavioural, and/or biological response to stress (distress) [7]. Psychological distress, however, is a state of psychological or emotional suffering characterized by features of mental disorder especially depression and anxiety [8]. Psychological distress in some situations is an end product of the stress process [8, 9].

Evidence suggests that the experience of stress and distress (mainly anxiety and depression) contributes to reproductive failure [10]. Biological responses to stress may lead to elevated levels of stress hormones, which impairs granulosa cell function and compromise follicular maturation, resulting in the eventual reduction in the number of oocytes available for harvest in In-vitro fertilization (IVF) cycles [4, 11]. Researchers also reported that psychological stress had a positive association with antral follicular count (AFC) in younger women, while the age-related reduction in AFC was accelerated by increased stress levels in all age groups [12]. Also, the investigators reported that depressive symptomatology (positive and negative affect) moderates the effects of psychological stress on this decline [13]. These findings support the psychosocial acceleration theory, which suggests that stress increases reproductive readiness in the short term at the expense of accelerating ovarian ageing [12, 14].

Anti-Müllerian hormone (AMH) is a glycoprotein secreted by the granulosa cells of primary and secondary pre-antral and small antral follicles; it is believed to inhibit follicle-stimulating hormone (FSH)-dependent oocyte recruitment [15]. Therefore, it prevents premature recruitment of oocytes before maturation [15]. Available evidence indicates that serum AMH is a reliable marker of ovarian reserve (quantity and quality of oocytes), function, and response in ovarian stimulation protocols [15–17]. Previous studies suggest that it is lower among infertile women when compared to fertile women [18, 19].

A cross-sectional study demonstrated a negative correlation between salivary alpha-amylase—a biomarker of psychological stress—and serum AMH in Chinese women with unexplained infertility [20]. However, another cross-sectional study found no relationship between self-reported fertility-related stress and serum AMH in infertile women with unexplained infertility [21]. Contrary to these two, Kudesia et al. reported a positive correlation between self-reported chronic stress and serum AMH in premenopausal women [22].

There are limited, yet contradictory evidence on the effect of stress and psychological distress on serum AMH, and available studies were mainly in developed nations outside of Africa. Therefore, this study aimed to determine if stress and psychological distress levels are related to serum AMH levels in infertile and fertile women attending the University of Ilorin Teaching Hospital, North-central Nigeria. We hypothesized that stress and psychological distress levels are associated with serum AMH in infertile and fertile women of reproductive age.

Methods

Study design and participants

This case-control study aimed to determine if stress and psychological distress are associated with serum AMH levels among 190 adult women of reproductive age: 81 infertile and 109 fertile women attending the gynaecology and immunization clinic of the University of Ilorin Teaching Hospital from March to July 2019. The power of this sample size was 91.52%, using the means and standard deviations for serum AMH levels (infertile women = 1.60 ± 2.51 ng/ml and fertile women = 2.71 ± 1.91 ng/ml) from the results of a study in Ile-Ife, Southwest of Nigeria [18].

We recruited study participants using purposive sampling following the study's inclusion and exclusion criteria. Study participants were adult women of reproductive age (18–45 years) with menstrual cycle length from 21 to 35 days. The infertile women were women attending the gynaecology clinic with 12-months history of inability to conceive despite adequate (2–3 times a week), unprotected sexual intercourse. The fertile controls were women attending the immunization clinic who had at least one pregnancy carried to term within the preceding 2 years, each pregnancy occurred spontaneously within 12 months of unprotected intercourse [18]. We excluded all women with a history of use of hormonal contraceptives or medications within 3 months before presentation, thyroid disorders, mental illness, use of chemotherapy and or radiotherapy, pelvic surgery(uterine or ovarian), and those who did not consent.

Data collection and analysis

We determined stress and psychological distress levels using the self-administered perceived stress scale-10 and Kessler psychological distress scale-10, respectively. The perceived stress scale-10 is a 10-item questionnaire used to determine the level of self-reported stress in a population. It is the most widely used scale to determine the level of perceived stress in a population in the past month on a 5-point Likert scale (0–4). Positively worded questions are reverse scored, and scores range from 0 to 40. The higher the score, the higher the stress level [23]. The reliability coefficient (Cronbach's alpha) in this population was satisfactory at 0.72 for the scale.

Kessler psychological distress scale-10 is a 10-item self-administered, short and simple measure of psychological distress. It measures anxiety and depressive symptoms that a person experienced in the past 4 weeks on a 5-point scale, and scores range from 10 to 50. The higher the score, the higher the psychological distress level [24]. The reliability coefficient (Cronbach's alpha) in the study population was satisfactory at 0.84.

We collected 3–5 ml of a random peripheral venous blood sample from each participant by venipuncture after the skin was prepared with a spirit swab. Blood samples were collected in sterile plain sample bottles and transported to the laboratory in ice packs at 4 °C. The sample was left to stand at room temperature for an hour, and after centrifuged for 10 min at 5000 rpm. Clear serum was pipetted into plain tubes in single-use aliquots and stored at -20 °C.

A consultant chemical pathologist analysed serum AMH concentration using human AMH enzyme-linked immunosorbent assay (ELISA) following the manufacturer's manual (Calbiotech Inc. El Cajon, CA, USA; 2018) at the chemical pathology research laboratory of the University of Ilorin Teaching Hospital. The sensitivity of the test was 0.039 ng/ml.

Independent variables were age, body mass index (BMI), psychological distress, and stress while the dependent variable was serum AMH. We conducted data analysis using SPSS version 25 for Windows (IBM Corp., Armonk, NY, USA, version 25.0). Normally distributed continuous variables were expressed in means ± standard deviation, and those not normally distributed were summarized as medians (interquartile range); furthermore, independent sample t test and Mann-Whitney U test were used as tests of difference, respectively. Categorical data were expressed in frequencies and percentages. Serum AMH was Log_{10}-transformed to meet the requirements for linear regression analysis. Tests of relationships between study variables were analysed using the Spearman correlation and linear regression. The level of significance was set at 5%.

Results

Table 1 summarizes the study variables by fertility status. There was a significant difference in age, body mass index (BMI), stress, and psychological distress between infertile and fertile women in this study. However, there was no difference in serum AMH levels between the two groups.

Among the infertile women, 37% had primary infertility while 63% had secondary infertility, with an average duration of infertility of 3.93 ± 2.36 years. The distribution of clinical diagnosis among the infertile women was: 40.7% unexplained infertility, 16% anovulation (including PCOS), and tubal factors respectively, 6.2% male-factor, 8.6% uterine factors and 12.5% more than one diagnosis.

There was a significant negative relationship between age and serum anti-Müllerian hormone level in infertile (B = − 0.042; p = 0.000) and fertile (B = − 0.026; p = 0.002) women (Figs. 1 and 2). However, linear regression showed a positive relationship with BMI in both groups, but this was only significant in the infertile women (B = 0.021; p = 0009) (Figs. 3 and 4).

There was no significant correlation between stress and serum AMH levels among infertile and fertile women. Also, correlation analysis showed no significant relationship between psychological distress and serum AMH in infertile and fertile women (Table 2).

Furthermore, in Tables 3 and 4, multivariate analysis controlling for age and BMI showed no significant relationship between stress and AMH level as well as psychological distress and serum AMH levels. However, the significant influence of age on serum AMH remained constant in both groups; although, an additional reduction of 0.1% in serum AMH with a year increase in age was noted among the fertile group (B = − 0.027; p = 0.002). BMI was only a predictor of serum AMH in infertile women, with a 0.1% decrease in the positive effect on serum AMH levels after adjusting for age, stress, and psychological distress.

Following the exclusion of women with polycystic ovarian syndrome (PCOS) in the infertile group, multivariate linear regression analysis demonstrated persistence in the significant negative relationship of AMH with age (B = − 0.035; p = 0.000), while the relationship between AMH and BMI (B = 0.015; p = 0.099) in infertile women (n = 74) became statistically insignificant. Stress (B = 0.000; p = 0.987) and psychological distress (B = − 0.001; p = 0.934) had no significant relationship with serum AMH.

Multivariate analysis demonstrated no significant difference in serum AMH levels between the infertile and fertile women (B = 0.043; p = 0.409) after controlling for age, BMI, stress, psychological distress, and excluding infertile women with PCOS.

Table 1 Summary of study variables among infertile and fertile women

Variables	Infertile mean ± S.D/median (IQR)	Fertile mean ± S.D/median(IQR)	***p value*** ***t*** test/Mann-Whitney ***U***
Age (years)	31.6 ± 4.5	29.9 ± 3.9	0.005*
BMI (kg/m^2)	25.4 (5.88)	23.8 (6.50)	0.007*
Stress	18.10 ± 4.95	15.53 ± 5.35	0.001*
Psychological distress	21 (10)	19 (8)	0.005*
Serum AMH (ng/ml)	5.50 (9.90)	5.59 (9.20)	0.494

*Statistical significance at 5% level of significance

Discussion

We found that infertile women reported significantly higher stress and psychological distress levels than fertile women, similar to findings from previous studies [1, 3]. This confirms that infertility is associated with high psychosocial strain and infertile women experience higher levels of stress and psychological distress compared to fertile women.

However, we found no significant difference in serum AMH levels between the two groups, which is contrary to previous studies comparing serum AMH among infertile and fertile women, in which authors reported a significantly lower serum AMH level among infertile women [18, 19]. The contradiction between our findings and that of other studies may be due to the inclusion of women with polycystic ovary syndrome (PCOS)—which is associated with significantly higher levels of serum AMH—in our study, contrary to previous studies. However, we found no significant difference in serum AMH levels between the two groups after the exclusion of women with PCOS from the infertile group and controlling for age, BMI, stress, and psychological distress. Furthermore, given the heterogeneous nature of infertility, a measure of ovarian function is not an exclusive marker of fertility. Therefore, findings may vary based on underlying factors responsible for infertility in the women in each study population.

Also, we found no significant correlation between stress and serum AMH level among infertile and fertile women. This is similar to findings by Vitek et al. in which psychosocial stress was not demonstrated to have any relationship with AFC or AMH among women with unexplained infertility [21]. Conversely, Dong et al. found a significant negative correlation between salivary amylase as a marker of psychological stress and serum AMH among Chinese women with unexplained

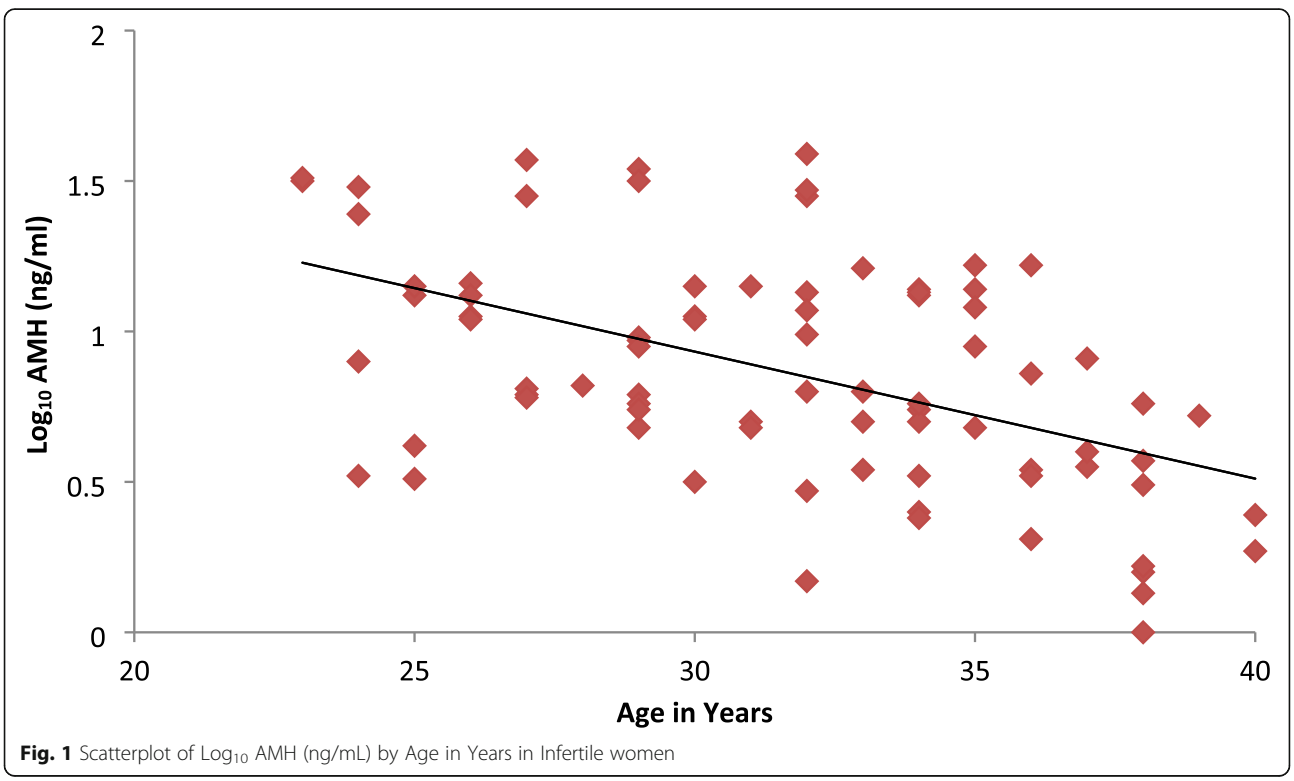

Fig. 1 Scatterplot of Log_{10} AMH (ng/mL) by Age in Years in Infertile women

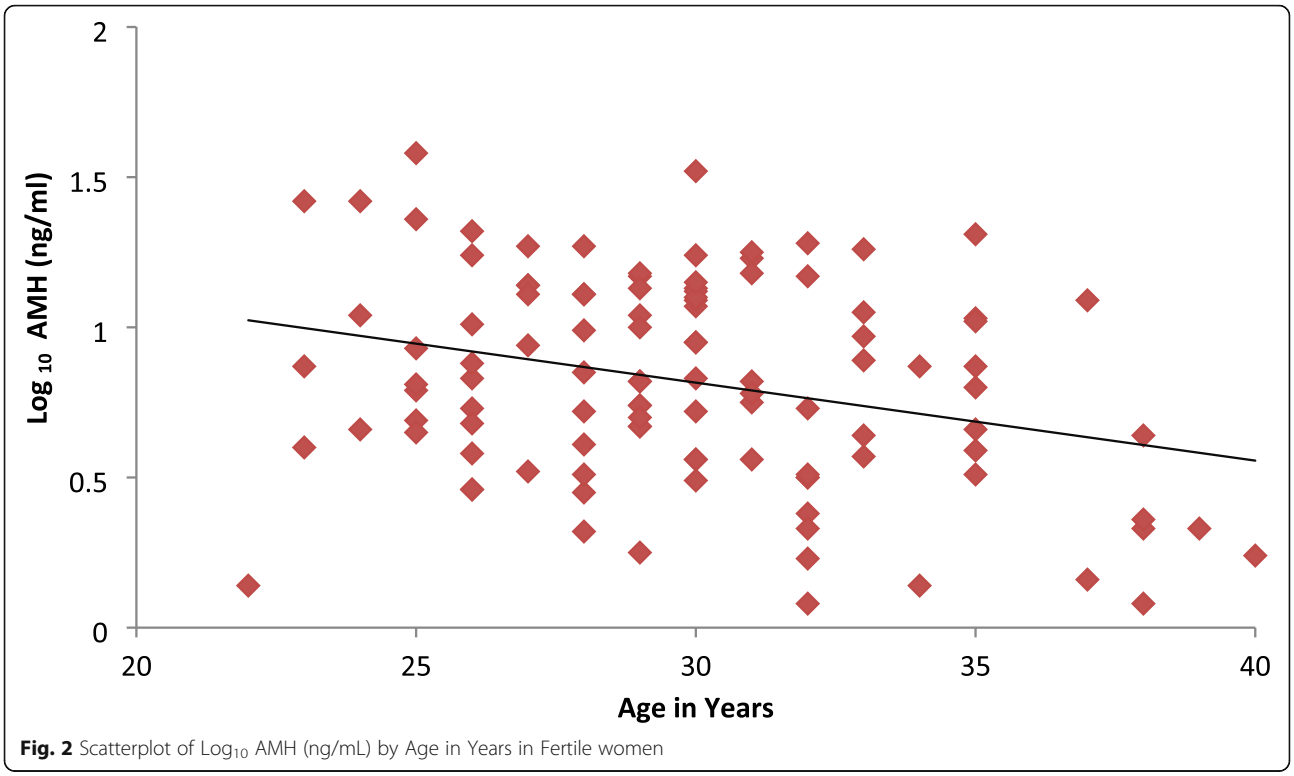

Fig. 2 Scatterplot of Log_{10} AMH (ng/mL) by Age in Years in Fertile women

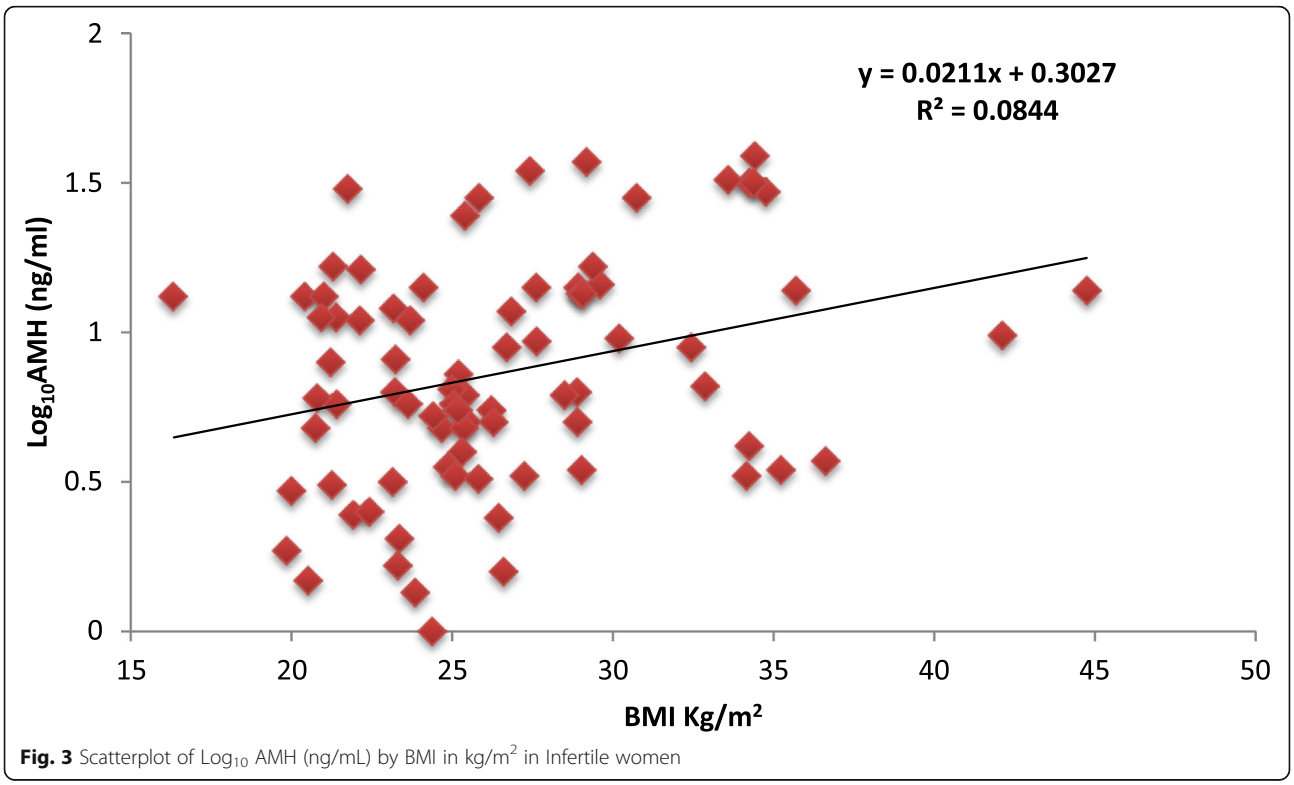

Fig. 3 Scatterplot of Log_{10} AMH (ng/mL) by BMI in kg/m^2 in Infertile women

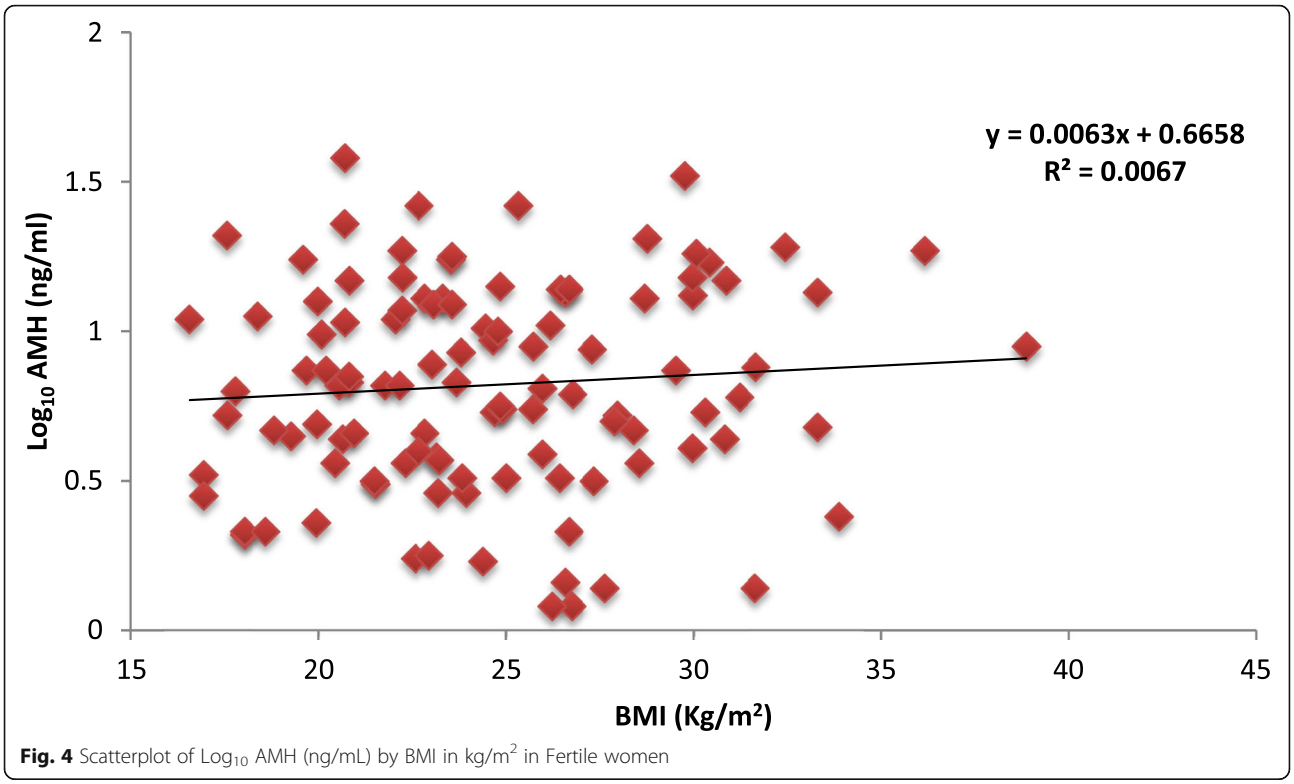

Fig. 4 Scatterplot of Log_{10} AMH (ng/mL) by BMI in kg/m^2 in Fertile women

infertility [20]. Contrary to previous findings, Kudesia et al. reported a positive correlation between chronic stress—measured with Wheaton's Chronic Stress Scale—and serum AMH in pre-menopausal women [22].

The contradictions could be because of the difference in the marker of psychological stress used and the overall sensitivity of perceived stress scale-10 used in our study to the biological response to stress. Since some of the theories surrounding the influence of psychological stress borders on the eventual influence the biological response to stress can have on human reproduction [11], a biomarker of this process will most likely be a more sensitive measure of an effect. Although perceived stress scale is a valid and reliable measure of stress [24, 25], it may not translate in all situations to the biological response or vulnerability to stress which could influence a significant change in serum AMH levels in these women.

We found no significant correlation between psychological distress—measured by the Kessler psychological distress scale-10—and serum AMH in infertile and fertile women. This finding is similar to those of Vitek et al. in which psychological distress—measured by the Patient Health Questionnaire (PHQ)—did not demonstrate any significant relationship with serum AMH or AFC [21]. Perhaps, the nonspecific and subjective nature of the instruments used affects their sensitivity to the biological distress that affects serum AMH levels.

We found that with an increase in age, there was a corresponding reduction in serum AMH among infertile and fertile women. This is similar to findings by other studies and lends credence to the use of serum AMH as a marker of ovarian ageing [26, 27]. However, infertile women showed almost twice the reduction in serum AMH with age, when compared to fertile women in this study population, suggesting that ovarian ageing is accelerated in women with infertility. Contrary to findings from previous studies, the rate of decrease in serum AMH with age remained constant in both groups despite controlling for stress and psychological distress,

Table 2 Correlation between stress, psychological distress, and AMH among infertile and fertile women

	Infertile		Fertile	
Variables	Spearman's correlation (***r***)	***p***	Spearman's correlation (***r***)	***p***
Stress level	0.041	0.719	− 0.090	0.353
Psychological distress level	− 0.020	0.860	− 0.049	0.636

Table 3 Multivariate linear regression among infertile women

Model: Log_{10} AMH			*p* value	95.0% confidence interval for *B*	
	B	Std. error		Lower limit	Upper limit
(Constant)	1.667	0.378	0.000	0.915	2.418
Stress	− 0.002	0.009	0.852	− 0.020	0.016
Psychological distress	0.001	0.009	0.914	− 0.012	0.013
Age (years)	− 0.042	0.008	0.000*	− 0.058	− 0.026
Body mass index (kg/m^2)	0.020	0.007	0.005*	− 0.006	0.034

*Statistical significance at 5% level of significance

which suggests that the acceleration in ovarian ageing in infertile women was not dependent on these psychological variables [12, 13]. However, the rate of reduction in serum AMH with age was 0.7% less following the exclusion of women with PCOS from the infertile group. This suggests that the inclusion of young women with PCOS with higher serum AMH levels contributed partly to the accelerated decrease in serum AMH with age in the infertile group. Also, previous studies reported that women with PCOS have an accelerated decrease in serum AMH with age compared to non-PCOS controls [28, 29].

There was a positive relationship between BMI and AMH in the infertile group, while none was found in the fertile group. This contradicts the findings by previous studies in southwest Nigeria, in which Okunola et al. found no relationship between BMI and random serum AMH in infertile and fertile women, while Oke and colleagues reported no significant relationship between BMI and day 3 AMH in all study participants [18, 30]. Outside Nigeria, Freeman et al. reported a negative relationship between BMI and serum AMH in Caucasians and African-American women of late reproductive age, while Moy et al. in a multi-ethnic study among infertile women also found a negative correlation between BMI and serum AMH levels in Caucasians, although no relationship was demonstrated in African-American, Hispanic, or Asian women [31, 32]. Dólleman et al., however, demonstrated no relationship between levels of BMI and serum AMH in a large population study of premenopausal women [33].

However, following the exclusion of women with PCOS in the infertile group, we found no significant relationship between BMI and serum AMH, suggesting that the significant positive relationship demonstrated in the infertile group was due to the inclusion of anovulatory women with PCOS in the infertile group of this study, which is often associated with high BMI and high serum AMH levels [34].

Strength and limitations

The case-control design of our study gave room for comparison and sub-analysis to further understand the effect of stress and psychological distress on serum AMH levels in infertile and fertile women, independently. This further removed the influence of infertility as a confounder, since stress, psychological distress, and abnormalities of serum AMH have all been found to be independently associated with infertility.

However, this study is not without its limitations. In determining the effect of stress and psychological distress on serum AMH, self-administered scales—which measured the perception of stress and psychological distress over a month—used are highly subjective and could be biased by the language barrier, concentration, and environment of the participants during the process of completing the questionnaires, although there is evidence that perceived-stress scale scores correlate with the biological response to stress [25]. The inclusion of a more objective measure of the biological response and vulnerability to stress would have increased the

Table 4 Multivariate linear regression among fertile women

Model: Log_{10} AMH			*p* value	95.0% confidence interval for *B*	
	B	Std. error		Lower limit	Upper limit
(Constant)	1.561	0.315	0.000	0.937	2.186
Stress	− 0.006	0.007	0.409	− 0.020	0.008
Psychological distress	− 0.003	0.005	0.625	− 0.013	0.008
Age (years)	− 0.027	0.008	0.002*	− 0.043	− 0.011
Body mass index (kg/m^2)	0.008	0.007	0.250	− 0.006	0.023

*Statistical significance at 5% level of significance

reliability of stress measurement and broadened the scope of the study.

Conclusion

The results of our study suggest that self-reported stress and psychological distress has no effect on serum AMH levels in infertile and fertile women within the reproductive age in North-central Nigeria. Serum AMH level is not exclusively a reliable marker of fertility; since the underlying pathology surrounding infertility in women are diverse and heterogeneous. Furthermore, age-related decline in serum AMH is accelerated in women with infertility; however, from our findings, this is not stress-related. Therefore, more research is required to determine factors that may influence this accelerated decline in infertile women as opposed to fertile women.

Abbreviations

AFC: Antral follicular count; AMH: Anti-Müllerian hormone; BMI: Body mass index; ELISA: Enzyme-linked immunosorbent assay; IQR: Interquartile range; PCOS: Polycystic ovary syndrome; PHQ: Patient health questionnaire; SD: Standard deviation

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Authors' contributions

O.I was involved in the study design, literature search, data collection, input and analysis, laboratory analysis, manuscript drafting, and review. J.O was involved in the study design, result interpretation, and manuscript review. J.O.Y contributed to the laboratory analysis, interpretation of results, and manuscript review. M.O was involved in the manuscript editing and review. The manuscript was read and approved by all authors for submission.

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Availability of data and materials

Data supporting findings are available from the corresponding author on reasonable request.

Ethics approval and consent to participate

This was gotten from the University of Ilorin institutional ethics review committee (ERC/PAN/2019/01/1869) in February 2019. Informed consent was signed by each participant before participation.

Consent for publication

Not applicable

Competing interests

The authors have no competing interest to declare.

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