Seminal Malondialdehyde (MDA) Levels in Iraqi Males with Infertility: Exploring the Relationship with Body Mass Index (BMI)

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KEYWORDS
Seminal Plasma, Male Infertility, Malondialdehyde (MDA), Lipid Peroxidation

ABSTRACT
Background: The investigation of the relationship between BMI and MDA levels in male infertility is pivotal for comprehending the underlying mechanisms of infertility, pinpointing potential therapeutic targets, and devising personalized approaches to enhance fertility outcomes in men. Therefore, the primary objective of this study was to explore the correlation between MDA levels in seminal fluid and infertility in Iraqi males, along with its potential association with Body Mass Index (BMI). Methodology: Ninety males aged between twenty and forty-five were included in this cross-sectional study, all diagnosed with infertility by specialists at the infertility unit of Al-Batool Teaching Hospital between February 2022 and February 2023. The participants were categorized into three groups: the Normozoospermic Group (G1), the Asthenospermia Group (G2), and the Oligozoospermic Group (G3). Results: The distribution of age and BMI across the study groups (G1, G2, and G3) appeared to be uniform and not statistically significant, indicating more noticeable variations among the groups. However, waist circumference (WC) analysis suggested statistically significant differences among the groups, highlighting a non-negligible change in the frequency of obesity within the groups participating in the study. Between the study groups, there was no statistically significant difference in smoking habits. The seminal fluid MDA levels for G1 were significantly lower than those for G2 and G3. Additionally, Seminal plasma MDA levels and BMI showed a non-significant positive correlation. Conclusion: There is no correlation between BMI and MDA levels in male infertility.

1. Introduction

Fertility refers to an individual's capacity to engage in normal sexual activity for the purpose of reproduction. Optimal fertility is contingent upon the adequate production of viable sperm. Any disruption in this process can result in infertility (1). Sperm chromatin that has been altered or degraded may have serious effects for fertilization or the risk of fetal developmental disturbances (2). Abnormal sperm morphology (teratospermia), low sperm numbers (oligospermia), low sperm motility (athenospermia), and an absence of spermatozoa (azoospermia) are all clinical manifestations of male infertility (3). Obstruction, testicular failure, hormone issues, drug and alcohol use, cryptorchidism, sperm agglutination, low semen volume, idiopathic infertility, ejaculatory dysfunction or erectile, varicocele, abnormal viscosity, high thickness of sperm, endocrine trouble, environmental causes, and genetic causes have all been identified as potential contributors to infertility (4). Obesity is widely acknowledged as a significant risk factor for various health issues, including infertility. However, its association with reduced sperm count was not officially recorded until 2004 (5).

Reputable infertile consultants estimate that male obesity accounts for 40% of male infertility; hence, male obesity (defined as an excessive buildup of white adipose tissues in the body) has a significant impact on infertility (6). The prevalence of obesity is increasing over the world, Body mass index (BMI) is a straightforward measure of the ratio of weight to height that is used to classify humans as obese or overweight both individually and in groups, men with a high body mass index have a greater risk of infertility, the risks of both chronic disease and reproductive issues are increased in the obese, male fertility is negatively impacted by obesity due to its pathophysiology (7). When lipids and polyunsaturated fatty acids undergo peroxidation, they become targets of oxidative stress, which is characterized as an imbalance between the body's production and clearance of reactive oxygen species (ROS), the chain reaction evolves in three steps: initiation, propagation, and termination. Each stage produces a different reactive product (8). Malondialdehyde (MDA) is a widely utilized marker for studying oxidative stress in biological systems. MDA accounts for approximately 20% of the end-products resulting from the oxidative degradation of lipids in vitro(9). Seminal levels of reactive oxygen species and malondialdehyde (MDA), a byproduct of lipid peroxidation caused by oxidative damage, also rose in tandem with these findings, during infection and tissue damage, semen contains high amounts of the same cytokines that play an important role in immune regulation for the male gonad. Leukocytospermia is intimately linked to their involvement in inflammation (10). The aim of this study was to investigate the relationship between MDA levels in seminal fluid and infertility in Iraqi males, as well as its potential correlation with Body Mass Index (BMI).

Materials and Methods

Study Population

Ninety males aged between twenty and forty-five including in a study (cross-sectional study) all were diagnosed with infertility in by specialists at the infertility unit of Al-Batool Teaching Hospital between February 2022 and February 2023. Subject were divided into three groups: the Normozoospermic Group (G1), the Asthenospermia Group (G2), and the Oligozoospermic Group (G3).

Anthropometric Measurements

Calculation of Body Mass Index (BMI)

Body mass index (BMI) is defined as the weight in kilograms divided by the square of the height: BMI = weight (kg)/height (m^2) BMI/the weight measuring by a scale and the height measuring by a stadiometer was categorized as follows:

- BMI less than 18.5 (underweight)
- BMI less than 24.9 (normal weight).
- BMI between 25-29.9 (overweight).
- More than 30 (obesity class I).
- More than 35 (obesity class II).
- More than 40 (obesity class III).

Measurement of Waist circumference

Waist circumference was measured in centimeters by using a tape measure.

Seminal fluid samples

All patients' semen samples were taken in sterile, clean cups, and placed in an incubator for 15-20 minutes to cause the semen to liquefy, throughout the course of three to four days of abstinence. The samples were then examined under a light microscope. Seminal plasma was obtained by centrifugation at 4000 rpm for 15 minutes were divided into two portions and kept until assay.

MDA Assay

Seminal MDA levels were measured by competitive Enzyme-linked immunosorbent assay (ELISA) according to the manufacturer (Cloud-Clone Corp/USA/Cat No.CEA597Ge).

Statistical Analysis

the statistical analysis conducted using Microsoft Excel for data input and preparation, which included organizing and cleaning the data for analysis, One-way ANOVA followed by multiple comparisons test was performed using GraphPad Prism version 19.5.1 for Windows, GraphPad Software, San Diego, California USA, and MedCalc® Statistical Software version 20.215was used to calculate and examine the strength and direction of relationships between variables, particularly between seminal plasma Malondialdehyde (SF-MDA) levels and other factors such as BMI. The statistical significance level, was set at 0.05 for most tests, indicating that results with a p-value less than 0.05 were considered statistically significant.

Results and Discussion

In an examination of anthropometric and demographic variables. Age, Body Mass Index, and Waist Circumference —across three groups (G1, G2, and G3).

Age and BMI appeared to be uniformly distributed across the three groups with mean ages of 29. 67±1.043, 29.86±0.896, and 29.96±0.752 years, and BMI, the mean values were 26.79±0.706, 28.77±0.866, and 26.96±0.934 for G1, G2, and G3, respectively. The P= 0.972=0.189 respectively, strongly suggests that the differences these groups are not statistically significant, demonstrated more discernible differences among the groups.

The mean waist circumference (WC) were 91.96±2.292 for G1, 99.86±2.598 for G2, and 92.60±2.504 for G3. The P= 0.047 suggests that the differences in WC among the groups are statistically significant.as seen in figure (1).

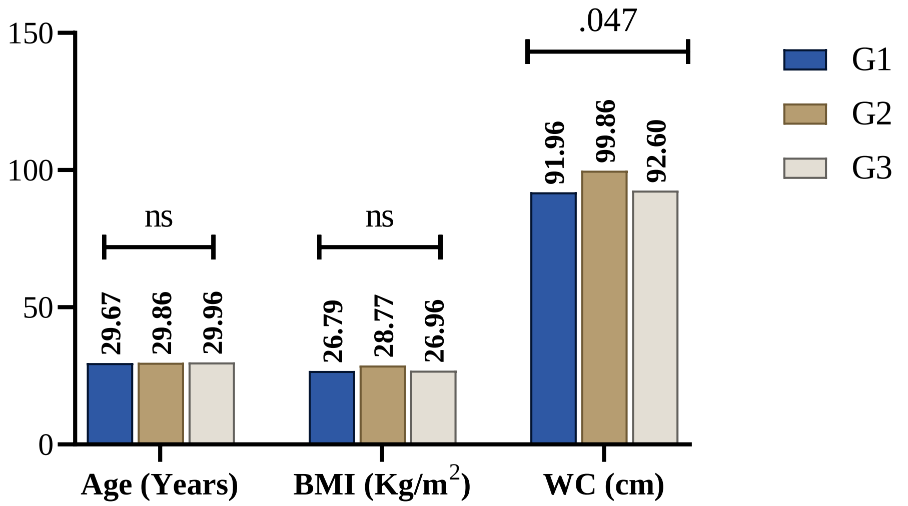

Figure 1: Characteristics of male in the study groups.

Figure (2) illustrates the results, which indicated a non-significant change in the study groups frequency of obesity (P = 0.3819).

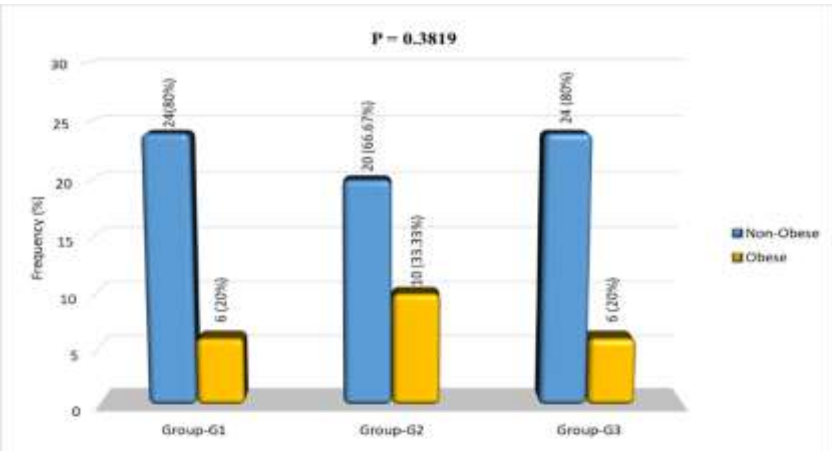

Figure 2: Frequency of obesity in the studied groups.

The figure (3) provides data on the participants' smoking habits. The P=459 indicates that there isn't a statistically significant variation in the research groups' smoking patterns.

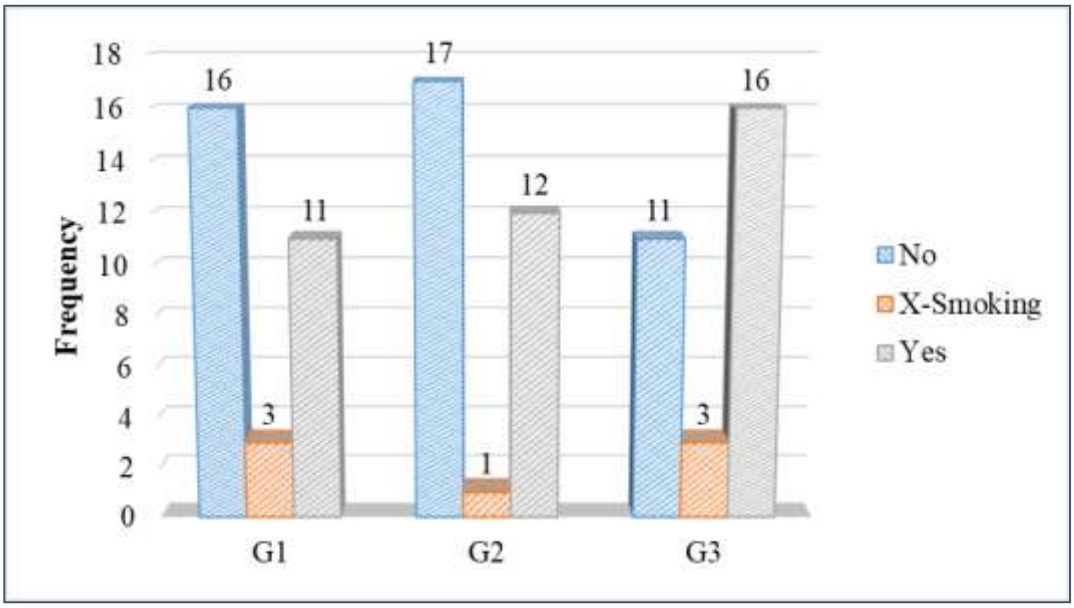

Figure 2: Frequency of smoking in the studied groups.

The mean of SF_MDA for G1 236.59 ± 105.30, was significantly lower than for G2 1904.08 ± 877.85, similarly, compared to G3 1,042.78 ± 339.03 with P= 0.001. As seen Table 1, and figure(4).

Table1: The Mean ± SE of SF-MDA for study groups.

Parameter	Mean ± SE			P-value
	G1	G2	G3	
SF-MDA (ng/ml)	236.59± 19.23	1904.08± 160.27	1042.78± 61.90	< .001

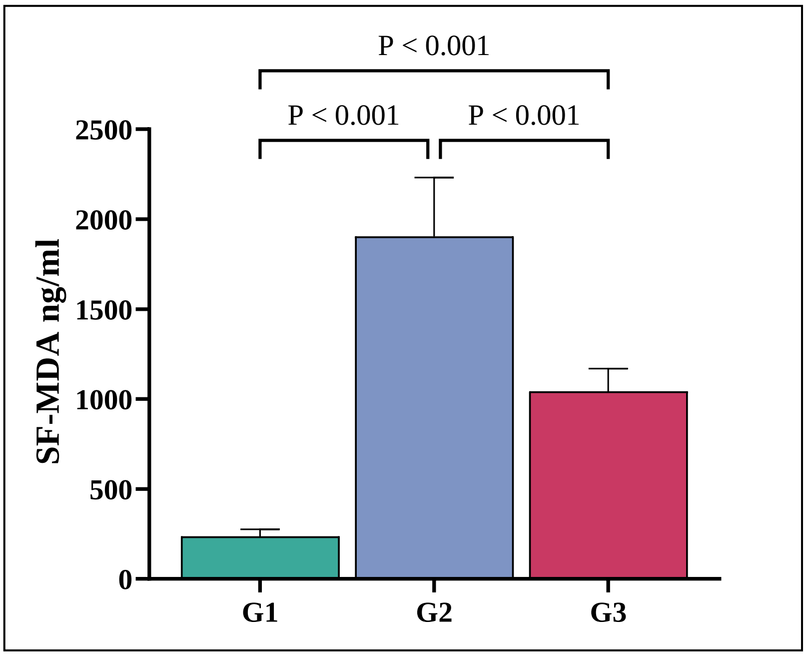

Figure 4: Means of SF-MDA by study groups.

A non-significant positive correlation was observed between seminal plasma MDA and BMI were (r = 0.001, 0.30, and 0.30, p =0.993,0.103,and 0.102) for G1, G2, and G3 respectively ,as seen in figure (5).

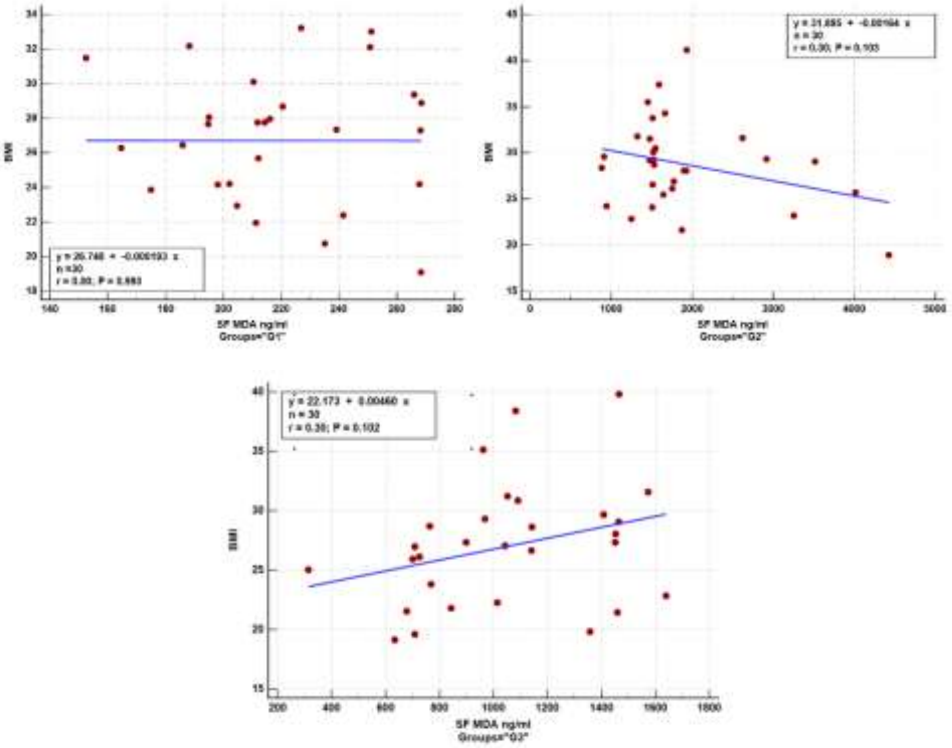

Figure 5: correlation of SF-MDA in the studied groups.

The interpretation that was provided suggests that in the context of study (Age, BMI, and WC) chosen to be matched among the three groups (G1, G2, and G3), there for no statistically significant differences when among study groups. This means that when comparing the mean or average age and BMI values of individuals in the three groups, the observed differences are not likely due to chance alone. In other words, any variations in age and BMI among the groups are not statistically significant (P= 0.972, and 0.189) respectively as shown in figure (1), this disagree with Chavarro, et al, (11) and Macdonald, et al, (12) and Kumar, et al, (13). Age and BMI are often considered as potential factors that might influence the outcomes of a study, the lack of significant differences in these factors among the three groups suggests that any observed variations in sperm characteristics are less likely to be attributed to differences in age or BMI Kadhem *et al.*,(14). This finding can be important for the study's validity and interpretation, it suggests that the differences in sperm characteristics observed among the groups are more likely to be related to the specific condition (G1, G2, and G3) rather than being confounded by age or BMI differences Hasan *et al.*,(15). The three groups' waist circumferences differ statistically significantly, as indicated by the p-value of 0.047, as shown in figure (1). There is evidence to suggest a relationship between waist circumference and male fertility issues, excess fat around the waist can be indicative of overall body fat accumulation, which can negatively affect both general health and fertility Hasan *et al.,* (16).

Higher waist circumference may be linked to insulin resistance and elevated blood sugar levels, negatively affecting fertility, increased fat can influence the production of sex hormones, potentially affecting sperm quality Zańko *et al.*,(17). Excess fat around the waist may lead to higher testicular temperatures, which is unfavorable for efficient sperm production, excess fat can contribute to systemic inflammation, affecting the immune system and potentially influencing sperm quality, as shown by Chavarro *et al*, (11). It appears in Figure (2) in this study depicts results related to the frequency of obesity within different sperm groups, namely (G1, G2, and G3). The key finding highlighted is a non-significant change obesity among these study groups.

A non-significant change typically suggests that there is no statistically significant difference(P = 0.3819) in the frequency of obesity between the specified groups, in the context of this study, this could mean that, based on the data and statistical analysis performed, there is no evidence to support the idea that the prevalence of obesity differs significantly among the Normozoospermic, Asthenospermia, and Oligozoospermic groups this disagree with Chavarro, et al, (11) and Macdonald, et al, (12). Certainly, there is some variability in research findings regarding the relationship between obesity and fertility, this variation can be attributed to factors such as differences in study design, study samples, and the variables analyzed Abdul-Rahman *et al.,* (18). Some studies suggest that obesity may not have a significant impact on fertility in men, Sperm parameters, in particular, including count, motility, and morphology, stay within the normal range, however, negative effects may be observed in cases of severe obesity or when obesity is associated with other health issues, this agrees with Kahn et al., (19)**.**

The statement suggests that Figure 3 presents data on the smoking habits of participants, and the corresponding p-value (P=0.459) suggests that the smoking behaviors of the G1, G2, and G3 groups do not differ statistically significantly. There is conflicting evidence regarding the association between male infertility and smoking some studies not found a significant association this agreement with study conducted by Saleh *et al.*,(20), might be was a sample size that's rather small in this study. While, there is scientific evidence indicating that smoking is associated with male fertility problems, smoking can negatively affect various aspects of male reproductive health, smoking may lead to a reduction in semen quality, including a drop in the number of sperm, motility, and alterations in the shape and structure of sperm Henriques et al. (21).

Some studies suggest that smoking can cause damage to the DNA in sperm, affecting their ability to fertilize properly, smoking can impact testicular function, resulting in a reduction in the production of male hormones responsible for sperm production, there is evidence suggesting that smoking may increase the likelihood of infertility and contribute to delayed conception, this agree with De Brucker, *et al*, (22),and Mostafa, *et al*, (23). As shown table (1) and figure (4)Seminal Plasma MDA levels for G1 were significantly lower (P<0.001) than those for both G2 and G3. Malondialdehyde (MDA) is a naturally occurring compound that serves as a marker for oxidative stress and lipid peroxidation in cells, it is a byproduct of the degradation of polyunsaturated fatty acids in cell membranes, when cells are exposed to oxidative stress, such as from reactive oxygen species (ROS) Hassan et al.,(2022), lipid peroxidation can occur, leading to the formation of MDA, this agree with Tavilani, et al (2005). Lipid peroxidation is a process where free radicals react with lipids, especially polyunsaturated fatty acids, in cell membranes, this process generates MDA as a final product, lower MDA levels in G1 may suggest reduced oxidative stress in the Normozoospermic Group (G1) compared to the Asthenospermia Group (G2) and Oligozoospermic Group (G3), oxidative stress is known to negatively impact sperm quality and function, high levels of ROS can lead to DNA damage, lipid peroxidation, and impaired sperm function. The observed difference in MDA levels between the groups may be associated with the differences in sperm parameters, especially considering that G2 is associated with asthenospermia (reduced sperm motility) (24). Elevated MDA levels are associated with various pathological conditions, including inflammation, cardiovascular diseases, neurodegenerative disorders, and reproductive health issues Singh etal.,(25). Studies often investigate MDA levels in relation to male fertility. Elevated MDA levels in seminal plasma or sperm cells may be linked to reduced sperm motility, viability, and overall sperm function, this agree with Kurkowska, et al, (26), and Bergsma et al, (27).

Conclusion

The absence of a relationship between BMI and MDA levels in male infertility may be attributed to the multifactorial nature of male infertility, the complexity of oxidative stress mechanisms, the limitations of the study design, and the heterogeneity of infertility causes. Further research considering these factors is necessary to fully understand the relationship between BMI and oxidative stress in male infertility.

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