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Influences of Semen Parameters and Malondialdehyde Levels on Male Infertility

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ABSTRACT

Seventy samples were gathered from males experiencing either primary or secondary infertility, while 35 samples were obtained from fertile individuals serving as the control group.

Throughout the study, the World Health Organization (WHO) criteria for semen parameters were consistently applied. These criteria encompassed aspects such as age, specimen volume, total sperm count/mL, sperm morphology, specimen pH, liquefaction time, and sperm activity. The impact of reactive oxygen species was assessed by measuring malondialdehyde concentration.

A computerized system (General Sperm Analyzer, Motic type) was employed to assess the semen parameters. Seminal plasma from liquefied samples was collected through centrifugation at 3000 rpm for 7 minutes, and the plasma was stored frozen (-80°C) until use. The concentration of malondialdehyde in seminal plasma was determined using the Enzyme-Linked Immunosorbent Assay (ELISA) technique.

Keywords: Malondialdehyde; Seminal parameters; Infertile males.

INTRODUCTION

Infertility is defined as the inability of a couple to conceive after 12 months of marriage with unprotected sex [1]. A significant proportion of infertility cases can be attributed to male factors. Numerous studies have highlighted the impact of Reactive Oxygen Species (ROS) on seminal parameters such as motility, count, volume, and activity [2]. ROS, free radicals naturally produced in controlled levels within the human body (physiological levels), play essential roles in processes like apoptosis and sperm maturation. Additionally, they function in destroying microbial agents infecting various sites in the human body [3]. Excessive ROS production, however, can have detrimental effects on several cell types, with spermatozoa being particularly vulnerable [4]. Due to their high instability, ROS requires immediate measurement. Free radicals, known for causing peroxidation of unsaturated lipids, target spermatozoa that contain a significant amount of unsaturated lipids in their heads, leading to the production of Malondialdehyde (MDA) [5].

Aim of the Study

This study aims to elucidate the influence of malondialdehyde on semen parameters in infertile males.

MATERIALS AND METHODS

Materials

- General Sperm Analyzer: Motic-China.
- Malondialdehyde: Eliza assay kit-Human Germany (used to detect MDA concentration).

Methods

Sample Collection

- Seventy semen samples were collected from males experiencing either primary or secondary infertility.
- Fertile individuals, who had conceived within the last 12 months or fathered a baby in this period, served as the control group.
- All individuals were instructed to collect semen samples in a wide-mouthed cup after a three-day absence.
- Sample collection occurred over three months, from April 2017 to June 2017.
- Semen quality and sperm parameters were examined using the General Sperm Analyzer (Motic/China).
- Semen samples were centrifuged at 3000rpm for 7 minutes, and seminal plasma was collected and frozen at -80°C until use.

Malondialdehyde Detection

The Eliza technique was employed for MDA detection, following the manufacturer's instructions.

A standard curve for MDA concentration was drawn using the provided standard concentration.

Statistical Analysis

Data tabulation, input, and coding were performed using IBM© SPSS© Statistics Version 22.

RESULTS

The samples were equally divided, with 50% (35 samples) in each of the primary and secondary infertile groups, and 35 samples from fertile males serving as the control group. The mean values and statistical correlations for all parameters are presented in Tables 1 and 2.

Age

The mean age across all groups fell within the anticipated range for this study, although the P-value indicated a significant difference in this regard.

Volume of Seminal Fluid

Table 2 indicated no significant difference in the volumes of seminal fluid among individuals in the study.

Total Sperm Count

The total count of spermatozoa in all three groups fell within the normal range. However, there was a notable difference between the control group and the primary or secondary groups (149.73 ± 87.09, 63.34 ± 61.8, 86.87 ± 88.161 sperm/mL, respectively).

Motility of Spermatozoa

Motility, as presented in Table 1, represented the mean total percentage of motility across all three groups. A clear statistically significant difference was observed between the control group and both infertile groups. Additionally, a difference was noted between the two infertile groups (primary and secondary infertility).

Abnormal Morphology

The study revealed a positive relationship between the ratio of teratozoospermia and male infertility. A significant difference was observed between the control group and other infertility groups.

pH of Seminal Fluid

No significant difference was found in the pH levels among the study groups. The study excluded samples with pH <7.2 or >8.6, neutralizing the potential impact of pH on male infertility.

Liquefaction Time of Seminal Fluid

The mean liquefaction time was within the normal range for all groups. According to WHO criteria, liquefaction time is considered abnormal if it exceeds 60 minutes.

Variables	Controls	Primary Infertility	Secondary Infertility
	Mean ± SD	Mean ± SD	Mean ± SD
Age	29.17 ± 5.5	28.49 ± 4.9	33.09 ± 5.88
Volume	3.23 ± 1.28	3.02 ± 1.21	2.95 ± 1.30
Total sperm	149.73 ± 87.09	63.34 ± 61.8	86.87 ± 88.161
Motility	50.30 ± 8.12	39.17 ± 15.31	29.80 ± 18.02
Abnormal Morphology	53.11 ± 11.69	33.69 ± 14.65	31.78 ± 13.19
PH	7.52 ± 0.22	7.65 ± 0.267	7.6 ± 0.264
Liquefaction Time	26.86 ± 11.76	23.43 ± 12.41	25.79 ± 11.54
Activity	70.22 ± 10.40	50.15 ± 20.26	44.06 ± 17.79
MDA	8.36 ± 2.97	8.13 ± 4.33	9.72 ± 3.66

Table 1: Semen parameters of study groups.

Activity of Spermatozoa

The computerized system used classified sperm activity into four classes (A-D). The mean activity of sperm in the control group was 70%, while it was 50% among primary infertile patients and further decreased to 44% among secondary infertile patients. This suggests a potential correlation between decreased sperm activity and infertility.

Lipid Peroxidation of Sperm Lipid Contents

Malondialdehyde levels showed no significant difference between the control group, primary infertile group, and secondary infertile group. The mean values were (8.36 ± 2.97, 8.13 ± 4.33, 9.72 ± 3.66) mmol/L, respectively.

Variables	Controls *vs.* PI	Controls *vs.* SI	PI *vs.* SI
Age	0.859	**0.01**	**0.002**
Volume	0.766	0.616	0.968
Total sperms count	**0**	**0.004**	0.438
Motility	**0.005**	**0**	**0.021**
Abnormal Morphology	**0**	**0**	0.818
PH	0.09	0.413	0.673
Liquefaction time	0.454	0.925	0.687
Activity	**0**	**0**	0.284
Key: PI: Primary Infertility; SI: Secondary Infertility.			

Table 2: Statistical significance of variables.

Correlation of MDA with Semen Parameters

The study revealed a statistically significant positive correlation of Malondialdehyde (MDA) with abnormal sperm morphology in all groups. This correlation was more pronounced in the primary infertile group. A negative correlation was found between MDA level and active sperm, with variations among the groups, indicating a somewhat controversial impact of MDA on semen parameters.

DISCUSSION

The findings of this study regarding the age of individuals align with recommendations from the American Fertility Society, which suggests that the age limit for good sperm quality is 50 years or younger [6-8]. While there is no specified age limit for male fertility in Iraq, this study observed no observable effect of age on male fertility at this stage.

No significant effect was noted on the volume of semen samples across all groups, and the results fell within the normal range defined by the World Health Organization (WHO) [9].

Although a clear difference in total sperm count was observed between fertile and infertile men, this difference did not appear to have an obvious impact on male fertility. Infertile men could still exhibit a normal sperm count according to the WHO report [9].

The interpretation of motility as a cause of infertility is deemed unfair in this study, as various factors, such as deficiencies in ions (calcium, magnesium, iron, vitamin D&C), and temperature fluctuations, may influence sperm motility [4, 10]. Additionally, the abnormality of sperm in all research groups did not exceed the acceptable ratio defined by the WHO, which considers a person infertile when the percentage

of normal sperm is less than 4% [1].

The liquefaction time of seminal fluid appeared to play no role in evaluating male infertility based on the present results and previous findings [11].

The study results on Malondialdehyde (MDA) concentration indicated similar activity in all study groups, suggesting that these groups may be under similar conditions. However, the correlation of MDA with semen parameters yielded conflicting results. While a positive effect with semen abnormality was observed in all groups, the impact was more pronounced among primary infertile individuals. A negative correlation was recorded between MDA concentration and sperm activity in the primary infertile group, while a positive correlation was noted in the control and secondary infertile groups. This inconsistency makes the influence of malondialdehyde on male infertility controversial [12].

CONCLUSION

In conclusion, infertility is a complex, multi-factorial process, and no single parameter can be considered a definitive cause of male infertility, whether primary or secondary. The study suggests that Malondialdehyde (MDA) does not have a significant effect as a factor for male infertility. The interplay of various factors and the complexity of male reproductive physiology highlight the need for comprehensive assessments when investigating male infertility.

RECOMMENDATIONSCONFLICT OF INTEREST

No conflicts of interest have been declared.

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