

Clinical Features and Sperm Count in a Sample of Males with Infertility: Study in Baghdad

Hassan Abdulreza Fayyad¹, Hind A. Abdulghafoor² and Noor Hani Al-Naji³

¹Imam Alkadhim University College, Baghdad, Iraq.

²Department of Pathology and Forensic Medicine, University of Fallujah, Iraq.

³Biology Department, Faculty of Science, University of Kufa, Najaf, Iraq.

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***Corresponding Author:** Hassan Abdulreza Fayyad, Imam Alkadhim University College, Baghdad, Iraq.

Abstract

Background: One of the global health problems that affects individuals is infertility, and it also affects different aspects of life. The current study was conducted to investigate the relationship of some factors with male infertility.

Methods and Results: The current study included 45 males with a mean age of 32.42 years and an age range of 19 to 50 years, to conduct sperm count examination, as they were present in a private clinic in Baghdad during the period from October 2022 to March 2023. They were asked to provide information about their age, occupation, medical history, and family history of infertility. The results obtained were that there was a large number of cases (70.6%, 29/45) with low sperm count, as more than a third of them had zero sperm count, as they had a history of mumps infection early in their lives, which is one of the factors causing infertility in males. Other health problems such as chronic asthma, testicular pain, undescended testicles and typhus were also reported at a higher frequency among the males studied who had infertility problems. The results of antisperm antibodies in spermMar test were significantly positive ($p > 0.0001$) in the patient group compared to the control group.

Conclusion: We found that lifestyle (occupation), some diseases and age have an effect on male fertility. The incidence of ASA in individuals with infertility was 8.2%. and about 50% in individuals with ASA-positive. The presence of acetic acid was associated with poor sperm parameters, but it did not affect pregnancy rates after assisted reproductive techniques.

Keyword: Male infertility, Semen quality, anti-sperm antibodies.

1. INTRODUCTION

Infertility is a health issue been evident when pregnancy could not be achieved after one year or more of regular unprotected sexual contact. This can influence different aspects of the affected individual emotionally, clinically and financially. In communities like ours with a strong emphasis on childbearing, the burden of infertility becomes more evident (1). It is estimated that approximately 10 % of the couples have infertility problems.

This has a negative impact on the couple's lives especially in communities where socioeconomic traditions and religious have imposed on married individuals to have children. In cases of primary male's infertility, an initial lack of the ability to impregnate of a woman is the main feature of this type of infertility. Secondary male's infertility is characterized by the loss of impregnation of a male after being able to impregnate a woman for some time of his life. Additionally, the reduction of the number, motility and viability of spermatozoa as well as abnormal sperm morphology could lead to either partial or complete infertility (2, 3).

A number of factors have been reported to be associated with an increased risk of male infertility (4). One such factor is the presence of antisperm antibodies (ASA). ASA are immunoglobulins directed

against sperm surface antigens (5). They can affect male fertility in several ways, causing abnormalities in conventional semen analysis or impaired function. The most common effects of ASA are poor sperm motility, sperm clumping, poor penetration of cervical mucus, poor capacitance, changes in acrosome reactivity, and disruption of sperm-egg interactions. (6). These risk factors have been divided into genetic and non-genetic factors (7, 8, 9). The genetic factors refer to the genomic instability that caused by numerical (aneuploidy) and structural (deletion, inversion and translocation) chromosomal aberrations which have been reported to be linked to men's sterility (10, 11). While non-genetic male's infertility-associated factors include life style such as tobacco smoking (12), drugs and alcohol consumption (13, 14), and extensive use of mobile phone (14). In addition to the exposure to heavy metals (15, 16) pesticides. The present study was designed to understand the role of age, jobs, some disease and antisperm antibodies (ASA) on fertility.

The following approaches will be adopted to achieve such aims:

1. Contact a semen analysis to evaluate sperm count.
2. Determine the ASA in semen samples.

2. METHOD

2.1. Study Population

The present study included 45 males, with a mean age of 32.42 years and an age range of 19 to 50 years. For sperm count examination, they attended a private clinic in Baghdad, Iraq, during the period from October 2022 to March 2023. They were asked to provide information about their age, occupation, medical history, and family history of infertility.

2.2. Semen Samples

2.2.1. Semen Collection

Fresh human semen samples were obtained by masturbation from males and collected directly into clean, dry and sterile disposable plastic Petri dishes in a dedicated room. Subjects had a known abstinence period of 3–5 days, and samples were immediately transferred for semen analysis after 30–60 minutes. Each semen sample was allowed to liquefy according to methods described by the World Health Organization (1999 and 2010). After complete liquefaction, semen was analysed by microscopic and macroscopic examination using the WHO 1999 and 2010 criteria (17).

2.2.2. Semen Analysis

Semen assessment was conducted according to the WHO recommended protocol (17). This analysis adds to determine the sperm count and dysfunctionality. Briefly, semen sample mixed thoroughly and aliquoted using Unopette capillary tube on a hemocytometer slide. Each semen sample was assessed within 15–20 mins after it loaded on the hemocytometer slide. The sperm count of consistent duplicate was used to calculate the spermatozoa's concentration per ml and the total number of sperm per ejaculate accordingly. Briefly, semen sample mixed thoroughly and aliquoted using Unopette capillary tube on a hemocytometer slide. Each semen sample was assessed within 15–20 mins after it loaded on the hemocytometer slide to avoid evaporation. The sperm count of consistent duplicate was used to calculate the spermatozoa's concentration per ml and the total number of sperm per ejaculate accordingly. Sperm's activity was estimated by Haemocytometer account for sperm.

2.3. Sperm Mar Test

2.3.1. Direct Sperm Test for IgA Antibodies

First, the reagents and samples were allowed to acclimatise to room temperature. Then, ten microliters of fresh semen and SpermMar test IgA latex particles were placed on a small slide, then a cover glass was placed on the mixture, and then the mixture was observed under a light microscope.

After 3 minutes, the result was read, where latex particles were observed attached to the motile sperm, and then 100 sperm cells were counted to determine the percentage of reactive sperm.

After 10 minutes, the results were read again, where the diagnosis of immunological infertility was suspected, as 10–39% of the motile sperm were attached to the latex particles; if 40% or more of the sperm were attached, immunological infertility was highly likely.

2.3.2. Direct SpermMar Test for IgG Antibodies

First, the reagents and samples were allowed to acclimatize to room temperature. Then ten microliters of fresh semen and SpermMar IgG latex particles were placed on a microslide, a cover glass was placed on the mixture, and the mixture was observed under a light microscope.

After 2-3 minutes, the result was read, where latex particles were observed adhering to the motile sperm, then 100 sperm cells were counted to determine the percentage of reactive sperm.

After 10 minutes, the results were read again, if the presence of anti-sperm antibodies reacting to the antigen(s) on the sperm was $\leq 10\%$, the sample was included in the study, if the percentage of clumping was more than this percentage, it was considered immunologically infertile.

3. STATISTICAL ANALYSIS

The excel software was used for presenting the data in figures and performing basic statistical analysis. The later included the calculation of mean, range and the means differences using T-test.

4. RESULTS

This pilot study was set with the aim of understanding some of the clinical features of males with infertility. The examined seminal fluid samples (n=45) were collected from males attended a private clinic in Baghdad- Iraq for their sperm quantity and quality test.

The results showed that the vast majority of the cases (70.6%, 29/45) had lower sperm count (lower than 20 million) . Interestingly, 39 %(16/45) of these cases had zero sperm count (Fig 1). Almost all of the cases have shown a significant number of inactive sperms (0-40%), **table (1)**.

Table 1. Age distribution and sperm count of the studied males with fertility problems.

Feature(s)	Mean	Range
Age	32.42 years	19-50 years
Sperm count	15.6 million	0-112 million
Sperm activity	23.43%	0-60%

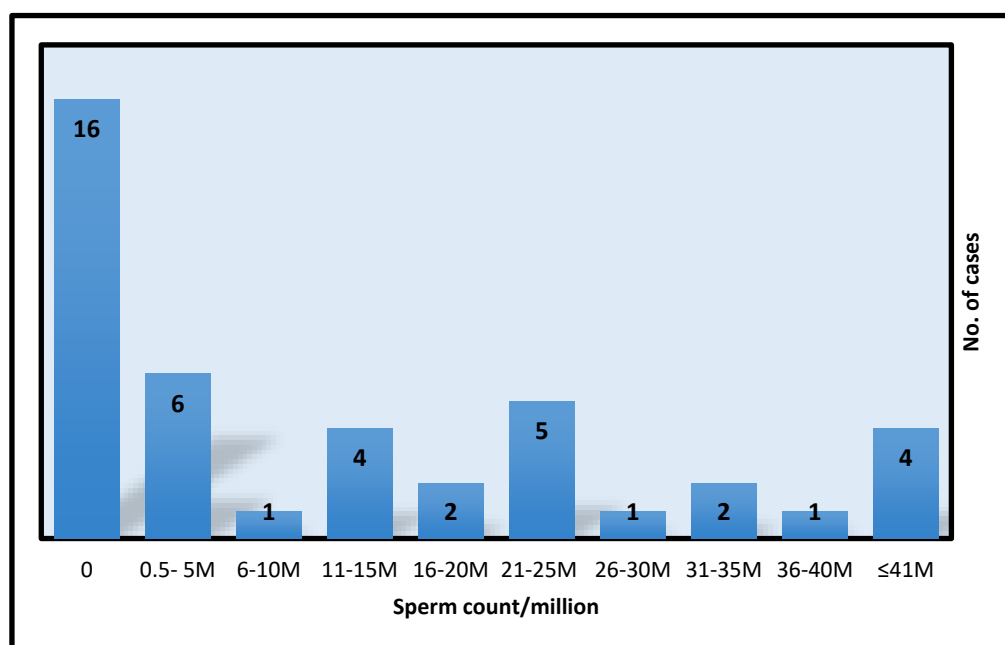
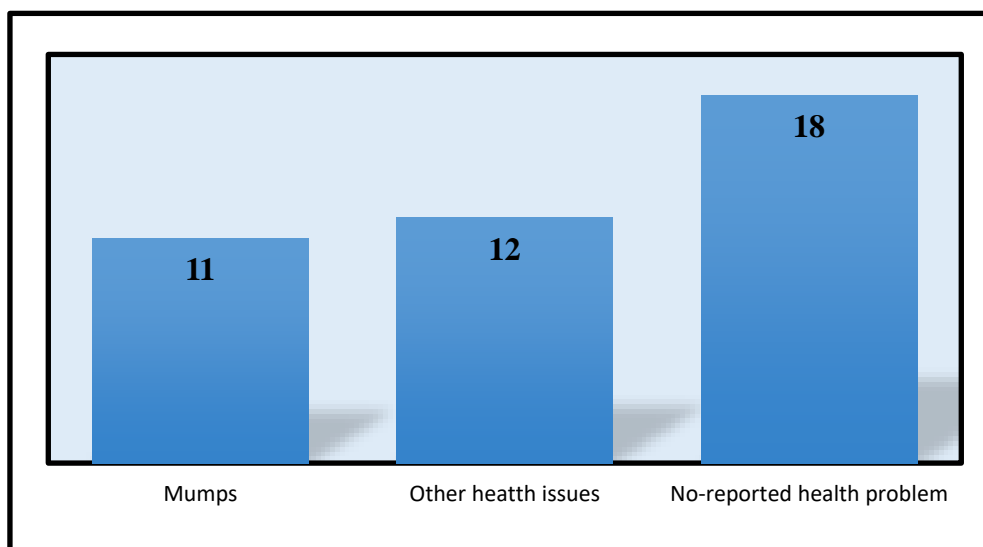


Figure 1. Sperm count of the studied samples

Looking at the risk factors associated with male's infertility, a slightly higher than a quarter of the studied cases (26.83%, 11/45) have a history of mumps viral infection. A similar proportion (29.27, 12/45) of the investigated males reported to have other health issues such as chronic asthma, testes pain, undescended testes and typhus. While the rest of the examined cases (43.9%, 18/46) were not having apparent health problems to be declared shown in **table (2)**.

Table 2. Health conditions might associate with male's infertility.

Health condition	No. (%)	Mean of sperm count	T-test
Mumps	11(26.83)	10.22	N.S.D.
Other health issues	12(29.27)	24.4	
No reported health issue	18 (43.9)	13.9	


Figure 2. The incidence of health issues reported to be associated with male's infertility in the studied cases.

In respect to the occupational impact on male's infertility, our pilot study results showed that approximately more than half of the studied males reported that they served in chemical/ constructing industries, military and cooking jobs **table (3)**.

Table 3. Job categories and sperm count of the studied cases

Job category	No. (%)	Mean of sperm count(million)
Chemical industries	13/41 (31.7)	22.20714
Military/police	9/41 (21.95)	12.64286
Other jobs	19/41 (46.34)	12.64286

The results of antisperm antibody (ASA) test showed in **table (4)**, the patients group highly significantly ($p > 0.0001$) positive compared with the control group.

Table 4. SpermMar test of the studied groups

Men group	IgA	IgG
Control group	100% negative	100% negative
Patients group	60% positive 40% negative	10% positive 90% negative
P value	0.0001	

5. DISCUSSION

One of the major health problems that affects a large number of individuals all over the world is infertility. This issue has an impact on wide health aspects of the affected individual's. Of interest, male's infertility has been reported to account for approximately 30% of the overall burden of infertility (18). The present study was set to investigate the potential causes might associate with male infertility in subjects attended private clinic in Baghdad-Iraq for their seminal examination.

The results obtained were that there was a large number of cases (70.6%, 29/45) with low sperm count, as more than a third of them had zero sperm count, as they had a history of mumps infection early in their lives, which is one of the factors causing infertility in males.. This is due to the significant number (26.83%, 11/45) of cases that reported to have mumps infection early in their life. This finding, if it confirmed by large scale studies, should draw the attention to the necessity of evaluation the efficiency of mumps vaccinations in early childhood. In this scenario further efforts are required to develop efficient and affordable mumps vaccine. There were also health problems with higher frequencies among males with infertility problems such as chronic asthma,

testicular pain, and typhus. These illnesses should also be considered in the strategies of tackling male's infertility problem. (19,20) In respect to the occupational impact on male's infertility, our pilot study results showed that approximately more than half of the half of the studied males reported that they served in heavy /chemical/ constructing industries, military and cooking job. In general, the current study is consistent with previous studies regarding reported risk factors associated with male infertility. (21,22).

The ASA result in the present study was very positive when compared to the control group. However, according to the current WHO guidelines (2010), two different types of ASA have been recognized; immunoglobulin A (IgA) and immunoglobulin G (IgG). SpermMAR and direct immunoglobulin bead tests are conventional techniques for determining the presence of ASA. SpermMAR test for $\geq 50\%$ motile sperm with adherent particles is positive (23).

When the blood-testis barrier is disrupted, the immune system can be exposed to sperm antigens (24). An immune response to ASA production can also occur when orchitis occurs, or mechanical injury to the testicle such as vasectomy or tubal ligation can subsequently damage the vas deferens leading to a negative impact on sperm quality (25). Despite the associations observed so far, the cause of positive ASA in subfertile men is not fully known. Overall, the present pilot study concluded that male infertility is a serious health problem affecting a wide range of people of all ages. Mumps viral infection and other health problems such as chronic asthma, testicular pain, undescended testicles, and typhus appear to influence this health problem. People serving in certain occupations such as chemical industries, construction, military, and cooking may be at increased risk of male infertility. An immune response including the production of ASA has also been observed. Therefore, large scale studies are recommended to establish such potential association between males' infertility and above mentioned risk factors.

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