THE ROLE OF BACTERIAL INFECTIONS ON MALE INFERTILITY IN AL-ANBAR PROVINCE OF IRAQ

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ABSTRACT
Background: Bacterial infections on male infertility has always been in the field of debate due to scarce analysis tools to examine seminal fluid specimens as a result of which these infectious processes leads to deterioration of spermatogenesis, impairment of sperm function and/or obstruction of the seminal tract.
Aims & Objective: In the current study we investigated the role of bacterial infections in male factor infertility in Al-Anbar Province, West of Iraq through detection of abnormal sperms and other factor pertains to male infertility.
Material and Methods: Seminal fluid from six hundred volunteer males was investigated for infertility by the detection of abnormal sperms using the WLJY-9000 TYPE WEILI Color Sperm Analysis System and the Neubauer counting chamber.
Results: From the six hundreds patients investigated for infertility, it was found that 408 (68%) patients had a positive culture for pathogenic bacteria, of different species. The results indicate that 32.0% had sperm density less than twenty million per millilitre. The oligospermic were 23.0%, severe oligospermic 0.17% and Azoospermia 8.83%. Asthenospermia was reported to be 76.33% and Teratospermia 86.16% respectively.
Conclusion: Seminal fluid infection increases with decreasing sperm density, motility and morphology. The prevalence of abnormal sperm indices and bacterial infection is high with Klebsiella spp. infection. Hence, treatment measures should be taken properly in the management of male factor infertility.
Key-Words: WLJY-9000 TYPE WEILI Color Sperm Analysis System; Seminal Fluid; Male Infertility; Bacterial Infection

Introduction
Role of bacterial infections on male infertility has always been in the arena of controversy due to lack of decisive analysis tools to examine seminal fluid specimens as a result of which these infectious processes leads to deterioration of spermatogenesis, impairment of sperm function and/or obstruction of the seminal tract.11 The associated problem related to male infertility is further elevated due to different populations studied and to the fact that leukocyte subtypes in semen may have different functions. Besides this, it is believed leukocytes may even have protective effects on spermatozoa. It has been reported that detection of bacteria in semen does not necessarily suggest infection since bacteria isolates in seminal fluid may represent contamination, colonization of the urethral orifice or infection.12 Among these infections, the most frequently isolated organisms in industrialized countries are Chlamydia trachomatis, Ureaplasma urealyticum and Enterobacteria as they also account for their influence due to sexual transmission resulting in tubal disease and subsequent infertility in the female partner rather than a direct influence on male reproductive functions. Reports obtained from eastern and southern parts of Nigeria stated that oligospermia and azoospermia are the common causes of male factor infertility which has been attributed to bacterial infections.3,4

However, according to the World Health Organization, 2010, seminal fluid infection was defined as the presence of significant bacteriospermia (≥10³ bacteria/ml ejaculate), which includes the detection of Neisseria gonorrhoeae, Chlamydia trachomatis, Ureaplasma urealyticum and significant leukocytospermia (10⁶ peroxidase positive leukocyte/ml ejaculate).5 It therefore follows that if some or all the conditions above are not met, the isolation of bacteria in semen are often regarded as contaminants by most increase.6 In this investigation we aimed at determining the prevalence and role of bacterial infection in male factor infertility in Al-Anbar Province, West of Iraq.

Materials and Methods
Six hundred seminal fluid specimens from men investigated for infertility over a period of 10 months were analyzed. These were seminal fluids of patients referred to the Al-Gailani Central Medical Laboratory-Ramadi, Al-Anbar province, Iraq from the infertility clinics. The
specimens were collected either by self or assisted masturbation into sterile bottle. Collection of the specimens and their transport to the laboratory within half an hour of production was done as reported earlier[7]. Briefly, they were told to first pass urine and then wash their hands and penis with soap, then rinse with water prior to masturbation and ejaculation into sterile container[8]. These men collected seminal fluid after they had abstained from coitus for at least 3 days. The semen was then cultured on Blood agar, Chocolate agar and MacConkey agar media and incubated for 24-48 hours at 37°C. The infective organisms were identified using Gram staining technique, biochemical reactions and sensitivity to a range of antibiotics[9,10]. The analysis was done using the WLJY-9000 TYPE WEILI Color Sperm Analysis by medical electronic system and a standard Neubauer counting chamber. The seminal fluid specimens were diluted 1:20 with 1% formalin and the spermatozoa counted under the microscope using 40X objective in Neubauer counting chamber. WLJY-9000 TYPE WEILI Color Sperm Analysis System was used to calculate the percentage of actively motile sperms, density, and the percentage of abnormal forms and to evaluate the presence or absence of pus cells[11]. All the experiments were performed in triplicate.

Results

Out of Six hundred seminal fluid specimens collected from men, 583 (97.16%) were reported to be infertile with abnormal seminal fluid sperm density, motility and morphology, while 17 (2.84%) of the patients showed seminal fluid cell density above twenty million/ ml (Figure 1). Data obtained from percentage of types of sperm density (Figure 2) revealed Oligospermic (23%), severe Oligospermic (0.17%) and Azoospermia (8.83%). Observation of the bacterial infection on sperm motility and morphology showed that Asthenospermia (motility less than 50%) 76.33% (Figure 3) and Teratospermia (abnormal morphology greater than 50%) 86.16% (Figure 4) respectively. Seminal fluid infection increases with decreasing sperm density, motility and morphology as above. Based on the biochemical reactions and antibiotic sensitivity tests it was found that 408 (68%) patients were positive for pathogenic bacteria (Figure 5). Among them, Klebsiella spp. was detected in 218 (37.39%) infected seminal fluids, Staphylococcus aureus was detected in 69 (11.83%), Escherichia coli was detected 65 (11.14%), Streptococcus pyogenes in 53 (9.09%), while Neisseria gonorrhoeae, Pseudomonas spp and Proteus vulgaris were also reported.
Discussion

Infertility is considered one of the main public health issues, as it affects about 15% of the couples of reproductive age. The male factor is involved in 40% - 50% of infertility cases. In 1992 a global decline in sperm density was reported; this was quickly followed by numerous critiques and editorials. Since the importance of this finding has direct influence on the public health, a detailed reanalysis of data was warranted to resolve these issues as reported by Swan et al, 1997. They also reported that the usage of multiple linear regression models (controlling for abstinence time, age, percent proven fertility, specimen collection method, study goal and location) were used to examine regional differences and the interaction between region (United States, Europe, and non-Western countries) and year. Nonlinear models and residual confounding were also examined in these data. In this study we showed that the computer analyses of semen have clinical significance in the diagnosis of infertile disease to substitute the routine skilled hand worked method. Onwudiegwu and Bako, 1993 during their studies have showed that the gross male contribution to infertility based on sperm density was 46%. Twelve per cent of the patients were azoospermic. Sperm quality was generally poor; 42.1% (128 patients) had poor sperm motility and in 43.8% (133 patients) there was significant abnormal sperm morphology. There was also a high incidence of necrospermia (53.3 %) occasioned by genital tract infections. Seminal fluid analysis is a generally accepted method of assessing male fertility potential. Macleod and Gold 1951, suggested that men with sperm counts above 20 million/ml or total count above 100 million per ejaculate should be considered fertile. Other investigators have revealed that sperm counts above 10 million or 25 million per ejaculate should be considered normal provided other parameters such as motility and morphology are normal. The concept of a minimal sperm count adequate for fertility has generated a lot of arguments since it was introduced in the 1920’s. It has been demonstrated that pregnancies was achieved by normal males who had spermatogenesis suppressed to about one million per millilitre as part of male contraceptive study. Seminal fluid infection contributed in no small measure to reduced sperm density, Asthenospermia and Teratospermia (abnormal sperm morphology of greater than 50%). Okon et al, 2005 in Africa, isolated S. aureus from 62.5% of the seminal fluids and reported it as a causative organism for 68.2% of seminal fluid infections. Most practitioners dismiss this infection as mere contamination which is assumed to be of no significance. The WHO definition of seminal tract infection does not clearly differentiate between infection, contamination and colonization of the genital tract. Semen that passes through the genital tract is routinely contaminated with Gram positive cocci such as Staphylococcus, Streptococcus and Diphtheroids. It is generally accepted that S. aureus which are coagulase positive is regarded as pathogenic and needs special attention as its persistence can cause damage and loss of germ cells. According to Bukharin et al, 2000 opportunistic microorganisms cause classical infections of the urogenital tract and sub clinical reproductive tract infections. Our result shows that 32% of the males had sperm density below 20 million/ml. Many bacterial species were considered as contaminants (coagulase negative Staphylococi, alpha-haemolytic Streptococci). These infections of the seminal fluid lead to decrease in the number of spermatozoa, the suppression of their motility, changes in their morphology and fertilizing capacity. The prevalence of abnormal sperm indices and bacterial infection is high with Klebsiella spp. infection which should be treated and no longer ignored in the management of male factor infertility.

Conclusion

This analysis demonstrated that the decline in sperm density is not likely to be an artifact of bias, perplexing or statistical analysis but based on modern computer and advanced image processing techniques for clinical test of sperm quality. The system has the ability to analyze the characters of sperm motions comprehensively through image processing of sperms in their dynamic or static status. Hence, it is strongly believed that special attention should be paid to sperm
quality in the overall assessment of semen analysis and routine semen culture, also sensitivity tests should be an integral part of semen analysis in infertility clinics.

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References