Role of Semen Culture in Infertility

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Summary

This prospective study was undertaken at Manipal Assisted Reproduction Centre (MARC) - the infertility centre of Kasturba Medical College, Manipal, to evaluate the relationship between seminal fluid analysis and seminal fluid microorganism in specimens with no significant evidence of pus cells. A total of 67 patients were studied. Fifty one (76%) patients did not show any significant evidence of bacteria in culture (Group I) and interestingly 16 (24%) patients showed significant evidence of bacteria in culture (Group II). There was significant difference in count (Group I – 52.63±37.78) & group II – 34.19±35.22 Million/ml) ($p<0.03$), motility (group I – 61.39±16.7) & group II – 54.25±10.13%) ($P<0.02$) and velocity (group I – 41.18±17.40 & group II – 36.75±9.94%) ($p<0.02$) between the two groups. Hence we conclude that the mere absence of WBC's in semen does not guarantee the absence of infection thus occurrence of silent subclinical infection of the semen could be one of the cause of male infertility.

Introduction

Infection of reproductive tract is a global problem and is estimated that about 250 million new cases get infected annually. The female reproductive tract is easily susceptible to microbial infection unlike the male where microbial colonisation is uncommon, therefore the clinical relevance of detection of bacteria in semen from exhibiting no overt signs of infection has remained unclear. However the incidence of presence of bacteria is higher in semen from infertile men as compared to fertile men. These observations have led to the view that silent subclinical infection of reproductive tract could be one of the cause of male infertility. However reports of correlation between infection and abnormal sperm count and motility have been conflicting. Nassens et al 1986 reported that the presence of aerobic and anaerobic bacteria in semen did not correlate with abnormal sperm motility, sperm count and morphologic features. Others have suggested that the presence of microorganisms in the seminal fluid does not effect male fertility as measured by sperm count or morphology (McGowan et al 1980). The purpose of our study was to investigate the association of bacteria without significant pus cells in semen with seminal fluid analysis.

Objective: To assess the relationship between seminal fluid microorganisms and seminal fluid analysis in a specimen with no significant evidence of pus cells.

Materials and methods

In this prospective study, undertaken at Manipal Assisted Reproduction Centre, Dept. of Obstetrics & Gynaecology, Kasturba Medical College, Manipal, during a 3 month period between March 1997 and May 1997.

Sixty seven (67) patients, attending Manipal Assisted Reproduction Centre for intertility treatment, were entered in this study. The criteria of exclusion were, patients with medical illness in the past three months or who had received antibiotics for past two weeks or presence of sexually transmitted disease. These men were instructed to abstain from ejaculation for 3 days before the visit to the hospital. After cleansing the hands and penis with bactericidal soap, semen samples were obtained by masturbation into a sterile container. The men were not allowed to use lubricants during collection of semen.

Within five minutes of collection, 0.5ml of semen was removed for microbiological examination. The remainder was allowed to liquefy for 20 minutes and was examined for WBC's, sperm concentration, motility, and velocity. The sample which was taken for microbiological examination was incubated and looked for both aerobic and anaerobic growth after 48 hours.

The association between evidence of infection in the semen and its relation to sperm count and motility was critically analyzed. Patients were divided into two groups. Group I consisted of men with sterile semen culture and Group II with semen culture showing significant infection.

Results

A total of 67 patients were studied. In Group I, 51 (76%) patients showed no evidence of infection and had a sterile semen culture. In group II (24%) patient had evidence of infection and had a count of 10^5 colonies (Table I).

Table I
Status of patients (n=67)

Total patients	Group I	Group II
67	51 (76%)	16(24%)

Table II shows the various types of microorganisms (bacteria) isolated by culturing the semen samples. E. coli were grown in 31.25% cases. Citrobacter, Pseudomonas, and Coagulase positive Staphylococci each were grown in 18.75% cases. In 12.15% men Klebsella were isolated (Table II).

Table II
Status of various microorganisms isolated from semen samples

	Type of organism	Percentage
1.	E.coli	31.25%
2.	Citrobacter	18.75%
3.	Psuedomonas	18.75%
4.	Coagulase positive Staph.	18.75%
5.	Klebsella	12.15%

This table shows the comparison of various semen parameters between the two groups. Patients in group II showed a decrease in sperm count (51.19 ± 35.22 million/ml), average motility (54.25 ± 10.23%), and grade III motility or velocity (36.75 ± 09.94%) when compared to group I (52.65±37.78, 61.39±16.7%, and 41.18±17.40% respectively), showing a correlation between infection and semen parameters ($p<0.05$). This indicates that inspite of WBC being less than 5 HPF in the semen (WBC/HPF in group I = 1.90±1.53 and in group II = 2.25±01.1), a subclinical infection may be present which can be detected by performing the culture studies (Table -III).

Table III
Comparison of semen parameters in two groups

	Group I (n=51)	Group II (n=16)	p Value
Count (in millions/ml)	52.65 ± 37.78	51.19 ± 35.22	p<0.03
Average motility	61.39 ± 16.7%	54.25 ± 10.23%	p<0.05
Grade 3 motility	41.18 ± 17.40%	36.75 ± 09.94%	p<0.02
WBC / HPF	1.90 ± 1.53	2.25 ± 01.1	p<0.57

Discussion

Aerobic as well as anaerobic bacteria have been isolated from semen (Hillier et al, 1990). Many species of bacteria are frequently isolated from semen but none of them appear to be unique to semen. Most of the bacterial species isolated are the normal inhabitants of the skin, GIT or the vagina. Bacterial species which are normally present in one part of the body can act as a pathogen in another. The normal bacterial flora in semen is not very well defined. The bacterial flora on external genitalia and in semen were different when studied simultaneously in the same individual suggesting that the bacteria isolated from semen are not mere contaminants (Lewis et al, 1981).

Presence of bacteria in semen is associated with a high incidence of immotile or sluggishly motile and morphologically abnormal sperms (Wolff et al, 1990) but other investigators observed that the presence of microorganism in seminal fluid is not associated with decreased motility or concentration (Nassens et al, 1986, Mackler et al, 1981). Presence of bacteria is not always accompanied by leukocytic infiltration especially when an individual does not exhibit signs of an infection, therefore there is no correlation between the number of leukocytes observed in semen and presence of bacteria in semen. Interestingly we observed that none of the patients had significant WBC's in semen but showed significant bacterial growth in 24% of the cases. The bacteria isolated in our study are Escherischia coli (31.25%), Pseudomonas (18.75%), Citrobacter (18.75%), and Klebsella (12.5%).

In this current study we observed a significant reduction in the average motility of sperms in 28% cases ($p<0.02$) and velocity in 10% patients ($p<0.05$) in the presence of bacteria in semen showing the statistical significance. Hence, we suggest that the male patients who have decreased sperm motility and velocity must undergo culture studies to detect the presence of bacterial infection. It may be of much significance to those who use donor sperms for intrauterine insemination (IUI) and sperm for in vitro fertilization (IVF) procedures as it may lead to infection to the recipient or may have decreased embryo cleavage.

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