Semen analysis parameters: Experiences and insight into male infertility at a tertiary care hospital in Punjab

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Abstract

Objective: To determine the prevalence of low sperm count including oligospermia and azoospermia in male infertile population, and to assess the pattern and distribution of abnormal semen parameters in infertile men.

Methods: The descriptive cross-sectional survey was carried out at the Department of Gynaecology and Obstetrics, Sharif Medical City Hospital, Lahore, from June 2009 to June 2010. A total of 500 consecutively consenting male partners of women fulfilling the inclusion criteria between 20 and 40 years of age were approached. Semen analysis was performed according to methods and standards defined by the World Health Organisation (WHO). Samples were categorised into normospermia, oligospermia and azoospermia on the basis of sperm count. After exclusion of azoospermic samples, normospermic and oligospermic samples were compared for ejaculated volume, pus cells, motility and morphology. SPSS 10 was used for statistical analysis.

Results: Out of the 500 males approached, 104 (20.8%) had to be left out either because of their unwillingness or inability to pass semen. The study sample comprised of 396 (response rate 79.2%); normospermia was observed in 293 (73.99%) males, azoospermia in 59 (14.89%), and oligospermia in 44 (11.11%). The oligospermic samples had low ejaculated volume, but significantly higher percentage of non-motile sperms 62%±23.9% and abnormal morphology 55%±15.6% in comparison to normospermic samples (p 0.0001). Asthenospermia was observed in 37 (25.81%), teratospermia in 11 (3.26%) and oligoasthenoteratospermia in 4 (9.09%) of samples.

Conclusion: Semen analysis is the cornerstone for the evaluation of infertility in men. Sperm concentration, motility and morphology are related to each other, factors that cause deterioration of one of them usually also have negative impact on the other two as well.

Keywords: Sperm, Motility, Morphology. (JPMA 63: 558; 2013)

Introduction

Approximately about 10-15% of couples suffer from infertility all over the world. Female factor is responsible in 35% and male factor in 45% of cases while the rest of the couples either have combination of factors or unexplained infertility. In more than 90% of cases male infertility is due to either low sperm count or poor semen quality or combination of the two. Recent data confirm the decline in semen quality and quantity all over the world probably due to increased prevalence of sexually transmitted diseases (STDs) and urogenital infections. A report in 1992 alarmed the world about the problem and led others to investigate the phenomenon. In our population such a decline has not yet been reported and the reported incidence of oligospermia is around 21% and azoospermia, 12.32%. In our population where infertility is considered a female issue, and husbands are not willing to go for too many tests, screening by semen analysis provides us with a baseline before going for extensive investigation.

This cross-sectional study aimed at finding the prevalence of low sperm count in the population and abnormal semen parameters in semen.

Methodology

The study was carried out at the Department of
Gynaecology and Obstetrics, Sharif Medical City Hospital, Lahore, from June 2009 to June 2010. A total of 396 consecutively consenting male partners of women attending the fertility clinic of the hospital between the ages of 20 and 40 years were recruited. Detailed history was taken from the male partners regarding age, duration of marriage, occupation, sexual history, infertility (primary or secondary infertility), first or second marriage, drugs, surgical and medical history for any illness. These men were referred for semen analysis to the lab as part of male infertility workup. Males excluded from study were those who were unable to pass specimen by masturbation and those who did not consent.

Detailed instructions were given before the collection of samples. These included abstinence from coitus for 3-4 days; samples were collected aseptically by masturbation into sterile wide-mouthed bottles within hospital premises or at home and delivered to the hospital within 1 hour of collection.

Semen analysis was performed according to the methods and standards outlined by the World Health Organisation (WHO). Parameters outlined included: Appearance: grey/opalescent; Volume: 2.0ml or more; PH: 7.2-7.8; Sperm concentration: >15x10^6 spermatozoa/ml; Total sperm count: 39x10^6 per ejaculate or more; Motility: 50% or more with forward progression; Morphology: 4% or more with normal form; White cell count: <1x10^6/ml.

Entire sample analysis was done by the same lab technician to avoid inter-laboratory variation. Semen analysis was performed within 60 minutes of their collection for volume, appearance, pH, liquefaction, concentration, motility, morphology, viability and the presence of pus cells. Semen volume was measured with a graduated disposable pipette; pH was checked with the help of pH paper. After liquefaction, the semen specimen was thoroughly mixed with the help of a pipette and a thin drop was spread on a glass slide by placing a cover slip on it. Sperm motility was assessed by microscopy appraisal of 200 spermatozoa from different fields. Counting of spermatozoa was done using Meckler's counting chamber. Semen samples were categorised on the basis of sperm count per milliliter of semen in accordance with WHO normal and pathological ranges i.e. normospermia (normal sperm count), oligospermia, and azoospermia. The samples categorised were compared for ejaculated volume, pus cells, motility and morphology (main outcome measures).

The following operational definitions were used:

Normospermia: Sperm count 20 million/ml to 120 million/ml; Oligospermia: Sperm count below 20 million/ml; Azoospermia: Absence of spermatozoa in the ejaculation; Astheno-spermia: Reduced sperm motility; Terato-zoospermia: Abnormal sperm morphology; Oligo-astheno-terato-spermia: All sperm variables abnormal; Hypospermia: Volume <2ml; and Hyperspermia: Volume >5ml.

The data was analysed using SPSS version 10. Mean ± Standard deviation (SD) were calculated for sperm count, volume, pus cells, motility and morphology; 95% Confidence interval was calculated for proportions and for means. Mean values were compared for statistical significance using t-value with level of significance <0.05 (p value).

**Results**

Of the total, 500 males who were approached, only 396 (79.2%) were willing to give semen sample for analysis, while 104 (20.8%) either did not consent or were unable to pass specimen. Among the 396 males, the mean age was 30±4.77 years; 260 (65%) males had primary infertility, and 136 (35%) suffered from secondary infertility. Mean duration of infertility was 5.136±4.60 years.

Using WHO standard for semen normality, 396 samples that were analyzed, normal sperm count (normospermia) was observed in 293 males (73.99%) (Table-1). The distribution of semen volume is shown in Table-2.

After excluding 59 samples with azoospermia, semen parameters were compared in oligospermic and normospermic samples for volume, pus cell, motility and morphology. The oligospermic samples had significantly higher percentage of non-motile sperms 62%±23.9%
and abnormal morphology 55%±15.6% compared to normospermia in which non-motile sperms were 43%±18%, and abnormal morphology was 35%±14.5% respectively (p <0.0001).

Comparison of volume showed mean volume of 2.97±1.35ml in normospermia vs 2.05±2.00ml in oligospermia (p <0.0001), and pus cells 10.80±7.69 in normospermia versus 11.16±10.16 in oligospermia. This was not statistically significant (p <0.782) (Table-3).

Normal motility was observed in 57%±18.3% of normospermic vs 38%±23% of oligospermic samples, and normal morphology of sperms was observed in 65%±14.5% of normospermic vs 45%±15.6% of oligospermic samples (p < 0.0001).

Proportion of multiple factor abnormalities defects is given in Table-4.

**Discussion**

Infertility is a subject of debate and females always remain a target of society for this problem, but advancing knowledge and development of assisted reproductive techniques prove males to be an equal contributor to this problem. Screening of males by semen analysis provides some insight about the underlying pathological problems occurring in the male genital tract.

As high as 90% of male infertility problems are related to count and there is a positive association between abnormal semen parameters and sperm count. Problem of sperm count, motility and morphology stems from disarray in control mechanism, including pre-testicular, testicular and post-testicular factors.

The results of the study were comparable to a study, in which mean sperm count was 86.8±7.5 million/ml, negating the fact that sperm count is declining in our part of the world. The results are also comparable to another study in UK, in which semen analysis was performed on a population of 1801 suspected infertile men, in which the mean sperm density was 84.3±78.3.

The original meta-analysis that sperm density has decreased globally by about 50% over the past 50 to 60 years attracted considerable attention and generated much controversy.1,2 The situation in Pakistan is not as devastating as in other parts of world where testicular cancer and urogenital infections are more prevalent. Most of the studies conducted on declining sperm count were European in origin; Asia is the region with highest population. The findings of this study provide some reflection about the situation in Pakistan although the duration of the study was only one year, and longer studies are needed to reflect the situation in Asia. A study was conducted at NIHFW (National Institute of Health and Family Welfare) Munirka, New Delhi, for 11 years to verify the claims and speculations on declining sperm counts in men. No significant decline in sperm counts was observed in any year during the entire study period.

Azoospermia affects only about 2% of the general male population and between 10% and 20% of men undergoing fertility treatments. Azoospermia stems from a problem with sperm production or a problem with sperm transport. There are variety of factors that may contribute to either of these causes. Prevalence of azoospermia in our study population was 14.89%, and of oligospermia 11.11% respectively. The results are comparable to a study in which the prevalence of azoospermia was 14.28% and that of oligospermia

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**Table-3: Comparison of semen parameters between normospermia and oligospermia.**

<table>
<thead>
<tr>
<th>Category</th>
<th>Count mean±SD</th>
<th>Volume mean±SD</th>
<th>Pus Cells mean±SD</th>
<th>Motile Sperm mean±SD</th>
<th>Nonmotile Sperm mean±SD</th>
<th>Normal Sperm mean±SD</th>
<th>Abnormal Sperm mean±SD</th>
</tr>
</thead>
<tbody>
<tr>
<td>Normospermia</td>
<td>135.41±70.6</td>
<td>2.97±1.35</td>
<td>10.80±7.69</td>
<td>57%±18.3</td>
<td>43%±18.3</td>
<td>65%±14.5</td>
<td>35%±14.5</td>
</tr>
<tr>
<td>95% CI</td>
<td>126.82-143.18</td>
<td>2.81-3.127</td>
<td>9.91-11.69</td>
<td>56.98-57.02</td>
<td>42.98-43.02</td>
<td>64.71-65.2</td>
<td>34.71-35.29</td>
</tr>
<tr>
<td>Oligospermia</td>
<td>6.07±5.60</td>
<td>2.05±2.00</td>
<td>11.16±10.16</td>
<td>38%±23</td>
<td>62%±23.9</td>
<td>45%±15.6</td>
<td>55%±15.6</td>
</tr>
<tr>
<td>95%CI</td>
<td>4.38-7.75</td>
<td>1.45-2.65</td>
<td>8.1-14.22</td>
<td>37.94-38.06</td>
<td>61.93-62.06</td>
<td>44.95-45.04</td>
<td>54.97-55.02</td>
</tr>
<tr>
<td>P value</td>
<td>0.0001(SS)</td>
<td>0.0001(SS)</td>
<td>0.782(NS)</td>
<td>0.0001(SS)</td>
<td>0.0001(SS)</td>
<td>0.0001(SS)</td>
<td>0.0001(SS)</td>
</tr>
</tbody>
</table>

P value <0.05; Statistically Significant (SS); Non Significant (NS).

**Table-4: Proportion of multiple factor abnormalities defects.**

<table>
<thead>
<tr>
<th>Pattern of Abnormality</th>
<th>Frequency</th>
<th>Percentage</th>
<th>95% CI</th>
</tr>
</thead>
<tbody>
<tr>
<td>Asthenospermia N=337</td>
<td>87</td>
<td>25.81%</td>
<td>(21.15-30.49)</td>
</tr>
<tr>
<td>Terato-spermia N=337</td>
<td>11</td>
<td>3.26%</td>
<td>(1.3-5.16)</td>
</tr>
<tr>
<td>Oligo-astheno-terato-spermia N=44</td>
<td>4</td>
<td>9.99%</td>
<td>(0.6-17.55)</td>
</tr>
</tbody>
</table>
Another study conducted by same personal showed the prevalence of azoospermia and oligospermia to be around 33%. The reported incidence of azoospermia in Pakistan is around 14% which is comparable to our study, but the incidence of oligospermia varies around 37%, the reason being the large sample size used in our study. Considering the percentage of azoospermia in Pakistan, it is comparable to USA and Kenya, with reported rates of 10% and 11.35% respectively. However, when the incidence rate of azoospermia was compared with Turkey and Zimbabwe, it was found to be on the lower side.

Low ejaculated volume can reflect abnormalities in accessory sex glands fluid synthesis i.e seminal vesical as 70% of seminal plasma contribution is from seminal vesical. It can also be indicative of a physical obstruction somewhere in the reproductive tract or in cases of incomplete retrograde ejaculation. Mean ejaculated volume in normospermia was 2.97±1.35 vs 2.05±2.00 in oligospermia and 2.43±1.19 in azospermic sample respectively. Majority of our patients had normal semen volume 74.24%, while 22% showed hypospermia (<2ml), and hyperspermia in 3.53%, the results were comparable to a study conducted in Sudan where majority of the subjects (89.7%) had adequate semen volume, while only 10.3% had abnormal semen volume. The results are also comparable to a study conducted in Nigeria in which majority of the subjects (91%) had adequate semen volume, while only 9% had abnormal semen volume i.e 7.3% hypospermia and 1.7% hyperspermia. The adequate semen volume obtained in our study may be a result of the 3-4 days of sexual abstinence. Our results suggest that seminal fluid volume plays little or no role in the etiology of male infertility and the role of sexual abstinence before seminal fluid sample collection for accurate semen analysis is important.

Infection of the male genital tract is an important morbidity factor. It is known that it may affect seminal quality through a direct action on spermatozoa or their environment, including local inflammatory reaction. When pus cells were compared, the results did not show any statistical significant difference.

Spermatozoa travel a long distance to meet and fertilize the oocyte. So sperm motility is a requisite for normal fertilisation. Motility comes with sperm maturation in their passage through the epididymis. The process of epididymal sperm maturation occurred under the influence of epididymal proteins and other substances which produce structural and biochemical changes in the sperms. Thus, motility is chiefly a parametre of post-testicular i.e. epididymal function.

The mean percentage of normal motile sperms were 57%±0.18 in normospermia samples as compared to oligospermia in which motile sperms were 38%±23%. Although advancing techniques had somewhat overcome the problems of sperm motility in infertile couples, but asthenospermia is still a common cause of human male infertility. In our study, asthenospermia was observed in 25% of samples and the results were comparable to a study conducted at the National Institute of Health, Islamabad, in which the prevalence was around 21.42%. In another study, the prevalence of asthenospermia was 18%.

Morphology i.e. the differential development of the head, midpiece and tail is a function of testes as well as the epididymis. In our study mean normal morphology in normospermia samples was 65%±14% vs. 45%±15.65% in oligospermic samples.

Oligospermic samples were associated with significant higher abnormal motility 62%±0.239 and abnormal morphology 55%±0.156 as compared to normospermic samples although we did not specify the type of abnormal morphology. The results are comparable to a study in which abnormal morphology was observed in 53% and abnormal motility in 60% oligospermic males. So sperm motility and morphology are changing parameters and their relative levels depend on the existing sperm count in an individual.

The prognosis of the infertile couple is inversely proportional to the number of abnormal patterns so one pattern of abnormality is better than two-pattern abnormality, and two is better than three-factor abnormality. When three-pattern abnormalities were identified in oligospermic sample population, the prevalence of oligoasthenoteratospermia was 9.09%. The results were comparable to a study in which prevalence of oligoasthenoteratospermia was 11%. The prevalence of teratospermia in our study population was 3.36%.

Conclusion
A high-quality basic semen analysis is the cornerstone of investigations related to infertile couple. The use of conventional semen parameters, such as sperm concentration, motility and morphology, are markers of male reproductive function. Sperm concentration in our population is not declining as in other parts of world and there is significant association between sperm

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concentration and sperm parameters.

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References