INTRODUCTION

It was Leeuwen Hoek who made the observation in the seventeenth century that men whose semen had no spermatozoa were incapable of begetting children, but it was not clear until 1928 that the sperm count was found to be associated with fertility potentials. Since that time a variety of sperm investigations and semen parameters have been developed with the hope of clarifying whether or not a man could impregnate his partner.

Glchen Russ warm (1779) expressed a view to the same effect that "in barren marriages the microscopes could settle the dispute between men and women". Benda et al (1891) developed the sophisticated microscopic technique, which has led to the deeper insight into spermatozoa. In the terminology used by them normozoospermia means the presence of not less than 60 million/ml spermatozoa with 70-80% of motility.

Macleod (1942), Macleod and Gold (1953), Eliasson (1971) and Hellinga (1976) have laid the scientific basis of conventional analysis of spermatozoa and the techniques recommended by them are still useful for reference. They attempted to demonstrate inexpensive investigation and showed that even a count of 20 million/ml spermatozoa might be regarded as normozoospermia.

In general, infertility is defined as failure to conceive after one year of regular unprotected intercourse with the same partner. However, the term "infertility" implies inability to conceive. Therefore, couples who do not conceive in > 1 year should be regarded as sub-fertile. To be more exact the term sub-fertile means a male who failed to conceive after one year of regular unprotected intercourse with the same partner and who had a sperm count of less than 20 million/ml. Semen analysis comprises a set of full descriptive measurements of spermatozoa and seminal fluid parameters that helps to estimate semen quality.

ABSTRACT

Background: To observe the variation in semen quality of infertile men, with the purpose of generating preliminary data in our population.

Material & Methods: Analysis of semen including volume, liquefaction time, pH, sperm concentration and quality was carried out over a period of 14 months from November 2003 to December 2004. A total of 350 men presenting as infertile couples were studied. Semen analysis was performed in Department of Reproductive Physiology/Health, Public Health Laboratories Division, National Institute of Health, Islamabad, as a part of fertility assessment.

Results: According to the sperm concentration, the samples were categorized in four different groups; (A) Azoospermic group having absence of sperms, (B) Oligozoospermic group with a sperm concentration of less than 20 million/ml, (C) Asthenozoospermic group with a sperm concentration greater than 20 million/ml but with less than 25% of progressive motility and (D) Normozoospermic group with a sperm concentration of more than 20 million/ml and sperm motility of 60% or greater, with more than 25% progressive motility. Linear regression analysis shows a decrease in semen volume in group A and B. The mean liquefaction time showing linear increase in group A and B. The pH did not vary too much among all the groups. The mean of sperm concentration was 0, 6.7 ± 1.7, 45.3 ± 8.8 and 86.8 ± 7.5 million/ml in groups A, B, C and D respectively.

Conclusion: Semen analysis provides important information about the quality and quantity of the sperm and is an integral part of infertility work up.

Key words: Infertility, Semen analysis, Liquefaction time, Sperm motility.
Generally in male dominated society and particularly in our society, where illiteracy and poverty are more prevalent, men hardly agree to investigate themselves for fertility potentials. Semen analysis is the gold standard and most valuable test that plays important role in an Andrology /Reproductive physiology laboratory. Although the computer assistant methodologies of women semen analysis and some other auxiliary methods have greatly enhanced the expectation of in-fertile patients, but however, the routine and conventional semen analysis plays a significant role world wide and the especially in the developing countries.7

This study was thus designed, in which a commonly performed sperm parameters of the male partners of infertile couples were studied, with the purpose of generating preliminary data in our population.

**MATERIAL & METHODS**

**Classification of patients:** A total of 350 subjects, referred for fertility assessment during November 20023 to December 2004, were included in the study. The subjects having no spermatozoa in the semen even after concentration by centrifugation at 3000 rpm for 15 minutes were noted as azoospermia, those having sperm count of less than 20 million/ml were noted as Oligozoospermia. The Oligozoospermia. Subject were further categorized into 3 categories have severe (count <5 million/ml), moderate (count between 5-10 million/ml), and mild (count >10 million/ml), according to the severity of sperm depletion.8 The subject having progressive motility of less than 25 % were termed as asthenozoospermia, while the subjects having progressive activity of more than 25%, with sperm concentration of 20 million/ml or more were classified as normozoospermia.

Semen analysis: This work was carried out at the department of reproductive physiology/health, Public health Laboratories Division, National Institute of Health, Islamabad. Total of 350 infertile males referred from gynecologist, urologist, and general practitioners for the semen analysis were included in the study. The age of the patient ranged from 20-55 years but however, the maximum number of patients was in the age range of 25-40 years, and duration of infertility between 1-20 years. A detailed history was taken, in order to determine any correlation of the problem of infertility faced by couples and the status of male partner.

Semen analysis was performed according to the procedure described in the WHO protocols,9 at the department of Reproductive Physiology/Health, Public Health Laboratories Division, National Institute of Health, Islamabad for the period of 14 months.

For each sample, the color and consistency of semen was visually ascertained and liquefaction time was recorded. 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 glass rod and a thin drop was spread on a glass slide by placing a cover slip on it. Sperm motility was assessed by microscope appraisal of 200 spermatozoa from different fields. They were classified as being actively motile, sluggish, and immotile. Total sperm count as million/ml was obtained by diluting 1:19 with semen diluting fluid in an improved Neubauer haemocytometer.10

**RESULTS**

The results of the study are given in the Table 1. It can be seen that perm were completely absent in the ejaculates of men belonging to group A (azoospermia), 6.7 + 1.7 million/ml in men belong-

<table>
<thead>
<tr>
<th>Group</th>
<th>Ejaculate volume (ml)</th>
<th>Liquefaction time (min)</th>
<th>pH</th>
<th>No. of sperms (x10/ml)</th>
<th>Sperm motility</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Active</td>
</tr>
<tr>
<td>A (50)</td>
<td>1.5 + 0.4a</td>
<td>37.5 + 0.7c</td>
<td>7.3+1c</td>
<td>0.00c</td>
<td>0.00c</td>
</tr>
<tr>
<td>B (75)</td>
<td>1.7+ 0.2a</td>
<td>28.7 + 3.7d</td>
<td>7.3+1c</td>
<td>6.7+ 1.7c</td>
<td>15.0+6.0c</td>
</tr>
<tr>
<td>C (75)</td>
<td>2.5+ 0.2a</td>
<td>18.5 + 0.7d</td>
<td>8.0+0.07d</td>
<td>45.3+ 8.8c</td>
<td>20.1+5.1c</td>
</tr>
<tr>
<td>D (150)</td>
<td>2.4+0.2</td>
<td>18.6 + 3.6</td>
<td>8.0+0.08</td>
<td>86.8+7.5</td>
<td>66.8+2.3</td>
</tr>
</tbody>
</table>

Values are mean + SE

Group A: Azoospermic, Group B: Oligozoospermic, Group C: Asthenozoospermic, Group D: Normozoospermic.

d = Not significant (p>0.05), a = Significant (p < 0.05), b = Significant (p < 0.01), c = Significant (p < 0.001).
ing to group B (oligozoospermia), 45.3 + 8.8 million/ml in men belonging to group C (asthenozoospermia) and 86.8+7.5 million/ml in men belonging to group D (normozoospermia). The means of ejaculates volume for group A, B, C and D were 1.5±0.4, 1.7+0.2, 2.5+0.1, and 2.4+0.2 ml respectively. The average liquefaction time recorded was 37.5±0.7, 28.7+3.7, 18.5+0.7, 18.6+0.7 minutes in groups A,B,C and D respectively. The means of pH were 7.8+0.1, 7.8+0.1, 8.0+0.07 and 8.0+0.08 in groups A, B, C and D respectively. It significant difference were observed for ejaculate volume, liquefaction time, and sperm concentration in azoospermic and oligozoospermic patients when compared with the normozoospermic group. The pH did not vary too much among all the groups.

DISCUSSION

Semen analysis is the most commonly advised test to estimate fertility assessment of the men. Although the physiological conditions and seasonal variations may affect the semen quality but it is proved that these does not cause azoospermia.

In the present study, data of 350 male partners of infertile couples was analyzed. The data indicates that 35.71% of subjects with low sperm density were either azoospermic (14.28%) or oligozoospermic (21.43%), where as in 21.43% of men, the sperm motility was low, although the sperm concentration was with in the normal range (20 million/ml). There were a large number of individuals (42.86%) having normal activity and normal concentration of sperms. The ratio between the normal and abnormal subjects evaluated on the basis of spermiogram quality was 0.75.

In Pakistan, the reported incidence of azoospermia is 12.32%, and in another study, the incidence rate was 14%, while in the present study it is 14.28%. The sample size in our study was a small, but the azoospermic percentage was similar to the earlier studies. 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 lower side. Comparing the incidence rate of oligozoospermia in our study and some other studies, it is found to be 21.43% in one study; and 53% in another one, which is in contrast to the present study. This high percentage in the previous studies might be due to large number of analyzed sample.

Our findings also suggest the increase of liquefaction time in group A and B. The difference in liquefaction time between azoospermic and Oligozoospermic was significant. The viscous specimens can impair the availability of motile sperm at the site of fertilization. Hence fertility is impaired in such cases, which is also in agreement with the previous studies.

The findings of the study show low ejaculate volume both in azoospermic and oligozoospermic males when compared with normozoospermic men. This low volume can reflect abnormalities in accessory sex glands fluid synthesis or secretion, it can be also be indicative of a physical obstruction somewhere in the reproductive tract or may occur in the cases of incomplete retrograde ejaculation. It can be seen that the case of azoospermia the liquefaction time exhibited significant decrease (P < 0.001), while the increase in oligospermic as well as asthenospermic patients was not significant. Similarly significant differences (p < 0.05 and p < 0.001) for total ejaculate volume and seminal pH was observed in azoospermic and oligospermic subjects in comparison with the normozoospermic subjects. The total sperm count and active motility were significant (p < 0.001) among the entire infertile groups, when compared with the normozoospermic group.

CONCLUSION

Semen analysis is the corner stone of testing for male infertility, which provides important information about the quality and quantity of the sperm and is an integral part of infertility work up. Although many infertile males are either azoospermic or oligozoospermic, a fair number of patients are normospermic and prompt us to look for the causes of male infertility other than described in this study.

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