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# SEMEN PARAMETERS;
## A DESCRIPTIVE OBSERVATIONAL STUDY ON SUB-FERTILE MALES PRESENTING AT A PRIVATE ASSISTED REPRODUCTION CLINIC IN LAHORE PAKISTAN.

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**ABSTRACT:** Semen is a pale whitish fluid secreted by male during ejaculation and contains spermatozoa which are male gametes essential of fertilizing the oocytes which are female gametes. In a quest to evaluate male's fertility potential semen is analyzed to look into some of its characteristics and of the sperms contained within the semen analyzed. Method of collection influences the results of Semen analysis as does the technique of analysis. Spermatozoa are examined for number (count), shape (morphology) and movement (motility) in order to assess their quality. Non sperm cells, volume, Fructose level, pH, liquefaction are also checked as a part of routine analysis. **Objectives:** To describe the pattern of semen parameters in sub-fertile males. To look into frequency and distribution of abnormal semen parameters in a group of Pakistani males in Lahore. **Methods:** In this Retrospective, cross sectional, observational study all males undergoing for evaluation and treatment for sub-fertility at a private Assisted Reproductive Technology clinic in Lahore, Pakistan were included. Approval of the IRB was sought and data collection instrument was a specially designed Performa which was validated by the biostatistician of LIFE research cell. Data was extracted from the files of LIFE (Lahore Institute of Fertility and Endocrinology) and entered in SPSS version 15. Sampling technique was non-probability, consecutive. Semen analysis was done by methods defined by the WHO (World Health Organization). **Results:** Of total patient (n=679) 92.2% (626) males passed sample at LIFE (Lahore institute of fertility and endocrinology) and (7.8%) 53 brought sample from home. Of the males who passed sample at LIFE (78.8%) 535 collected semen by masturbation, (11.9%) 81 by coitus; the source of sample of (9.3%) 63 males was not known. As 2-6 ml semen was consider to be normal by WHO criteria, (80.6%) 547 males were in normal range (14.1%) 96 found to be less than 2-6 ml and (5.3%) 36 found to be more than normal range. According to WHO criteria 15 million/ml count is said to be normal, in our research (82.0%) 557 were found to be normal, in (2.9%) 20 count was found to be less than 15 million/ml and in (5.9%) 40 count was less than 1 million/ml. In (9.1%) 62 counts was found to be abnormally low. In this research (66.1%) 449 had normal sperm motility, (21.8%) 148 had less than 40% and abnormally low sperm motility was found in (12.1%) 82 males. **Conclusion:** The results of the single semen analysis are of limited utility and no decision should be taken on the bases of these results in term of diagnosis and treatment strategies.

**Key words:** Semen analysis, subfertility, count, morphology, motility

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## INTRODUCTION

Semen is a pale whitish fluid secreted by male during ejaculation and contains spermatozoa which are male gametes essential for fertilizing the oocytes which are female gametes.[1] In a quest to evaluate male's fertility potential semen is analyzed to look into some of its characteristics and of the sperms contained within the semen analyzed.[2,3] Method of collection influences the results of Semen analysis as does the technique of analysis.[1] The results of semen analysis not only point towards the diagnosis but also helps in deciding about the interventions to be planned.[4,5] In order to assess their quality spermatozoa are examined for number (count), shape (morphology) and movement (motility).[6] Non sperm cells, volume, Fructose level, pH, liquefaction are also checked as a part of routine analysis. The values of semen

analysis's results are compared with WHO reference values.

Sperms are stored in the lower portion of the epididymis before they pass through the ejaculatory ducts to reach the seminal vesicle.[7] Fluids from this simple tubular gland are added to it and so are the secretions from the prostate and the bulbourethral glands.[8] Composition of seminal fluid is complex and consist of various organic and inorganic components. Transport of spermatozoa through the female reproductive tract is essential to achieve fertilization the chances of which are maximized if intercourse occurs a few days prior to ovulation.[9,10]

The seminal plasma provides a nutritive and protective medium for the spermatozoa during their journey through the female reproductive tract.[11] Seminal fluid provides nutrients to the spermatozoa; it is also protective in the hostile environment of the female reproductive tract.[6] Semen analysis is a very simple, basic and non-invasive laboratory procedure that provides some important information about fertility potential of the male. This information may not be complete but may be useful to give a direction to further investigation.

## OBJECTIVES

To describe the pattern of semen parameters in sub-fertile males

To look into frequency and distribution of abnormal semen parameters in a group of Pakistani males in Lahore

## METHODOLOGY

In this Retrospective, cross sectional, observational study all males undergoing for evaluation and treatment for sub-fertility at a private Assisted Reproductive Technology clinic in Lahore, Pakistan during the study period were included. Approval of the IRB was sought and data collection instrument was a specially designed Performa which was validated by the biostatistician of LIFE research cell. Data was extracted from the files of LIFE (Lahore Institute of Fertility and Endocrinology) and entered in SPSS version 15. Sampling technique was non-probability, consecutive. Semen analysis was done by methods defined by the WHO (World Health Organization). Variables included color, volume, liquefaction, viscosity, sperm concentration, sperm motility, rapid progressive motility, slow progressive motility, poor progressive motility, sperm morphology, abnormal sperm morphology, abnormal sperm heads, abnormal neck/mid piece, tail defects. After making table and graphs descriptive analysis was done i.e. frequencies and percentages of categorical variables and mean, standard deviation and standard error for numerical variables.

## RESULTS

Of total patient (n=679) 92.2% (626) males passed sample at LIFE (Lahore institute of fertility and endocrinology) and (7.8%) 53 brought sample from home. Of the males who passed sample at LIFE (78.8%) 535 collected semen by masturbation, (11.9%) 81 by coitus; the source of sample of (9.3%) 63 males was not known. Color of the sample was found to be normal in (99.7%) 677 patients where as only (0.3%) 2 had reddish colored semen.

As 2-6 ml semen was considered to be normal by WHO criteria, (80.6%) 547 males were in normal range (14.1%) 96 found to be less than <2 ml and (5.3%) 36 found to be more than normal range (>6 ml).

Majority of the male's i.e (99.4%) 675 had normal liquefaction and in (0.6%) 4 liquefaction function was not normal. Viscosity was found to be normal in (97.9%) 665 where as in (2.1%) 14 viscosity was not normal.

According to WHO criteria 15 million/ml count is said to be normal, in our research (82.0%) 557 males were found to be normal, in (2.9%) 20 males count was found to be less than 15 million/ml and in (5.9%) 40 patients, count was less than 1 million/ml. In (9.1%) 62 males count was found to be abnormally low (Fig-1).

According to WHO criteria 40 % motility is said

to be normal, in our research (66.1%) 449 males had normal sperm motility, (21.8%) 148 males had less than 40% and abnormally low sperm motility was found in (12.1%) 82 males(Fig-2). Rapid progressive motility was found to be normal in (5.6%) 38 males, less than 20 mic/second in (66.4%) 451 and in (28.0%) 190 males rapid progressive motility was necrosopic (Fig-3). Slow progressive motility was normal in (13.3%) 90 males, less than 5-20 mic/second in (3.4%) 23, more than 5-20 mic/second in (68.6%) 466 and in (28.0%) 190 males Slow Progressive Motility was azoospermic (Fig-4). In (7.2%) 49 males poor progressive motility was normal, more than 5 mic/second in (80.9%) 549 and in (11.9%) 81 males Poor Progressive Motility was necrospermic (Fig-5).

(81.6%) 554 males had normal sperm morphology, (9.3%) 63males had less than 4% morphology and (9.1%) 62 males had abnormal morphology (Fig-6). The mean and S.D of abnormal sperm morphology was (56.57±0.959, 24.99), Abnormal sperm heads (20.35±0.509, 13.275), Abnormal neck/mid piece (13.88±0.344, 8.975) and Tail defects (22.29±0.458, 11.929) Table-II.

| Semen parameters | WHO 1980 | WHO 1987 | WHO 1992 | WHO 1999 | WHO 2010a |
|---|---|---|---|---|---|
| Volume (mL) | ----- | ≥2 | ≥2 | ≥2 | 1.5 |
| Sperm count(106/mL) | 20-200 | ≥20 | ≥20 | ≥20 | 15 |
| Total Sperm count(106 ) | ----- | ≥40 | ≥40 | ≥40 | 39 |
| Total motility (%motile) | ≥60 | ≥50 | ≥50 | ≥50 | 40 |
| Progressive motility | ≥2c | ≥25% | ≥25%(Grade A) | ≥25%(Grade A) | 32% (A+B) |
| Vitality (% alive) | ----- | ≥50 | ≥75 | ≥75 | 58 |
| Morphology (% normal) | 80.5 | ≥50 | ≥30d | 14o | 4’ |
| Leukocyte count(106/mL) | <4.7 | <1.0 | <1.0 | <1.0 | <1.0 |

**Table-I. Standards for Semen Examination as Published in Consecutive World Health Organization Manuals**

Table-I. (Permission granted by Dr. Esteves toads his table in this article). WHO has gradually over years lowered the criteria of normal semen analysis.

Group table of all variables

| Sr. No | Variable | Categories | frequencies | Percentages |
|---|---|---|---|---|
| 1 | sample produced at | LIFE (clinic) | 636 | 92.2 |
| | | Home | 53 | 7.8 |
| 2 | source of sample | Masturbation | 535 | 78.8 |
| | | Coitus | 81 | 11.9 |
| | | not known | 63 | 9.3 |
| 3 | Color | Normal | 677 | 99.7 |
| | | Reddish | 2 | 0.3 |
| | | Reddish Yellowish | 0 | 0 |
| 4 | Volume | Less than 2-6 ml | 96 | 14.1 |
| | | 2-6 ml (normal) | 547 | 80.6 |
| | | More than 2-6 ml | 36 | 5.3 |
| 5 | Liquefaction | 30-60 mints (normal) | 675 | 99.4 |
| | | Abnormal | 4 | 0.6 |
| 6 | Viscosity | Normal | 665 | 97.9 |
| | | Abnormal | 14 | 2.1 |
| 7 | sperm concentration | 15 mil/ml or more (normal) | 557 | 82.0 |
| | | less than 15 mill/ml | 20 | 2.9 |
| | | less than 1mil/ml | 40 | 5.9 |
| | | Azoospermia | 62 | 9.1 |
| 8 | Sperm Motility | 40% or more (normal) | 449 | 66.1 |
| | | less than 40% | 148 | 21.8 |
| | | Necrospermia | 82 | 12.1 |

| Sr. No | Variable | Categories | Frequencies | Percentages |
|---|---|---|---|---|
| 9 | Rapid progressive Motility | less than 20 | 451 | 66.4 |
| | | > 20 mic/second | 38 | 5.6 |
| | | Nil | 190 | 28.0 |
| 10 | Slow progressive Motility | Less than 5-20 mic/sec | 23 | 3.4 |
| | | 5-20 mic/sec (normal) | 90 | 13.3 |
| | | More than 5-20 mic/sec | 466 | 68.6 |
| | | Nil | 100 | 14.7 |
| 11 | Poor progressive Motility | < 5 mic/sec | 49 | 7.2 |
| | | More than 5 | 549 | 80.9 |
| | | Nil | 81 | 11.9 |
| 12 | Sperm morphology | 4% or more (normal) | 554 | 81.6 |
| | | not in range | 63 | 9.3 |
| | | Abnormal | 62 | 9.1 |

**Descriptive characteristics of semen parameter**

Analysis of numeric data (table-III) macphological

| Sr. No | Numeric variables | Mean | Std. Error of Mean | Std. Deviation |
|---|---|---|---|---|
| 1. | Abnormal sperm morphology | 56.57 | .959 | 24.995 |
| 2. | Abnormal sperm heads (%) | 20.35 | .509 | 13.275 |
| 3. | Abnormal neck/mid piece (%) | 13.88 | .344 | 8.975 |
| 4. | Tail defects (%) | 22.29 | .458 | 11.929 |

**Morphological characteristics of sperm parameters**

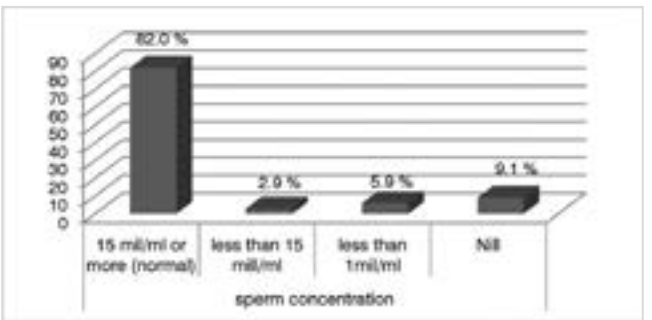


**Figure-1. Frequency of sperm concentration**

Legend: sperm concentration is shown in four categories and their respective frequency

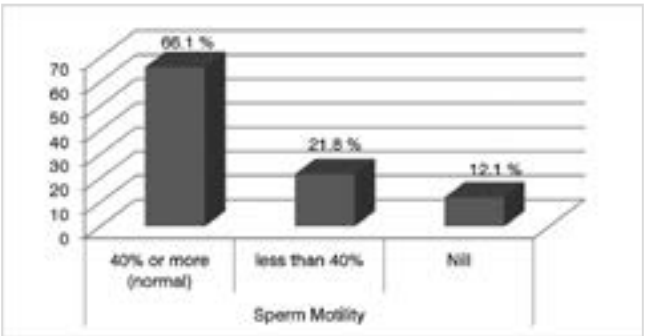


**Figure-2. Frequency of sperm Motility**

Legend: Sperm Motility is shown in three categories and their respective frequency

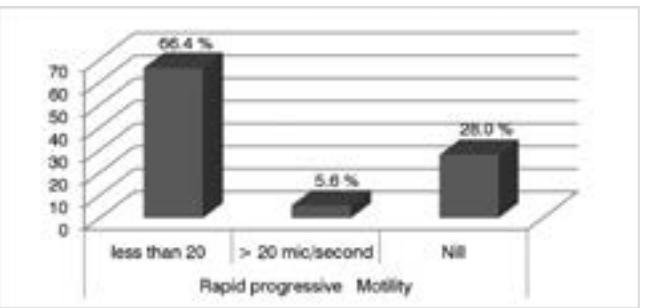


**Figure-3. Frequency of sperm Motility Rapid Progressive Motility**

Legend: Sperm Rapid Progressive Motility is shown in three categories and their respective frequency

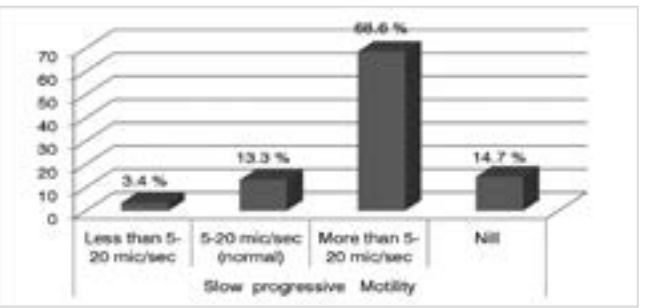


**Figure-4. Frequency of sperm Motility Slow Progressive Motility**

Legend: Sperm Slow Progressive Motility is shown in four categories and their respective frequency

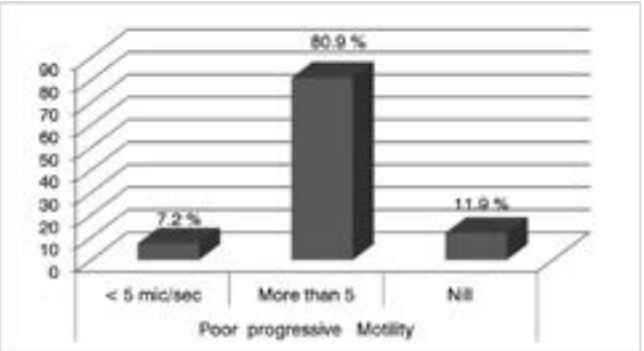


**Figure-5. Frequency of sperm Motility poor Progressive Motility**

Legend: Sperm poor Progressive Motility is shown in three categories and their respective frequency

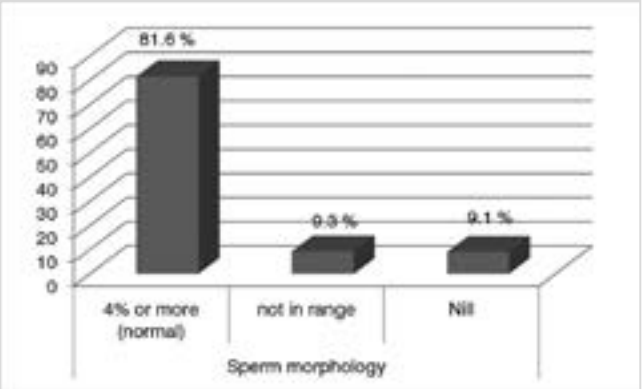


**Figure-6. Frequency of sperm Morphology**

Legend: Sperm Morphology is shown in three categories and their respective frequency

## DISCUSSION

In this research count was more than 15 million/ml in 82% (n=679), less than 15 million/ml in 2.9%, less than one million in 5.9% and abnormally low count in 9.1%. Motility was more than 40% in 66.1% (n=679), less than 40% in 21.8% and abnormally low sperm motility in 12.1%. Morphology was more that 4% in 81.6% (n=679), less than 4% in 9.3% and 9.3% had abnormal morphology.

Semen analysis has been regarded as a single most important test for male subfertility.[12,1,13] Semen analysis also furnishes information about functioning of accessory sex glands, epididymis and seminiferous tubules.[14-18] Despite phenomenal advancement in assisted reproductive technology role of a robust semen analysis is still not diminished.[19] As clinicians use the results of semen analysis in crucial decision making, like diagnosis and treatment, it must be reliable.[20]

Sperm parameters which are normal according WHO criteria may misguide the clinician and a sub-fertile male with "normal" sperm parameters may be misdiagnosed as a case of unexplained subfertility.[21] Special sperm function tests such as DNA fragmentation, oxidative stress, will help to finalize the diagnosis.[22]

Though standards set by WHO have been accepted by most of the labs performing semen evaluation. Several reports of sub-fertile males will be regarded as normal which were said to be abnormal before. So referral of these sub-fertile males to the fertility center may be delayed for separated semen analysis.[23,24] It has been also proposed that assessing the sperm parameters like morphology and motility may not be sufficient.[25] Good practice should include, a thorough history taking and complete clinical examination, followed by relevant endocrine and genetic studies.[14] Medical evaluation may also be essential in males of higher age group.[22]

Importance of sperm morphology and motility in relation to occurrence of pregnancy in sub fertile couples undergoing ART investigation like IVF/ ICSI was confirmed in a number of studies and the value of sperm concentration was reduced.[26]

Sperm motility is critical at the time of fertilization because as the sperm passes through the ZonaPellucida motility helps and increases the chance of conception. Decreased sperm motility not only reduces the movement of sperm in the cervical mucus but also hampers the transport of sperm towards the egg.[27,28,29]

Sperm concentration is associated with fecundity. Sperm count less than $20x10^6$/mL has been associated with reduced fecundity.[30]

It was found that all the sperm factors do not have equal significance in predicting male fertility potential in couples with higher the percentage

of motile sperms with normal morphology, pregnancy rate was significantly higher.[31]

In a study SandroEsteves compared cut off references value of characteristics of semen which were published in various WHO manuals in between 1980-2010. This table (table-I) shows semen characteristics which were labeled as normal by WHO in various manuals.[14]

Anne Jequier suggested three decades ago that quality assurance in the laboratories performing semen analysis was necessary.[32] About ten years ago she proposed that quality assurance was no more needed for laboratories performing semen analysis. The notion behind this statement was the results of research in the last three decades which proved that though semen analysis was very important investigation to start the evaluation of male subfertility but was not sufficient to make a final diagnosis. Even sperm functions tests had little prognostic value in terms of fertility assessment[33] Jequier concluded that a competently performed semen analysis was sufficient and there was no need of the expense, time and effort being wasted in the quality assurance.[34]

Duration of abstinence, individual biological variability and activities of accessory glands and error of analysis are, to some extent responsible, for high variability of semen characteristics from same individuals.[35,14] It adds to the uncertainty about the semen analysis results and their clinical importance. The results of the single semen analysis are of limited utility and no decision should be taken on the bases of these results in term of diagnosis and treatment strategies.[14]

## CONCLUSION

The results of the single semen analysis are of limited utility and no decision should be taken on the bases of these results in term of diagnosis and treatment strategies. Semen analysis still remains an important investigation regarding male sub fertility. A complete history and thorough clinical examination is essential to reach final diagnosis.



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*"Price is what you pay.*
*Value is what you get."*

WARREN BUFFETT

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| 2 | Prof. Dr. Yousaf Latif Khan | Conceptualization and designing the study and helped in writing introduction | |
| 3 | Irfan Mehfooz | Helped in data collection | |
| 4 | Muhammad Burhan | coordination and eiterative search | |
| 5 | Saba Sardar | Data analysis and interpretation of the results | |
| 6 | Dr. Abdul Rahman Khawaja | Write up of eiterative review discussion and concusion | |