Quest Journals
Journal of Research in Humanities and Social Science
Volume 12 ~ Issue 8 (2024) pp: 145-153
ISSN(Online):2321-9467
www.questjournals.org

Research Paper

Assessment of Morphological Abnormality in Spermatozoa

Deepanshi[1], Dr. Minakshi Ghosh[2], Dr. K. Manoj kumar[3]
[1] *Research Scholar, BMLT 6th Semester, STAREX UNIVERSITY*
[2] *Sr. Faculty, Department of Microbiology, Institute of Public Health and Hygiene, Mahipalpur, New Delhi*
[3] *Associate Professor, Department of Clinical Pathology, Institute of Public Health and Hygiene, Mahipalpur, New Delhi*
**Corresponding Author: (K. Manoj Kumar)*

ABSTRACT
The spermatozoa were first seen in ejaculates in the 17th century. However, the basic mechanisms of human fertilization have been only fully understood after the discovery of ovum in 1827. As a result, the interest in developing technologies for semen analysis arose from the early 1900s. Indeed, standard methodologies for semen analysis were designed mostly along the first half of the 20th century. Before the 1930s, semen analysis was nearly unavailable clinically, since there were still no robust methodologies for assessing sperm characteristics, as well as to set up standard references that could be able to assess the reproductive capacity of men. We demonstrate a deep learning to classify sperm abnormality. Our deep learning approach classifies sperm at accuracy and performs well. A retrospective analysis of semen sample of patients received in andrology laboratory. A total of 602 semen samples were processed for analysis. We see that the adult (26-32) and younger (33-39) patient are lower rate of fertility as compared to middle age (40-47) of patient. As we can see awful lifestyle can effect of the sperm quality. We compare three data of age (26-32, 33-39, 40-47) and we can clearly seen that a middle age (40- 47) have good quality of sperm as compared to younger and adult age.

***R**eceived 01 Aug., 2024; **R**evised 08 Aug., 2024; **A**ccepted 10 Aug., 2024 © The author(s) 2024.*
P**ublished with open access at **www.questjournas.org

I. Introduction

Over 30% of documented cases of human infertility, which affects more than 10% of spouses globally and is serious to the reproduction process, are related to men. Semen analysis, heavily rely on sperm morphology, or the form and manually and heavily dependent on clinician expertise, is an important stage in the assessment of male infertility and sub fertility. Families are increasingly turning to medicine to help them get conceived because infertility is a global health concern. In order to classify sperm abnormalities, we demonstrate deep learning. With accuracy and effectiveness, our deep learning method helps to find out the highest quality sperm. Since 15-20% of spouses worldwide had difficulty getting pregnant, infertility has emerged as a major global health issue. With about 15-20% of spouses experiencing difficulty getting pregnant worldwide, infertility has emerged as a major health concern. About 20– 70% of cases of infertility are caused by male aspects, and about 30–40% of these factors directly affect conception. In 1677, the first focusing and description of infertility was given. Infertility should first be categorised as primary or secondary. A male patient identified as infertile undergoes a preliminary targeted assessment. The patients' full medical, sexual, and reproductive history were recorded. For this investigation, the results of physical examination reports were also taken into account. According to the World Health Organization's (WHO) recommendations, investigations on the examination of sperm were carried out. For every infertile person, a semen analysis forms the basis of the diagnostic process. An evaluation of the sperm quality as a whole and the probable need for secondary diagnosis are both made possible only after preliminary analysis of the sperm.

II. Objective and Scope of Work

The main objective of the present project work was focused on the following:

1) Describe the morphology of male infertility.
2) Create awareness of male infertility.
3) Create awareness bad lifestyle impact of patient fertility.

III. Literature Review

More than three centuries ago, in 1677, the finding of sperm was reported for the first time. Van Leeuwenhoek and his assistant described animalcule in human semen solely out of human curiosity and without any consideration of scientific purpose.

Eliasson (1971) made a significant contribution when he found out that the mid-piece and tail of human spermatozoa should also be rated morphologically, and that the entire spermatozoon must therefore be taken into account when evaluating sperm morphology. For the morphological classification of spermatozoa, Eliasson was the first to emphasize the significance of sperm size and dimensions. Eliasson divided sperm defects into three major categories: the head, the middle, and the tail. The head, mid-piece, and tail of a spermatozoon must all be normal for the spermatozoon to be categorized as normal. A typical head had to have an oval form. Forms on the edge have to be counted as typical.

Mantegazza (1886) was the first to link male fertility with semen parameters. However, he only focused on the examination of semen volume as a measure of men's fertility. He discovered readings between 0.85 and 6.0 ml. Mantegazza also studied the impact of temperature on sperm motility and found that exposing semen to temperatures between 37°C and 47°C had a progressive detrimental effect on sperm motility. This was the first study to look into how temperature affects sperm motility.

James Marion et al. (1868) In this regard, made a significant contribution. They invented a laboratory technique known as the postcoital test, which analyses the gradually motile sperm in the cervical mucus after coitus. The inspection was planned for the ovulation since there was an abundance of mucus that made it easier for sperm to penetrate and be evaluated. He noted that a man was not infertile if motile sperms were found in cervical mucus, and that the cervix was not the reason for female infertility. Sims claims that the post-coital test had the following functions: (1) to confirm the presence of spermatozoa in the semen; (2) to detect whether or not spermatozoa entered the utero-cervical canal; and (3) to ascertain whether or not the secretions of this canal were favourable.

Gustaf Retzius et al. (1995) has also made significant remarks regarding the sperm analysis. Using samples from the epididymis, seminiferous tubules, and semen that had been fixed using osmium tetroxide and Zenker's fixation, he analysed a wide variety of sperm from more than 400 species, including humans. Drawings of Retzius that were published in the journal Biologische Untersuchungen, which he founded to publish his works, were incredibly detailed. He began the publication in the 11th volume in 1904 and continued it until the 19th volume. Retzius contributed too little to the development of the semen analysis, but his notes and sketches made a significant contribution to our understanding of the morphological traits of sperm from various species. Doctors and researchers unanimously agreed that a male partner examination was necessary in the diagnostic workup of infertility once it became clear that males can also have difficulty becoming pregnant with their partner. Men were only looked into, though, if their companion did not exhibit sterility.

Martin et al. (1902) researched azoospermic men with obliterating epididymitis, they stressed for the first time the significance of looking into male sterility by semen analysis. Accordingly, should a marriage fail, the semen should be inspected before the lady is treated by a gynaecologist; more importantly, the treatment should continue until microscopic analysis reveals that spermatozoa are once more present in the semen. In his work, they used semen smears stained with iron-hematoxylin, whose characteristics were portrayed in drawings, to analyse the sperm motility following surgery and to carry out a thorough investigation of the sperm morphology. He believed that the lack of spermatozoa in the seminal fluid was conclusive evidence of infertility. Although the existence of moving spermatozoa in these ejaculates is typically taken as a strong indication of creative ability, there isn't enough data to support this claim. It should be noted that Martin was one of the first to demonstrate that spermatogenic failure or ductal occlusion was the root cause of azoospermia.

John Adolph Detlefsen's (1914) investigation of the sperm motility in samples aspirated from the epididymis, hybrid animals (wild cavies vs. regular guinea pigs) showed that it has a higher chance of producing offspring. They also conducted a thorough investigation into male and female fertility and sterility and concluded that, in addition to sperm motility and frequency, the vitality of sperm in the cervix, vagina, and fundus of the uterus also has a positive correlation with men's reproductive ability. The five types of sperm motion described by them—progressive vibratile, undulatory tactile, stationary bunting, rotatory swimming, and pendulum swimming- provided a quick way for evaluating sperm motility. They believed the first three to be the subsequent regular phases of spermatozoic development. They believed that the first three spermatozoic phases were successively normal and that the latter two were aberrant. This appears to be the first effort to categorise sperm motility patterns. They placed special emphasis on the evaluation of sperm vitality based on the duration (endurance) of sperm motility, first by noting the amount of time that motility lasts in fresh semen, second by noting the amount of time that motility lasts in the natural medium (secretions of the female genital tract), and third by noting the amount of time that motility lasts in various artificial media. He said that

comparing these data seems to accurately demonstrate the vitality of semen, but the technique is labor-intensive and too onerous for most people. The process was actually too complicated to be used in the semen analysis method. He came to the conclusion that the animal was undoubtedly sterile if there were no motile sperm. As the percentage of motile sperm rises, so does the likelihood of conception. He presum-ed the significance of sperm motility analysis for sterility research.

Cary (1916) paved the stage for semen analysis to be seen as a useful method for examining men in barren marriages, but it also became the object of criticism once it was published.
Huhner (1921) criticised the procedure for collecting and transporting semen samples from home to the workplace as well as the suggestions for keeping the temperature constant during the entire laboratory evaluation process. He asserted that the challenging semen collection system and laboratory procedures deterred doctors from eventually implementing semen analysis in offices.
Macomber and Sanders (1929) revealed that semen analysis should be formally implemented in clinical and laboratory practise, with serving as its foundation. A correlation between pregnancy rates and sperm counts greater than 60- 106/cc was found by Macomber and Sanders after studying 294 males. This was the first citation for sperm count in the routine of the semen analysis, and it was undisputed for at least three decades. This cut-off point has to be adjusted since pregnancy could also be obtained by males whose sperm counts were less than 60- 106/cc.
Samuel Raynor Meaker (1934) Semen analysis reveals importance for determining male sterility highlighting the importance of rigorous procedures for the evaluation of semen, he also came to the conclusion that correct evaluations could reveal a notable degree of infertility, and he suggested that the male bears a major portion of the responsibility for a childless union.
Hotchkiss (1931) came to the conclusion that the passage of time leaves it up to future generations to judge the true worth of this significant notion as evidence gathered to support or contradict the prevailing theories of sperm morphology. According to Moench, a man needs more than 80% of his typical sperm morphology to produce one pregnancy. In actuality, this figure is higher than the one that the current WHO guidelines for semen analysis recommend.
WHO (2010) Finally, it may be worthwhile collecting data using this method on the total number per ejaculate of each of the abnormal sperm forms to provide a maximum number associated with infertility, just as the total number of motile and morphologically "normal" spermatozoa per ejaculate is more important than their percentages in providing a minimum number of such cells compatible with fertility (WHO in 2010).

IV. Methodology

A retrospective analysis of semen sample of patients received in andrology laboratory. A total of 602 semen samples were processed for analysis.
Sample collection
The samples were collected in a private room near the laboratory, in order to limit the exposure of the semen to fluctuations in temperature and to control the time between collection and analysis
1) The samples were collected after a minimum of 2 days and a maximum of 7 days of sexual abstinence. If additional samples are required, the number of days of sexual abstinence should be as constant as possible at each visit.
2) The people were given clear written and spoken instructions concerning the collection of the semen sample. These should emphasize that the semen sample needs to be complete and that the man should report any loss of any fraction of the sample.
3) The samples were labelled with the date and time of collection, the completeness of the sample, any difficulties in producing the sample, and the interval between collection and the start of the semen analysis were also recorded.
4) The sample should be obtained by masturbation and ejaculated into a clean, wide- mouthed container made of glass or plastic, from a batch that has been confirmed to be non-toxic for spermatozoa.
5) The specimen container should be kept at ambient temperature, between 20 °C and 37 °C, to avoid large changes in temperature that may affect the spermatozoa after they are ejaculated into it. It must be labelled with the person's name and identification number, with date and time of collection.
7) The specimen container is placed on the bench or in an incubator (37 °C) while the semen liquefies.
8) In case of any incomplete sampling, especially if the first, sperm-rich fraction is missed out. The patient should pass urine. After a while, a second sample should be collected, again after an abstinence period of 2–7 days.
9) Wash hands and penis with soap, to reduce the risk of contamination of the specimen with commensal organisms from the skin.
10) Rinse away the soap.

11) Dry hands and penis with a fresh disposable towel. Ejaculate into a sterile container.

Liquefaction

Immediately after ejaculation into the collection vessel, semen is typically a semi-solid coagulated mass; however, within a few minutes at room temperature, the semen usually starts to liquefy (become thinner), at which point heterogeneous mixture of lumps will be seen in the fluid; as liquefaction continues, the semen becomes more homogeneous and quite watery; and in the final stages, only small areas of coagulation remain.

Semen viscosity

After the sample has liquefied, its viscosity can be determined by gently aspirating it into a wide- bore, disposable plastic pipette with an approximate 1.5 mm diameter, letting the semen fall by gravity, and measuring the length of any threads that form. A typical sample comes out of the pipette in discrete little drops.

Semen volume

The seminal vesicles and prostate gland make up the majority of the ejaculate's volume, with only a minor contribution from the epididymides and bulb urethral glands. Any evaluation of semen requires accurate volume measurement since it enables estimation of the total number of spermatozoa and non-sperm cells in the ejaculate. Reference limit: If the sample is less than the needed quantity, it is referred to as hypo-spermia, and if it is more than 6.0 ml, it is referred to as hyper-spermia. The lower reference limit for semen volume is 1.5 ml to 6 ml.

Measurement of pH

The balance between the acidic prostatic secretion and the alkaline seminal vesicular secretion, the two main secretions from the accessory glands, is represented by the pH of semen. The pH should be measured after liquefaction at a consistent period, preferably after 30 minutes, but in any case within 1 hour of ejaculation, as it is affected by the loss of CO2 that occurs after production. For typical samples, paper with a pH of between 6.0 and 10.0 should be used. The steps were as follows:

1) Thoroughly mix the semen sample and evenly distribute a drop of semen onto the pH paper. Hold off until the impregnated zones color is uniform (around 30 seconds).
2) To determine the pH, compare the color to the calibration strip. Reference helps to compare the color with the calibration strip, refers pH value of given sample. Reference Limits- There is currently few reference values for the pH of semen from fertile men (consensus value of 7.2 as a lower threshold value to 7.8).

Sperm motility

To minimize the detrimental effects of dryness, changes in pH, or temperature on motility, it is important to test sperm motility inside semen as soon as possible after liquefaction of the sample, preferably within 30 minutes, but in any event within 1 hour, following ejaculation. Mix the semen sample completely. Take a sample of the semen as soon as it has finished mixing to keep the spermatozoa in suspension. Before taking an aliquot of duplicate material, stir the semen sample. For each replicate, create a moist preparation that is 20 micrometres deep. No more than 60 seconds should be given for the sample to wander. Utilise phase-contrast optics to analyse the slide at a 200 or 400x magnification. Verify the proportion of the different motile groups among the 200 or so spermatozoa produced by each replication. Compare the replicate values to see if they are sufficiently close. If so, perform the calculations; if not, generate new samples. To distinguish between spermatozoa with progressive or non-progressive motility and those that are immotile, it is advisable to utilise a simple approach for grading motility. 40% is the reference level for overall motility (PR + NP).

1) Each spermatozoon's level of motility is rated as follows:

i. **Progressive motility (PR):** spermatozoa moving actively, either linearly or in a large circle, regardless of speed.

ii. **Non-progressive motility (NP):** all other patterns of motility with an absence of progression, e.g. swimming in small circles, the flagellar force hardly displacing the head, or when only a flagellar beat can be observed.

iii. **Immotility (IM):** no movement.

Sperm vitality

It is possible to routinely verify the vitality of the sperm by evaluating the cell membranes from all samples; however those with less than 40% progressively motile spermatozoa are very crucial. Since the percentage of dead cells shouldn't exceed (within sampling error) the percentage of immotile spermatozoa, this test can serve as a check on the evaluation of motility. Ordinarily, more viable cells than motile ones are present. By counting the number of spermatozoa with an unbroken cell, the proportion of living spermatozoa is calculated. Membrane swelling caused by hypotonicity or dye exclusion was seen. Based on the idea that defective plasma membranes, such as those found in non-vital (dead) cells, allow the admission of membrane-impermeant stains, the dye exclusion method was developed. The test for hypo- osmotic swelling presumes that

only cells with intact membranes (live cells) will swell in hypotonic solutions. Sperm vitality should be assessed as soon as possible after liquefaction of the semen sample, preferably at 30 minutes, Vitality test using eosin–nigrosin.

Preparing the reagents

1) Eosin Y: dissolve 0.67 g of eosin Y (color index 45380) and 0.9 g of sodium chloride (NaCl) in 100 ml of purified water with gentle heating.

1) Eosin Y: Gently heat 100 ml of filtered water while dissolving 0.67 g of eosin Y (colour index 45380) and 0.9 g of sodium chloride (NaCl).

2) Eosin-nigrosin: combine 100 ml of eosin Y solution with 10 g of nigrosin (colour index 50420).

3) Bring the suspension to a boil before letting it cool to room temperature. Remove gritty and gelatinous pre-capitates by filtering through filter paper (90 g/m2, for example), then store in a sealed dark-glass bottle.

The semen sample should be thoroughly mixed. Take a 50-microliter portion of semen and combine it with an equivalent amount of eosin-nigrosin suspension. Before removing a replicate aliquot, combining the semen sample with eosin-nigrosin, and treating as in step 2 above, mixes the sample once more. Make a smear of each suspension on a glass slide, and then let it dry in the air. Examine as soon as drying is complete or later after mounting with a durable non- aqueous mounting medium. Bright field optics with a magnification of 1000 and oil immersion should be used to inspect each slide. Count the quantity of stained (dead) or unstained (vital) cells using a lab counter. To get a sample inaccuracy that is acceptable, evaluate 200 spermatozoa in each replicate. Calculate the two percentages of important cells from the replicate slides, and then determine their average and difference was determined.

Sperm numbers

The indicators of fertilisation include the total quantity of spermatozoa per ejaculate and the sperm concentration, which are connected to both time to pregnancy and pregnancy rates. To decide the proper dilution and chambers to employ, examine a well-mixed, undilutable mixture of liquid semen on a glass slide under a cover slip. This is often the moist preparation used for motility testing combining semen and preparing fixative dilutions. Adding material to the Makler chamber and letting spermatozoa congregate there, evaluating the samples in ten to fifteen minutes per replication, at least 200 spermatozoa should be counted. Calculating the number of spermatozoa present per millilitre estimating the overall spermatozoa production per ejaculate is done by equation (1) given below:

Number of sperm counted x dilution factor/volume x 1000 = sperm/ml… (1)

Staining methods by Diff- Quik stain

Standard testing protocols recommends a quick staining method to assess sperm morphology. Rapid staining techniques are especially helpful in clinical laboratories when results must be available the same day as the analysis. There are several differential staining set options. Some smears stained quickly have a lot of background colour and might not be as good as those stained with Papanicolaou stain. Fixative reagent (triarylmethane dye soaked in methanol); staining solution 1 (eosinophilic xanthene); and staining solution 2 (basophilic thiazine) are all components of the Reagents-Diff-Quik fast staining kit. The steps are as follows: after dipping slides in triarylmethane fixative (as supplied in the Diff-Quik kit or made as above) for 15 seconds or 95% methanol alone for 1 hour, the air-dried semen smear is fixed. Placing slides will help drain the surplus solution. Slides should be placed upright on absorbent paper to drain any excess solution. Once the fixed semen smear has been stained, dip the slides in rapid stain solutions for 1- 10 seconds and 2- 5-second intervals. Drain the extra solution at each stage by setting slides upright on absorbent paper while running the tap ten to fifteen times to remove any remaining stains.

V. Results and Discussion

The review of 602 semen sample of male patient of infertile couple showed different percentage of sperm vitality. Figure 1 (a), 1(b) and 1(c) shows semen collection and liquefaction and pH measurement. All the semen parameters in different age groups are represented in table: 1 (a), 1(b) and 1(c) given below. Figure 2 (a), 2 (b) shows sperm vitality and sperm count and figure 3 shows morphology of stained healthy sperm. The graphical representation given below in graph 1(a), 1 (b) and 1 (c) represents that the upper middle age group produce better quality of sperm, followed by middle aged group and young individuals. The males of age group 38- 40 have been observed to form good quality of sperm that has high vitality, motility and ability to fertilize ova. The current observations and findings of the present study reveal the following as given below:

(a) (b) (c)

Fig 1 (a) Semen sample in a sterile container, fig 1 (b) Liquefaction of semen sample and fig 1 (c) Evaluate of pH in semen sample

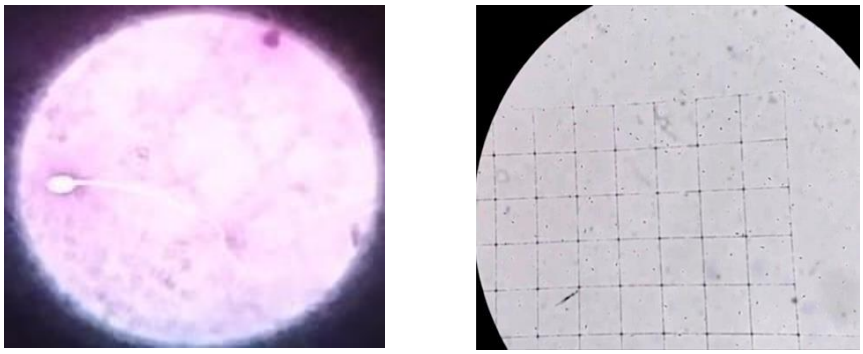


Fig 2 (a) Evaluation of sperm vitality in semen sample, fig 2 (b) Counting of spermatozoa number in makler chamber

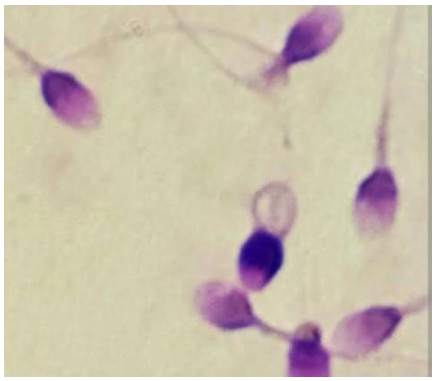


Fig 3 Morphology of stained healthy sperm

Less than 1.5 ml of semen constitutes hypospermia. To transport sperm and allow it to fertilise an egg, enough semen volume is required. So, one of the elements looked at in a semen analysis to gauge fertility is semen volume. A multitude of medical disorders, including as ejaculatory duct obstruction, missing seminal vesicles, and congenital diseases can all be caused by decreased semen volume, including those. In male patients between the ages of 26 and 32, there are 29% more hypospermic cases than normal. Between the ages of 33 and 39, 28% of the male patients had hypospermia. Men between the ages of 40 and 47 who were treated had a 23% hypospermia rate.

Oligospermia (sperm count of semen sample less than 15 million per millilitre), is a defect in sperm production and may be caused by surgically correctable causes like varicocele, exposure to environmental factors such as heat or chemicals that inhibit spermatogenesis, hormonal factors, intrinsic testicular defect, or idiopathic condition. To determine the reason of oligospermia and develop a specific treatment plan, a patient must undergo a thorough evaluation. 35% of male patients between the ages of 26 and 32 were found to have oligospermia. 35% of male patients between the ages of 33 and 39 had oligospermia. 33% of male patients between the ages of 40 and 47 had oligospermia.

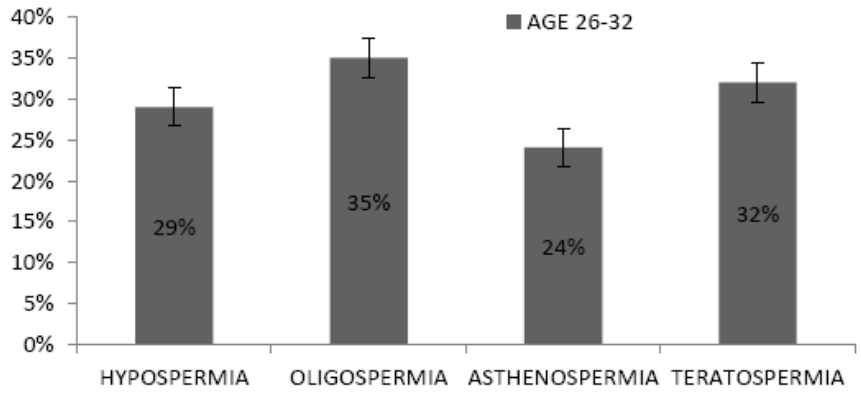
According to WHO standards, asthenospermia is a condition in which a person has sperm that is normally mobile, with values equal to or greater than 40% of both progressive and non- progressive motile

sperm.Sperm with a progressive motility value greater than 32% can only move forward. Asthenozoospermia has a variety of different and poorly understood reasons that can impair sperm motility. It could be brought on by the environment, infections, genetic or immunological changes, etc. However, it is understood that the following conditions, including the presence of Anti-spermatozoa Antigen (ASA), can affect sperm motility. It was discovered that 24% of male patients between the ages of 26 and 32 had asthenospermia. In male patients aged 33 to 39, asthenospermia affected 17% of them. In male patients between the ages of 40 and 47, asthenospermia was present in 9% of cases.

When sperm in an individuals' semen are malformed or faulty, it is known as teratozoospermia or teratospermia. Teratozoospermia is a condition in which sperm exhibits less than 4% of their typical morphology in semen samples. It causes infertility by lowering the sperm's capacity to fertilise the egg or by making it difficult for the sperm to swim to the egg inside a fallopian tube. 32% of male patients between the ages of 26 and 32 had teratospermia. Between the ages of 33 and 39, 26% of the male patients had teratospermia. 20% of male patients between the ages of 40 and 47 had teratospermia.

Table 1.0 (a) Table shows the age wise abnormality of sperm (26-32)

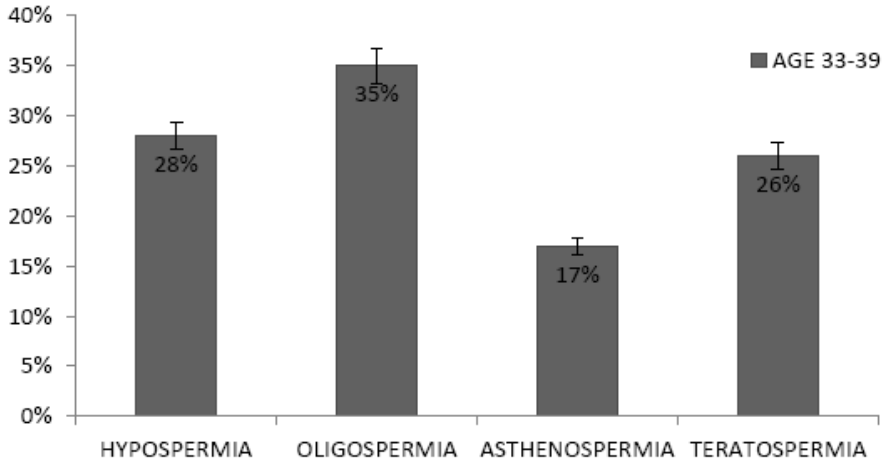
Parameters	Lower Limit	Percentage (%)
1. Volume	1.5 ml	28%
2. pH	7.4	Normal
3. Sperm Count	15 mil/ml	35%
4. Sperm Motility	40%	17%
5. Sperm Morphology	04%	26%



Graph 1.0 (a) Graph shows the percentage of occurrence of Hypo, Oligo, Astheno, Teratospermia patient of age 26-32

Table 1.0(b) Table shows the age wise abnormality of sperm (33-39)

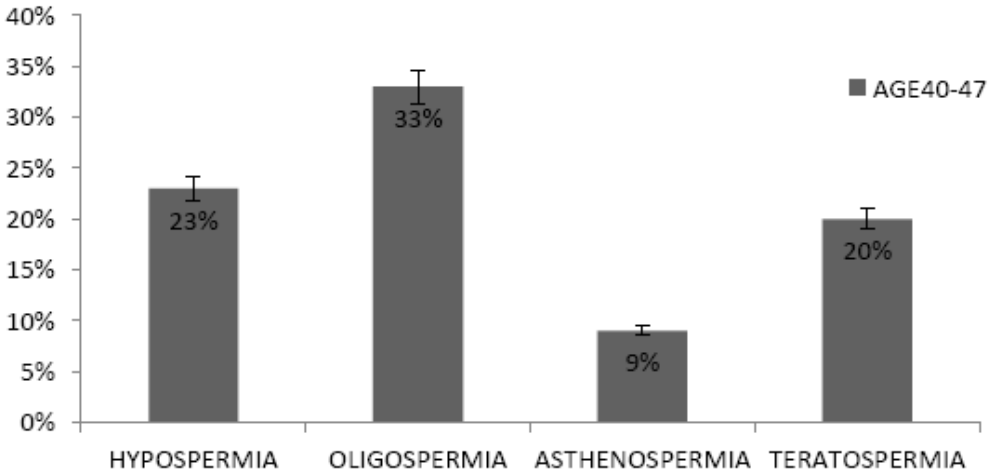
Semen Parameters	Lower Limit	Percentage
1. Volume	1.5 ml	28%
2. pH	7.4	Normal
3. Sperm Count	15 mil/ml	35%
4. Sperm Motility	40%	17%
5. Sperm Morphology	04%	26%



Graph 1.0 (b) Graph shows the percentage of occurrence of Hypo, Oligo, Astheno, Teratospermia patient of age 33-39

Table 1.0 (c) Table shows the age wise abnormality of sperm (40-47)

Semen Parameters	Lower Limit	Percentage
1. Volume	1.5 ml	23%
2. pH	7.4	Normal
3. Sperm Count	15 mil/ml	33%
4. Sperm Motility	40%	09%
5. Sperm Morphology	04%	20%



Graph 1.0 (c) Graph shows the percentage of occurrence of Hypo, Oligo, Astheno, Teratospermia patient of age 40-47

VI. Conclusion

According to the present study, male infertility has a significant impact on psychology and physiology of spouses and is a significant contributor to infertility. It might be for a variety of reasons. Additionally, current research indicates that India is seeing an increase in this trend. As a result, it is urgent to investigate the causes of the rise in male infertility and to take steps to have these causes under control in the near future. Numerous studies have demonstrated that sperm quality declines with ageing. According to our research, sperm quality rises with age. There are many reasons why adult and younger sperm are now of higher quality than middle-

aged sperm. Taking steroids also lowers the quality of sperm. Most males in their 20s and 30s work out in the gym and often consume steroids. Steriod consumers are unhealthy diets that impact sperm and might also have a negative impact on sperm quality. Another significant factor affecting sperm quality is stress. Alcohol consumption and smoking both contribute to sperm loss. Unhealthy lifestyle affects sperm production and motility [11]. A very serious problem for the next generation is declining sperm quality. According to our research, middle-aged men had better sperm quality than adult and younger males. This indicates increase in future male infertility cases and will be a significant problem for infertility as a result, which is clearly understood from the present study.

References

[1]. Cary WH, Hotchkiss RS. Semen appraisal. A differential stain that advances the study of cell morphology.
[2]. Fredricsson B, Sennerstam R. Morphology of live seminal and postcoital cervical spermatozoa and its bearing on human fertility.
[3]. World Health Organization WHO Laboratory Manual for the Examination and Processing of Human SemenWorld Health Organisation Press; 2010
[4]. Hotchkiss RS. Factors in stability and variability of semen specimens—observations on 640 successive samples from 23 men. 1941;45:875–88. [Google Scholar]
[5]. Mantegazza P. Research on human sperm. 1866;25:289–92. [Google Scholar]
[6]. Hühner M. Sterility in the Male and Female and its Treatment. New York: Rebman Company; 1911. [Google Scholar]
[7]. Macomber D, Sanders MB. The spermatozoa count. Its value in the diagnosis, prognosis and treatment of sterility. N Engl J Med. 1929;200:981–4. [Google Scholar]
[8]. Eliasson R Standards for investigation of human semen Andrologie1971:3:49–64.
[9]. Menkveld R, Swanson RJ, Kotze TJ, Kruger TF. Comparison of a discontinuous Percoll gradient method versus a swim-up method: effects on sperm morphology and other semen parameters. Andrologia. 1990;22:152–8. [PubMed] [Google Scholar]
[10]. Kruger TF, Menkveld R, Stander FS, Lombard CJ, Van der Merwe JP, et al. Sperm morphologic features as a prognostic factor in in vitro fertilization. Fertil Steril. 1986;46:1118–23. [PubMed] [Google Scholar]
[11]. World Health Organization. WHO Laboratory Manual for the Examination of Human Semen and Sperm- Cervical Mucus Interaction4th edn. Cambridge: Cambridge University Press; 1999
[12]. Enginsu ME, Dumoulin JC, Pieters MH, Bras M, Evers JL, et al. Evaluation of human sperm morphology using strict criteria after Diff-Quik staining: correlation of morphology with fertilization in vitro. Hum Reprod. 1991;6:854–8. [PubMed] [Google Scholar]
[13]. Kobayashi T, Jinno M, Sugimura K, Nozawa S, Sugiyama T, et al. Sperm morphological assessment based on strict criteria and in-vitro fertilization outcome. Hum Reprod. 1991;6:983–6. [PubMed] [Google Scholar]
[14]. Jeulin C, Feneux D, Serres C, Jouannet P, Guillet-Rosso F, et al. Sperm factors related to failure of human in-vitro fertilization. J Reprod Fertil. 1986;76:735–44. [PubMed] [Google Scholar].