



Seminal parameters among patients visiting infertility clinic at the National Referral Hospital, Bhutan

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ABSTRACT

Introduction: Seminal fluid analysis is the initial investigation for any couple presenting with either primary or secondary infertility. In this study, we aim to study the seminal fluid parameters among Bhutanese patients who presented at the infertility clinic for an initial seminal fluid examination. **Methods:** This retrospective descriptive study was conducted at the Infertility Clinic of the National Referral Hospital, Bhutan from January to December 2022. Using the census sampling technique, data on seminal parameters of 304 patients was extracted from the clinic's register. A descriptive analysis of frequencies and percentages on the above data was performed using IBM Corp's SPSS version 26.0, Armonk, NY). **Results:** The result showed that the majority 217(70.4%) of the patients who came for semen analysis were in the age range between 30 and 39 years. Among 304 patients examined, 72(23.7%) showed normal semen parameters while 232(76.3%) displayed abnormalities in their semen parameters. These abnormalities were further categorised as follows: Azoospermia 14(4.6%), Oligozoospermia 109(35.9%), Asthenozoospermia 23(7.6%) and 86(28.2%) cases were Oligoasthenozoospermia. **Conclusions:** This study provides a scientific data for the first time regarding seminal parameters of patients that visited infertility clinics. This data can serve as a reliable reference and valuable source of evidence for clinicians and future researchers. As the pioneering study of its kind in the country, this study generates fundamental evidence-based data for the global scientific community as well, promoting sustained research involvement in the field of reproductive medicine over the long run.

Keywords: Infertility; Parameters; Semen.

INTRODUCTION

Seminal fluid examination, also known as semen analysis, is a crucial diagnostic tool in the field of reproductive medicine and healthcare¹. It involves the thorough assessment of a man's semen, which contains spermatozoa and various other components present in a man's ejaculate, to evaluate fertility potential and overall reproductive health². The analysis involves a comprehensive assessment of various sperm parameters, including volume, pH, appearance, liquefaction time, total sperm count, sperm concentration, motility, and other characteristics that can impact fertility³.

Indeed, semen analysis is considered as the initial diagnostic test that the clinicians will advise when the infertile couple come to outpatient department⁴. Semen analysis is

also performed for patients undergoing wide range of medical conditions, including fertility assessment, infertility diagnosis, evaluation of treatment effectiveness, reproductive health assessment, post-vasectomy confirmation, and support for clinical research studies⁴. Due to its pivotal role in reproductive and diagnostic medicine, it is considered one of the most important tests for men's health^{5,6}.

According to the register maintained at the outpatient department of Jigme Dorji Wangchuck National Referral Hospital, lots of infertile couples visited outpatient department for fertility issues. However, there was no facility available to perform semen analysis in the country. This deprived the people from availing infertility treatment services within the country. Therefore, the Ministry of Health felt an urgent need to set up a clinic to cater to infertile couples. Finally infertility clinic was established in 2016.

When patients come with fertility issues at outpatient department, the Gynaecologist first send the male for semen analysis. This is to rule out azoospermia. If a male is detected with azoospermia, further evaluation is unnecessary as without

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donor’s semen, fertilization is impossible^{7,8}. Also unnecessary invasive investigations for the female can be prevented which has benefit for patients as well as reduce financial burden for the country. Following semen analysis, the reports will be reviewed by the gynaecologist. If semen reports are normal, then the female partners has to undergo further investigation. Unfortunately, no study was ever conducted to utilize this data and generate basic data for the country.

Hence, this study was conducted to prepare and present our nation’s data to the scientific community, thereby fostering additional research efforts that could ultimately pave the way for the development of preventive measures to address infertility issues within the country.

METHODS

This retrospective descriptive study was conducted at the infertility clinic of Jigme Dorji Wangchuck National Referral Hospital (JDWRH), Bhutan, between January and December 2022, covering a duration of one year. As per the register maintained at the clinic, a total of 304 patients underwent semen analysis for one year. Therefore, using the census sampling technique, all 304 patients recorded in the register were included in our study. From the register, the patient’s seminal parameters including semen volume, pH, appearance, viscosity, liquefaction time, total sperm count, progressive motility, non-progressive motility, immotile and azoospermia were noted and copied into the data documentation sheet. Later on this data was exported to SPSS IBM version 26.0, Armonk, NY.

To maintain anonymity and protect the privacy of the patients, no names or any other types of identifications were used in the data collection process. A unique code number was assigned to each patients and only the code number was used during data entry. Subsequently, only the code number was used if any reference was required and same is practised throughout the analysis and publication of the study. A descriptive analysis was done using frequency distribution and percentages of seminal parameters.

Approval was sought from the Institutional Review Board (IRB) at Khesar Gyalpo University of Medical Sciences of Bhutan vide letter no. IRB/Approval/PN21-021/2022/519. Additionally, study site clearance was obtained from Jigme Dorji Wangchuk National Referral Hospital vide letter no. JDWRH/MERU/01/2021-2022/599.

RESULTS

Three hundred and four patients had undergone semen analysis from January to December 2022 at the Infertility clinic of JDWRH.

The study showed that 214(70.4%) of the patients who underwent semen analysis were in the age range of 30 to 39 years followed by 57(18.8%) between 20 to 29 years and 2(0.7%) patients above the age of 60 years (Table 1).

Annexure I

At the infertility clinic, we adhered to WHO guidelines⁸ for semen preparation, analysis, and reporting, including reference values for semen parameters. The clinic maintains a registry to document the seminal parameters of all individuals who underwent semen analysis.

The seminal fluid examination consisted of the following steps:

Step 1: Semen Collection: On the day of insemination, semen samples were collected in a sterile container at the infertility clinic by masturbation. Immediately after production, semen containers were kept in a hot air oven at 37°C for around 30 minutes to one hour for liquefaction.

Step 2: Macroscopic semen examination: After liquefaction for 30 minutes or a maximum of 60 minutes, a macroscopic examination of semen samples was performed. The volume of the ejaculate was determined by aspirating the liquefied sample into a graduated disposable pipette. Liquefaction time, viscosity, appearance, PH and volume of semen were all recorded in a register.

Step 3: Microscopic examination: With the use of microscope, progressive motility, non-progressive motility and immotile spermatozoa were examined and noted down in the register.

Table 1. Age and occupation of patients who underwent semen analysis at the National Referral Hospital of Bhutan from January to December 2022 (n=304)

Variables	%(n)
Age	
20-29	18.7(57)
30-39	70.4(214)
40-49	8.9(27)
50-59	1.3(4)
60 or above	0.7(2)
Total	100.0(304)
Occupation	
Civil services	28.9(88)
Arm forces	6.3(19)
Agriculture farming	6.9(21)
Private Business	42.4(129)
Private company	8.2(25)
NGO*/INGO†/CSO‡	7.3(22)
Total	100.0(304)

*Non-governmental organization; †International Non-governmental Organization; ‡Civil Society Organization

Amongst different occupational groups, private businesses 129(42.4%) had the highest number coming for semen analysis followed by civil servants 88(28.9%). Whereas from other occupations it was found to be less than 9% (Table 1).

Amongst the patients who came for semen analysis, 203 (66.8%) of them maintained the recommended three to five days of sexual abstinence while 63(20.7%) had less than two days and 38(12.5%) had more than six days of abstinence (Table 2)

Regarding macroscopic semen profile, 241(79.0%) of them had normal semen volume whereas 63(21.0%) had low semen volume. The grey opalescent appearance of semen was the highest 264(86.8%) followed by brown appearance 24(7.9%). The remaining were found to be less than 3% (Table 3). However, the pH of all the samples was found to be normal. Viscosity of semen was found to be normal in 243(91.1%) of the patients. The majority 277(91.1%) of the patients had a normal liquefaction time of less than or equal to 60 minutes and 27(8.9%) of them had more than 60 minutes (Table 2).

According to the microscopic semen profile, 197(64.8%) of the patients had a low sperm count (<35 million/ml) and 93(30.6%) of them had normal sperm count (>35 million/ml) whilst no spermatozoa were found in 14(4.6%) of them. Out of 304 semen analyses, 183(60.2) had normal progressive motility and 107(35.2%) had less sperm motility while 14(4.6) had no spermatozoa in their semen. Regarding the diagnosis, the highest

Table 2. Macroscopic semen profile of patients who underwent semen analysis at the National Referral Hospital of Bhutan from January to December 2022 (n=304)

Variables	% (n)
Volume	
Normal	79.0(241)
Low	21.0(63)
Total	100.0(304)
Appearance	
Grey opalescent	86.8(264)
Brown	7.9(24)
Red	1.6(5)
Yellow	0.7(2)
White	3.0(9)
Total	100.0(304)
pH	
Normal	100.0(304)
Total	100.0(304)
Liquefaction time	
≤ 60 min	91.1(277)
> 60 min	8.9(27)
Total	100.0(304)

was Oligozoospermia found in 109(35.9%) patients followed by Oligoasthenozoospermia 86(28.2%), Normozoospermia 72(23.7), Asthenozoospermia 23(7.6) and the least was Azoospermia 14(4.6%) (Table 3).

Table 3. Microscopic semen profile of patients who underwent semen analysis at the National Referral Hospital of Bhutan from January to December 2022 (n=304)

Variables	% (n)
Total sperm count (million/ml)	
0	4.6(14)
<35	64.8(197)
≥35	30.6(93)
Total	100.0(304)
Progressive motility (PR in %)	
0 %	4.6(14)
<32%	35.2(107)
≥32%	60.2(183)
Total	100.0(304)
Diagnosis	
Normozoospermia	23.7(72)
Oligozoospermia	35.9(109)
Asthenozoospermia	7.6(23)
Oligoasthenozoospermia	28.2(86)
Azoospermia	4.6(14)
Total	100.0(304)

DISCUSSION

The present study included 304 patients who came for semen analysis and their seminal parameters were evaluated from January to December 2022 at the infertility clinic, National Referral Hospital, Bhutan.

In our study, a majority of patients who underwent semen analysis fell within the age range of 30 to 39 years. This particular age group has consistently appeared as a common finding in various male fertility studies in the region^{3,4,6}. Our findings closely resembled the findings from previous research, where it has been noted that the maximum number of patients coming to avail semen analysis services fall within this age range^{4,6}. The factors affecting the semen parameters in the country were not considered in our study, the significance of this age range in fertility issues cannot be correlated with the semen parameters at the moment. Interestingly, there exists a lot of evidence supporting the association between male age and reduced fertility after the age of 60 in other countries^{9,10}, but no such evidence existed for the age range 30 to 39.

Regarding the requirement of certain number of days for sexual abstinence, our study found a majority of the patients following the recommended abstinence days of three to five days. Sexual abstinence before performing semen analysis is an important factor to be considered for standardization of semen reports around the globe⁸. It has been proven by previous studies that sexual abstinence of less than two days, the semen volume might become low, lowering the total sperm count. More than five days of sexual abstinence, most of the spermatozoa might become immotile affecting progressive and non-progressive percentage¹⁰. Hence, the recommended duration of sexual abstinence is fixed at three to five days⁸. Out of many possible factors contributing to non-adherence to recommended abstinence duration, one potential factor of absence of clear guidance from the healthcare providers stood out. The other factor might be attributed to inconveniences and lack of responsibility on the part of the patients⁵.

We categorized the patients in our study into six different occupational groups, considering the common professions available within the country. Our study found that a majority of patients were from the private business sector followed by civil servants. These findings were similar to other studies^{3,4,5}, where maximum patients came from private business sectors. The plausible reasons they cited were stress at work, less free time together, unfavourable working condition and exposure to hazardous agents at work^{3,4}. But the reason was not explored in our study and therefore cannot elaborate more. Hence a comprehensive study on this topic could be an interesting area for future research.

Regarding macroscopic semen analysis, the majority of the patients in our study had normal semen volume, appearance, pH, viscosity, and liquefaction time. Our findings in this regard closely resembled the findings from various other studies conducted in the regions^{9,10,12}. These studies concluded that the semen volume is one of the most important factors associated with infertility. Numerous other studies have also proven that abnormalities in semen volume can impact fertility^{13,14}. Similarly, the abnormal appearance, pH, viscosity and liquefaction time of the semen can indicate certain underlying pathologies which must be treated accordingly¹², as these might be the only reasons preventing successful fertilization^{12,13}.

Our study revealed that a maximum number of patients had low total sperm count. Total sperm count is derived by multiplying semen volume by sperm concentration⁸. So the low total sperm count can be attributed to either low volume or concentration. We found that the majority of the patients in our study had normal semen volume, and therefore the low total sperm count in our study is due to low sperm concentration alone. It's worth noting that the total sperm count is a significant factor in determining male fertility, with a higher count correlating to a greater likelihood of achieving pregnancy, as reported in previous studies¹⁴. Furthermore, the assessment of sperm motility, another crucial parameter, is closely associated with successful pregnancy outcomes⁸.

The study revealed only about one-fourth (23.7%) of patients had normal semen parameters. This low rate of normal semen parameters was anticipated, given that all the male participants in our research were patients with fertility issues. These findings warrant a broader investigation into the general population of the country so that an overall idea of seminal parameters in the population can be known.

Strength of the study

This is the first study of its kind in the country is the major strength of this study. The study produced valuable evidence-based basic data for the country's patients, with potential implications for future reference within the scientific community. Building upon the foundational reference data, a more detailed and comprehensive investigation can be conducted. This, in turn, would offer momentum for informing policy planning, resource allocation, and establishing a centre of excellence in fertility treatment and prevention in the country.

LIMITATIONS

There were several limitations to consider in this study. The primary limitation is the clinic's inability to conduct a comprehensive semen examination. This was mainly attributed to the absence of a state-of-the-art facility and adequately trained staff. The absence of cutting-edge laboratory, diagnostic equipment, and trained staff may have impacted the precision and reliability of the results obtained. Furthermore, the study did not account for other confounding factors that have the potential to impact semen parameters, such as diabetes, hypertension, smoking, alcohol consumption, and history of sexually transmitted infections. Given the limitations mentioned above, the findings and conclusions of this study may not be fully comparable with those of other studies that had access to more advanced equipment and well-trained staff. Future research should prioritize the development of robust research designs to facilitate meaningful comparisons with findings from other studies.

CONCLUSIONS

In conclusion, the data on the seminal parameters of patients who visited the infertility clinic over the past year represents a valuable resource for clinicians and researchers alike. This dataset not only provides a reference point for current clinical practices but also serves as a foundational source of evidence for future studies in the field of reproductive health. The significance of this data extends beyond its immediate utility. It underscores the importance of addressing infertility issues in Bhutan and highlights the commitment to improving healthcare services in the country through continued research.

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AUTHORS CONTRIBUTION

Following authors have made substantial contributions to the manuscript as under:

- GD:** Concept, design, data collection and analysis, manuscript editing, writing and review
- KW:** Concept, design, data collection and analysis, manuscript editing, writing and review
- TJ:** Concept, design, data collection and analysis, manuscript editing, writing and review
- ND:** Manuscript editing, writing and review
- ND:** Manuscript editing, writing and review
- MD:** Manuscript editing, writing and review
- NW:** Manuscript editing, writing and review

Author agree to be accountable for all respects of the work in ensuring that questions related to the accuracy and integrity of any part of the work are appropriately investigated and resolved.

CONFLICT OF INTEREST

None

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