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Original Article

Effect of COVID-19 Infection on Semen Parameters of Males

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ABSTRACT

Background: Corona Virus Disease 2019 [COVID-19] is a pandemic caused by a new coronavirus named severe acute respiratory syndrome coronavirus 2 [SARS-CoV-2; previously known as 2019-nCoV]. Other effects on systems other than respiratory system are reported. However, the effects on semen parameters is not well-addressed.

The aim of the work: The aim of this investigation was to detect the effect of COVID 19 infection on different semen parameters.

Patients and Methods: This six-months prospective case-control investigation on 66 cases. They were selected from Helwan University Hospitals and approved by research and ethics committee on December 23, 2021 [Approval No. 79-2021]. Participants have been divided into two groups, with COVID-19 confirmed via reverse transcription-polymerase chain reaction [RT-PCR] on nasal and pharyngeal swabs. Semen analysis was conducted at the start and after three months.

Results: After three months of COVID-19 infection, cases showed a statistically significant increase in semen volume [p=0.03], sperm concentration [p=0.001], total motility [p=0.001], and rapid progressive motility [p=0.001], along with a significant reduction in abnormal forms [p=0.002], while non-progressive motility demonstrated insignificant change [p=0.7]. Comparisons among COVID-19 cases and the control group revealed insignificant distinctions in semen volume [p=0.10] or sperm motility [p=0.06, 0.4, 0.6]. However, semen concentration was significantly higher in the control group [p=0.001], and abnormal forms were significantly increased in the COVID-19 group [p=0.000].

Conclusion: COVID-19 infection has negative impact in most of cases. This effect is reversible after three months after infection.

Keywords: SARS-CoV-2; Fertility; Semen analysis; Testicular function.

INTRODUCTION

Corona Virus Disease 2019 [COVID-19] is a pandemic caused by a new coronavirus known as severe acute respiratory syndrome coronavirus 2 [SARS-CoV-2; previously designated as 2019- nCoV]. The primary route of SARS-CoV-2 transmission is direct person-to-person contact through respiratory droplets. Research indicates that SARS-CoV-2 utilizes angiotensin-converting enzyme 2 [ACE2] as a receptor in host cells to initiate infection [1].

Fever, drugs and prolonged sexual abstinence could elucidate the changes in semen parameters. The impacts may be reversible and are not exclusive to COVID-19. Consequently, it has been advised from the onset of the pandemic to postpone assisted reproductive technology procedures for three months following SARS-CoV-2 infection, particularly in instances of fever. In fever absence, impact of the novel coronavirus on semen remains uncertain [2].

SARS-CoV-2 gains host cells by attaching its spike protein [S1] to severe acute respiratory syndrome coronavirus 2 receptors prevalent on respiratory epithelial cells, particularly type II alveolar epithelial cells. In addition to respiratory epithelium, severe acute respiratory syndrome coronavirus 2 receptors are present in other organs, including the upper esophagus, ileal enterocytes, cardiac cells, proximal tubular cells of the kidney, and urothelial cells of the bladder [3].

An online database study revealed that severe acute respiratory syndrome coronavirus 2 is prominently expressed in Leydig cells and cells within the seminiferous ducts of the testes. Consequently, it has been hypothesized that severe acute respiratory syndrome coronavirus 2 could be targeting the testes, leading to possible damage to testicular tissues and heightening the possibility of male sexual dysfunction and infertility [4].

The involvement of the testicles diminishes testosterone secretion, a hormone that has recently been shown to offer protection against COVID-19 [5,6].

This research aimed to assess the impact of COVID-19 infection on several semen parameters.

PATIENT AND METHODS

This six-months prospective case-control investigation included 66 cases at Helwan University Hospitals. It was approved on December 23, 2021 [Approval No. 79-2021] by the research and ethics committee. COVID-19 infection had been confirmed via RT-PCR on nasal and pharyngeal swabs. Semen analysis was conducted at the start and after three months.

Categorization:

The investigated population has been stratified into two groups: Group A [33 males with confirmed COVID-19] and Group B [33 males without COVID-19 as controls].

Inclusion criteria: Sexually active men, aged between 18 and 65, confirmed previous diagnosis with COVID-19 in the past 3 months and proven recovery from COVID-19 [negative NP-swab].

Exclusion criteria: Any male with hormonal disturbances, malignancy, immunosuppressive disease, azoospermia, COVID 19 cases more than 6 months and cases with palpable varicocele, males who were unable to express an informed consent, males with drug history that may affect results and pyospermia.

Sample size calculation: The sample size has been determined utilizing Open Epi program version 3, referencing a previous investigation by **Li *et al.*** [7], which reported that SARS-CoV-2 has been identified via qualitative real-time polymerase chain reaction [RT-PCR] in the semen of six out of thirty-eight cases [15.8%]. With a confidence interval set at ninety-five percent a test power of eighty percent, and a group ratio of one to one, the minimum required sample size for this investigation has been calculated to be sixty-six cases, divided into two groups of thirty-three cases each.

All patients were subjected to the following:

The evaluation included a comprehensive history covering personal, past medical, surgical, and family history, along with a complete physical examination comprising general assessment [including body proportions and secondary sexual characteristics] and a genital examination [penile and testicular evaluation].

Investigation to confirm past COVID 19 infection:

The World Health Organization's guidelines have been followed in defining laboratory confirmation for COVID-19 as a positive result for SARS-CoV-2 in a RT-PCR assay of pharyngeal and nasal swabs. The swabs have been retested based on the National Health Care System criteria till the NP [nasal-pharyngeal]-swab was negative.

Semen analysis:

Masturbation has been utilized to gather semen samples following a recommended period of abstinence of three to five days. Following gathering, the specimens have been permitted to liquefy at room temperature for thirty minutes prior to analysis. Each semen sample underwent microscopy. All individuals who exhibited significant pus cells [not less than five pus cells/high power field] or excessive white blood cells [more than one million per milliliter] on microscopy, which might indicate that an infection, have been eliminated from the investigation. The World Health Organization [WHO] recommended that sperm motility, count, and morphology percentages be objectively assessed through microscopic examination. All data has been recorded, analyzed, and compared.

Ethical consideration:

The institutional review board approved the investigation's protocol. Each participant who participated in the investigation provided written informed consent. The investigation has been conducted with respect for personal privacy and confidentiality. The data that has been collected hasn't been utilized for any other purpose.

Statistical Analysis: Data have been gathered, refined, encoded, and input into Statistical Package for Social Sciences [IBM SPSS] version 23. The quantitative data have been given as means, standard deviations, and ranges for parametric data, and as medians and inter-quartile ranges [IQR] for non-parametric data. Qualitative variables have been additionally provided as numbers and percentages. The comparison of groups of qualitative data has been conducted utilizing Chi-square test and/or Fisher's exact test where the expected count in any cell was below five. The comparison of two independent groups with quantitative data and parametric distribution has been conducted utilizing the Independent t-test, whereas the comparison with non-parametric distribution has been carried out utilizing the Mann-Whitney test. P-value more than 0.05: Not significant [NS], P-value less than 0.05: Significant [S], P-value less

than 0.01: Highly significant [HS]. A smaller P value indicates greater significance of the outcomes.

RESULTS

Socio-demographic characteristics: A statistically insignificant distinction has been observed among both groups as regard age, education, and residence [**Table1]**.

Clinical characteristics: Most of the case group [78.8%] had COVID-19 symptoms, [81.8%] had more than 3 months of symptom duration and [78.8%] were isolated at home [**Table 2]**.

Comparing semen parameters at the beginning between the two studied groups: As regard semen parameters among the two studied groups at the beginning of the study, a statistically significant lower concentration, total motility, rapid progressive and non-progressive motility has been observed among the COVID 19 cases than the control group p= 0.001, 0.001,0.001 0.02 correspondingly, while regarding the abnormal forms and immotile sperms, they were statistically significant greater in the COVID cases than the control group p= 0.000, 0.001 correspondingly. But an insignificant distinction regarding volume p=0.5 has been observed [**Table 3]**.

Semen parameters in COVID 19 cases at the beginning of the infection and after 3 months of infection: A statistically significant increase in semen volume, sperm concentration, total and rapid progressive motility has been observed in COVID-19 cases after 3 months of infection p= 0.03, 0.001, 0.001, 0.001 respectively. While a statistically significant reduction in abnormal forms p=0.002 has been observed. Where, non-progressive motility had statistically insignificant distinction p=0.7 [**Table 4]**.

Comparing between COVID 19 group [case group] following 3 months of the infection and the control group: A statistically insignificant distinction in semen volume, total motility, rapid progressive and non-progressive motility has been observed among the COVID 19 group [case group] following 3 months of infection and control group p= 0.10, 0.06, 0.4, 0.6 correspondingly. On the other hand, a statistically significant elevation in semen concentration has been observed in the control than the COVID 19 group [case group] after 3 months of infection p=0.001. Moreover, a highly statistically significant increase in abnormal forms has been observed in the COVID group following 3 months than the control group p=0.000 [**Table 5].**

Table [1]: Socio-demographic characteristics of the studied group

Variables		Case [n=33]	Control [n=33]	Test	p-value
Age [years]	Mean ± SD Median [Range]	32.4 ± 8.1 31 [20-50]	34.5 ± 6.8 33 [21-51]	1.1	0.2
Education	Low [number=1] Moderate [number=11] High [number=54]	1 [3.03%] 6 [18.18%] 26 [78.79%]	0 [0.0%] 5 [15.15%] 28[84.85%]	1.1	0.6
Residence	Rural [number =27] Urban[number =39]	11[33.33%] 22 [66.67%]	16[48.48%] 17[51.52%]	1.6	0.2

Table [2]: Clinical characteristics of the case group

Variables		Case number=33 [%]
Symptoms presence	No Yes	7 [21.2%] 26 [78.8%]
Symptoms duration	Within 3 months More than 3 months	27 [81.8%] 6 [18.2%]
Type of quarantine	Home Hospital	26 [78.8%] 7 [21.2%]

Table [3]: Comparing semen parameters at the beginning of the study between the two studied groups

Variables		Case [n=33]	Control [n=33]	Test	p-value
Volume	Mean ± SD Median [Range]	3.03 ± 1.3 3 [0.6-7]	3.1 ± 0.49 3 [2.5-4]	0.6	0.5
Concentration	Mean ± SD Median [Range]	30.4 ± 25.1 20 [4-97]	92 ± 43.2 90 [19-237]	7.1	**0.001****
Total motility	Mean ± SD Median [Range]	44.7 ± 16.1 47 [10-80]	62.6 ± 8.1 60 [45-80]	5.7	**0.001****
Rapid Progresssive	Mean ± SD Median [Range]	23.3 ± 11.7 25 [0.0-50]	37.3 ± 8.4 40 [23-60]	5.5	**0.001****
Non-progressive	Mean ± SD Median [Range]	21.4 ± 6.7 20 [10-38]	24.9 ± 5.7 25 [15-40]	2.2	**0.02***
Immotile sperms	Mean ± SD Median [Range]	55.1 ± 16.3 53 [20-90]	37.6 ± 8.3 40 [20-55]	35.3	**0.001****
Abnormal forms	Mean ± SD Median [Range]	94.84 ± 13.04 98 [25-100]	87.96 ± 6 91 [75-95]	5.5	**<0.001****

Table [4]: Semen parameters in COVID 19 cases at the beginning of the infection and after 3 months of infection

Variables		At the beginning	After 3 months	p-value
Volume	Mean ± SD Median [Range]	3.03 ± 1.3 3 [0.6-7]	3.7 ± 1.7 3.5 [0.8-9]	0.03*
Concentration	Mean ± SD Median [Range]	30.4 ± 25.1 20 [4-97]	56.9 ± 26.2 50 [15-142]	**0.001****
Total motility	Mean ± SD Median [Range]	44.7 ± 16.1 47 [10-80]	57.9 ± 10.7 55 [44-80]	**0.001****
Rapid progressive	Mean ± SD Median [Range]	23.3 ± 11.7 25 [0.0-50]	35.6 ± 8.4 35 [20-60]	**0.001****
Non-progressive	Mean ± SD Median [Range]	21.4 ± 6.7 20 [10-38]	21.9 ± 6.6 20 [10-35]	**0.7**
Immotile sperms	Mean ± SD Median [Range]	55.1 ± 16.3 53 [20-90]	42.5 ± 11.1 45 [20-56]	**0.001****
Abnormal forms	Mean ± SD Median [Range]	94.84 ± 13.04 98 [25-100]	93.06 ± 14.37 96 [15-99]	**0.002***

Table [5]: Comparing between COVID 19 group [case group] after 3 months of the infection and the control group

	Variables	Case [Number=33]	Control [Number=33]	test	p-value
Volume	Mean ± SD Median [Range]	3.7 ± 1.7 3.5 [0.8-9]	3.1 ± 0.49 3 [2.5-4]	1.5	0.10
Concentration	Mean ± SD Median [Range]	56.9 ± 26.2 50 [15-142]	92 ± 43.2 90 [19-237]	3.9	**0.001****
Total motility	Mean ± SD Median [Range]	57.9 ± 10.7 55 [44-80]	62.6 ± 8.1 60 [45-80]	1.9	**0.06**
Rapid progressive	Mean ± SD Median [Range]	35.6 ± 8.4 35 [20-60]	37.3 ± 8.4 40 [23-60]	0.8	**0.4**
Non-progressive	Mean ± SD Median [Range]	21.9 ± 6.6 20 [10-35]	24.9 ± 5.7 25 [15-40]	1.9	**0.6**
Immotile sperms	Mean ± SD Median [Range]	42.5 ± 11.1 45 [20-56]	37.6 ± 8.3 40 [20-55]	2.1	**0.4***
Abnormal forms	Mean ± SD Median [Range]	93.06 ± 14.37 96 [15-99]	87.96 ± 6 91 [75-95]	5.400	**<0.001****

DISCUSSION

There are several potential mechanisms by which COVID-19 might have an impact on the male reproductive system**:** [i] Interaction of severe acute respiratory syndrome coronavirus 2 and ACE2 might negatively impact testicular/sperm function**.** [ii] Immune reactions triggered by COVID-19 that harm the male reproductive system**.** [iii] Impact of hormone levels on male fertility [8].

In our investigation, a statistically significant decreased semen concentration, total motility, rapid progressive and non-progressive motility has been observed among the case than the control group. Additionally, the abnormal form and immotile sperms showed statistically significant increasing among the case in comparison to the control group. Multiple studies have repeatedly confirmed that SARS-CoV-2 infection has negative effects on male fertility and semen parameters [9,10].

The findings of **Pazir *et al.*** [11] were different**.** Their findings suggested that cases with a history of mild COVID-19 had significantly reduced sperm motility and total motile sperm counts. It implies that COVID-19 may cause some spermatogenesis damage, which is supported by autopsies performed on COVID-19 cases [12]**.** It should be noted that the SARS-CoV-2 virus might not have a direct effect on the impairment of semen parameters. Stress and other psychopathological factors can have an adverse effect on fertility**.** Additionally, sperm quantity and quality, as well as morphology and motility, can all be negatively impacted by fever**.** Long-term COVID sequelae are likely influenced by additional factors, some of which are unknown at this time [13].

Additionally, **Holtmann *et al.*** [14] revealed that cases with a moderate infection had a sperm quality impairment. However, their sample size was small [20 cases and 18 control]. Their case group involved only an inadequate cases number in mild and moderate conditions and they only had two cases in life-threatening condition**.**

Regarding the distinction between the case group at the beginning of the study and after 3 months, our study shows that a statistically significant increase in semen volume, semen concentration, total motility and rapid progressive motility has been observed. While a statistically significant reduction in immotile and abnormal forms has been observed. Additionally, non-progressive motility had no statistically significant distinction. This is in line with **Dipankar *et al.*** [15] who found that semen quality during COVID attack showed poor quality regarding semen viscosity, agglutination and vitality.

We found that a statistically insignificant distinction in semen volume, total motility, rapid progressive motility and non-progressive has been observed between case group after 3 months of the investigation and control group. Also we found that a statistically significant increase in semen concentration has been observed among the control than the case group following 3 months of infection. Moreover, a statistically significant elevation in abnormal forms has been observed among the case group after 3 months than the control group. Agreeing with **Stigliani *et al.*** [16] who stated that when semen samples are taken three months or more after infection, COVID-19 disease has no detrimental ;8effects on the quality of the semen or the male reproductive potential. However, **Gacci *et al.*** [17] concluded that one-fourth of men who recovered from COVID-19 showed

oligo-crypto-azoospermia.

In our study, regarding relation between symptoms duration and semen analysis after 3 months of the study among the case group; a statistically significant elevation in rapid progressive has been observed among cases with an illness period of more than 3 months than cases with an illness period within 3 months. Regarding other semen parameters, there was no statistically significant distinction.

Agreeing with **Guo *et al.*** [18] who showed that, in comparison to controls, the sperm concentration, motility, and progressive motility of the cases were all significantly diminished at the 1st sampling, conducted a median of fifty-six days' post-hospital release. The overall count of sperms, sperm concentration, and the quantity of motile and progressively motile spermatozoa were significantly elevated in the 2nd sampling, conducted at a median of eighty-four days' post-hospital discharge, compared to the 1st sampling in the twenty-two cases analyzed. This indicates a potential recovery of sperm numbers over time after COVID-19 recovery.

These results suggest that the COVID-19 aftereffects might last for one spermatogenic cycle given that the human spermatogenic cycle is thought to last roughly 74 days. However, **Ruan *et al.*** [19] who classified the cases, regardless of the severity of the disease into groups with long [>90 days] and short [< 90 days] recovery times, and the semen was analyzed accordingly. Cases who took longer to recover had significantly fewer total sperm than those who took less time. Additionally, although not statistically significant, the total motility and progressive motility both demonstrated a declining trend in between these two teams**.** They hypothesize that the normal time course of semen turnover may be the cause of the decline in semen quality with a lag time**.**

Limitations: The sample size was insufficient, the monitoring duration was inadequate, and the investigation has been conducted at a single center, therefore we can't do generalization to the information.

Conclusion: COVID-19 infection has negative effect on male fertility and semen parameters which is reversible after one cycle of spermatogenesis.

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