How to Cite:
Sunnu, C. C., Tribowo, J. A., Juwita, W., Rezano, A., I'tishom, R., Supardi, S., & Margiana, R. (2022). The long-term impact of COVID-19 infection on semen quality of the COVID-19 survivors. *International Journal of Health Sciences*, *6*(S6), 925–936. https://doi.org/10.53730/ijhs.v6nS6.10501

The long-term impact of COVID-19 infection on semen quality of the COVID-19 survivors

Christian Christopher Sunnu
Andrology Specialist Program, Faculty of Medicine, Universitas Airlangga/Dr. Soetomo General Hospital Surabaya, Indonesia | Dr. Soetomo General Academic Hospital, Surabaya, Indonesia
0000-0002-6062-8778

Jefry Albari Tribowo
Andrology Specialist Program, Faculty of Medicine, Universitas Airlangga/Dr. Soetomo General Hospital Surabaya, Indonesia | Dr. Soetomo General Academic Hospital, Surabaya, Indonesia
0000-0001-7865-7051

Widya Juwita
Andrology Specialist Program, Faculty of Medicine, Universitas Airlangga/Dr. Soetomo General Hospital Surabaya, Indonesia | Dr. Soetomo General Academic Hospital, Surabaya, Indonesia
0000-0001-8454-0327

Andri Rezano
Andrology Specialist Program, Faculty of Medicine, Universitas Airlangga/Dr. Soetomo General Hospital Surabaya, Indonesia | Dr. Soetomo General Academic Hospital, Surabaya, Indonesia | Department of Biomedical Sciences, Faculty of Medicine, Universitas Padjadjaran, Sumedang, Indonesia
0000-0003-2550-8794

Reny I'tishom
Department of Biomedical Sciences, Faculty of Medicine, Universitas Airlangga, Surabaya, Indonesia
*Corresponding author email: ritishom@fk.unair.ac.id
0000-0002-9971-7786

Supardi Supardi
Dr. Soetomo General Academic Hospital, Surabaya, Indonesia
0000-0001-8497-3536

Manuscript submitted: 9 March 2022, Manuscript revised: 27 May 2022, Accepted for publication: 18 June 2022

Ria Margiana
Andrology Specialist Program, Faculty of Medicine, Universitas Airlangga/Dr. Soetomo General Hospital Surabaya, Indonesia | Dr. Soetomo General Academic Hospital, Surabaya, Indonesia | Department of Anatomy, Faculty of Medicine, Universitas Indonesia, Jakarta, Indonesia
0000-0002-6747-0117

***Abstract*---**Background. Several studies have shown the short-term effects of COVID-19 on semen quality, but not as many for the long-term effects. This follow-up study aims to explore the long-term effects of COVID-19 on male fertility by comparing semen parameters of patients recovering from COVID-19 infection. Methods. Fourteen subjects aged >18 years old who had a history of resolved COVID-19 infection with previous evidence of laboratory-confirmed positive SARS-CoV-2 nasopharyngeal PCR swab result, and had a previous history of infertility after COVID-19 infection based on the evidence of the semen analysis within 6 months or longer were recruited in this study. Analysis of the semen parameter of all fourteen subjects was performed, including semen volume, pH, sperm concentration, total, progressive, non-progressive, and immotile motility percentage, morphology percentage, leukocyte, erythrocyte, and immature sperm cells parameters. The semen analysis obtained from this current semen analysis was compared with individual baseline semen analysis results, which were obtained in the resolution phase of COVID-19 infection, 6 months or longer from the current semen analysis. Results. Mean pH decreased significantly in follow-up semen analysis, with baseline vs. follow-up 7.9 vs 7.4 (p=0.002; 95%CI=0.3-0.67). Mean sperm concentration also decreased significantly, with baseline vs. follow-up 78.5 million/mL vs. 37.4 million/mL (p=0.013; 95%CI=7.5-74.5). Lastly, mean immature sperm count is also decreased significantly, with baseline vs. follow-up 2.5 million/mL vs. 0.4 million/mL (p=0.006; 95%CI=0.37-4.03). No significant difference was documented for other semen parameters. Conclusion. After 6-7 months of recovery from COVID-19 symptoms, follow-up semen analysis showed reduced sperm pH, sperm concentration, and immature sperm count. These findings indicate that COVID-19 infection could have a longer-term impact on male infertility.

***Keywords*---**COVID-19, semen parameter, sperm concentration, resolution phase, long-term.

Introduction

In December 2019, a series of viral pneumonia cases emerged in Wuhan, China. Further research shows evidence that the cause of the pneumonia cases was a novel coronavirus, which at the time was named 2019-nCoV.[1] Later on February 2021, WHO renamed the disease as coronavirus disease 2019 (COVID-19). The virus also was immediately given a new name, severe acute respiratory syndrome

coronavirus 2 (SARS-CoV-2), by the Coronavirus Study Group of the International Committee on Taxonomy of Viruses (ICTV) based on phylogenetic analysis of related coronaviruses.[2,3] As of 3 October 2021, according to WHO, global confirmed cases of COVID-19 reached 234,551,981 cases with 4,796,171 confirmed deaths.[4] Since the appearance of COVID-19 in Indonesia in March 2020, there have been many cases, reaching their peak in July 2021 with 350,723 weekly cases. As of 4 October 2021, 4,227,932 confirmed cases were reached with cumulative confirmed deaths of 142,641.[5] Further epidemiological studies showed that in Jakarta, as the capital city, the incidence of COVID-19 was the highest, reaching 34.3% during the period of observation. East Java, with Surabaya as the capital city, followed in second place with 12.3% incidence rate. The occurrence seemed highest in males (56.5%) on productive ages (31-59 years old) (57.5%).[6]

Recently, various COVID-19 variant of concern (VOC) has been detected, one of them being the B.1.1.529 variant, widely known as the Omicron variant. The Omicron variant was firstly reported on November 24th, 2021 in South Africa.[7] The first case of the Omicron variant in Indonesia was reported on December 16th, 2021, widely spreading across the country reaching its peak on February 2022. As of March 16th, 2022, 5,927,550 confirmed cases were reached with cumulative confirmed deaths of 152,975 deaths in Indonesia.[8] East Java still showed a high occurrence of weekly COVID-19 incidence reaching 57,9 incidence per 100,000 citizens, though not as many as previously documented. As of March 17th, 2022, 114,342 cumulative confirmed cases in Surabaya were reported.[8,9] Though most of the Omicron variant cases contributed to mild cases of COVID-19, its communicability was significantly higher than previous variants. Numerous studies have shown multiple long-term effects of COVID-19, including fatigue, pain syndromes, respiratory and cardiovascular symptoms, and even gastrointestinal symptoms such as nausea and ageusia. Long-term hypercoagulability due to COVID-19 may also lead to coronary thrombosis, increasing the risk of death even after the acute phase of COVID-19.[10-11] SARS-CoV-2 appeared to be capable of persistence in certain tissues, confirmed by detection of SARS-CoV-2 RNA in various body tissues including neural cells, even after the active phase of the disease.[12]

Regarding the effects of COVID-19 on male reproductive functions, there were also reports of decreasing testosterone levels and sperm quality in the patients during active disease.[13] Multiple mechanisms of testiculopathy caused by COVID-19 have been postulated, including direct infection of testicle via ACE2 receptors, long-term fever leading to increased testicle temperature, steroid therapy, autoimmune orchitis caused by inflammatory cytokines in seminiferous tubules, and oxidative stress injury caused by the inflammatory processes.[14-15] Recent evidence from Belgium showed that sperm abnormality strongly correlates with SARS-CoV-2 IgG antibody against spike 1 and the receptor-binding domain of spike 1 in serum. The study also reported 3 subjects developed high levels of anti-sperm antibodies (ASA).[16] As for SARS-CoV-2 in the sperm itself, there have been contradictory results with several studies reporting that SARS-CoV-2 RNA is undetectable in the sperm both in active infection and post-infection, but some others reporting high levels of RNA.[17] While the vertical communicability of SARS-

CoV-2 is currently inconclusive, it is apparent that COVID-19 was quite impactful to the quality of the semen.[16,17]

Several studies have shown the short-term effects of COVID-19 on semen quality. A multicenter study consisting of 69 cases conducted in Turkey showed that during the recovery phase of COVID-19, it was observed that the semen quality of the patients was significantly decreased compared to before COVID-19 infection.[18] Analysis of 38 men semen samples in Shangqiu Municipal Hospital showed that 6 out of 38 men had results positive for SARS-CoV-2 in semen during acute or recovery phase of COVID-19 infection.[19] Similar result was found in a prior study conducted in Surabaya, that from 34 men diagnosed with infertility in the resolution phase of COVID-19 infection, one of them showed positive results in seminal SARS-CoV-2 PCR, after 121 days since diagnosis of COVID-19 based on nasopharyngeal PCR swab.[20] Longer-term effects of COVID-19 on males have also been reported in several case reports. A 36 years-old man with no history of any kind of infertility pre-COVID-19 was diagnosed with oligoasthenoteratozoospermia one month post-COVID-19, only improving after four months post-COVID-19 without reaching pre-COVID-19 semen analysis values.[21] A similar case was observed in a 33 years-old man experiencing reversible cryptoozoospermia 4 months after COVID-19 infection. The condition reversed to normal 3 months afterward.[22] Many studies have reported short-term effects of COVID-19 on male fertility but not as many for the long-term effects. This study aims to explore the long-term effects of COVID-19 on male fertility further with a larger study sample.

Materials and Methods

Study Design & Participants

COVID-19 survivors previously evaluated by semen analysis were included in this follow-up study. All patients were outpatients treated in the Andrology Unit of Dr. Soetomo General Hospital and Professor Nidom Foundation, both located in Surabaya, Indonesia, and recently diagnosed with infertility based on the semen analysis by the time of the resolution of COVID-19 symptoms by Supardi, et al.[20] Inclusion criteria for the subjects are patients aged >18 years old, had a history of resolved COVID-19 infection with previous evidence of laboratory-confirmed positive SARS-CoV-2 nasopharyngeal PCR swab result and had a previous history of infertility after COVID-19 infection based on the evidence of the semen analysis within 6 months or longer. Patients with any history of primary or secondary infertility before COVID-19 infection, history of testicular infection or malignancy, and patients currently using hormonal drugs which could affect spermatogenesis are excluded.

Data Collection

Patients who met the inclusion criteria were contacted by phone number obtained from the hospital. Informed consent was obtained before the study. All patients were asked to abstain for 2-7 days before the sampling. The sampling was performed 2 times for each participant. The baseline sampling, which was already performed before by Supardi, et al.[20], was performed immediately by the time of resolution of COVID-19 symptoms. The follow-up sampling was performed within

6 months or more from the previous baseline sampling. Semen samples obtained were then to be examined manually by single study personnel according to WHO 2010 criteria.[23]

Statistical Analysis

The patients were grouped according to COVID-19 severity based on WHO classification.[24] Statistical analysis was performed globally and within each group, respectively, if sufficient. Semen volume, pH, sperm concentration, total, progressive, non-progressive, and immotile motility percentage, morphology percentage, leukocyte, erythrocyte, and immature sperm cells parameters obtained from the current semen analysis were to be statistically compared with individual previous results, which were obtained by Supardi, et al.[20] All statistical analyses were performed using SPSS version 25.0. Normality test was based on the result of the Shapiro-Wilk Test. Comparison between current and previous sperm analysis results performed with Paired-T Test or Wilcoxon Test according to the result of normality test.

Results

Out of 34 subjects from the previous study[20], 14 subjects agreed to participate further in this study. Twelve of the subjects had mild symptoms of COVID-19, while the remaining 2 subjects had asymptomatic COVID-19 infection. The semen analyses were obtained 6 months or more, ranging 6-7 months, after the previous analyses for all subjects. Due to the small number of subjects in the asymptomatic group, the final statistical analysis was only performed globally. Epidemiologic characteristic data is provided in Table 1.

Table 1
Demographic and Clinical Characteristic of Subjects

Parameter	Subjects (n=14)	Range
Age, mean±SD (years)	36.1±6.3	27-48
Marriage status (n, %)		
Married	14 (100%)	
Not married	0	
Spouse's age, mean±SD (years)	34.4±6.1	26-45
Duration of marriage, mean±SD (years)	6.7±4.8	0.16-14
Infertility (n, %)		
Primary	5 (35.7%)	
Secondary	2 (14.3%)	
Hormone Profile		
LH, mean±SD (mIU/mL)	3.92±1.77	1.61-7.31
FSH, mean±SD (mIU/mL)	5.85±4.17	1.51-16.32
Testosterone, mean±SD (ng/dL)	478.76±183.08	248.70-950.50

Semen analysis results could be observed in Table 2. There was no significant difference between volume, motility, morphology, leukocyte count, and erythrocyte count parameter between baseline and follow-up sperm analysis.

However, a slight decrease in morphology parameter is observed between baseline vs. follow-up (1.2% vs. 0.7%). Only a single subject with erythrocyte was documented in follow-up sperm analysis, previously not documented in the baseline. A similar result is observed with the leukocyte count parameter. Only a single subject in each group (baseline and follow-up each in different subjects) with positive leukocyte in the semen samples.

Table 2
Baseline vs. Follow-up Semen Analysis Result

Parameter	Baseline	Follow-up	Mean difference (95% CI)	p
Volume (mL)	3.1±1.1	3.1±1.4	0.06 (-0.55-0.67)	0.944
pH	7.9±0.3	7.4±0.2	0.5 (0.3-0.67)	0.002†
Motility				
PR (%)	49.6±26.5	46.8±25.7	2.8 (-6.4-11.9)	0.528*
NP (%)	7.0±5.4	13.6±9.6	-6.5 (-13.2-0.1)	0.053*
Total Motility (%)	56.6±28.4	60.4±24.6	-3.8 (-13.2-5.5)	0.345
IM (%)	29.1±19.6	32.4±19.8	-3.3 (-17.1-10.5)	0.612*
Concentration (million/mL)	78.5±85.0	37.4±55.2	41.1 (7.5-74.5)	0.013†
Morphology (%)	1.2±1.4	0.7±0.8	0.5 (-0.2-1.2)	0.142
Leukocyte (cells/mL)	0.07±0.27	0.02±0.08	0.05 (-0.11-0.2)	0.655
Erythrocyte (cells/mL)	0	0.03±0.11	-0.03 (-0.09-0.03)	0.317
Immature Sperm (million/ml	2.5±3.1	0.4±0.3	2.1 (0.37-4.03)	0.006†

All statistical analyses were performed using Wilcoxon signed-rank test, due to abnormal distribution according to Shapiro-Wilk Test, except for PR, NP, and IM motility parameters.
*Statistically analysed using Paired-T Test.
†Significant difference observed with p<0,05

Mean pH decreased significantly in follow-up semen analysis, with baseline vs. follow-up 7.9 vs 7.4 (p = 0.002; 95% CI = 0.3-0.67). Mean sperm concentration also decreased significantly, with baseline vs. follow-up 78.5 million/mL vs. 37.4 million/mL (p = 0.013; 95% CI = 7.5-74.5). Lastly, mean immature sperm count is also decreased significantly, with baseline vs. follow-up 2.5 million/mL vs. 0.4 million/mL (p = 0.006; 95% CI = 0.37-4.03).

Discussion

In regards to the effect of COVID-19 on semen quality of men recovering from COVID-19 infection, multiple shreds of evidences of short-term effects on semen quality have been documented in several cases.[21,22]. Mannur, et al. reported a case of decreased sperm concentration, decreased progressive motility, and also a decrease in normal morphology at 1 month after COVID-19 infection. Sperm concentration and motility recovered only partially at 4 months after COVID-19 infection, while sperm morphology recovery is not yet observed. A similar case was reported by Basourakos, et al. The patient suffered from cryptozoospermia with no sperm in the semen analysis, 3 months after COVID-19 infection. Even at 7 months after COVID-19 infection, there was only partial recovery, showing

improvement in sperm concentration and motility, but with poorer sperm motility compared with pre-COVID-19 infection sperm analysis.[22]

Interestingly, our findings on sperm-analysis of 14 subjects at 6-7 months after COVID-19 infection could bring a new perspective on the longer-term impact of COVID-19 infection on semen analysis. We observed a significant decrease of sperm concentration even after 6-7 months, with a slight decrease of progressive sperm motility and normal sperm morphology, albeit being not statistically significant. These findings are similar to Basourakos, et al. case. A meta-analysis conducted by Tiwari, et al. involving 7 studies with 934 participants reported a statistically-significant decrease in total sperm count accompanied by the decrease of sperm concentration and progressive motility, albeit statistically insignificant.[25] These observed decreases in the semen parameters could be due to the testicular injury from the SARS-COV-2 orchitis triggered by the immune response in opposition to the seminiferous epithelium, as previously documented on Severe Acute Respiratory Syndrome (SARS) cases.[26]

Another interesting finding is that the subjects' semen samples pH decreased significantly in follow-up semen analysis with mean pH 7,9 from baseline to 7,4 at follow-up. We hypothesized that the inflammation caused by COVID-19 could play a part in this. The alkaline pH of semen is mainly sourced from the secretion of the seminal vesicles. Decreased secretion of the semen samples due to various causes could be related to lower semen samples pH.[27] It is found that ACE2 receptor is abundantly expressed in the seminal vesicle, which indicates the possibility of the infection of the seminal vesicle.[28] Direct damage to the seminal vesicles or secondary inflammatory response might contribute to the decrease of pH. Meanwhile, the decreased pH is still above the WHO referenced range (≥7,2), hence this current finding might not clinically be the cause of the poorer quality of semen. However, it would be important to note that oligozoospermia and asthenozoospermia could be observed in seminal pH lower than 7,2.[29]

As mentioned before, we observed a statistically insignificant decrease in progressive sperm motility. From 5 studies of the meta-analysis conducted by Tiwari, et al., the progressive sperm motility of those subjects recovered from COVID-19 was 1,73% less than those without COVID-19. One of the included studies in the meta-analysis, conducted by Guo, et al., mentioned that the significant influence of COVID-19 in decreased motility was only observed in COVID-19 infection needing hospitalization.[30] Regarding decreased motility, our findings are also consistent with the meta-analysis result. We observed progressive sperm motility at follow-up decreased by 2,8% from the baseline while being statistically insignificant, which might be because the enrolled subjects are with mild and asymptomatic cases only. Although we observed higher total motility at follow-up, it is due to the increased non-progressive sperm motility, while the progressive sperm motility is decreased.

Guo, et al., also reported significantly higher abnormal sperm morphology at 84 days after discharge, with a mean difference of 2% from 56 days after discharge.[30] We observed 0.5% decrease in sperm morphology from baseline after 6-7 months. The difference that was not statistically significant might be due to the smaller sample size and due to the normal sperm morphology of the baseline being

already low (1,2%). Our findings might prove that the decrease of normal sperm morphology is already occurred and persevered through the 6-7 months period.

We also hypothesized that decreased sperm concentration, progressive motility, and normal sperm morphology observed in this study might be connected to hormonal factors. Leydig cells, which produce testosterone, have high expressions of ACE2. Direct SARS-CoV-2 infection of Leydig cells may lead to testicular hypofunction, especially in severe diseases.[31] In a postmortem examination of the testes from 12 COVID-19 patients conducted by Yang M, et al., the mean number of Leydig cells per tubule cross-section in COVID-19 testes was significantly lower compared with 5 non-COVID-19 patients' testes (2,2 vs. 7,8; p<0,001). In addition, only 3 out of 12 COVID-19 patients' testes showed normal signs of spermatogenesis.[32] The transient decrease of testosterone secretion in COVID-19 patients could be related to damage to the Leydig cells, as observed in a study involving 40 males aged 21-65 years with COVID-19 infection.[33]

Regarding the severity of COVID-19, it seems that hypogonadism is positively related to the severity of infection.[34,35] In one study involving 55 COVID-19 pneumonia-recovered, post-hospitalized male patients with an average of 60 days of discharge, the prevalence of testosterone deficiency was 50,9%, and the cause was mainly hypogonadotropic (60,7%). Recovered COVID-19 pneumonia patients had significantly lower total testosterone and calculated free testosterone and higher testosterone to LH ratio than the control group.[34] Longer-term observation of 7 months cohort study of 121 men who recovered from COVID-19 showed improvement of testosterone levels from the majority of the patients (87,6%) compared to hospital admittance. However, 55% of the patients still had total testosterone levels suggestive of hypogonadism, with 9,9% of them having even lower testosterone levels than admittance.[35] In this study, one of the limitations is that we couldn't measure hormone levels that could be compared between baseline and 6-7 months follow-up. As the long-term hormonal impact was yet to be elucidated, we also suggest that further research should incorporate hormonal analysis comparing between before and after infection of COVID-19, and also with the follow-up results.

Besides Leydig cells, Sertoli cell's involvement with SARS-CoV-2 could also produce a potential problem of ASA formation. Sertoli cells damage can lead to a breakdown of the blood-testicular barrier which functions to preserve the spermatozoa from immune responses. Disruption of the blood-testicular barrier could be the trigger to the development of ASA which can lead to fertility impairment.[36] IL-6 was thought to be responsible for the infiltration of leukocytes to the testes which disrupted of the blood-testicular barrier.[37] As mentioned before, recent evidence from Belgium reported 3 subjects of recovered COVID-19 infection developed high levels of anti-sperm antibodies (ASA).[16] As with the hormonal impact, the risk of blood-testicular barrier disruption positively correlated with viral load.[38] While there is not much evidence yet about ASA, it is one of the possible longer-term impacts of COVID-19 on male fertility.

A thorough proteomics comparative analysis was performed by Ghosh S, et al. This analysis compared semen protein profile between 10 healthy fertile men and 17 COVID-19 recovered men. Of the 48 expressed proteins, 21 proteins were

downregulated and 27 were upregulated in COVID-19-recovered males. The consequence of proteomic profile changes to the major reproductive functions includes the downregulation of sperm-oocyte recognition, testosterone response, cell motility regulation, adhesion regulation, extracellular matrix adhesion, and endopeptidase activity in COVID-19 recovered men. Specifically, downregulation of semenogelin 1 (SEMG1) from the seminal vesicle decreased sperm motility, downregulation of prosaposin (PSAP) could correlate with decreased size of the testis, epididymis, and prostate gland, and lastly, downregulation of Outer dense fiber protein-2 (ODF-2) could lead to asthenozoospermia, due to lack of functionality of the flagellum.[39] Prosaposin is also a protein with a contribution to sperm-oocyte binding, fertilization, and embryo development. According to a study involving 166 participants, high seminal prosaposin concentrations were significantly associated with fertilization rate and good embryo proportion in IVF.[40] Summarized from this evidence, COVID-19 could also impact male fertility due to changes in the regulation of protein expression.

The limitation of our study is due to the small sample size included. While 34 subjects initially participated in the baseline analysis. Only 14 subjects agreed to participate in the follow-up analysis. A larger sample size could provide higher quality data. Numerous hints of evidence of short-term impacts on semen quality have been observed in various situations about the impact of COVID-19 on the semen quality of men recovering from COVID-19 infection. This essay provides compelling evidence that Follow-up semen study after 6-7 months of recovery from COVID-19 symptoms revealed decreased sperm pH, sperm concentration, and immature sperm count. These results suggest that male infertility may be affected by COVID-19 infection over a longer period of time. Immature sperm have a different morphology from mature spermatozoa, which can explain why the number of immature sperm increased after 6-7 months.[21,22,41,42,43] The duration between baseline and follow-up analysis of 6-7 months could shed a light on the lingering effects of COVID-19 infection on male infertility. Longer observation time would provide the time needed to recover from the COVID-19 impact on male infertility.

Conclusion

After 6-7 months of recovery from COVID-19 symptoms, follow-up semen analysis showed reduced sperm pH, sperm concentration, and immature sperm count, while no significant difference is observed with volume, motility, morphology, leukocyte count, and erythrocyte count parameter. These findings indicate that COVID-19 infection could have a longer-term impact on male infertility.

Data Availability

The data used to support this study are available from the corresponding author on reasonable request.

Conflicts of Interest

The authors declare that there was no conflict of interest.

Acknowledgments

The authors would like to express gratitude to all staff of the Andrology Unit, Dr. Soetomo General Hospital Surabaya, and Department of Biomedical Science, Faculty of Medicine, Universitas Airlangga, especially all participants, colleagues, and acquaintances involved in this study for their help to accomplish this project. This study funded by Universitas Airlangga grant for COVID-19 Research (Grant Number: 664/UN.3.14/PT/2020) and Professor Nidom Foundation, Surabaya, Indonesia (Grant Number: 006/PNF-RF/10/2020).

References

1. Huang C, Wang Y, Li X, Ren L, Zhao J, Hu Y, et al. Clinical features of patients infected with 2019 novel coronavirus in Wuhan, China. Lancet. 2020; 395: 497-506.
2. Jiang S, Shi Z, Shu Y, Song J, Gao GF, Tan W, et al. A distinct name is needed for the new coronavirus. Lancet. 2020; 395: 949.
3. Wu Y, Ho W, Huang Y, Jin DY, Li S, Liu SL, et al. SARS-CoV-2 is an appropriate name for the new coronavirus. Lancet. 2020; 395; 949-50.
4. WHO. Weekly operational update on COVID-19 issue no. 74. [Internet]. 2021 [cited 2021 Oct 14]. Available from: https://www.who.int/publications/m/item/weekly-operational-update-on-covid-19---4-october-2021
5. WHO. Indonesia situation. [Internet]. 2021 [cited 2021 Oct 14]. Available from: https://covid19.who.int/region/searo/country/id
6. Hikmawati I, Setiyabudi R. Epidemiology of COVID-19 in Indonesia: common source and propagated source as a cause for outbreaks. J Infect Dev Ctries. 2021; 15: 646-52.
7. Amalia H. Omicron penyebab COVID-19 sebagai variant of concern. Jurnal Biomedika dan Kesehatan. 2021; 4: 139-41.
8. WHO. Coronavirus Disease 2019 (COVID-19) Situation Report, 88. [Internet]. 2022 [cited 2022 Mar 18]; Available from: https://cdn.who.int/media/docs/default-source/searo/indonesia/covid19/external-situation-report-88_16-march-2022.pdf?sfvrsn=3409040d_5
9. Satgas COVID-19 Kota Surabaya. Peta sebaran COVID-19 di Kota Surabaya. [Internet]. 2022 [cited 2022 Mar 18]; Available from: https://lawancovid-19.surabaya.go.id/
10. Lopez-Leon S, Wegman-Ostrosky T, Perelman C, Sepulveda R, Rebolledo PA, Cuapio A, et al. More than 50 long-term effects of COVID-19: a systematic review and meta-analysis. Scientific Reports. 2021; 11: 16144.
11. Arevalos V, Ortega-Paz L, Fernanez-Rodriguez D, Jimenez-Diaz VA, Rius JB, Campo G, et al. Long-term effects of coronavirus disease 2019 on the cardiovascular system, CV COVID registry: a structured summary of a study protocol. Plos One. 2021; 16: e0255263.
12. Fave RFD, Polisini G, Giglioni G, Parlavecchio A, Dell'Atti L, Galosi AB. COVID-19 and male fertility: taking stock of one year after the outbreak began. Archivio Italiano di Urologia e Andrologia. 2021; 93: 115-9.

13. Proal AD, VanElzakker MB. Long COVID or Post-acute Sequelae of COVID-19 (PASC): An overview of biological factors that may contribute to persistent symptoms. Front Microbiol. 2021; 12: 1-24.
14. Fathi M, Vakili K, Aliaghaei A, Nemattolahi S, Peirovi T, Shalizar-Jalali A. Coronavirus disease and male fertility: a systematic review. Middle East Fertil Soc J. 2021; 26: 26.
15. Bhattacharya K, Mukhopadhyay LD, Goswami R, Dutta S, Sengupta P, Irez T, et al. SARS-CoV-2 infection and human semen: possible modes of contamination and transmission. Middle East Fertil Soc J. 2021; 26: 18.
16. Donders GGG, Bosmans E, Reumers J, Donders F, Jonckheere J, Salembier G, et al. Sperm quality and absence of SARS-CoV-2 RNA in semen after COVID-19 infection: a prospective, observational study and validation of the SpermCOVID test. Fertility and Sterility. 2022; 117: 287-96.
17. Nguyen TT, Hulme J, Tran HD, Vo TK, Vo GV. The potential impact of COVID-19 on male reproductive health. J Endocrinol Invest. 2022; Available from: https://doi.org/10.1007/s40618-022-01764-z
18. Erbay G, Sanli A, Turel H, Yavuz U, Erdogan A, Karabakan M, et al. Short-term effects of COVID-19 on semen parameters: a multicenter study of 69 cases. Andrology. 2021; 9: 1060-5.
19. Li D, Jin M, Bao P, Zhao W, Zhang S. Clinical characteristics and results of semen tests among men with coronavirus disease 2019. JAMA Network Open. 2020; 3: e208292.
20. Supardi S, Nidom RV, Sisca EM, Tribowo JA, Kandar PS, Budiharto JM, et al. Novel investigation of SARS-CoV-2 in COVID-19 survivors' semen in Surabaya, Indonesia. [Internet]. 2021 [cited 2021 Oct 14]. Available from: https://www.medrxiv.org/node/420760.external-links.html
21. Mannur S, Jabeen T, Khader MA, Rao LSS. Post-COVID-19-associated decline in long-term male fertility and embryo quality during associated reproductive technology. QJM Int J Med. 2021; 114: 328-30.
22. Basourakos SP, Schlegel PN. Impact of coronavirus disease 2019 on semen parameters. Asian J Urol. 2021; https://doi.org/10.1016/j.ajur.2021.06.004
23. WHO. WHO laboratory manual for the examination and processing of human edition. 5th ed. Geneva: WHO; 2010.
24. WHO. COVID-19 clinical management: living guidance. WHO; 2021.
25. Tiwari S, Niranjan KC, Thapa S, Ghimire A, Bijukchhe S, Sah GS, et al. Semen parameters in men recovered from COVID-19: a systematic review and meta-analysis. Mid East Fertil Soc J. 2021; 26: 44.
26. Xu J, Qi L, Chi X, Yang J, Wei X, Gong E, et al. Orchitis: a complication of severe acute respiratory syndrome (SARS). Biol Reprod. 2006; 74(2):410–6.
27. Aziz N. The importance of semen analysis in the context of azoospermia. Clinics. 2013; 68: 35-38.
28. Hesari FS, Hosseindazeh SS, Sardroud MAAM. Review of COVID-19 and male genital tract. Andrologia. 2021; 53: e13914.
29. Zhou J, Chen L, Li J, Li H, Hong Z, Xie M, et al. The semen pH affects sperm motility and capacitation. PLoS ONE. 2015; 10: e0132974.
30. Guo T, Sang M, Bai S, Ma H, Wan Y, Jiang X, et al. Semen parameters in men recovered from COVID-19. Asian J Androl. 2021; 23: 479-83.
31. Nassau DE, Best JC, Kresch E, Gonzalez DC, Khodamoradi K, Ramasamy R. Impact of the SARS-CoV-2 virus on male reproductive health. BJU Int. 2021. Available from: doi:10.1111/bju.15573

32. Yang M, Chen S, Huang B, Zhong J, Su H, Chen Y, et al. Pathological findings in the testes of COVID-19 patients: clinical implications. Eur Urol Focus. 2020; 6: 1124-9.
33. Claude EEM, Ghislaine BS, Vicky AM, Anne OB, Arnaud NMJ, Mesmin DY, et al. Transient hypogonadism induced by SARS-CoV-2 during COVID-19 in men aged under 65 years. OJEMD. 2022; 12: 1-8.
34. Aboelnaga MM, Abdelrazek A, Abdullah N, Shaer ME. Late impact of COVID-19 pneumonia on testosterone levels in recovered, post-hospitalized male patients. J Endocrinol Metab. 2021; 11: 76-82.
35. Salonia A, Pontillo M, Capogrosso P, Gregori S, Carenzi C, Ferrara AM, et al. Testosterone in males with COVID-19: a 7-month cohort study. Andrology. 2021; 2021: 1-8.
36. Haghpanah A, Masjedi F, Alborzi S, Hosseinpour A, Dehghani A, Malekmakan L, et al. Potential mechanisms of SARS-CoV-2 action on male gonadal function and fertility: current status and future prospects. Andrologia. 2020; 00: e13883
37. Montano L, Donato F, Bianco PM, Lettieri G, Guglielmo A, Motta O, et al. Air pollution and COVID-19: a possible dangerous synergy for male fertility. Int J Environ Res Public Health. 2021; 18: 1-21.
38. Moshrefi M, Ghasemi-Esmailabad S, Ali J, Findikli N, Mangoli E, Khalili MA. The probable destructive mechanisms behind COVID-19 on male reproduction system and fertility. J Assist Reprod Genet. 2021; 38: 1691-708.
39. Ghosh S, Parikh S, Nissa MU, Acharjee A, Singh A, Patwa D, et al. Semen proteomics of COVID-19 convalescent men reveals disruption of key biological pathways relevant to male reproductive function. ACS Omega. 2022; 7: 8601-12.
40. Xu C, Cai J, Liu L, Ren J. Prosaposin in seminal plasma on the day of oocyte retrieval is associated with normal fertilization and embryo development in in vitro fertilization cycles. PeerJ. 2019; 7: e8177
41. Tribowo JA, Tanojo TD, Supardi S, Pakpahan C, Siswidiyanto EB, Rezano A et al. The impact of the coronavirus disease-19 pandemic on sexual behavior of marriage people in Indonesia. Open Access Macedonian Journal of Medical Sciences. 2021;9:440-445. https://doi.org/10.3889/oamjms.2021.6277
42. Sisca EM, Rezano A, Pakpahan C, Supardi S, Agustinus A, Darsini N, Wen MD, Kirana T, Margiana R. Preliminary report of profile FSH and LH of the COVID-19 survivors in Surabaya, Indonesia. ijhs [Internet]. 2022 Apr. 15 [cited 2022 Jul. 3];6(S3):2433-51. Available from: https://sciencescholar.us/journal/index.php/ijhs/article/view/6054
43. Budiharto JM, Margiana R, William W, Supardi S, Narulita P. Relationship between spermatogenesis, DNA fragmentation index, and teratozoospermia index. ijhs [Internet]. 2022 Apr. 10 [cited 2022 Jul. 3];6(S3):1827-3. Available from: https://sciencescholar.us/journal/index.php/ijhs/article/view/5808
44. Rinartha, K., & Suryasa, W. (2017). Comparative study for better result on query suggestion of article searching with MySQL pattern matching and Jaccard similarity. In *2017 5th International Conference on Cyber and IT Service Management (CITSM)* (pp. 1-4). IEEE.
45. Suryasa, I. W., Rodríguez-Gámez, M., & Koldoris, T. (2022). Post-pandemic health and its sustainability: Educational situation. *International Journal of Health Sciences*, *6*(1), i-v. https://doi.org/10.53730/ijhs.v6n1.5949