Original Article

HEALTH SCIENCES
MEDICINE

DOI: 10.32322/jhsm.1138691
J Health Sci Med 2022; 5(5): 1371-1377

Examining the long-term effects of COVID-19 on the umbilical cord

Fatih Taş[1], Mehmet Yılmaz[2], Fikri Erdemci[3], Fırat Aşır[3], Engin Deveci[3]

[1] Siirt University, Faculty of Medicine, Department of Histology and Embryology, Siirt, Turkey
[2] Siirt University, Faculty of Medicine, Department of Obstetrics and Gynecology, Siirt, Turkey
[3] Dicle University, Faculty of Medicine, Department of Histology and Embryology, Diyarbakır, Turkey

Cite this article as: Taş F, Yılmaz M, Erdemci F, Aşır F, Deveci E. Examining the long-term effects of COVID-19 on the umbilical cord. J Health Sci Med 2022; 5(5): 1371-1377.

ABSTRACT

Introduction: It is known that COVID-19 in pregnancy causes some negative consequences. Although some studies have been conducted on the possible effects of COVID-19 seen in late pregnancy, its effects in the previous trimesters are not clearly known. This study aimed to examine the umbilical cords of pregnant women who did not have COVID-19 and those who had in the second and third trimesters, after delivery using histopathological and immunohistochemical methods.

Material and Method: The study included 27 pregnant women who had never had COVID-19 (n:9), who had had COVID-19 in the second trimester (n:9) and had had COVID-19 in the third trimester (n:9). After delivery, sections were taken from the umbilical cords of the pregnant women and examined with histopathological and immunohistochemical (VEGF and vimentin antibodies) methods. H-scores were determined for statistical analysis of immunohistochemical staining results. Group means were analyzed using the non-parametric Kruskal Wallis Test.

Results: In cases that had COVID-19 in the third trimester of pregnancy, histopathological findings were more significant than in the other groups. Hemorrhage, thinning of the tunica intima layer, and deterioration in its integrity were observed in the umbilical vascular structures of this group. VEGF and vimentin expression levels were higher in the third-trimester group than in the other groups.

Conclusion: The COVID-19 disease has both acute and long-term effects. The presence of histopathological and immunohistochemical findings in the umbilical cord during the third trimester of pregnancy supports this information. Moreover, the high levels of expression of VEGF and vimentin in the umbilical cords of pregnant women may contribute to the understanding of the pathogenesis of COVID-19 and the post-acute effects of these proteins.

Keywords: COVID-19, umbilical cord, VEGF, vimentin, long COVID

INTRODUCTION

The novel coronavirus disease (COVID-19) has emerged due to the Sars-CoV2 virus and has spread to many countries, causing an outbreak (1). The disease may progress asymptomatically as well as may lead to serious life-threatening conditions such as sepsis (2). Although the most frequently affected system due to this disease is the respiratory system, organs in many systems such as nervous, circulatory, digestive, and urogenital systems are also adversely affected. Damages that occur in the organs may occur in the acute period as well as in the later periods (3). The COVID-19 disease results in recovery in many cases whereas in some cases, although the PCR test result turns negative, some symptoms may persist. This condition is defined as "post-COVID-19 syndrome" or "long COVID" (4).

Pregnant women are among the risk groups affected by the COVID-19 disease. When this disease occurs during pregnancy, it can cause negative effects on both maternal and infant health (5). The COVID-19 infection during the active period of pregnancy leads to immunohistochemical and histopathological degenerative effects on the placenta (6). In addition, it is known that pregnant women experience mental complications such as anxiety, depression, and obsessive-compulsive disorder during the Covid-19 pandemic (7). In order to prevent these effects, necessary measures should be taken to reduce the rate of Sars-CoV2 infection during pregnancy and perinatal periods. Also, pregnant patients should be monitored closely (5, 8).

Corresponding Author: Fatih Taş, ftas85@yahoo.com
Received: 30.06.2022 **Accepted:** 02.08.2022

The umbilical cord has an important role in fetal development (9). It is stated that perinatal and intrapartum complications are associated with umbilical cord abnormalities (10). Inflammation in the umbilical cord is defined as funisitis (11). Infection in the umbilical cord may cause neonatal mortality or morbidity and unhygienic practices performed during delivery lead to the development of the disease (12). COVID-19 infection can also cause perinatal transmission if the correct hygienic measures are not taken (13).

Vascular endothelial growth factor (VEGF) is a specific mitogen for vascular endothelial cells. VEGF, also known as vascular permeability factor, increases the proliferation of vascular endothelial cells, microvascular permeability, and angiogenesis (14). Angiogenesis is involved in many physiological and pathological processes. The VEGF family plays a significant role in the development of the vascular system through angiogenesis and is also involved in tissue repair (15). Umbilical cord cells widely express VEGF and these proteins play a role in the growth of the umbilical cord (16). It is known that VEGF levels increase in feline infectious peritonitis (FIP), an infection caused by coronaviruses (17).

Vimentin is an intermediate filament (IF) protein of mesenchymal cells and plays a significant role in organ homeostasis (18). One of the organs in which this protein is expressed and takes part is the umbilical cord (19). Vimentin is also a regulator of proteins involved in cell signaling and migration (18). It is thought that these proteins work as a cytoprotective structure that prevents the movement of some viruses into the cytoplasm (20). Despite this information, the functions of vimentin are not fully known and further studies are needed.

We could not find any information in the literature on how the umbilical cord is affected in pregnant women who have had COVID-19 infection in the second and third trimesters. This research aims to examine the umbilical cords of pregnant women who had not had COVID-19 and those who had had it in the second and third trimesters, after delivery with histopathological and immunohistochemical (VEGF and vimentin antibodies) methods.

MATERIAL AND METHOD

The study was carried out with the permission of Siirt University Non-Interventional Clinical Research Ethics Committee (Date: 13.04.2022, Decision No: 42191). The procedures applied in the study were carried out in line with the ethical rules and the principles of the Declaration of Helsinki. Participants included in the research were informed and a written informed consent form was taken.

A total of 27 pregnant women, nine who had had and recovered from COVID-19 in the second trimester of pregnancy, nine who had had and recovered from COVID-19 in the third trimester, and nine who had never had a COVID-19 infection, were included in the study. Pregnant women with a secondary disease or any chronic disease were not included in the study. Tissues (umbilical cord) were taken from pregnant women who applied to the Department of Obstetrics and Gynecology of Siirt Training and Research Hospital, right after delivery.

Histological Follow-up

Tissues taken from the groups were put into 10% buffered neutral formalin and kept for 16 hours for fixation. Then, the fixed tissues were washed with running water for about 12 hours and the formalin solution was eliminated. Then, the tissues were incubated in alcohols of 30%, 50%, 70%, 80%, 90%, and 96%, respectively, for 8 hours. The dehydration process of the tissue was completed after they were kept in absolute alcohol (99.9%) for 2X20 minutes. After the dehydration process, the tissues were kept in xylol for 2X15 minutes to make the tissues transparent and discard the alcohol from the tissues. The tissues then were incubated in melted paraffin containing 50% xylol in an incubator at 58°C for approximately 1.5 hours. Then the tissues were put into pure paraffin and left for approximately 4 hours for paraffin to penetrate the tissues. Then, the tissues embedded in liquid paraffin were kept at room temperature to dry and sections of 5 µm were taken from the dried tissues using a microtome (Leica RM 2265, Germany).

Hematoxylin-Eosin Staining Protocol

Sections kept in xylol in two series of 15 minutes were deparaffinized. Then, the sections were passed through the decreasing alcohol series for about 5 minutes, ending with the distilled water step. The sections were then incubated in Harris hematoxylin for 8 minutes. Sections were taken under the running water and kept here for 5 minutes, then immersed in 1% acid-alcohol solution to ensure differentiation and washed with distilled water for 5 minutes. After washing, the sections were kept in eosin-floxin solution for 2 minutes for counterstaining. Then, the sections were passed through increasing alcohol series, kept in xylol for two series of 15 minutes, and mounted with entellan.

VEGF and Vimentin Immune Staining Methods

Sections with a thickness of 5 µm were first kept in xylol for 2x15 minutes. Sections recovered from paraffin in xylol were then kept in decreasing alcohol series for 5 minutes each and taken into distilled water for a while. Then, the sections were put into EDTA solution and kept in the microwave oven for approximately 3

minutes. Sections taken from the microwave were left at room temperature for 15 minutes for cooling. At the end of incubation, the sections were placed in distilled water again and then dried. The parts with the tissues were drawn with hydrophobic pencils. Then, PBS-T was added and the sections were kept for 3x15 minutes. During these procedures, special care was taken to keep the immunohistochemistry box moist. After the PBS-T on the sections was discarded, hydrogen peroxide solution was dripped onto them and the sections were kept for an average of 20 minutes. Tissues were divided into two sections: some of the sections were incubated with 1/100 diluted with VEGF primary antibodies (cat no: MA5-13182, Thermofisher Scientific, US) and the remaining sections were incubated in 1/100 diluted Vimentin primary antibodies (cat no: MA5-11883, Thermofisher Scientific, US) overnight at +4°C. The sections were kept at room temperature for approximately 1 hour the next day and were washed with PBS-T for 3x5 minutes. The sections were then incubated with anti-rabbit secondary antibody for approximately 15 minutes. Streptavidin-peroxidase was dripped onto the sections washed with PBS-T for 3x5 minutes and the sections were kept for 15 minutes. They were washed with PBS-T for 3x5 minutes and DAB was dripped onto the sections. The reactions of the tissues were monitored with a light microscope and the reaction of those showing involvement was terminated with PBS-T. The sections were re-washed with PBS-T for 3x5 minutes, counterstained with Gill III hematoxylin for approximately 45 seconds, and washed in tap water for 5 minutes. Then, the sections passed through increasing alcohol series were kept in xylol for 2x15 minutes and mounted with entellan.

Immunohistochemical staining results were analyzed with H scoring (HS). The formula used is HS= Σ (1 + i) x pi. Here, 'pi' represents the intensity of the percentage of staining; 'i' corresponds to the staining intensity (0=no expression, 1=mild, 2=moderate, 3=intense, and 4=very intense) (6).

Statistical Analysis

Statistical analysis was made in the IBM SPSS program. Non-parametric Kruskal Wallis Test was performed to analyze the group means. Data were presented as mean and standard deviation (SD). Statistically, $p<0.05$ was considered significant.

RESULTS

Pregnant women who participated in the study did not develop any complications and no fetal abnormality was detected in their babies.

Histopathological Findings

Hematoxylin-eosin staining of umbilical cord tissues of the control and experimental groups (who had had COVID-19 in the second and third trimesters) are shown in **Figure 1**. **Figure 1A** represents the control group; **Figure 1B** represents the group who had had COVID-19 in the second trimester; **Figure 1C** represents the groups who had had COVID-19 in the third trimester. In the histopathological images of the umbilical cords of the control group, the muscles in the tunica media layer of the umbilical vessel were a regular structure. The histological structure of the Wharton gel was normal. There were occasional ruptures between the muscle fibers in the tunica media of the umbilical vessel in the umbilical cords of pregnant women who had had COVID-19 in the second trimester of pregnancy. The integrity of the tunica intima layer was lost. Hemorrhagic findings were detected in the umbilical vascular structure of the umbilical cords of pregnant women who had had COVID-19 in the third trimester of pregnancy. Furthermore, the tunica intima layer was thinner and the integrity partly deteriorated.

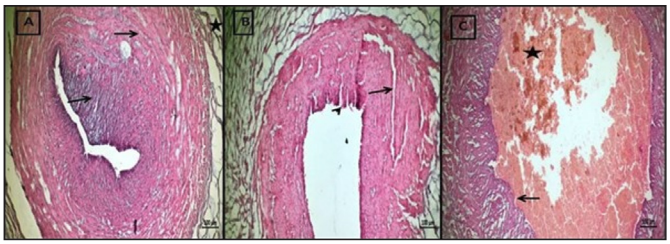

Figure 1. HE staining of sections of the umbilical cord (A) Normal histological appearance in the tunica intima, media (arrows), and adventitia (star) in the control group (B) Structural deterioration in the tunica media (arrow) and intima (arrowhead) layers in the group who had had COVID-19 in the second trimester (C) Hemorrhage (star) and thinning of the tunica intima (arrow) and deterioration in the integrity in the group who had had COVID-19 in the third trimester (X10)

Immunohistochemical Findings

VEGF immunohistochemical staining of the umbilical cords of the control and experimental groups are shown in **Figure 2**. In the umbilical cords of the control group, the expression of VEGF in the tunica intima and adventitia layers of the umbilical vessel was mild (**Figure 2A**). VEGF expression was positive in endothelial cells in the tunica intima of the umbilical vessel and at the border of the tunica media and tunica adventitia in the umbilical cord tissues of pregnant women who had had COVID-19 in the second trimester of pregnancy (**Figure 2B**). VEGF expression was higher in the second-trimester group than in the control group. In umbilical cord tissues of pregnant women who had had COVID-19 in the third trimester of pregnancy, VEGF expression was positive in endothelial cells in the tunica intima of the umbilical vessel, at the border of the tunica media and adventitia, and in the

tunica adventitia (**Figure 2C**). VEGF expression was higher in the group who had had COVID-19 in the third trimester than in both the control group and the group who had had COVID-19 in the second trimester.

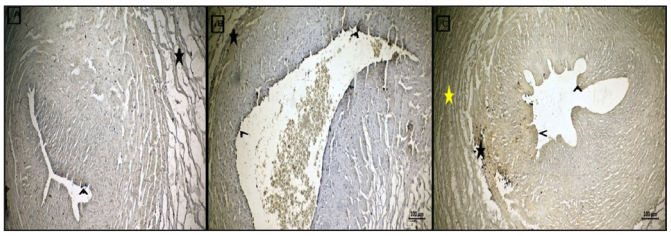

Figure 2. Immunohistochemical staining of VEGF in umbilical cord sections (A) Control group: Mild expression in the tunica intima (arrowhead) and adventitia (black star) of the umbilical vessel (B) Group who had had COVID-19 in the second trimester: Positive expression in the tunica intima (arrowheads) of the umbilical vessel, at the border of the tunica media and adventitia (star) (C) Group who had had COVID-19 in the third trimester: Positive expression in the tunica intima (arrowheads), at the border of the tunica media and adventitia (black star), and tunica adventitia (yellow star) of the umbilical cord (X10)

Vimentin immunohistochemical staining of the umbilical cords of the control and experimental groups are shown in **Figure 3**. In the umbilical cord of the control group, vimentin expression was positive in endothelial cells in the tunica intima of the umbilical vessel and mild in the tunica media layer (**Figure 3A**). Vimentin expression was positive in the tunica intima and media layers of the umbilical vessel and mild in the tunica adventitia layer in the umbilical cord tissues of pregnant women who had had COVID-19 in the second trimester of pregnancy (**Figure 3B**). Vimentin expression was higher in the second-trimester group than in the control group. In the umbilical cord tissues of pregnant women who had had COVID-19 in the third trimester of pregnancy, vimentin expression was positive in the tunica media and adventitia layers of the umbilical vessel and mild in the tunica intima layer (**Figure 3C**). Vimentin expression was higher in the third-trimester group than in both the control group and second-trimester group.

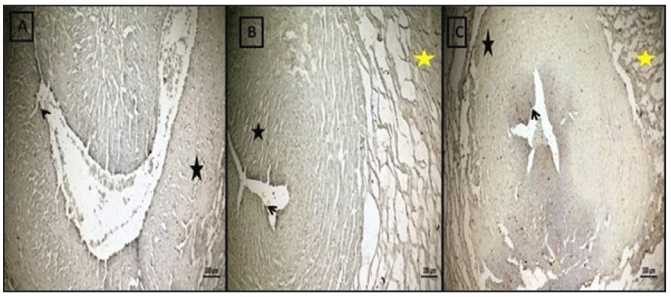

Figure 3. Immunohistochemical staining of vimentin in umbilical cord sections (A) Control group: Positive expression in the tunica intima (arrowhead), mild expression in the tunica media (star) of the umbilical vessel (B) Group who had had COVID-19 in the second trimester: Positive expression in the tunica intima (arrow) and media (black star) of the umbilical vessel and mild expression in the adventitia (yellow star) (C) Group who had had COVID-19 in the third trimester: Positive expression in the tunica media (black star) and adventitia (yellow star) of the umbilical vessel and mild expression in the intima (arrow) (X10)

Immunohistochemical Statistics

Statistical analysis of immune activities of VEGF and vimentin was shown in **Table 1**. The change in VEGF expression was statistically significant between groups. The significance of VEGF in the second group compared to the control group was less than the significance of the third group compared to the second group. The expression was the highest in the third-trimester group. Similarly, the change in vimentin expression was significant between control, second and third-trimester groups. Vimentin expression was significantly higher in the second-trimester group than in the control group. Vimentin reaction was higher in the third-trimester group than in the control and second-trimester group, this increase was statistically significant.

Table 1. Comparison of VEGF and Vimentin expression between groups

Parameters	Control group	2nd trimester	3rd trimester	p value
	Median (min-max)	Median (min-max)	Median (min-max)	
VEGF	0.00 (0.00-1.00)	1.00 (0.00-2.00)	2.00 (1.00-3.00)	*p=0.0019 ** p<0.001 ***p<0.001
Vimentin	0.00 (0.00-1.00)	2.00 (0.00-3.00)	3.00 (2.00-3.00)	*p<0.001 ** p<0.001 ***p=0.027

* control vs second, **control vs third, ***second vs third

DISCUSSION

The clinical findings of pregnant women with COVID-19 are generally similar to other COVID-19 patients. However, it is known that COVID-19 in pregnancy causes some negative consequences such as preterm birth, fetal distress, and neonatal asphyxia. Although COVID-19 does not usually cause fetal and neonatal adverse effects, COVID-19 infection, especially seen in the third trimester, may cause more risky and negative consequences (21). Furthermore, there is a need for further comprehensive studies, including other trimesters to understand the long-term effects of COVID-19 on maternal and infant health during pregnancy. In our study, we examined the long-term effects of COVID-19 in the second and third trimesters of pregnancy, using histopathological and immunohistochemical methods on the umbilical cord.

Immunohistochemical and histopathological disorders are seen in the placentas of pregnant women with active COVID-19. In a study, it was seen that IL-6 and Bax expression levels were higher in the postpartum placentas of pregnant women with active COVID-19 compared to the control group (6). In another study, fetal and maternal vascular malformation, villitis, thrombus formation, and chorangiosis were histopathologically reported in

the placentas of pregnant women with COVID-19 (22). This information suggests that COVID-19 should be evaluated not only in terms of clinical outcomes but also in terms of histopathological outcomes. Although there are studies on the possible effects of COVID-19 seen in the last period of pregnancy, its effects in the previous trimesters are not fully known (21). Studies examining the relationship of this disease with the umbilical cord are limited (23). Previous studies mostly reported that mesenchymal stem cells taken from the umbilical cord are successful in the treatment of this disease (24, 25).

A study in which the umbilical cords of pregnant women with COVID-19 were examined immunohistochemically reported antibody involvement against Sars-CoV-2 N protein. It was stated that the cells with involvement were macrophages and fibroblast-like cells in Wharton gel (23). The fact that there was no involvement in the umbilical cords in the control group whereas there was an involvement in the active COVID-19 group is important. This shows that the umbilical cord is affected by the Sars-CoV-2 virus, like many other organs. Accordingly, the presence of the Sars-CoV-2 virus in the umbilical cords and its long-term effects should be considered when evaluating the effects of COVID-19 on the mother-placenta-fetus system.

In our study, structural deteriorations were histopathologically observed in the layers of the umbilical vessels in the umbilical cords of pregnant women who had had COVID-19 in the second and third trimesters. There was more histopathological damage with hemorrhage in the third-trimester group. Given that the Sars-CoV-2 virus can be transmitted to the umbilical cord, the apparent histopathological findings, especially in the third trimester, can be explained by the histopathological effects of this infection. As a matter of fact, the pregnant women in the third-trimester group included in the study mostly consisted of cases who were near delivery. This suggests that the effects of the virus on the umbilical cord remain in the post-acute period. On the other hand, milder histopathological findings seen in the second-trimester group can be explained by the fact that the effects of COVID-19 infection seen in earlier periods on the umbilical cord decrease. At this point, it is important to ensure new pathogenic mechanisms to provide fetal protection against COVID-19 infections, especially near delivery.

VEGF is one of the major factors for fetal and placental development (26). A study comparing VEGF expressions between normal placenta and placenta increata cases found that VEGF expression levels were higher in placenta increata cases. Overexpression of VEGF and abnormal villous vessel formation seen in placental adhesion anomalies were found to be associated with the pathogenesis of placenta increata (27). In another study examining normal pregnancies and pregnancies with severe preeclampsia, VEGF expression levels in the umbilical cord were compared and VEGF expression was found to be higher in the preeclampsia group. It is known that VEGF affects NO synthesis by regulating eNOS production (28). Therefore, the increase in VEFG expression in the preeclampsia group may be a tolerant mechanism to dilate blood vessels and improve fetal blood flow against developing hypoxic conditions. In another study, it was stated that VEGF reduces the expression of proinflammatory cytokines such as TNF-α and IL-1. Moreover, VEGF was reported to reduce tissue damage by inhibiting inflammation (29).

In our study, VEGF expression in the vascular structures that form the basic structure of the umbilical cords and are embedded in the Wharton gel was higher in the group who had had COVID-19 in the third trimester. This suggests that VEGF may be involved in the pathogenesis of the COVID-19 infection, especially in the period near delivery. COVID-19 is known to cause arterial hypoxia due to pulmonary involvement (30). The increase in VEGF expression, especially seen in the third-trimester group, might be a defense mechanism that supports the blood circulation of the fetus against hypoxic conditions. Another finding in COVID-19 infection is the increase of proinflammatory cytokines such as TNF-α (31). Considering the effects of VEGF such as reducing the expression of proinflammatory cytokines such as TNF-α and inhibiting inflammation (29), it can be suggested that this protein has a regulatory and protective role in COVID-19 infection.

Vimentin has many functions such as cell physiology, cellular interactions, and tissue homeostasis. Another important function of vimentin is that it takes part in cases of infection (18). It was reported that vimentin has a cytoprotective role that prevents the entry of viral components into the cytoplasm. Indeed, this effect of vimentin was demonstrated in a study conducted on African swine fever virus (ASFV) infection (20). The increase in vimentin expression observed in the third-trimester group in our study may be due to the cytoprotective role of vimentin against viral infections. The decreasing vimentin expression from the second-trimester group to the control group can be explained by the fact that the effect of the disease and the viral load gradually decrease.

In a study in which the placentas of preeclamptic and normotensive pregnant women were compared in terms of vimentin expression, it was found that the vimentin expression was higher in the preeclamptic group and that vimentin has a role in the pathogenesis of preeclampsia (32). In another study, it was stated that the expression levels of

vimentin and VEGF in these patients were correlated with each other. Moreover, both proteins were held responsible for trophoblast invasion, angiogenesis, and vascular permeability (26). In our study, the expression levels of vimentin and VEGF in the umbilical cord were found to be higher in the third-trimester group. This suggests that both proteins can be co-evaluated in the pathogenesis of COVID-19. On the other hand, new studies are required to explain the molecular mechanisms by which vimentin and VEGF play a role in the relationship between COVID-19 and the umbilical cord.

Limitations

This study has some limitations. Primarily, each group included in the study consisted of nine cases. The decreasing trend of the number of COVID-19 cases in our region made it difficult to find pregnant patients who had COVID-19, especially in the third trimester. Comprehensive studies can be conducted with a larger number of pregnant patients who have had COVID-19 by increasing the period for the study. Secondly, cases who had had the disease in the second and third trimesters were included in the study to investigate the long-term effects of COVID-19. Further studies to examine postpartum umbilical cords can be planned with cases who have had COVID-19 in the first trimester and even before pregnancy. Lastly, the use of different primary antibodies may provide important results when evaluating the expression of other proteins important for umbilical cord development besides VEGF and vimentin primary antibodies.

CONCLUSION

Today, both the acute and long-term effects of the COVID-19 disease are still a matter of discussion. One of the most researched subjects among these effects is maternal and infant health. Histopathological and immunohistochemical findings in the umbilical cord, especially in the third trimester of pregnancy, were more prominent, suggesting that more attention should be paid to COVID-19 infection near delivery. Furthermore, the high levels of VEGF and vimentin expression in the umbilical cords of pregnant women in this period may be useful in understanding the post-acute effects of these proteins in the pathogenesis of COVID-19.

ETHICAL DECLARATIONS

Ethics Committee Approval: The study was carried out with the permission of Siirt University Non-Interventional Clinical Research Ethics Committee (Date: 13.04.2022, Decision No: 42191).

Informed Consent: All patients signed the free and informed consent form.

Referee Evaluation Process: Externally peer-reviewed.

Conflict of Interest Statement: All authors state that there is no conflict of interest.

Financial Disclosure: The authors declared that this study has received no financial support.

Author Contributions: All of the authors declare that they have all participated in the design, execution, and analysis of the paper, and that they have approved the final version.

REFERENCES

1. Velavan TP, Meyer CG. The COVID-19 epidemic. Trop Med Int Health 2020; 25: 278.
2. Wiersinga WJ, Rhodes A, Cheng AC, Peacock SJ, Prescott HC. Pathophysiology, transmission, diagnosis, and treatment of coronavirus disease 2019 (COVID-19): a review. JAMA 2020; 324: 782-93.
3. Jain U. Effect of COVID-19 on the Organs. Cureus 2020; 12: e9540.
4. Suvvari TK, Kutikuppala LS, Tsagkaris C, Corriero AC, Kandi V. Post-COVID-19 complications: Multisystemic approach. J Med Virol 2021; 93: 6451-5.
5. Liu Y, Chen H, Tang K, Guo Y. Clinical manifestations and outcome of SARS-CoV-2 infection during pregnancy. J infect 2020; 82: e9-10.
6. Taş F, Erdemci F, Aşır F, Maraşlı M, Deveci E. Histopathological examination of the placenta after delivery in pregnant women with COVID-19. J Health Sci Med 2022; 5: 868-74.
7. Gürbüz T, Gökmen O, Kaptan G, İnanlı E, Karadeniz SB, Söylemez NE, Söylemez İ. Investigating anxiety, depression and obsessive-compulsive disorders among the pregnant women during Covid-19 pandemic. J Health Sci Med 2021; 4: 7-12.
8. Chen H, Guo J, Wang C, et al. Clinical characteristics and intrauterine vertical transmission potential of COVID-19 infection in nine pregnant women: a retrospective review of medical records. The Lancet 2020; 395: 809-15.
9. Küçük Ş. Placenta and Umbilical Cord Histopathologies in Pathologic Spesmen and Relationship Between Pregnancy. Selcuk Med J 2019; 35: 1-8.
10. Tantbirojn P, Saleemuddin A, Sirois K, et al. Gross abnormalities of the umbilical cord: related placental histology and clinical significance. Placenta 2009; 30: 1083-8.
11. Yoon BH, Romero R, Park JS, et al. The relationship among inflammatory lesions of the umbilical cord (funisitis), umbilical cord plasma interleukin 6 concentration, amniotic fluid infection, and neonatal sepsis. Am J Obstet Gynecol 2000; 183: 1124-9.
12. Mullany LC, Darmstadt GL, Katz J, et al. Risk factors for umbilical cord infection among newborns of southern Nepal. Am. J. Epidemiol 2007; 165: 203-11.
13. Salvatore CM, Han JY, Acker KP, et al. Neonatal management and outcomes during the COVID-19 pandemic: an observation cohort study. Lancet Child Adolesc. Health 2020; 4: 721-7.
14. Yim HE, Bae IS, Yoo KH, et al. Genetic control of VEGF and TGF-β1 gene polymorphisms in childhood urinary tract infection and vesicoureteral reflux. Pediatr. Res 2007; 62: 183-7.
15. Vural P. Fizyolojik ve Patolojik Anjiogenezde Vasküler Endotelyal Büyüme Faktörünün Rolü. Türk Klinik Biyokimya Derg 2018; 16: 53-62.
16. Olaya-C M, Michael F, Fabian G, et al. Role of VEGF in the differential growth between the fetal and placental ends of the umbilical cord. J. Neonatal. Perinatal Med 2019; 12: 47-56.

17. Kuruçay HN, Gümüşova S. Feline İnfeksiyöz Peritonitise Genel Bakış ve Antiviral Yaklaşımlar. Turk Vet J 2021; 3: 4-12.
18. Ivaska J, Pallari HM, Nevo J, Eriksson JE. Novel functions of vimentin in cell adhesion, migration, and signaling. Exp Cell Res 2007; 313: 2050-62.
19. Kadner A, Zund G, Maurus C, et al. Human umbilical cord cells for cardiovascular tissue engineering: a comparative study. Eur J Cardiothorac Surg 2004; 25: 635-41.
20. Stefanovic S, Windsor M, Nagata KI, Inagaki M, Wileman T. Vimentin rearrangement during African swine fever virus infection involves retrograde transport along microtubules and phosphorylation of vimentin by calcium calmodulin kinase II. J Virol 2005; 79: 11766–75.
21. Yang Z, Wang M, Zhu Z, Liu Y. Coronavirus disease 2019 (COVID-19) and pregnancy: a systematic review. J Matern Fetal Neonatal Med 2022; 35: 1619-22.
22. Motwani R, Deshmukh V, Kumar A, Kumari C, Raza K, Krishna H. Pathological involvement of placenta in COVID-19: a systematic review. Infez Med 2022; 30: 157.
23. Nizyaeva NV, Lomova NA, Dolgopolova EL, et al. Detection of SARS-Cov-2 N-protein in Mesenchymal Cells of Umbilical Warton's Jelly in Women with COVID-19. Biomed Chem Res Methods 2022; 5: e00165.
24. Ciccocioppo R, Gibellini D, Astori G, et al. The immune modulatory effects of umbilical cord-derived mesenchymal stromal cells in severe COVID-19 pneumonia. Stem Cell Res Ther 2021; 12: 1-10.
25. Rodriguez HC, Gupta M, Cavazos-Escobar E, El-Amin SF, Gupta A. Umbilical cord: an allogenic tissue for potential treatment of COVID-19. Hum Cell 2021; 34: 1-13.
26. Irtegun S, Agacayak E, Deveci E. Preeklamptik ve normotansif plasentalarda VEGF ve Vimentin ekspresyon düzeylerinin immunohistokimya ve Western Blot yöntemleri ile incelenmesi. Dicle Med J 2016; 43: 400-5.
27. Yan J, Wu X, Zhang Q. Relationship between vascular endothelial growth factor and microvessel density in placenta and placenta increta. Chin J Perinat Med 2016; 12: 608-13.
28. Bhavina K, Radhika J, Pandian SS. VEGF and eNOS expression in umbilical cord from pregnancy complicated by hypertensive disorder with different severity. BioMed Res Int 2014; 1-6.
29. Wang H, Wang Y, Li D, et al. VEGF inhibits the inflammation in spinal cord injury through activation of autophagy. Biochem Biophysic Res Commun 2015; 464: 453-8.
30. Aktoz M, Altay H, Aslanger E, et al. Türk Kardiyoloji Derneği uzlaşı raporu: COVID-19 pandemisi ve kardiyovasküler hastalıklar konusunda bilinmesi gerekenler. Turk Kardiyol Dern Ars 2020; 48: 1-48.
31. Hu B, Huang S, Yin L. The cytokine storm and COVID-19. J. Med. Virol 2021; 93: 250-6.
32. Yılmaz EPT, Topdağı YE, Aşkın, S. Comparison of Vimentin Levels Between Preeclamptic and Normotensive Pregnant Women. J Obstet Gynecol Neonatal 2020; 17: 610-4.