Since January 2020 Elsevier has created a COVID-19 resource centre with free information in English and Mandarin on the novel coronavirus COVID-19. The COVID-19 resource centre is hosted on Elsevier Connect, the company's public news and information website.

Placenta 126 (2022) 209–223

Contents lists available at ScienceDirect

Placenta

journal homepage: www.elsevier.com/locate/placenta

Coronavirus disease 2019 and the placenta: A literature review

Samwel R. Gesaka [a,*], Moses M. Obimbo [a,b,c], Anthony Wanyoro [a,d]

[a] Basic Clinical and Translational (BCT) Research Laboratory, University of Nairobi, Nairobi, Kenya
[b] Department of Human Anatomy, University of Nairobi, Nairobi, Kenya
[c] Department of Obstetrics and Gynecology, University of Nairobi, Nairobi, Kenya
[d] Department of Obstetrics and Gynecology, Kenyatta University, Nairobi, Kenya

ARTICLE INFO

Keywords:
SARS-CoV-2
ACE2
Placental infection
Placental pathology
Single-cell analysis

ABSTRACT

Coronavirus disease 2019 (COVID-19) caused by the severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) virus has been implicated in the clinical pathology of multiple organs and organ systems. Due to the novelty of the disease, there is a need to review emerging literature to understand the profile of SARS-CoV-2 in the placenta. This review sought to evaluate the literature on the mediators, mechanism of entry, pathogenesis, detection, and pathology of SARS-CoV-2 in the placenta. Systematic literature searches found 96 eligible studies.

Our review revealed that SARS-CoV-2 canonical mediators, angiotensin-converting enzyme-2 (ACE2), and transmembrane serine protease-2 (TMPRSS2) are variably expressed in various placenta compartments, including the villous cytotrophoblasts, syncytiotrophoblasts (STBs), and extravillous trophoblasts (EVTs) throughout pregnancy. Placental SARS-CoV-2 and coronavirus-associated receptors and factors (SCARFs), including basigin (BSG/CD147), dipeptidyl peptidase-4 (DPP4/CD26), cathepsin B/L (CTL B/L), furin, interferon-induced transmembrane protein (IFITM1-3), and lymphocyte antigen 6E (LY6E) may increase or reduce the permissiveness of the placenta to SARS-CoV-2. EVTs express genes that code for proteins that may drive viral pathogenesis in the placenta. Viral RNA, proteins, and particles were detected primarily in the STBs by *in situ* hybridization, immunohistochemistry, electron microscopy, and polymerase chain reaction. Placental pathology in SARS-CoV-2-infected placentas included maternal and fetal vascular malperfusion and a generally nonspecific inflammatory-immune response.

The localization of SARS-CoV-2 receptors, proteases, and genes involved in coding proteins that drive viral pathogenesis in the placenta predisposes the placenta to SARS-CoV-2 infection variably in all pregnancy trimesters, with antecedent placental pathology. There is a need for further studies to explicate the mechanism of entry and pathogenesis of SARS-CoV-2 in the placenta.

1. Introduction

Coronavirus disease 2019 (COVID-19), caused by the severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) virus, emerged in China in December 2019. It quickly spread throughout the world and was declared a pandemic by the World Health Organization (WHO) in March 2020 [1]. The virus belongs to the genus beta-coronaviruses alongside the Middle East respiratory syndrome coronavirus (MERS-CoV) and severe acute respiratory syndrome human coronavirus (SARS-CoV), which have been implicated in past pandemics [2]. Beta-coronaviruses are enveloped, single-stranded, positive-sense RNA (+ssRNA) viruses [2]. Transmission of SARS-CoV-2 is primarily through infected respiratory droplets, with other transmission routes being fecal-oral, contaminated fomites, and potentially vertical transmission [3,4]. Angiotensin-converting enzyme-2 (ACE2) is the target receptor for SARS-CoV-2. The viral spike (S) surface protein binds to the host ACE2 receptors, followed by priming the S1/S2 polybasic cleavage site by the host proteases. The two host proteases include transmembrane serine protease 2 (TMPRSS2) and cathepsin B and L (Cat B/L), which enable membrane fusion, implying the virus utilizes two independent

Abbreviations: ACE2, angiotensin-converting enzyme-2; EVTs, extravillous trophoblasts; SCARFs, SARS-CoV-2 and coronavirus-associated receptors and factors; STBs, syncytiotrophoblasts; TMPRSS2, transmembrane serine protease-2.

* Corresponding author. Basic Clinical, and Translational (BCT) research laboratory, University of Nairobi, Kenya P.O. Box 30197, 00100, Nairobi 14 Riverside Drive, Nairobi, Kenya.

E-mail addresses: gesakarema@gmx.com (S.R. Gesaka), obimbomad@gmail.com (M.M. Obimbo), Wanyoro.Anthony@ku.ac.ke (A. Wanyoro).

https://doi.org/10.1016/j.placenta.2022.07.007
Received 29 December 2021; Accepted 13 July 2022
Available online 19 July 2022

mechanisms to enter host cells [5]. However, SARS-CoV-2 can also be preactivated by furin, which reduces its dependence on proteases for entry [6]. Other receptors (basigin (BSG/CD147), dipeptidyl peptidase-4 (DPP4/CD26)), and proteases (elastase, tyrosine, membrane-associated serine proteinases (MASPs)) may enhance viral infectivity [7–9]. The spike protein undergoes a structural change in which the viral envelope fuses to the cell membrane to release its RNA. Viral proteases cleave the resulting polypeptide into virions released through vesicles into the extracellular space [10]. The recognition of virions triggers an immune response by the pattern recognition receptors (PRRs), such as toll-like receptors (TLRs) [5]. The infection and apoptosis of type 2 pneumocytes secrete mediators like C-X-C motif chemokines (CXCL) that attract macrophages and neutrophils. The sustained inflammatory response may lead to systemic inflammatory response syndrome (SIRS), which may develop into septic shock, causing multi-organ damage and even death [10]. The occurrence of ACE2 receptors in other organs increases the tissue tropism of SARS-CoV-2 [2].

The shift of maternal immunity from predominantly type 1 T helper (Th1) to type 2 T helper (Th2) effector during pregnancy significantly predisposes the mother to viral respiratory infections with potential adverse maternal and neonatal outcomes [11,12]. In general, COVID-19-positive pregnant women present with similar presentations as non-pregnant adults [13]. However, COVID-19-positive pregnant women may be asymptomatic compared to the general population [13]. COVID-19 infection during pregnancy is associated with adverse fetal and maternal outcomes, including fetal distress, intrauterine growth retardation (IUGR), preterm delivery, miscarriage, stillbirth, maternal depression, and mortality. A significant disparity has been observed between high and low-resource settings [14]. Evidence for placental involvement and vertical transmission has been evolving since the onset of the pandemic, with studies often presenting conflicting findings [15–17]. Studies during the onset of the pandemic reported that the placenta was not susceptible to SARS-CoV-2 infection [18–20], and vertical transmission and neonatal infection were not entertained [21,22]. However, recent studies have since elucidated the expression of SARS-CoV-2 mediators in various placental compartments, leading to the virus implication in placental infection and pathology [23–30]. Additionally, a few cases of neonates born to COVID-19-positive mothers have tested positive for the virus and IgM antibodies shortly after birth, implying transplacental infection [24,31].

2. Methods

2.1. Literature search

The search strategy was as follows: "((2019 AND coronavirus) OR COVID-19 OR SARS-CoV-2 OR HCoV-19) AND (placenta OR placenta factors OR placenta mediators OR placental infection OR placenta pathogenesis OR vertical transmission OR placenta pathology)" in PubMed and Cochrane library COVID-19 databases. The search period was unlimited. Two authors independently screened the abstracts and full articles for eligibility, and a third author resolved disagreements. The last search on all databases was on November 19, 2021.

2.2. Eligibility criteria

Studies were considered eligible if they reported on SARS-CoV-2 mediators, mechanism of entry and pathogenesis, detection, and pathology in the placenta. Studies that did not report the outcomes of interest were excluded. No language limits were applied.

2.3. Data extraction

Data were extracted into tables. The information extracted included the first author's name, sample size, investigations, SARS-CoV-2 entry mediators, detection, and placental pathology. Non-English studies were translated into English. The extracted data were double-checked for accuracy. The third author verified a sample of screened records and exclusions.

3. Results

A total of 1498 studies were identified; of these, 96 met the inclusion criteria. Results are summarized in the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) flowchart (Fig. 1). Characteristics of included studies are described in Tables 1 and 2.

3.1. SARS-CoV-2 mediators in the placenta

Angiotensin-converting enzyme 2 is expressed in the placenta throughout pregnancy and is involved in physiological regulation, including blood pressure [32]. Placental ACE is actively responsive to SARS-CoV-2 infection, and afterward, this results in its downregulation, which may be heightened during severe infection [33]. The downregulation of ACE may lead to dysregulation of the renin-angiotensin system (RAS), resulting in elevated maternal blood pressure and dysfunctional placental vascularization. This may lead to comorbidities associated with infection in pregnancy, such as preeclampsia [33]. In contrast, Lu-Culligan et al. reported that placental ACE2 levels were elevated in mothers with COVID-19 at term [34]. On the other hand, Lye et al. reported an increase in ACE2 levels in preterm placentas compared with term placentas [28].

Microarray, single-cell RNA sequencing (scRNA-seq), and single-nuclear RNA sequencing (snRNA-seq) analyses of various datasets found variable expression of both canonical and non-canonical SARS-CoV-2 mediators, including ACE2, TMPRSS2, DPP4, CTSL, furin, CD147, interferon-induced transmembrane protein (IFITM1-3) and lymphocyte antigen 6E (LY6E) in almost all decidual, placental, and trophectoderm (TE) cells throughout pregnancy [20,24,30,35–37]. Analysis of the Human Protein Atlas (HPA), Genome-based Tissue Expression (GTEx), and FANTOM5 datasets reported high ACE2 protein levels in first-trimester placental tissues despite low mRNA levels [23,25]. ACE2 mRNA was abundantly expressed in the syncytiotrophoblasts (STBs) (39%) in the first trimester, while in the second trimester, it was abundantly expressed in the extravillous trophoblasts (EVTs) (62%) [24]. Immunofluorescence, immunohistochemical, polymerase chain reaction (PCR), and Western blot analyses of placental tissues of different gestational ages reported ACE2 protein in the villi (cytotrophoblasts (CTBs), STBs, villous blood vessels endothelium, and vascular smooth muscle cells of the primary villi), maternal stroma (invading, intravascular and EVTs) and decidual cells), and the umbilical cord (arterial, venous endothelial and smooth muscle cells) [26,27,37,38]. Placental neutrophils and M1/M2 macrophages express ACE2 mRNA and protein while macrophages also express silaoadhesin (CD169), an adhesion molecule thought to enhance SARS-CoV-2 infection [28,29]. TMPRSS2 mRNA is expressed in low levels in a few placental compartments, including EVTs (19%) and villous stromal cells (STRs) (23%) in the first and second trimester, respectively, but increased a term [24]. The expression of TMPRSS2 in decidual and placental cells during COVID-19 infection is insignificant [34]. While some authors have shown an increase in the co-expression of ACE2 and TMPRSS2 in the EVTs, STBs, CTBs increases along with pregnancy trimester [24,30], Pique-Regi et al. reported minimal ACE2/TMPRSS2 co-expression in all pregnancy trimesters [20].

Other mediators may increase or decrease the permissiveness of the placenta SARS-CoV-2 infection depending on the pregnancy trimester at infection [39,40]. Cathepsin was widely expressed in fibroblasts, immune cells, and trophoblasts and was increased in decidual antigen-presenting cells and stromal cells in COVID-19 infection [34]. The co-expression of ACE2 and CTSL in the placenta varies throughout pregnancy [37]. Basigin is expressed in nearly all placenta compartments (96–100%) [25]. First-trimester ACE+ trophoblast subtypes

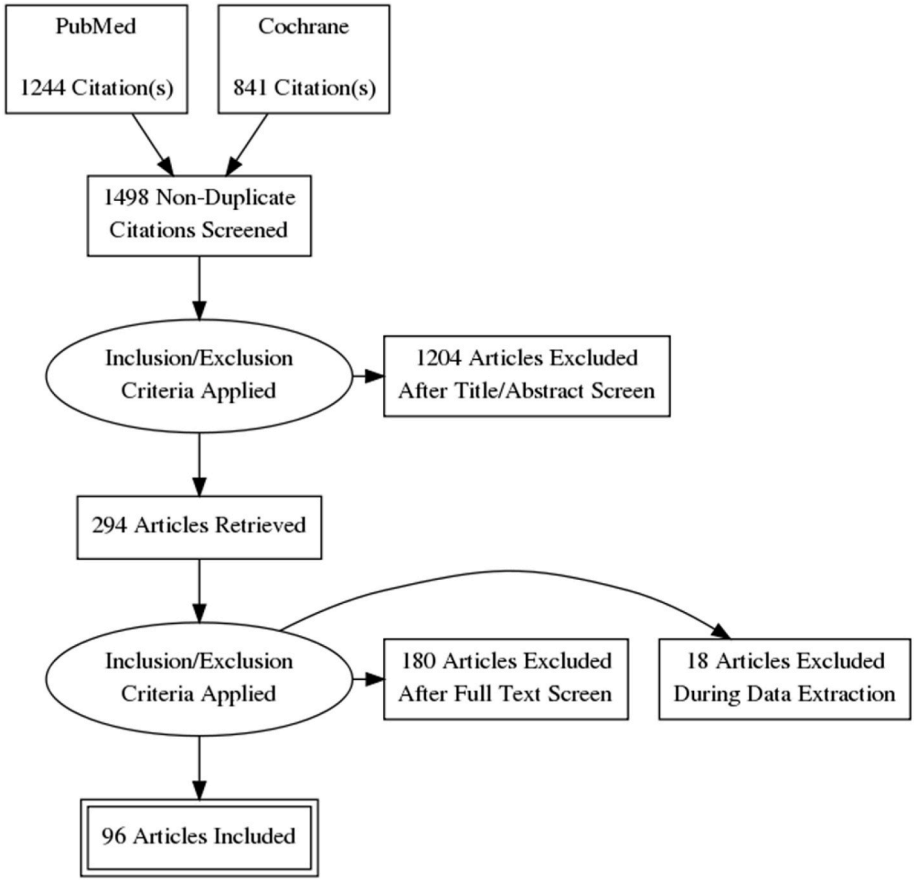

Fig. 1. PRISMA flowchart of literature search and study selection.

coexpress CD147 and CTSL, and all CD147+ cells coexpress CTSL, whereas second-trimester ACE + EVTs coexpress CD147 and CTSL, and all CD147+ cells coexpress CTSL [24]. Furin is expressed in the fetal and maternal cells throughout gestation [20,24,36,38]. A scRNA expression map identified SARS-CoV-2 and coronavirus-associated receptors and factors (SCARFs) that may promote or restrict the entry of SARS-CoV-2 and other human coronaviruses [36]. The coexpression of basigin and dipeptidyl peptidase-4 (DPP4) with TMPRSS2 and furin may increase the permissiveness of placenta cells to viral entry [36]. In contrast, basigin and DPP4 co-expression with IFITM1-3 and LY6E reduce the permissibility of the placenta to viral entry [36]. The expression of ACE2 and TMPRSS2, and other coronavirus receptors in the trophectoderm (TE) and placenta tissues (EVTs, STBs, CTBs), combined with low levels of IFITMs, significantly predispose the developing placenta to SARS-CoV-2 infection [36]. Neurolipin 1 (NRP1) is also an alternative receptor for SARS-CoV-2 entry in the placenta [39].

3.2. *Placental barrier and immune response against pathogens, including SARS-CoV-2*

The placenta barrier plays a significant role in preventing the entry of pathogens into the placenta provided by a dense network of branched microvilli that cover the syncytiotrophoblasts at the fetomaternal interface and the underlying villous cytotrophoblast layer [41]. However, the STBs lack intercellular gap junctions and barely express pattern recognition receptors (PRRs) such as toll-like receptors (TLRs) and internalization receptors (E-Cadherin) that can be exploited by pathogens or modulated by inflammatory signals [42]. Besides, caveolin is barely expressed in the STBs but in the endothelium, pericytes, stroma, and smooth muscle cells [43]. Since SARS-CoV-2 does not use caveolins to enter cells, the clustering of the virus outside the cells may trigger local inflammation [19].

The syncytiotrophoblasts are still susceptible to pathogen invasion, especially in early gestation, late gestation, high viral load, and when there are underlying maternal diseases [43]. Placental immune response against viruses is mediated by type 3 IFN (IFN-λ), nuclear factor kappa-light-chain-enhancer of activated B cells (NF-κB) pathway, and miRNA-triggered autophagy [43]. Caveolin-1 (Cav-1) initiates inflammation through the NF-κB pathway upregulating the release of cytokines IL-6 and tumor necrosis factor-alpha (TNF-α) [44]. Trophoblasts mediate the delivery of specific micro RNA (miRNA), inducing autophagy, thus conferring viral resistance [45,46]. The EVTs express genes involved in the Janus kinases and signal transducers and activators of a transcription (JAK-STAT) system, which is involved in inflammatory response through IFN-λ [24,43]. The localization of ACE2 and genes involved in the JAK-STAT pathway may predispose the EVTs to an inflammatory response in SARS-CoV-2 placental infection [24]. This could be attributed to the inflammatory-immune responses in the placenta [24]. Placental immunomodulation during COVID-19 infection may mitigate cytokine storm and lessen cellular and tissue damage [43]. Through apoptosis, necroptosis, and pyroptosis, programmed cell death may also play an antiviral role against viruses that require host cells for replication [47]. Placental IFITM3 reduces the permissibility of the placenta to SARS-CoV-2 entry [36]. Higher IFITM3 expressions were reported in mothers with severe COVID-19 disease [48].

3.3. *Pathogenesis of SARS-CoV-2 in the placenta*

Many microorganisms, including viruses, can infect the placenta either via the ascending transvaginal route or hematogenously due to a breach in the placenta's integrity or immune responses [49,50]. The mechanisms through which pathogens traverse the placenta include (i)

Table 1
SARS-CoV-2 mediators in the placenta.

Author	Study Characteristics and Materials	Methods	Receptors/ Proteases	Main results
Ashary N et al., 2020	Placentas: first-trimester (8 w, n = 7), second-trimester (n = 1, 24 w); second trimester and term placentas (n = 4).	scRNA-seq	ACE2 TMPRSS2 BSG/ CD147 CTSB CTSL Furin	ACE2 mRNA was most expressed in the STB (39%) in first trimester and 62% EVTs in second trimester. TMPRSS2 mRNA was expressed in the STR (23%) and EVTs (19%) in first trimester and second trimester, respectively. ACE2 and TMPRSS2 mRNA was co-expressed in STBs (14%) and EVTs (15%) in first and second trimester, respectively. ACE+/ TMPRSS2+ EVTs and STB expressed Furin alongside mRNA for proteins in SARS-CoV-2 budding and replication. BSG mRNA was expressed in all cells (96–100%) of EVTs, CTBs, STBs, and STRs in the first trimester. CTSL mRNA was expressed in nearly all CTBs, EVTs, STBs, and STRs but was higher in EVTs in second trimester.
Constantino et al., 2021	Villous trophoblast tissues of first (6–8 w, n = 4), second (15–16 w, n = 4) and third (n = 4) trimesters of uncomplicated pregnancies; 11 deciduas and 5 placentas (6–14 w).	Microarray scRNA-seq	ACE2 TMPRSS2 DPP4 CTSL	ACE2 and TMPRSS2 were less expressed in the CTBs, EVTs, and STBs, but DPP4 and CTSL were abundantly co-expressed in these cells throughout pregnancy.
Cui D et al., 2021	11 deciduas and 5 placentas (6–14 w); Placentas from healthy women: 8 w (induced abortion (n = 3), 17 w (n = 1), 18 w (n = 1), 24 w (n = 1) and 39 w (n = 1).	scRNA-seq IHC	ACE2 TMPRSS2	TE: ACE2 was expressed in TE1 cells (54%), CTBs (9%), EVTs (3.2). and STBs (29.5%) in first and second trimesters, whereas TMPRSS2 was expressed in TE1 cells (90.7%), CTBs (31.5%), EVTs (3.2%), and STBs (29.5%) in first and second trimesters. Placental cells: ACE2 was expressed in the CTBs (20.4%, 8 w), EVTs (3.4% 8 w vs 63% 24 w), and STBs (44.1%, 8 w), whereas TMPRSS2 was expressed in CTBs (1.6%), EVTs (1.9% 8 w vs 20.1% 24 w), and STBs (26.5%).
Faure-Bardon V et al., 2021	Placentas from medically terminated pregnancies: mothers negative for SAR-CoV-2 (n = 5, 15–38 w); non-infected spontaneous miscarriage (n = 1, 7 w); and SARS-CoV-2 symptomatic pregnancy (n = 1, 34 w).	IHC	ACE2	ACE2 was expressed in CTBs and STBs but absent in the vascular endothelium and amnion from 7 weeks onwards. ACE2 staining was comparable between SARS-CoV-2 non-infected and SARS-CoV-2 symptomatic cases.
Li M et al., 2020	Early pregnancy placentas (6–14 w); placenta villi (8 w) and decidua (24 w).	scRNA-seq	ACE2 TMPRSS2	ACE2 gene was expressed in the decidual perivascular and stromal cells, and CTBs and STBs. TMPRSS2 was expressed in epithelial glandular cells and CTBs and low in STBs. ACE2 was low at 8 w but increased at 24 w. TMPRSS2 had a similar trend in the STBs. ACE2 and TMPRSS2 were co-expressed in EVTs, CTBs and STBs.
Lye P. et al., 2021	Placentas from pregnancies complicated with PTB (n = 12, 31.7 ± 0.93 wks); PTB with ChA (n = 13,	IHC RT-qPCR	ACE2	ACE2 mRNA expression was increased with PTM pregnancies complicated with ChA than

(*continued on next page*)

Table 1 (*continued*)

Author	Study Characteristics and Materials	Methods	Receptors/ Proteases	Main results
	29.3 ± 1.01 wks); and term pregnancies (ELCS (n = 13 38.6 ± 0.30 wks); and SVD (n = 13, 39.0 ± 0.32 wks)).			PTM alone, SVD or ELCS (at term) but ACE2 protein was not increased within ChA than age-matched PTB. ACE2 protein was increased in PTB placentas than in term placentas. The localization of ACE2 protein was in decidual stromal cells, endothelium of fetal capillaries, EVTs, STBs, and immune cells within maternal blood in the inter-villous space.
Pique-Regi et al., 2020	11 deciduas and 5 placentas (6–14 w); 9 placentas (basal plate (including decidua basalis), villi) and chorioamniotic membranes (including decidua parietalis)) from women with or without labor at term (38–40 w) or peterm labor (33–35 w); placental samples from placenta accreta (n = 1, 18 w), third-trimester samples from mothers with different conditions (n = 32); placentas from women who delivered after spontaneous labor without (n = 10) and with VUE (n = 10); PE (n = 10, 34.2 ± 2.5), SGA (n = 8, 33.9 ± 2.0), and PE + SGA (n = 10, 34.5 ± 3.8).	scRNA-seq snRNA-seq Microarray	ACE2 TMPRSS2 CD147 CTSL Furin	Placental cells and chorioamniotic membrane neglibly co-expressed ACE2 and TMPRSS2. CD147 was highly expressed in the chorioamniotic membranes and placenta throughout pregnancy. CTSL and Furin were highly abundant in placental tissue throughout gestation.
Singh M et al., 2020	11 deciduas and 5 placentas (6–14 w).	scRNA-seq	ACE2 DPP4 Furin TMPRSS2 IFITM1-3 LY6E	ACE2 and DPP4 were expressed in the CTBs and STBs Furin was broadly expressed in fetal and maternal cells but abundantly in fetal cells. EVTs expressed low levels of ACE2 or TMPRSS2 but moderate to high levels of IFITM1-3 and LY6E
Valdez et al., 2006	Placentas from spontaneous abortions (n = 5, 9.5 ± 2.2 w); ectopic pregancies (n = 6, 7.4 ± 1.9 w); normal pregancies (n = 15, 38.7 ± 0.9 w); and PE (n = 10, 35.0 ± 2.9 w).	IHC	ACE2	ACE2 was expressed in the endothelium, STBs, CTBs, and vascular smooth muscle, and in maternal stroma decidual cells and invading and intravascular trophoblasts. ACE2 expression was similar in normal term amd PE pregancies but increased in umbilical cord endothelium in PE.

ACE2: angiotensin-converting enzyme 2; CTB: cytotrophoblast; CTSB/L: cathepsin B/L; BSG/CD147: basigin/cluster of differentiation 147; ChA: chorioamnionitis; ELCS: elective cesarean section; EVT: extravillous trophoblasts; IF: immunofluorescence; IHC: immunohistochemistry; w: weeks; PE: pre-eclampsia; PTB: preterm birth; scRNA-seq: single-cell RNA sequencing; snRNA-seq: single-nuclear RNA sequencing; STB: synctiotrophoblast; SVD: spontaneous vaginal delivery; STR: villous stromal cells; TE: trophectoderm; vs: versus.

cell-to-cell spread involving infected maternal immune cells to placental trophoblasts, (ii) transcytosis of virions through immune-mediated receptors, (iii) maternal endothelial microvasculature to the endovascular trophoblasts, or (iv) as transvaginal ascending infection [51]. SARS-CoV-2 may gain access into the placenta through direct interaction of the virus with the STBs and resident immune cells that express viral receptors in peripheral maternal blood surrounding the syncytia, be trafficked by immune cells, or via endovascular trophoblasts [24,28,52]. Placental transmission via the hematogenous route may occur in maternal viremia and the interaction of the virus with its mediators in the placenta and maternal decidua to facilitate its entry [17,24,53,54]. Viremia in maternal COVID-19-positive disease is low (1–27%) [17,55]. The likelihood of viremia is increased in severe diseases [56]. The colocalization of ACE2 with early endosomes in trophoblasts suggests that ACE2 and its interacting SARS-CoV-2 are likely to enter endocytic pathways [38]. Placenta access by the virus has also been demonstrated in studies that demonstrated the entry of the virus to be through the ACE2 receptors, in which the virus appeared in all compartments of the fetal-maternal unit [33,38,57]. SARS-CoV-2 infections of the cytotrophoblast, the stroma of the villi, and Hofbauer cells were also demonstrated in placenta explants [57]. Furthermore, the susceptibility of the placenta increased with ACE2 levels in these explants. Neonatal Fc receptor (FcRn-) mediated transcytosis was also hypothesized as a possible entry route [58].

An *ex vivo* study in which ACE2 receptor was blocked demonstrated infection of the STBs, suggesting that SARS-CoV-2 may utilize other receptors, such as neurolipin 1 [57]. The expression of ACE2 and BSG in maternal endothelial cells in the decidual may be another route of entry of the virus into the placenta [24]. The destruction of trophoblasts may

Table 2
SARS-CoV-2 detection in the placenta, placental pathology and vertical transmission in COVID-19-positive mothers.

Author	No. of cases	Patient Characteristics (Gestational age at infection/Symptoms (weeks); Disease severity; Days of infection/Symptoms until delivery (range); Delivery mode	Investigations	SARS-CoV-2 Placental Infection	Placental Pathology	Vertical Transmission
Baud D et al., 2020	1	19 w; Mild; 4 days, VD.	Placenta RT-PCR, histology and IHC, and fetal autopsy.	Placental submembranes and cotyledons positive for SARS-CoV-2.	Acute subchorionitis (macrophages and neutrophils) and increased intervillous fibrin deposition and synctial knots. Funisitis and umbilical cord vasculitis.	Maternal blood, urine and vaginal swab were negative for SARS-CoV-2. Fetal swabs (axillae, blood, meconium, mouth), lung, liver, and thymus were negative for SARS-CoV-2.
Penfield CA et al., 2020	32	26-41 w (range); 6 mild, 2 severe, 3 Mild; 0–15 days; 4 CS, 7 VD.	RT-PCR (11 placentas).	3 placentas from women with severe to critical COVID-19 were positive for SARS-CoV-2.	N/A	Fetal swabs were negative for SARS-CoV-2.
Kirtsman M et al., 2020	1	35 w, Mild, 1 day; CS.	Placenta PCR and histology.	Placental swabs and placental parenchymal and chorion tissues were positive for SARS-CoV-2.	Multiple areas of infiltration by inflammatory cells (mainly CD68-positive macrophages) and extensive early infarction. Only scattered T-cells (CD3), B-cells (CD20) and neutrophils (CD15) were identified on IHC. The inflammatory infiltrate was largely confined to the intervillous space, consistent with chronic histiocytic intervillositis.	Breast milk and vaginal swab were positive for SARS-CoV-2. All 3 of the neonate's nasopharyngeal swabs were positive for SARS-CoV-2 gene targets via RT-PCR testing; neonatal plasma tested positive on day 4, and stool was positive on day 7.
Patanè L et al., 2020	22	Third trimester; 1 case Mild; 1 CS, 1 VD.	Placenta RT-PCR ISH.	2 positive placentas; In situ hybridization for SARS-CoV-2 spike protein mRNA, highlighting the presence of SARS-CoV-2 spike antigens in villous STBs.	Case 1: Chorionic villi showing chronic intervillositis with macrophages; Macrophages in the intervillous spaces highlighted by anti-CD68 IHC in 2 cases that were positive for SARS-CoV-2.	Case 1: The newborn had a positive result for COVID-19 from NP swabs that were obtained immediately after birth, 24 h later, and after 7 days. Case 2: The NP swab obtained at birth from the neonate was negative for SARS-CoV-2, but a follow-up test of an NP swab that was obtained on day 7 was positive for SARS-CoV-2.
Schoenmakers S et al., 2021	1	31 + 4 w; Mild, 8 days; CS.	Maternal, placental, and neonatal swabs for RT-PCR, serology, IHC, ISH, TEM.	Positive placenta RT-qPCR; SARS-CoV-2 RNA ISH demonstrated predominant localization of SARS-CoV-2 in the STBs; Electron microscopy confirmed the presence of SARS-CoV-2 particles in the STBs, whereas villous and fetal parenchyma showed no evidence of SARS-CoV-2 infection on IHC, ISH, or TEM.	Presence of SARS-CoV-2 particles with generalized inflammation characterized by histiocytic intervillositis with diffuse perivillous fibrin depositions with damage to the STBs associated with an intervillous inflammatory infiltrate, characterized by IHC as M2 macrophages (CD163+ and CD68^{+}), cytotoxic (CD8), and helper T-cells (CD4) as well as activated B-lymphocytes (PAX5 and CD38)	RT-qPCRs of the maternal blood, vagina, and urine were all positive over a period of 6 days, while breast milk, feces, and all neonatal samples (umbilical cord blood, urine, feces, blood, nasopharynx, and sputum from a deep tracheal aspirate) tested negative over a period of 9 days. Paediatric multisystem inflammatory syndrome—temporally associated with SARS-CoV-2 (PIMS-TS).
Hosier H et al., 2020	1	22 w; Severe; 10 days; Termination.	Placenta, umbilical cord, fetal heart and lungs RT-PCR, Histology, IHC, ISH, TEM and WGS, and maternal serology.	Placenta and umbilical cord positive for SARS-CoV-2 RNA; SARS-CoV-2 localized predominantly to STBs on IHC and ISH; viral particles seen within the cytosol of placental cells.	Marginal adherent clot with focal placental infarct; Diffuse pervillous fibrin and inflammatory infiltrate (macrophages and T cells) demonstrated by IHC for CD68 and CD3, consistent with histiocytic intervillositis	Maternal saliva and urine positive but oral and nasal swabs tested negative postoperatively. Fetal heart and lung were negative for SARS-CoV-2.
Pulinx B et al., 2020	1	22 w; Mild; 15 days; VD.	Placenta and amniotic fluid RT-PCR, IHC, and histology.	Positive placenta RT-PCR; viral localization in the placental STBs confirmed by IHC.	Chronic intervillositis and extensive intervillous fibrin depositions with ischemic necrosis of the surrounding	Amniotic fluid was positive for SARS-CoV-2.

(*continued on next page*)

Table 2 (*continued*)

Author	No. of cases	Patient Characteristics (Gestational age at infection/Symptoms (weeks); Disease severity; Days of infection/Symptoms until delivery (range); Delivery mode	Investigations	SARS-CoV-2 Placental Infection	Placental Pathology	Vertical Transmission
					villi. Aggregates of histiocytes and cytotoxic T lymphocytes in the intervillous space were also present and confirmed with IHC stainings for CD68, CD3, and CD8; chronic intervillositis and extensive intervillous fibrin depositions with ischemic necrosis of the surrounding villi. In the fetal circulation, there was nuclear debris and an increase in erythroblasts, as can be seen in fetal hypoxia.	
Richtmann R et al., 2020	5	21-38 w (range); Mild-Moderate; 1–22 days (range); 2 CS 3 VD.	Placenta and amniotic fluid for RT-PCR and histology, and fetal autopsy (1).	SARS-Cov-2 was detected by RT-PCR in placental specimens in two cases.	Acute chorioamnionitis in all five cases. Two cases had massive deposition of intervillous fibrin associated with mixed intervillitis and villitis, and intense neutrophil and lymphocyte infiltration. These findings are described as villitis of unknown etiology (VUE).	SARS-Cov-2 was detected by RT-PCR in amniotic fluid. Fetal deaths. One fetus had neutrophils inside alveolar spaces, suggestive of fetal infection.
Vivanti AJ et al., 2020	1	35 + 2 w; Mild; 3 days; CS.	Placenta and amniotic fluid RT-PCR and histology.	RT-PCR on the placenta was positive for both SARS-CoV-2 genes. An intense cytoplasmic positivity of peri-villous trophoblastic cells was diffusely observed performing immunostaining with antibody against SARS-CoV-2 N-protein.	Diffuse peri-villous fibrin deposition with infarction and acute and chronic intervillositis.	Amniotic fluid collected before rupture of membranes tested positive for both the E and S genes of SARS-CoV-2. Blood and non-bronchoscopic bronchoalveolar lavage fluid were collected for RT-PCR and both were positive for the E and S genes of SARS-CoV-2. NP and rectal swabs were first collected after having cleaned the baby at 1 h of life, and then repeated at 3 and 18 days of postnatal age: they were tested with RT-PCR and were all positive for the two SARS-CoV-2 genes. Neurological symptoms and inflammatory findings in CSF.
Hsu AL et al., 2021	1	40 + 4 w; Mild; 2 days; VD.	Placenta for IHC, histology.	Placenta was positive for SARS-CoV-2 antigens in the umbilical cord, central and peripheral disc, chorionic villi endothelial cells and rarely in trophoblasts.	Normal gross morphology. Acute uterine hypoxia (hypertrophic arteriopathy, subchorionic laminar necrosis) superimposed on chronic uterine hypoxia (extravillous trophoblasts and focal chronic villitis).	Neonate RT-PCR negative.
Facchetti F et al., 2020	101	37 + 5 w; Moderate; 0 days; VD.	Placentas for IF, IHC, ISH, RT-PCR, and TEM.	SARS-CoV-2 N and S proteins in the STB; viral RNA detected in the cytoplasm of the STB, fetal capillaries endothelium, fibroblasts, and fetal intravascular mononuclear cells; S protien and intense signal detected in the STB on IHC and ISH respectively; viral particles detected in the cytoplasm of the STB, fetal capillaries	COVID-19-positive placenta: Gross: weight 448 g (25–50 percentiles for age), no significant abnormalities were detected on cord insertion, cord vessels, fetal surface and membranes, the chorionic parenchyma was dark brown and contained multiple small pale areas Histology: histiocytic-neutrophilic intervillositis; avascular and fibrotic villi	Newborn tested positive for viral RNA at 36 and 72 h, and 17 d and developed COVID-19 pneumonia and severe respiratory distress 24 h after birth.

(*continued on next page*)

Table 2 (*continued*)

Author	No. of cases	Patient Characteristics (Gestational age at infection/Symptoms (weeks); Disease severity; Days of infection/Symptoms until delivery (range); Delivery mode	Investigations	SARS-CoV-2 Placental Infection	Placental Pathology	Vertical Transmission
				endothelium, fibroblasts, and cytoplasm of intra-capillary cells, likely corresponding to monocytes (1 placenta).	and stroma-vascular karyorrhexis, accelerated villous maturation; chorangiosis.	
Mongula JE et al., 2020	1	31 + 4 w; Moderate; 1–13 days; CS.	Placenta for PCR, IHC.	Fetal side of the placenta was positive for SARS-CoV-2.	Abnormal gross (not described). Histology: Increased perivillous fibrin deposition leading to trophoblast necrosis and intervillositis (chronic (histiocytes) and acute (granulocytes)); increased villi maturation	Vaginal swab positive. Neonatal NP swabs negative on days 4 and 7.
Menter T et al., 2021	5	35 + 5–40 + 5 w (range); Asymptomatic-Mild; 35-(-1) days; 2 CS 3 VD.	Placenta and umbilical cord for RT-qPCR, histology, and ISH.	1 placenta and umbilical cord positive; viral RNA visualized in the decidua by ISH.	Umbilical cord hypercoling in 3 placentas. Prominent lymphohistiocytic villitis and intervillositis; signs of maternal and fetal malperfusion in 100% and 40% of cases, respectively.	Amniotic fluid, breast milk and umbilical cord blood negative for SARS-CoV-2 in all cases.
Fenizia C et al., 2020	31	34.4–41.4 w (range); 26 mild, 4 severe; 1–17 days; 6 CS, 25 VD.	Maternal and newborn NP swabs, vaginal swabs, maternal and umbilical cord plasma, placenta and UC biopsies, amniotic fluid and milk for serology, inflammatory gene expression analysis (RT-PCR).	2 positive term placentas. Inflammatory hyperactivation upregulated for effector cytokines and chemokines (CXCL10, CXCL8, CCL5, CCL3, and CCL2), adaptive immunity mediators, downstream signaling molecules and TLRs.	N/A	SARS-CoV-2 genome detected in 1 UC blood and 1 vaginal swab. Anti-SARS-CoV-2 IgM and IgG antibodies in 1 milk specimen and UC blood (whose placenta tested positive). 2 fetuses NP swabs tested positive but one tested negative after 48 h.
Schueda Stonoga ET et al., 2021	1	26 + 5 w; Severe; 8 days; CS.	Placenta for RT-PCR, IHC and histology, and fetal autopsy.	SARS-CoV-2 RNA in cotyledons and membranes.	The placental disc was round, and had tan and glistening membranes peripherally attached. The umbilical cord had 3 vessels; it was 28 cm long, inserted eccentrically, and under coiled. The fetal surface was gray with normal chorionic plate vessels. The trimmed placental disc weighed 135 g and measured 12 × 12 cm (<3rd percentile). Chronic histiocytic intervillositis, maternal and fetal vascular malperfusion (infarcts, decidual vasculopathy, accelerated villous maturation).	SARS-CoV-2 RNA in umbilical cord blood aspirate. Fetal tissue samples tested negative for SARS-CoV-2. Microglial hyperplasia, mild lymphocytic infiltrate, and edema in skeletal muscle.
Shende P et al., 2021	1	8 w; Asymptomatic; 35 days; termination.	Amniotic fluid and placenta (villi) for FISH, RT-PCR, Histology. POC for routine examination and SARS-CoV-2 testing.	SARS-CoV-2 RNA for E, N, and RdRp genes detected in the placenta villli; S1 and S2 proteins localized in the cytoplasm of CTB and STB cells and fetal membranes.	Avascular villi with extensive perivascular fibrin deposition; Decidua had fibrin deposition with extensive leucocyte inflitration in the intravillous spaces and decidua, and lysis of the STB of the villous cells.	Viral RNA in amniotic fluid. Hydrops fetalis (non-autoimmune and placenta disomic for chromosomes 13, 18, 21 and X).
Zhang P et al., 2020	74	38-40 w; 20 CS, 54 VD.	Placenta for ISH, IHC and histology.	Viral particles in the STB, atrophic endometrial glandular epithelium, and subchorionic plate (Langhan's fibrinoid) 2 (ISH) placenta.	Weight mean 437 (390–519); Vasculopathy (classic type (33.8%), mixed type (9.5%), and mural hypertrophy (5.4%)); Infarcts (9.5%); chorioamnionitis (64.9%);	No viral signals in maternal or fetal tissues. Newborn tested (whose placenta tested positive for SARS-CoV-2 by ISH) positive by swab PCR at 24 h, 48 h and 7 days, but was asymptomatic.

(*continued on next page*)

Table 2 (*continued*)

Author	No. of cases	Patient Characteristics (Gestational age at infection/Symptoms (weeks); Disease severity; Days of infection/Symptoms until delivery (range); Delivery mode	Investigations	SARS-CoV-2 Placental Infection	Placental Pathology	Vertical Transmission
					meconium (32.4%); thrombosis (24.3%); villitis (23%); cord issue (1.4%); MFI (97.3%); avascular villi (93.2%).	
Feitosa Machado SS et al., 2021	1	35 w; Mild, 0 days; CS.	Placenta for RT-PCR, and fetal autopsy.	Positive placental cut surface	Placenta had an irregular shape, weighed 245g, below the percentile 3, about one eighth of fetal weight, and measured 17.5 × 14.5 × 1 cm. The maternal surface of the placenta had some lobes with little distinct borders, presenting an area of previous infarction and an area of recent infarction, peripheral, making up about 20% of the organ. The fetal membranes had thickened and yellowish areas and umbilical cord had peripheral implantation.	NP swab of the fetus was negative for SARS-CoV-2. Histological sections of the lung was impaired due to autolysis, but the presence of a mixed inflammatory infiltrate was noticeable.
Michel AS et al., 2021	1	16 + 4 w; Moderate; 7 days; VD.	Amniotic fluid and placenta for RT-PCT, IHC and histology, and fetal autopsy.	Placenta tested positive (twice same day).	Reactive villitis (histiocytes) specifically in infected villi but no intervillitis.	Amniotic fluid positive for SARS-CoV-2. Negative RT-PCR for SARS-CoV-2 in the fetal liver and lung.
Cribiù FM et al., 2021	21	37 + 3 w (median); Mild-Moderate; 9 days; 9 CS, 12 VD.	Placenta for RT-PCR, ISH, and gene expression.	10 placentas positive for SARS-CoV-2. Signature enrichment and differential expression analyses showed that genes implicated in innate antiviral immunity, chemotactic and inflammatory response, and adaptive response (such as CXCL9, CXCL10, CXLC11, CCL2, CCL7, IL6, IL21R, CD8A, GZMA, PRF1, CD68, and CD163.) were highly expressed in the placenta in 1 patient and the infected lungs, compared with the rest of the placenta samples.	Non-specific features indicative of maternal inflammation rather than direct viral infection except in one placenta with massive fibrin deposition and necrosis of the STB layer of the villi.	N/A
Sanchez J et al., 2021	2	Case 1: 36 w, Case 2: 37 w; Asymptomatic; 1–2 days; 2 CS.	Placenta for IHC, RT-PCR, and WGS.	2 placentas positive in the fetal side, endothelial cells of chorionic villi vessels proximal to both maternal and fetal sides (A1 and B1 lineage with A to G nucleotide mutation at position A23403G (D614G) and G11803T in both genomes).	Dense infiltrate of lymphoid cells around decidual vessels and endothelial cells with cytopathic changes karyomegaly and hyperchromia, especially on the maternal side.	NP swabs from the ingants negative for SARS-CoV-2 at 24 h.
Lu-Culligan A et al., 2021	39	22-40 w (range); Asymptomatic-Severe; 0–27 days; 13 CS.	Maternal plasma and placentas for IF, IHC, hstology, RT-qPCR, serology, and single-cell transcriptomics.	Viral RNA found in 1 placenta on RT-qPCR and CTB *in vitro*.	Increased intervillous fibrin (33%) but none in controls.	Noenate NP negative (whose placenta tested positive).
Zaigham M et al., 2021	1	34 + 1 w; Mild; 0 days; CS.	Maternal blood, neonatal NP swab and placenta for RT-PCR, serology, WGS, histology, IHC, and ISH.	SARS-CoV-2 RNA was positive in the cytoplasm and nucleus of CTBs and STBs in areas with intervillositis and fibrinoid depositions, with positive staining in	Gross: membranes had normal color without signs of meconium staining, placental weight 342 g, within the 10th-90th percentile for GW 34 + 0–34 + 6; UC: 3 vessels;	Maternal blood positive for SARS-CoV-2. Neonate positive for SARS-CoV-2 at 48 h and IgM- and IgG-seropositive at 14 days.

(*continued on next page*)

Table 2 (*continued*)

Author	No. of cases	Patient Characteristics (Gestational age at infection/Symptoms (weeks); Disease severity; Days of infection/Symptoms until delivery (range); Delivery mode	Investigations	SARS-CoV-2 Placental Infection	Placental Pathology	Vertical Transmission
				the villous stromal cells; ds-RNA detected in the vCTB and STB by ISH (genetic seq placenta (clade 20B/GR/B.1.1 with 11 SNPs and 1 MNPs from the reference Wuhan genome).	fibrinoid depositions (50%). Intervillous fibrinoid deposition accompanied by denudation of the villi from trophoblasts and STB with dislocated STB visible in the fibrinoid; multiple regions of dense intervillous infiltrates of neureophilic granulocytes and macorphages. Chorangiosis in areas devoid of fibrinoid depositions. IHC confirmed inflammatory cell component of the intervillitis was dominated by myeloperoxidase-positive granulocytes and CD-68-positive macrophages with sparse amounts of $CD3^{-}$ and CD-20-positive lymphocytes	
JAK B et al., 2021	1	Asymptomatic; 10 days; VD.	Placenta for ISH, RT-qPCR, nested PCR, and fetal autopsy.	Positive placenta on nested RT-PCR.	Gross: placenta and umbilical cord weighed 416g and measured 42*12*4 cm, discoid in shape with a firm reddish-colored maternal surface, spongy in appearance, and adherent clots and diffuse whitish areas; fetal surface smooth and opaque and evident vessels Intervillous and subchorionic fibrin; recent infarcts, villous agglutination, and narrowing of the intervillar spaces, an increase in synctial nodes; fetal thrombotic vasculopathy.	Fetal brain, heart, liver, and lung positive for SARS-CoV-2 on nested RT-PCR. Left fetal lung showed intense and extensive bronchopneumonia with numerous neureophis and pyocytes filling the alveoli, along with abundant amniotic fluid, fibrin deposits, and Gram-positive bacterial growth in blood culture. Right lung showed intense capillary and vascular congestion, with amniotic fluid and meconium in the alveoli.
Valdespino-Vazquez MY et al., 2021	1	13 w; Mild; 5 days; VD.	Placental and fetal autopsy for IF, IHC, histology RT-qPCR, and TEM.	SARS-CoV-2 N protein and viral particles found in the placenta; viral dsRNA.	Gross: bichorial, biamniotic. The placental weight was 25 g with measurements of 8 × 7 × 1cm with parenchymal infarcts in 25% of the examined surface. Placental infarction, with diffuse perivillous fibrin, active chronic intervillositis, and subchorial inflammation; CD163 positivity in the villous stroma and intervillous space	Positive signal to N protein detected in the placenta; dsRNA detected in the fetal and kidney; viral particles in the cytoplasmic vacuoles of lung cells.
Sinaci S et al., 2021	48	38 w (median); Mild-Moderate; 38 CS, 10 VD.	Amniotic fluid, cord blood, placenta, and vaginal secretions for serology, and RT-PCR.	1 positive placental sample.	N/A	Vaginal secretions positive for SARS-CoV-2 in two cases. Two newborns screened positive for IgG-IgM at 24 h but RT-PCR were negative. 1 newborn had a positive NP RT-PCR but the amniotic fluid, cord blood, placenta and vaginal secretions were negative.
Garcia-Ruiz I et al., 2021	45	Median: 34.7 w (range: 14–41.3 w); 29 Mild, 12 Severe, 3 Critical;	Amnitoic fluid, UC blood and placenta for IHC,	1 positive placenta on IHC and ISH.	N/A	1 case positive for SARS-CoV-2 in amniotic fluid and UC blood but negative NP.

(*continued on next page*)

Table 2 (*continued*)

Author	No. of cases	Patient Characteristics (Gestational age at infection/Symptoms (weeks); Disease severity; Days of infection/Symptoms until delivery (range); Delivery mode	Investigations	SARS-CoV-2 Placental Infection	Placental Pathology	Vertical Transmission
		Median: 21.5 days (range: 0–141 days); 15 CS, 29 VD.	ISH, RT-PCR, and serology.			1 case negative at birth returned positive at 24–48 h.
Karade S et al., 2021	1	39 w; Moderate; 2 days; VD.	Amniotic fluid, breast milk and placenta swab for RT-PCR.	Placenta positive for SARS-CoV-2 ORF 1b and RDRp.	Grossly normal. Histology not done.	Amniotic fluid positive for SARS-CoV-2 ORF 1b and RdRp. Neonate NP positive RT-PCR but returned negative on DOL 5.
Husen MF et al., 2021	36	269.5 days (39 median); 28 Asymptomatic/Mild, 8 Moderate/Severe; 19.5 days (median 33); 22 CS 14 VD.	Fetal, maternal blood, feces, and urine, placental samples for serology, IHC, RT-PCT, and histology.	Positive SARS-CoV-2 on RT-PCR and IHC.	Perivillous fibrin (23.1%), fibrinoid necrosis (10.3%); maternal $CD20^{+}$ B-cell infiltrate (12.8%).	N/A

CS: cesarean section; CSF: cerebsrospinal fluid; CTBs: cytotrophoblasts; DOL: day of life; dsRNA: double-stranded RNA; IF: immunofluoresence; IHC: immunohistochemistry; ISH: *in situ* hybridization; MFI: massive perivillous fibrinoid deposit; N/A: not available; NP: nasopharyngeal; POC: products of conception; RdRp: RNA-dependent RNA polymerase; RT-PCR: reverse transcriptase polymerase chain reaction; STBs: synciotrophoblasts; TEM: transmission electron microscopy; UC: umbilical cord; VD: vaginal delivery; WGS: whole genome sequencing.

also permit virus entry into the chorionic villi reaching the fetal vessels and eventually into fetal circulation [59]. Ischemic injury sustained during COVID-19 infection may increase the permeability of the placenta to SARS-CoV-2 [40]. Based on studies on other pathogens, it is hypothesized that vertical transmission of SARS-CoV-2 may occur once sufficient time has elapsed for the virus to breach the placenta, usually 6–8 weeks [60]. *However, in vitro* studies have demonstrated SARS-CoV-2 pseudovirus entry into placental biopsies and explants in approximately 72 h [33,38].

Genes for proteins involved in viral budding and replication and host immune response are expressed in first-trimester STBs and second-trimester EVTs that coexpress ACE2+/TMPRSS2+ [24]. The capacity for viral invasion, cell-adhesion-molecule binding, and epithelial-cell proliferation is increased in early ACE2+/TMPRSS2+ TE1 cells [37]. The EVTs and STBs are enriched in genes that encode for endosomal sorting complexes required for transport (ESCRT) and viral replication proteins and can potentially be utilized by SARS-CoV-2 [24]. Viral S protein was detected in the cytoplasm of the CTBs, STBs, and stroma cells in all pregnancy trimesters [61–63]. Besides, SARS-CoV-2 dsRNA was detected in the placenta *in vivo*, thus implying active viral replication in the placenta [64,65]. Maturation of the viral nucleocapsid (N) and spike (S) proteins in the STBs was also demonstrated *ex vivo* [57]. However, the inflammatory response was minimal, attributed to the lack of a functional endothelium and immune system in an *ex vivo* system [57]. Failure to activate postentry systems, including endosomal escape and lysosomal deacidification, may limit viral replication in the placental [38]. This may be supported by demonstrating the restriction of SARS-CoV-2 replication by IFITM proteins through its amphipathic helix domain [66]. Whole-genome sequencing (WGS) of placentas that tested positive for SARS-CoV-2 found no remarkable virus adaptation for placental infection [64,67].

Placenta immune response and dysregulation of the renin-angiotensin system during maternal COVID-19 infection may compromise placenta function and result in placental insufficiency, pathology, potential *in utero* transmission, and adverse pregnancy outcomes [24]. Dysregulation of the RAS pathway in maternal COVID-19 infection results in an imbalance of the angiogenic and antiangiogenic factors in the placenta [33]. The ACE2 receptors are downregulated, with an increase in antiangiogenic factors (soluble fms-like tyrosine kinase-1 (sFlt-1) and endoglins, nitric oxide modulators, and vasoconstrictive peptides), and reduction of the proangiogenic factors (placental growth factor (PIGF)) [33,68]. Preeclampsia in SARS-CoV-2 infection can occur via multiple mechanisms, including angiogenesis, hypoxia, imbalance of vasoactive peptides, inflammatory signaling, and platelet or thrombin activation [69]. Besides, EVTs that coexpress ACE2 and TMPRSS2 consist of invasive endovascular trophoblasts and spiral artery remodeling into the myometrium vital for fetal growth and development. Failure in remodeling of arteries results in disordered perfusion and nutrition and oxygen supply, resulting in IUGR, placental abruption, preeclampsia, and preterm labor [24]. Placental inflammatory-immune response evidenced by the infiltration of immune cells in placenta compartments may be attributed to some preterm deliveries [33,70].

A study that examined inflammatory changes at the maternal-fetal interface reported activation of the immune system at the materno-fetal interface in the absence of SARS-CoV-2 placental infection [34]. However, inflammation at the maternal-fetal interface was likely to increase the risk of complications. Genes for complement factors, interferon-stimulated, and heat shock protein family A (Hsp70) member 1A (HSPA1A) were upregulated in maternal SARS-CoV-2 infection compared to controls [34]. Genes that encode cytotoxic proteins (GNLY, GZMA, and GZMB) in NK cells, ribosomal proteins (RPL36A and RPS10) in T cells, and interferons (ISG15), and the NF-kB pathway (NFKBIA and NFKBIZ) regulators were also upregulated [34,71]. Besides, T cells' interactions with monocytes and NK cells suggest placental localized innate-to-adaptive immune cell communication [34,71].

3.4. Detection of SARS-CoV-2 placental infection

SARS-CoV-2 placental infection is defined as the detection and localization of the virus in the placenta using *in-situ* hybridization (ISH) with anti-sense and sense probes to detect replication and viral genome respectively, or immunohistochemistry to detect viral nucleocapsid (N) and spike (S) proteins [72]. Where IHC and ISH tests are not available, reverse-transcriptase polymerase chain reaction (RT-PCR) to detect and quantify viral RNA in placental homogenates or electron microscopy (EM) may be used [72]. A graded classification was proposed to determine the possibility of placenta infection with criteria such as definitive, probable, possible, unlikely, and no testing [72]. Definite diagnosis is when there is evidence of active viral replication in the placenta whereas, a probable diagnosis is when viral RNA or protein is detected in the placenta without evidence of active replication. Possible infection is when viral RNA or viral-like particles are detected in placental

homogenates by electron microscopy. An unlikely diagnosis is when the above methods report a negative result [72]. The proposed classification also gave a recommendation on placenta handling, processing, and examination.

The prevalence of placental infection in COVID-19-positive women is low (7.7–21%) [73]. A few studies have demonstrated SARS-CoV-2 placental infection diagnosis by ISH, IHC, RT-PCR, and transmission electron microscopy (TEM) [11,15,31,34,52,53,58,61,62,64,65,67,71,74–92]. Viral RNA, proteins, and particles were mainly localized in the syncytiotrophoblasts [52,58,61,62,64,80,81,86]. The infection of other placenta compartments, including the CTBs, chorionic villi, decidua, membranes, fetal capillaries endothelium, fibroblasts, fetal intravascular mononuclear cells, umbilical cord, and rarely the trophoblasts was also reported [34,58,62,64,67,76,82,83,85,86,89]. Accurate estimation of SARS-CoV-2 placental infections is limited by the fact that most studies did not use standardized diagnostic criteria and lacked all the data, such as duration of infection and maternal and viral loads [24,77]. The detection of the virus in fetal membranes, amniotic fluid, from neonates' nasopharyngeal swabs of neonates and SARS-CoV-2 IgM antibodies are implicative of placental infection and *in utero* transmission [24].

3.5. Placental pathology in COVID-19 infection

Placenta pathology in maternal COVID-19 disease can be attributed to the systemic, localized, or direct impact of SARS-COV-2 on the placenta [18,40]. Gross and microscopic examination are essential in evaluating placenta pathology [93]. Placenta pathologies in this review were categorized according to the Redline Classification of Placental Disorders, 2015 [94].

3.5.1. Gross evaluation of the placenta

Gross examination of the placenta may reveal significant pathological features, including hypoplasia, infarction, and retroplacental hemorrhage, reflecting maternal vascular malperfusion [93]. Placenta weight is a surrogate for placental function, whereas the fetoplacental weight ratio and placental disk dimensions are indicators of adequacy of placental reserve capacity in fetal growth restriction [93,95]. Gross investigations of placentas from COVID-19-positive mothers were rarely or incompletely reported. A few studies reported normal gross placenta morphology [52,64,76,92], but others found adherent clots and focal infarcts [58,65,74,89]. Weights of second and third-trimester placentas from COVID-19 positive mothers had conflicting results. Some studies reported smaller and lower weights for the expected gestational age [69,87,96], and some reported no difference [97]. Both old and new infarcts and fibrinoid deposition were also reported in a few cases [64,87]. Gross umbilical cord pathology in placental SARS-CoV-2 infection was rarely reported [98].

3.5.2. Placental vascular processes

Vascular processes in the placenta reflect a state of hypercoagulability and may be systematic or localized [93]. Placental production of plasminogen activator inhibitor-2 (PAI-2) and trophoblast tissue factor, mediated by the NF-κB pathway, disposes the placenta to thrombosis [99].

i. Maternal vascular malperfusion

SARS-CoV-2 infection results in hypoxia, and if the infection happens during pregnancy, maternal hypoxia may lead to maternal vascular malperfusion (MVM) [73]. MVM features, including acute villous maturation, decidual arteriopathy characterized by subchorionic laminar necrosis and extravillous trophoblasts, maternal vessel injury or abnormality, intervillous thrombi, were reported in SARS-CoV-2-infected placentas [11,52,74,76,84–86,89,96]. Maternal vascular malperfusion may increase with disease severity [55]. However, a prospective case-control study in which cases were managed with pharmacotherapy and oxygen therapy found no difference in placenta histopathology compared to the controls; a finding that could have been due to modification of the pathology due to the therapies used [100].

ii. Fetal vascular malperfusion

The obstruction of fetal blood flow due to umbilical cord lesions, hypercoagulability, and complications of fetal cardiac dysfunction (hypoxia) may result in fetal vascular malperfusion (FVM) [93]. FVM features including avascular villi, intramural fibrin deposition, thrombosis, and villous-stromal karyorrhexis were widely reported in placentas infected with SARS-CoV-2 [52,62,84–86,89,97,98,101]. In one study, the placenta's viral S protein and RNA staining were negative, implying systemic thrombosis, which may explain the increased likelihood of thrombosis in the fetal circulation in maternal COVID-19 positive pregnancy [101].. Placentas from mothers with mild and severe COVID-19 had higher von Willebrand factor (vWf) levels with reduced levels of VE-cadherin and claudin-5 in the chorionic villi and decidua, this revealing infarction, thrombosis, and vascular wall remodeling [102]. A meta-analysis of SARS-CoV-2 infected placentas' pathology reported comparable maternal and fetal vascular changes with non-infected placentas [103].

3.5.3. Inflammatory-immune processes

Placental infections may promote localized immune response, including inflammation resulting from an infection or immune-mediated response [94]. Though not widely reported, maternal and fetal inflammatory features in COVID-19 positive placentas included acute chorioamnionitis, subchorionitis, funisitis, and umbilical cord vasculitis [35,86,96,104]. Chronic infection-associated inflammatory lesions in SARS-CoV-2 infected placentas included chronic villitis with increased inflammatory lymphocytic infiltrate in the absence of other morbidities and chronic intervillositis with T-lymphocytes and macrophages ($CD68^{+}$) infiltrate associated with viral RNA [53,61,65,82,105]. Immune or idiopathic inflammatory features (villitis of unknown etiology (VUE) and associated lesions such as chronic chorioamnionitis, chronic villitis, and lymphoplasmacytic deciduitis were reported in placentas positive for SARS-CoV-2 [62,64,67,78,88]. VUE complicated by COVID-19 was rarely reported [97,106]. Chronic histiocytic intervillositis (CHI), which is rarely repeatedly etiologically associated with any infectious agent, was reported in placentas infected with SARS-CoV-2 [58,59,74,81,84,85]. B-cell infiltration in CHI was rarely reported and is thought to be a signature that may differentiate SARS-CoV-2 associated histiocytic intervillositis from CHI of unknown etiology (CHIUE) [80,82,107]. However, further inquiry is necessary to validate these findings. A meta-analysis cited above also found comparable inflammatory lesions between SARS-CoV-2 infected and non-infected placentas [103]. Other placental pathologies that were rarely reported in placentas from mothers with COVID-19 infection were calcifications [108], meconium-associated changes, and fibrin deposition in variable degrees within the villi and surrounding area with increased local syncytial nodules [11,34,53,62,64,65,74,81,97,108].

3.6. In utero transmission of SARS-CoV-2

The prevalence of vertical transmission in COVID-19-positive pregnant mothers is between 3 and 5% [4,13]. Vertical transmission can occur *in utero*, intrapartum, or the early postnatal period [109]. *In utero* transmission can occur via the ascending route or hematogenously, whereas intrapartum and postnatal transmission occurs when the neonate is exposed to the offending pathogen during delivery and breastfeeding. Infection to the neonate may also occur through respiratory droplets or contact with contaminated fomites [49,109]. *In utero* transmission of SARS-CoV-2 is possible owing to the possible occurrence

of viremia in maternal COVID-19 disease, localization of SARS-CoV-2 mediators in the placenta, and vascular injury in SARS-CoV-2 placental infection [109]. The criteria for the resolution of vertical transmission include diagnosis of maternal infection, choice of tests to evaluate the viability of *in utero* or intrapartum exposure, and tests to ascertain the persistence of the virus, later exposure, or fetal/neonatal immune response to the virus [109]. Vertical transmission timing is classified as confirmed, possible, unlikely, or indeterminate based on the above criteria. Confirmed *in utero* transmission is when the maternal SARS-CoV-2 is positive anytime during pregnancy, the fetus tests positive for SARS-CoV-2 at <24 h, a positive molecular test (ISH or RT-PCR) in sterile samples such as amniotic fluid, neonatal blood, lower respiratory tract samples, cerebrospinal fluid or placenta. Positive IgA or IgM serology and positive RT-PCR of sterile samples [109]. A few confirmed cases of *in utero* transmission of SARS-CoV-2 have been reported in the literature [15,53,64,91]. SARS-CoV-2 infection of the placenta does not always translate to vertical transmission but may cause placenta insufficiency and pathology, resulting in perinatal morbidity and mortality without infection [72,90]. Studies that addressed the mode of delivery found that the positivity rate of SARS-CoV-2 neonatal infection in cesarean and vaginal deliveries is relatively similar (5.3 vs. 2.7%) [110], despite the significantly higher risk of contamination during vaginal delivery [109].

3.7. Pregnancy outcomes in SARS-CoV-2 placental infection

More preterm deliveries were observed in COVID-19-positive pregnant women with active disease than those with a previous infection or recovered [11]. SARS-CoV-2 placental infection changes, such as RAS dysregulation and inflammation, can result in placental deficiency and possible adverse maternal and neonatal outcomes, such as preeclampsia and premature births [33,80,111,112]. Intervillositis may be associated with high IUGR, severe early-pregnancy preeclampsia, and high rates of miscarriage [58]. Besides, severe placental pathological changes have been demonstrated with the neonates born of such pregnancies showing neurological deficits and fetal inflammatory multisystem-like syndrome with coronary artery ectasia temporarily associated with SARS-CoV-2 [71,80]. This has particularly been associated with the high viral load of SARS-CoV-2. However, SARS-CoV-2 infection of the placenta is not associated with any particular pathological or adverse maternal or neonatal outcomes [71]. Both the infected and uninfected neonatal outcomes are the same [113], implying that the placenta provides an effective maternal-neonatal barrier against the virus [43,71].

4. Conclusions and next steps

In conclusion, SARS-CoV-2 pathogenesis and pathophysiology in the placenta are inadequately examined, and only a few case studies have elucidated the mechanism of entry, immune response, and pathology in SARS-CoV-2 placental infection. The lack of standardization may limit drawing clear-cut conclusions on the profile of SARS-CoV-2 in the placenta. The expression of SARS-CoV-2 receptors and proteases in the placenta predisposes the placenta to SARS-CoV-2 infection. However, the impact of the placental barrier and immune response in resisting or increasing the susceptibility of the placenta to SARS-CoV-2 infection is unclear. Generally, placental morphological and histological changes in SARS-CoV-2 infected placentas are comparable to non-infected placentas. This review found very few studies on umbilical cord pathology and acute inflammatory features in placentas from COVID-19-positive mothers. Therefore, there is a need to conduct more studies to look at the acute inflammatory response and cord pathology specifically. To accurately evaluate the impact of SARS-CoV-2 on the placenta, researchers need to adopt current standardization and critically evaluate placental immune response, resistance mechanisms, and entry mechanisms during SARS-CoV-2 infection of the placenta. Therefore, studies need to examine the histological and morphological changes associated with SARS-CoV-2 infections and their impact on pregnancy outcomes. Other factors that may influence placental response to infection such as ACE2 expression variation, maternal clinical characteristics such as nutritional status, parity, previous infections with tropical viruses, and comorbidities such as preeclampsia, as well as effects on timing and use of different therapeutic interventions on fetal-maternal outcomes should also be evaluated.

Funding

No external funding.

Conflict of interest

No conflict of interest to declare.

References

[1] T.K. Burki, Coronavirus in China, Lancet Respir. Med. 8 (3) (2020) 238, https://doi.org/10.1016/S2213-2600(20)30056-4.
[2] H. Harapan, N. Itoh, A. Yufika, et al., Coronavirus disease 2019 (COVID-19): a literature review, J Infect Public Health 13 (5) (2020) 667–673, https://doi.org/10.1016/j.jiph.2020.03.019.
[3] M. Cevik, K. Kuppalli, J. Kindrachuk, M. Peiris, Virology, transmission, and pathogenesis of SARS-CoV-2, BMJ 371 (2020) m3862, https://doi.org/10.1136/bmj.m3862. Published 2020 Oct 23.
[4] A.M. Kotlyar, O. Grechukhina, A. Chen, et al., Vertical transmission of coronavirus disease 2019: a systematic review and meta-analysis, Am. J. Obstet. Gynecol. 224 (1) (2021) 35–53, https://doi.org/10.1016/j.ajog.2020.07.049, e3.
[5] A.G. Harrison, T. Lin, P. Wang, Mechanisms of SARS-CoV-2 transmission and pathogenesis, Trends Immunol. 41 (12) (2020) 1100–1115, https://doi.org/10.1016/j.it.2020.10.004.
[6] J. Shang, Y. Wan, C. Luo, et al., Cell entry mechanisms of SARS-CoV-2, Proc. Natl. Acad. Sci. U. S. A. 117 (21) (2020) 11727–11734, https://doi.org/10.1073/pnas.2003138117.
[7] P. Fuentes-Prior, Priming of SARS-CoV-2 S protein by several membrane-bound serine proteinases could explain enhanced viral infectivity and systemic COVID-19 infection, J. Biol. Chem. 296 (2021), 100135, https://doi.org/10.1074/jbc.REV120.015980.
[8] J.K. Millet, G.R. Whittaker, Host cell proteases: critical determinants of coronavirus tropism and pathogenesis, Virus Res. 202 (2015) 120–134, https://doi.org/10.1016/j.virusres.2014.11.021.
[9] C.S. Stevens, K.Y. Oguntuyo, B. Lee, Proteases and variants: context matters for SARS-CoV-2 entry assays, Curr Opin Virol 50 (2021) 49–58, https://doi.org/10.1016/j.coviro.2021.07.004.
[10] P.M. Varghese, A.G. Tsolaki, H. Yasmin, et al., Host-pathogen interaction in COVID-19: pathogenesis, potential therapeutics and vaccination strategies, Immunobiology 225 (6) (2020), 152008, https://doi.org/10.1016/j.imbio.2020.152008.
[11] M.F. Husen, L.E. van der Meeren, R.M. Verdijk, et al., Unique severe COVID-19 placental signature independent of severity of clinical maternal symptoms, Viruses 13 (8) (2021) 1670, https://doi.org/10.3390/v13081670. Published 2021 Aug 23.
[12] G. Reinhard, A. Noll, H. Schlebusch, P. Mallmann, A.V. Ruecker, Shifts in the TH1/TH2 balance during human pregnancy correlate with apoptotic changes, Biochem. Biophys. Res. Commun. 245 (3) (1998) 933–938, https://doi.org/10.1006/bbrc.1998.8549.
[13] M. Jafari, A. Pormohammad, S.A. Sheikh Neshin, et al., Clinical characteristics and outcomes of pregnant women with COVID-19 and comparison with control patients: a systematic review and meta-analysis, Rev. Med. Virol. 31 (5) (2021) 1–16, https://doi.org/10.1002/rmv.2208.
[14] B. Chmielewska, I. Barratt, R. Townsend, et al., Effects of the COVID-19 pandemic on maternal and perinatal outcomes: a systematic review and meta-analysis [published correction appears in Lancet Glob Health. 2021 Jun;9(6):e758], Lancet Global Health 9 (6) (2021) e759–e772, https://doi.org/10.1016/S2214-109X(21)00079-6.
[15] C. Fenizia, M. Biasin, I. Cetin, et al., Analysis of SARS-CoV-2 vertical transmission during pregnancy, Nat. Commun. 11 (1) (2020) 5128, https://doi.org/10.1038/s41467-020-18933-4. Published 2020 Oct 12.
[16] M. Karimi-Zarchi, H. Neamatzadeh, S.A. Dastgheib, et al., Vertical transmission of coronavirus disease 19 (COVID-19) from infected pregnant mothers to neonates: a review, Fetal Pediatr. Pathol. 39 (3) (2020) 246–250, https://doi.org/10.1080/15513815.2020.1747120.
[17] A. Lamouroux, T. Attie-Bitach, J. Martinovic, M. Leruez-Ville, Y. Ville, Evidence for and against vertical transmission for severe acute respiratory syndrome coronavirus 2, Am. J. Obstet. Gynecol. 223 (1) (2020) 91.e1–91.e4, https://doi.org/10.1016/j.ajog.2020.04.039.
[18] S. Komine-Aizawa, K. Takada, S. Hayakawa, Placental barrier against COVID-19, Placenta 99 (2020) 45–49, https://doi.org/10.1016/j.placenta.2020.07.022.

[19] O. Celik, A. Saglam, B. Baysal, et al., Factors preventing materno-fetal transmission of SARS-CoV-2, Placenta 97 (2020) 1–5, https://doi.org/10.1016/j.placenta.2020.05.012.

[20] R. Pique-Regi, R. Romero, A.L. Tarca, et al., Does the human placenta express the canonical cell entry mediators for SARS-CoV-2? Elife 9 (2020), e58716 https://doi.org/10.7554/eLife.58716. Published 2020 Jul 14.

[21] A.C. Simões E Silva, C.R.V. Leal, Is SARS-CoV-2 vertically transmitted? Front Pediatr 8 (2020) 276, https://doi.org/10.3389/fped.2020.00276. Published 2020 May 15.

[22] K. Grimminck, L.A.M. Santegoets, F.C. Siemens, P.L.A. Fraaij, I.K.M. Reiss, S. Schoenmakers, No evidence of vertical transmission of SARS-CoV-2 after induction of labour in an immune-suppressed SARS-CoV-2-positive patient, BMJ Case Rep. 13 (6) (2020), e235581, https://doi.org/10.1136/bcr-2020-235581. Published 2020 Jun 30.

[23] Y. Wang, Y. Wang, W. Luo, et al., A comprehensive investigation of the mRNA and protein level of ACE2, the putative receptor of SARS-CoV-2, in human tissues and blood cells, Int. J. Med. Sci. 17 (11) (2020) 1522–1531, https://doi.org/10.7150/ijms.46695. Published 2020 Jun 18.

[24] N. Ashary, A. Bhide, P. Chakraborty, et al., Single-cell RNA-seq identifies cell subsets in human placenta that highly expresses factors driving pathogenesis of SARS-CoV-2, Front. Cell Dev. Biol. 8 (2020) 783, https://doi.org/10.3389/fcell.2020.00783. Published 2020 Aug 19.

[25] F. Hikmet, L. Méar, Å. Edvinsson, P. Micke, M. Uhlén, C. Lindskog, The protein expression profile of ACE2 in human tissues, Mol. Syst. Biol. 16 (7) (2020), e9610, https://doi.org/10.15252/msb.20209610.

[26] V. Faure-Bardon, P. Isnard, N. Roux, et al., Protein expression of angiotensin-converting enzyme 2, a SARS-CoV-2-specific receptor, in fetal and placental tissues throughout gestation: new insight for perinatal counseling, Ultrasound Obstet. Gynecol. 57 (2) (2021) 242–247, https://doi.org/10.1002/uog.22178.

[27] G. Valdés, L.A. Neves, L. Anton, et al., Distribution of angiotensin-(1-7) and ACE2 in human placentas of normal and pathological pregnancies, Placenta 27 (2–3) (2006) 200–207, https://doi.org/10.1016/j.placenta.2005.02.015.

[28] P. Lye, C.E. Dunk, J. Zhang, et al., ACE2 is expressed in immune cells that infiltrate the placenta in infection-associated preterm birth, Cells 10 (7) (2021) 1724, https://doi.org/10.3390/cells10071724. Published 2021 Jul 8.

[29] N. Azinheira Nobrega Cruz, D. Stoll, D.E. Casarini, M. Bertagnolli, Role of ACE2 in pregnancy and potential implications for COVID-19 susceptibility, Clin. Sci. (Lond.) 135 (15) (2021) 1805–1824, https://doi.org/10.1042/CS20210284.

[30] M. Li, L. Chen, J. Zhang, C. Xiong, X. Li, The SARS-CoV-2 receptor ACE2 expression of maternal-fetal interface and fetal organs by single-cell transcriptome study, PLoS One 15 (4) (2020), e0230295, https://doi.org/10.1371/journal.pone.0230295. Published 2020 Apr 16.

[31] C.A. Penfield, S.G. Brubaker, M.A. Limaye, et al., Detection of severe acute respiratory syndrome coronavirus 2 in placental and fetal membrane samples, Am J Obstet Gynecol MFM 2 (3) (2020), 100133, https://doi.org/10.1016/j.ajogmf.2020.100133.

[32] R. Ferrer-Oliveras, M. Mendoza, S. Capote, et al., Immunological and physiopathological approach of COVID-19 in pregnancy, Arch. Gynecol. Obstet. 304 (1) (2021) 39–57, https://doi.org/10.1007/s00404-021-06061-3.

[33] S. Verma, C.S. Joshi, R.B. Silverstein, M. He, E.B. Carter, I.U. Mysorekar, SARS-CoV-2 colonization of maternal and fetal cells of the human placenta promotes alteration of local renin-angiotensin system, Med (N Y). 2 (5) (2021) 575–590, https://doi.org/10.1016/j.medj.2021.04.009, e5.

[34] A. Lu-Culligan, A.R. Chavan, P. Vijayakumar, et al., Maternal respiratory SARS-CoV-2 infection in pregnancy is associated with a robust inflammatory response at the maternal-fetal interface, Med (N Y). 2 (5) (2021) 591–610, https://doi.org/10.1016/j.medj.2021.04.016, e10.

[35] F.B. Constantino, S.S. Cury, C.R. Nogueira, R.F. Carvalho, L.A. Justulin, Prediction of non-canonical routes for SARS-CoV-2 infection in human placenta cells, Front. Mol. Biosci. 8 (2021), 614728, https://doi.org/10.3389/fmolb.2021.614728. Published 2021 Nov 8.

[36] M. Singh, V. Bansal, C. Feschotte, A single-cell RNA expression map of human coronavirus entry factors, Cell Rep. 32 (12) (2020), 108175, https://doi.org/10.1016/j.celrep.2020.108175.

[37] D. Cui, Y. Liu, X. Jiang, et al., Single-cell RNA expression profiling of SARS-CoV-2-related ACE2 and TMPRSS2 in human trophectoderm and placenta, Ultrasound Obstet. Gynecol. 57 (2) (2021) 248–256, https://doi.org/10.1002/uog.22186.

[38] Y. Ouyang, T. Bagalkot, W. Fitzgerald, et al., Term human placental trophoblasts express SARS-CoV-2 entry factors ACE2, TMPRSS2, and furin, mSphere 6 (2) (2021), e00250, https://doi.org/10.1128/mSphere.00250-21, 21. Published 2021 Apr 14.

[39] L.B. Argueta, L.A. Lacko, Y. Bram, et al., SARS-CoV-2 infects syncytiotrophoblast and activates inflammatory responses in the placenta, Preprint. *bioRxiv*. 2021 (2021), https://doi.org/10.1101/2021.06.01.446676, 06.01.446676. Published 2021 Jun 17.

[40] A.P. Mahyuddin, A. Kanneganti, J.J.L. Wong, et al., Mechanisms and evidence of vertical transmission of infections in pregnancy including SARS-CoV-2s, Prenat. Diagn. 40 (13) (2020) 1655–1670, https://doi.org/10.1002/pd.5765.

[41] J.R. Robbins, A.I. Bakardjiev, Pathogens and the placental fortress, Curr. Opin. Microbiol. 15 (1) (2012) 36–43, https://doi.org/10.1016/j.mib.2011.11.006.

[42] T.W. Lyden, C.L. Anderson, J.M. Robinson, The endothelium but not the syncytiotrophoblast of human placenta expresses caveolae, Placenta 23 (8–9) (2002) 640–652, https://doi.org/10.1053/plac.2002.0847.

[43] N.N. Kreis, A. Ritter, F. Louwen, J. Yuan, A message from the human placenta: structural and immunomodulatory defense against SARS-CoV-2, Cells 9 (8) (2020) 1777, https://doi.org/10.3390/cells9081777. Published 2020 Jul 25.

[44] X.J. Lv, Y.Y. Li, Y.J. Zhang, M. Mao, G.S. Qian, Over-expression of caveolin-1 aggravate LPS-induced inflammatory response in AT-1 cells via up-regulation of cPLA2/p38 MAPK, Inflamm. Res. 59 (7) (2010) 531–541, https://doi.org/10.1007/s00011-010-0157-9.

[45] E. Delorme-Axford, R.B. Donker, J.F. Mouillet, et al., Human placental trophoblasts confer viral resistance to recipient cells, Proc. Natl. Acad. Sci. U. S. A. 110 (29) (2013) 12048–12053, https://doi.org/10.1073/pnas.1304718110.

[46] E. Delorme-Axford, A. Bayer, Y. Sadovsky, C.B. Coyne, Autophagy as a mechanism of antiviral defense at the maternal-fetal interface, Autophagy 9 (12) (2013) 2173–2174, https://doi.org/10.4161/auto.26558.

[47] E.F. Cornish, I. Filipovic, F. Åsenius, D.J. Williams, T. McDonnell, Innate immune responses to acute viral infection during pregnancy, Front. Immunol. 11 (2020), 572567, https://doi.org/10.3389/fimmu.2020.572567. Published 2020 Sep. 30.

[48] M. Mourad, T. Jacob, E. Sadovsky, et al., Placental response to maternal SARS-CoV-2 infection, Sci. Rep. 11 (1) (2021) 14390, https://doi.org/10.1038/s41598-021-93931-0. Published 2021 Jul 13.

[49] A. Spinillo, A.D. Iacobone, I.G. Calvino, I. Alberi, B. Gardella, The role of the placenta in feto-neonatal infections, Early Hum. Dev. 90 (Suppl 1) (2014) S7–S9, https://doi.org/10.1016/S0378-3782(14)70003-9.

[50] S.E. Ander, E.N. Rudzki, N. Arora, Y. Sadovsky, C.B. Coyne, J.P. Boyle, Human placental syncytiotrophoblasts restrict *Toxoplasma gondii* attachment and replication and respond to infection by producing immunomodulatory chemokines, mBio 9 (1) (2018), e01678, https://doi.org/10.1128/mBio.01678-17, 17. Published 2018 Jan 9.

[51] M. León-Juárez, M. Martínez-Castillo, L.D. González-García, et al., Cellular and molecular mechanisms of viral infection in the human placenta, Pathog Dis 75 (7) (2017) ftx093, https://doi.org/10.1093/femspd/ftx093.

[52] F. Facchetti, M. Bugatti, E. Drera, et al., SARS-CoV2 vertical transmission with adverse effects on the newborn revealed through integrated immunohistochemical, electron microscopy and molecular analyses of Placenta, EBioMedicine 59 (2020), 102951, https://doi.org/10.1016/j.ebiom.2020.102951.

[53] A.J. Vivanti, C. Vauloup-Fellous, S. Prevot, et al., Transplacental transmission of SARS-CoV-2 infection, Nat. Commun. 11 (1) (2020) 3572, https://doi.org/10.1038/s41467-020-17436-6. Published 2020 Jul 14.

[54] C. Egloff, C. Vauloup-Fellous, O. Picone, L. Mandelbrot, P. Roques, Evidence and possible mechanisms of rare maternal-fetal transmission of SARS-CoV-2, J. Clin. Virol. 128 (2020), 104447, https://doi.org/10.1016/j.jcv.2020.104447.

[55] A.G. Edlow, J.Z. Li, A.Y. Collier, et al., Assessment of maternal and neonatal SARS-CoV-2 viral load, transplacental antibody transfer, and placental pathology in pregnancies during the COVID-19 pandemic, JAMA Netw. Open 3 (12) (2020), e2030455, https://doi.org/10.1001/jamanetworkopen.2020.30455. Published 2020 Dec 1.

[56] M.I. Andersson, C.V. Arancibia-Carcamo, K. Auckland, et al., SARS-CoV-2 RNA detected in blood products from patients with COVID-19 is not associated with infectious virus, Wellcome Open Res 5 (2020) 181, https://doi.org/10.12688/wellcomeopenres.16002.2. Published 2020 Oct 12.

[57] A. Fahmi, M. Brügger, T. Démoulins, et al., SARS-CoV-2 can infect and propagate in human placenta explants, Cell Rep Med 2 (12) (2021), 100456, https://doi.org/10.1016/j.xcrm.2021.100456.

[58] H. Hosier, S.F. Farhadian, R.A. Morotti, et al., SARS-CoV-2 infection of the placenta, J. Clin. Invest. 130 (9) (2020) 4947–4953, https://doi.org/10.1172/JCI139569.

[59] D.A. Schwartz, M. Baldewijns, A. Benachi, et al., Chronic histiocytic intervillositis with trophoblast necrosis is a risk factor Associated with placental infection from coronavirus disease 2019 (COVID-19) and intrauterine maternal-fetal severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) transmission in live-born and stillborn infants, Arch. Pathol. Lab Med. 145 (5) (2021) 517–528, https://doi.org/10.5858/arpa.2020-0771-SA.

[60] C. Gengler, E. Dubruc, G. Favre, G. Greub, L. de Leval, D. Baud, SARS-CoV-2 ACE-receptor detection in the placenta throughout pregnancy, Clin. Microbiol. Infect. 27 (3) (2021) 489–490, https://doi.org/10.1016/j.cmi.2020.09.049.

[61] L. Patanè, D. Morotti, M.R. Giunta, et al., Vertical transmission of coronavirus disease 2019: severe acute respiratory syndrome coronavirus 2 RNA on the fetal side of the placenta in pregnancies with coronavirus disease 2019-positive mothers and neonates at birth, Am J Obstet Gynecol MFM 2 (3) (2020), 100145, https://doi.org/10.1016/j.ajogmf.2020.100145.

[62] P. Shende, P. Gaikwad, M. Gandhewar, et al., Persistence of SARS-CoV-2 in the first trimester placenta leading to transplacental transmission and fetal demise from an asymptomatic mother, Hum. Reprod. 36 (4) (2021) 899–906, https://doi.org/10.1093/humrep/deaa367.

[63] E. Taglauer, Y. Benarroch, K. Rop, et al., Consistent localization of SARS-CoV-2 spike glycoprotein and ACE2 over TMPRSS2 predominance in placental villi of 15 COVID-19 positive maternal-fetal dyads, Placenta 100 (2020) 69–74, https://doi.org/10.1016/j.placenta.2020.08.015.

[64] M. Zaigham, A. Holmberg, M.L. Karlberg, et al., Intrauterine vertical SARS-CoV-2 infection: a case confirming transplacental transmission followed by divergence of the viral genome, BJOG 128 (8) (2021) 1388–1394, https://doi.org/10.1111/1471-0528.16682.

[65] M.Y. Valdespino-Vázquez, C.A. Helguera-Repetto, M. León-Juárez, et al., Fetal and placental infection with SARS-CoV-2 in early pregnancy, J. Med. Virol. 93 (7) (2021) 4480–4487, https://doi.org/10.1002/jmv.26965.

[66] G. Shi, A.D. Kenney, E. Kudryashova, et al., Opposing activities of IFITM proteins in SARS-CoV-2 infection, EMBO J. 40 (3) (2021), e106501, https://doi.org/10.15252/embj.2020106501.

[67] J. Sanchez, P. Vigil-De Gracia, E. Guerrero, et al., Severe acute respiratory syndrome coronavirus 2 detected in placentas of 2 coronavirus disease 2019-positive asymptomatic pregnant women-case report, AJOG Glob Rep 1 (1) (2021), 100001, https://doi.org/10.1016/j.xagr.2020.100001.

[68] W.O. Beys-da-Silva, R.L. da Rosa, L. Santi, et al., The risk of COVID-19 for pregnant women: evidences of molecular alterations associated with preeclampsia in SARS-CoV-2 infection, Biochim. Biophys. Acta, Mol. Basis Dis. 1867 (3) (2021), 165999, https://doi.org/10.1016/j.bbadis.2020.165999.

[69] A. Conde-Agudelo, R. Romero, SARS-CoV-2 infection during pregnancy and risk of preeclampsia: a systematic review and meta-analysis [published online ahead of print, 2021 Jul 21], Am. J. Obstet. Gynecol. (2021), https://doi.org/10.1016/j.ajog.2021.07.009. S0002-9378(21)*00795*-X.

[70] D.J. Jamieson, S.A. Rasmussen, An update on COVID-19 and pregnancy [published online ahead of print, 2021 Sep 14], Am. J. Obstet. Gynecol. (2021), https://doi.org/10.1016/j.ajog.2021.08.054. S0002-9378(21)00991-1.

[71] F.M. Cribiù, R. Erra, L. Pugni, et al., Severe SARS-CoV-2 placenta infection can impact neonatal outcome in the absence of vertical transmission, J. Clin. Invest. 131 (6) (2021), e145427, https://doi.org/10.1172/JCI145427.

[72] D.J. Roberts, A.G. Edlow, R.J. Romero, et al., A standardized definition of placental infection by SARS-CoV-2, a consensus statement from the national institutes of health/eunice kennedy shriver national institute of child Health and human development SARS-CoV-2 placental infection workshop, Am. J. Obstet. Gynecol. 225 (6) (2021) 593.e1–593.e9, https://doi.org/10.1016/j.ajog.2021.07.029.

[73] M.C. Sharps, D.J.L. Hayes, S. Lee, et al., A structured review of placental morphology and histopathological lesions associated with SARS-CoV-2 infection, Placenta 101 (2020) 13–29, https://doi.org/10.1016/j.placenta.2020.08.018.

[74] J.E. Mongula, M.W.E. Frenken, G. van Lijnschoten, et al., COVID-19 during pregnancy: non-reassuring fetal heart rate, placental pathology and coagulopathy, Ultrasound Obstet. Gynecol. 56 (5) (2020) 773–776, https://doi.org/10.1002/uog.22189.

[75] J.L. Hecht, B. Quade, V. Deshpande, et al., SARS-CoV-2 can infect the placenta and is not associated with specific placental histopathology: a series of 19 placentas from COVID-19-positive mothers, Mod. Pathol. 33 (11) (2020) 2092–2103, https://doi.org/10.1038/s41379-020-0639-4.

[76] A.L. Hsu, M. Guan, E. Johannesen, et al., Placental SARS-CoV-2 in a pregnant woman with mild COVID-19 disease, J. Med. Virol. 93 (2) (2021) 1038–1044, https://doi.org/10.1002/jmv.26386.

[77] J.C. Watkins, V.F. Torous, D.J. Roberts, Defining severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) placentitis, Arch. Pathol. Lab Med. 145 (11) (2021) 1341–1349, https://doi.org/10.5858/arpa.2021-0246-SA.

[78] R. Richtmann, M.R. Torloni, A.R. Oyamada Otani, et al., Fetal deaths in pregnancies with SARS-CoV-2 infection in Brazil: a case series, Case Rep Womens Health 27 (2020), e00243, https://doi.org/10.1016/j.crwh.2020.e00243. Published 2020 Jul 12.

[79] G.N. Algarroba, P. Rekawek, S.A. Vahanian, et al., Visualization of severe acute respiratory syndrome coronavirus 2 invading the human placenta using electron microscopy, Am. J. Obstet. Gynecol. 223 (2) (2020) 275–278, https://doi.org/10.1016/j.ajog.2020.05.023.

[80] S. Schoenmakers, P. Snijder, R.M. Verdijk, et al., Severe acute respiratory syndrome coronavirus 2 placental infection and inflammation leading to fetal distress and neonatal multi-organ failure in an asymptomatic woman, J Pediatric Infect Dis Soc 10 (5) (2021) 556–561, https://doi.org/10.1093/jpids/piaa153.

[81] B. Pulinx, D. Kieffer, I. Michiels, et al., Vertical transmission of SARS-CoV-2 infection and preterm birth, Eur. J. Clin. Microbiol. Infect. Dis. 39 (12) (2020) 2441–2445, https://doi.org/10.1007/s10096-020-03964-y.

[82] M. Kirtsman, Y. Diambomba, S.M. Poutanen, et al., Probable congenital SARS-CoV-2 infection in a neonate born to a woman with active SARS-CoV-2 infection, CMAJ (Can. Med. Assoc. J.) 192 (24) (2020) E647–E650, https://doi.org/10.1503/cmaj.200821.

[83] D. Baud, G. Greub, G. Favre, et al., Second-trimester miscarriage in a pregnant woman with SARS-CoV-2 infection, JAMA 323 (21) (2020) 2198–2200, https://doi.org/10.1001/jama.2020.7233.

[84] T. Menter, K.D. Mertz, S. Jiang, et al., Placental pathology findings during and after SARS-CoV-2 infection: features of villitis and malperfusion, Pathobiology 88 (1) (2021) 69–77, https://doi.org/10.1159/000511324.

[85] E.T.S. Stonoga, L. de Almeida Lanzoni, P.Z. Rebutini, et al., Intrauterine transmission of SARS-CoV-2, Emerg. Infect. Dis. 27 (2) (2021) 638–641, https://doi.org/10.3201/eid2702.203824.

[86] P. Zhang, C. Salafia, T. Heyman, C. Salafia, S. Lederman, B. Dygulska, Detection of severe acute respiratory syndrome coronavirus 2 in placentas with pathology and vertical transmission, Am J Obstet Gynecol MFM 2 (4) (2020), 100197, https://doi.org/10.1016/j.ajogmf.2020.100197.

[87] S.S. Feitosa Machado, Analysis of placenta with SARS-CoV-2 and fetal death: case report, J Psychiatry Res Rev Reports 3 (1) (2021) 1–4.

[88] A.S. Michel, V. De Logiviere, A. Schnuriger, M. Lefebvre, E. Maisonneuve, G. Kayem, Description of a late miscarriage case at 16 Weeks of Gestation associated with a SARS-CoV-2 infection, J Gynecol Obstet Hum Reprod 50 (3) (2021), 102064, https://doi.org/10.1016/j.jogoh.2021.102064.

[89] B. Jak, G. Zanirati, F.V.F. Rodrigues, et al., Case report: placental maternal vascular malperfusion affecting late fetal development and multiorgan infection caused by SARS-CoV-2 in patient with PAI-1 4G/5G polymorphism, Front. Med. 8 (2021), 624166, https://doi.org/10.3389/fmed.2021.624166. Published 2021 Feb 17.

[90] S. Sinaci, D.F. Ocal, B. Seven, et al., Vertical transmission of SARS-CoV-2: a prospective cross-sectional study from a tertiary center, J. Med. Virol. 93 (10) (2021) 5864–5872, https://doi.org/10.1002/jmv.27128.

[91] I. Garcia-Ruiz, E. Sulleiro, B. Serrano, et al., Congenital infection of SARS-CoV-2 in live-born neonates: a population-based descriptive study, Clin. Microbiol. Infect. 27 (10) (2021) 1521.e1–1521.e5, https://doi.org/10.1016/j.cmi.2021.06.016.

[92] S. Karade, A.K. Vishal, S. Sen, N. Bewal, R.M. Gupta, Probable vertical transmission of severe acute respiratory syndrome coronavirus 2 infection from mother to neonate, Med. J. Armed Forces India 77 (Suppl 2) (2021) S490–S493, https://doi.org/10.1016/j.mjafi.2020.11.026.

[93] T.Y. Khong, E.E. Mooney, I. Ariel, et al., Sampling and definitions of placental lesions: amsterdam placental workshop group consensus statement, Arch. Pathol. Lab Med. 140 (7) (2016) 698–713, https://doi.org/10.5858/arpa.2015-0225-CC.

[94] R.W. Redline, Classification of placental lesions, Am. J. Obstet. Gynecol. 213 (4 Suppl) (2015) S21–S28, https://doi.org/10.1016/j.ajog.2015.05.056.

[95] M.C. Roland, C.M. Friis, N. Voldner, et al., Fetal growth versus birthweight: the role of placenta versus other determinants, PLoS One 7 (6) (2012), e39324, https://doi.org/10.1371/journal.pone.0039324.

[96] E.D. Shanes, L.B. Mithal, S. Otero, H.A. Azad, E.S. Miller, J.A. Goldstein, Placental pathology in COVID-19, Am. J. Clin. Pathol. 154 (1) (2020) 23–32, https://doi.org/10.1093/ajcp/aqaa089.

[97] E.T. Patberg, T. Adams, P. Rekawek, et al., Coronavirus disease 2019 infection and placental histopathology in women delivering at term [published correction appears in Am J Obstet Gynecol. 2021 Jun 10;:]. Am. J. Obstet. Gynecol. 224 (4) (2021) 382.e1–382.e18, https://doi.org/10.1016/j.ajog.2020.10.020.

[98] R.N. Baergen, D.S. Heller, Placental pathology in covid-19 positive mothers: preliminary findings, Pediatr. Dev. Pathol. 23 (3) (2020) 177–180, https://doi.org/10.1177/1093526620925569.

[99] A.J. Chu, Tissue factor, blood coagulation, and beyond: an overview, Int. J. Inflamm. 2011 (2011), 367284, https://doi.org/10.4061/2011/367284.

[100] C. Tasca, R.S. Rossi, S. Corti, et al., Placental pathology in COVID-19 affected pregnant women: a prospective case-control study, Placenta 110 (2021) 9–15, https://doi.org/10.1016/j.placenta.2021.04.002.

[101] J.J. Mulvey, C.M. Magro, L.X. Ma, G.J. Nuovo, R.N. Baergen, Analysis of complement deposition and viral RNA in placentas of COVID-19 patients, Ann. Diagn. Pathol. 46 (2020), 151530, https://doi.org/10.1016/j.anndiagpath.2020.151530.

[102] A. Flores-Pliego, J. Miranda, S. Vega-Torreblanca, et al., Molecular insights into the thrombotic and microvascular injury in placental endothelium of women with mild or severe COVID-19, Cells 10 (2) (2021) 364, https://doi.org/10.3390/cells10020364. Published 2021 Feb 10.

[103] J.T. Suhren, A. Meinardus, K. Hussein, N. Schaumann, Meta-analysis on COVID-19-pregnancy-related placental pathologies shows no specific pattern [published online ahead of print, 2021 Oct 19], Placenta 117 (2021) 72–77, https://doi.org/10.1016/j.placenta.2021.10.010.

[104] H.S. Rad, J. Röhl, N. Stylianou, et al., The effects of COVID-19 on the placenta during pregnancy, Front. Immunol. 12 (2021), 743022, https://doi.org/10.3389/fimmu.2021.743022. Published 2021 Sep. 15.

[105] L. Bertero, F. Borella, G. Botta, A. Carosso, S. Cosma, M. Bovetti, et al., Placenta in SARS-CoV-2 Infection: a New Target for Inflammatory and Thrombotic Events, 2020.

[106] E. Ozer, E. Cagliyan, R.I. Yuzuguldu, M.C. Cevizci, N. Duman, Villitis of unknown etiology in the placenta of a pregnancy complicated by COVID-19. Villitis of unknown etiology in the placenta of a pregnancy complicated by COVID-19, Turk Patoloji Derg 37 (2) (2021) 167–171, https://doi.org/10.5146/tjpath.2020.01506.

[107] L. Linehan, K. O'Donoghue, S. Dineen, J. White, J.R. Higgins, B. Fitzgerald, SARS-CoV-2 placentitis: an uncommon complication of maternal COVID-19, Placenta 104 (2021) 261–266, https://doi.org/10.1016/j.placenta.2021.01.012.

[108] M.E. Brien, D. Bouron-Dal Soglio, S. Dal Soglio, et al., Pandemic stress and SARS-CoV-2 infection are associated with pathological changes at the maternal-fetal interface, Placenta 115 (2021) 37–44, https://doi.org/10.1016/j.placenta.2021.09.007.

[109] Covid- TWHO, Synthesis LE, Definition and Categorization of the Timing of Mother-To-Child Transmission of SARS-CoV-2 vols. 1–14, 2021. February.

[110] K.F. Walker, K. O'Donoghue, N. Grace, et al., Maternal transmission of SARS-COV-2 to the neonate, and possible routes for such transmission: a systematic review and critical analysis, BJOG 127 (11) (2020) 1324–1336, https://doi.org/10.1111/1471-0528.16362.

[111] E. Celik, C. Vatansever, G. Ozcan, et al., Placental deficiency during maternal SARS-CoV-2 infection [published online ahead of print, 2021 Oct 23], Placenta 117 (2021) 47–56, https://doi.org/10.1016/j.placenta.2021.10.012.

[112] A. Bouachba, F. Allias, B. Nadaud, et al., Placental lesions and SARS-Cov-2 infection: diffuse placenta damage associated to poor fetal outcome, Placenta 112 (2021) 97–104, https://doi.org/10.1016/j.placenta.2021.07.288.

[113] E.H. Adhikari, W. Moreno, A.C. Zofkie, et al., Pregnancy outcomes among women with and without severe acute respiratory syndrome coronavirus 2 infection, JAMA Netw. Open 3 (11) (2020), e2029256, https://doi.org/10.1001/jamanetworkopen.2020.29256. Published 2020 Nov 2.