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Case Report

Possible intrauterine SARS-CoV-2 infection: Positive nucleic acid testing results and consecutive positive SARS-CoV-2-specific antibody levels within 50 days after birth

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ABSTRACT

Whether severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) affects the fetus in utero is important to the well-being of the mother and neonate. We report the case of a full-term neonate born to a mother who developed symptoms of coronavirus disease 2019 (COVID-19) at 32 weeks of gestation. The placental pathology showed slight local inflammation. Serial quantitative antibody measurements in the neonate showed elevated levels of IgM on the day of birth and a gradual decline to negative levels within 28 days of life; the levels of IgG declined gradually, but IgG was still positive on day 50 of life. The sequential dynamic changes in antibody levels in the neonate were consistent with those in his mother. One-step reverse transcriptase droplet digital PCR testing for SARS-CoV-2 nucleic acid in throat and anal swabs showed positive results (750 and 892 copies/ml) on day 7 of life and negative results on day 14 of life. The neonate had no symptoms of COVID-19. This report enables us to re-evaluate the significance of IgM detection in intrauterine SARS-CoV-2 infection and presents a favorable prognosis for the neonate with long-term exposure to maternal COVID-19, despite a high possibility of intrauterine infection.

Introduction

Pregnancy with coronavirus disease 2019 (COVID-19) is a special scenario that needs a good understanding of the pathophysiology of this disease (Sahu et al., 2020), in particular whether severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) affects the fetus in utero. Neonates are susceptible to SARS-CoV-2 infection and vertical transmission is possible (Alzamora et al., 2020; Zeng et al., 2020b). Evidence has shown positive SARS-CoV-2 nucleic acid testing results for blood samples and the fetal surface of the placenta (Baud et al., 2020; Wang et al., 2020). However, previous reports have shown no evidence of SARS-CoV-2 genome in the amniotic fluid or umbilical cord blood, as well as no detection in samples from newborns immediately after birth (Chen et al., 2020). Minimal symptoms have been observed in neonates when their mothers have delivered soon after the onset of COVID-19 in the third trimester (Alzamora et al., 2020; Zhu et al., 2020).

We report the case of a full-term neonate born to a mother who developed symptoms of COVID-19 at 32 weeks of gestation. She underwent an elective cesarean section at 38 weeks of gestation due to a previous cesarean section delivery. The mother was infected long before her due date and chose to continue the pregnancy to full term, which might have caused long-term exposure to inflammatory factors and/or viremia for the fetus. The effect of maternal COVID-19 on the fetus was evaluated by placental pathology, nucleic acid testing, sequential specific SARS-CoV-2 antibody titers, and clinical features during the first 50 days after birth.

Methods

Real-Time PCR (RT-PCR) (Daan Gene, Guangzhou, China) for SARS-CoV-2 nucleic acid testing was performed on throat swabs and placenta of the mother, and on feces, throat, and anal swabs from the neonate from day 0 to day 14 of life. Details of the method used are given in a previous report (Liu et al., 2020). On days 7 and 14 of life, one-step reverse transcriptase droplet digital PCR (one-step RT-ddPCR) (QX200 droplet generator and QX200 droplet reader; BioRad, USA) was used to detect nucleic acid in throat and anal swabs from the neonate. A copy number > 50 copies/ml was considered positive. Details of the one-step RT-ddPCR method are provided in the Supplementary Material Appendix.

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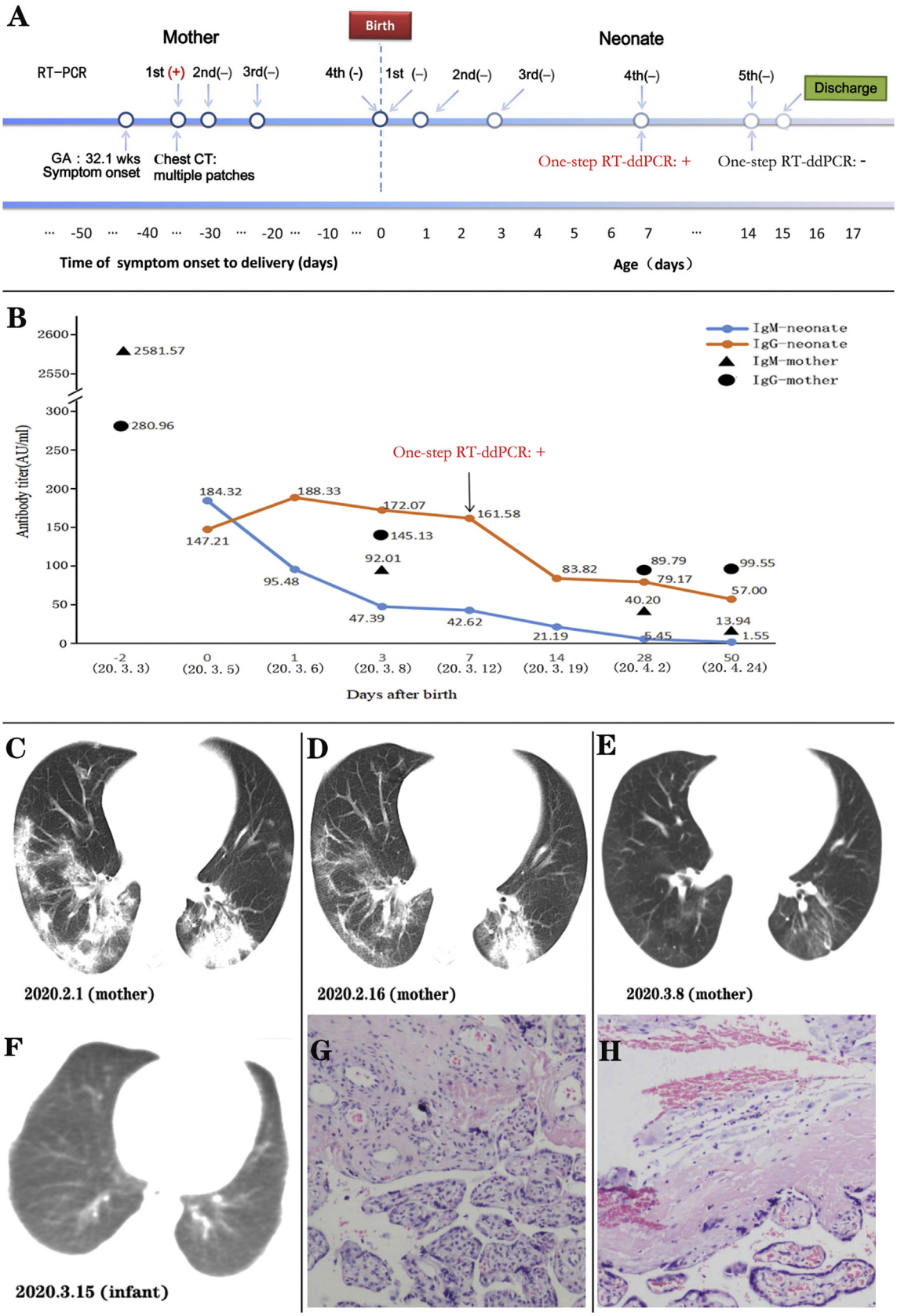

Figure 1. Details of the sequential antibody measurements, nucleic acid tests, clinical features, and placental pathology in COVID-19 dyads. (A) Timeline of symptom onset, serological tests, and nucleic acid tests. (B) Sequential dynamic changes in antibody levels within 50 days in COVID-19 dyads. (Note: IgM to SARS-CoV-2 > 10 AU/ml was positive; IgG to SARS-CoV-2 > 10 AU/ml was positive.) (C) Maternal lung CT image before treatment (7 days after the onset of COVID-19). (D) Maternal lung CT image after 15 days of treatment (22 days after the onset of COVID-19). (E) Maternal lung CT image after 37 days of treatment (44 days after the onset of COVID-19). (F) Lung CT scan of the infant (10 days after birth). (G) H&E staining images of placenta tissue (100 ×): normal placenta villi. (H) H&E staining images of placenta tissue (100×): slight fibrin deposition and lymphocyte infiltrate (COVID-19, coronavirus disease 2019; SARS-CoV-2, severe acute respiratory syndrome coronavirus 2; CT, computed tomography; H&E, hematoxylin and eosin).

Quantitative assessments of IgG and IgM were performed by chemiluminescence immunoassay (IFlash3000 Chemiluminescence Immunoassay Analyzer and chemiluminescence immunoassay kits; YHLO Biotech Co., Ltd, Shenzhen, China); this immunoassay has been shown to be a highly accurate method for the evaluation of anti-SARS-CoV-2 antibody profiles (Infantino et al., 2020). Titers > 10 AU/ml were considered positive.

The placenta was fixed and then stained with hematoxylin and eosin for pathological analysis. The timeline of symptom onset, serological testing, and nucleic acid testing is detailed in Figure 1A.

Case report

The mother in this case was on the medical staff of the hospital and had a history of close contact with COVID-19 patients. She was 30 years old (G2P1) and developed the symptoms of COVID-19 at 32 weeks of gestation. Seven days later, she was confirmed to have COVID-19 by positive nucleic acid testing result. Her condition gradually improved after therapy (Figure 1C–E). She underwent an elective cesarean section at 38 weeks of gestation due to a previous cesarean section delivery. SARS-CoV-2 IgM and IgG titers were high on the day of delivery, while the nucleic acid testing result was negative (Figure 1B).

The male neonate had a birth weight of 2700 g. His Apgar score was 8 at 1 min and 9 at 5 min. The amniotic fluid and umbilical cord were normal. There was no symptom of COVID-19, although blurred lung computed tomography images showed a few small speckles in the bilateral lungs (Figure 1F). During follow-up until day 50 of life, all health indices were normal.

The placental pathology showed slight local fibrin deposition and lymphocyte infiltrates (Figure 1G and H). Several repeat RT-PCR tests for SARS-CoV-2 nucleic acid in the placenta, amniotic fluid, and throat and anal swabs from the neonate from day 0 to day 14 of life all showed negative results. However, one-step RT-ddPCR tests performed on throat and anal swabs were positive (750 and 892 copies/ml) on day 7 of life, while RT-PCR was still negative at the same time (Figure 1A). In fact, the sensitivity of the one-step RT-ddPCR is much higher than that of RT-PCR for virus nucleic acid testing (Pinheiro-de-Oliveira et al., 2019). However, the one-step RT-ddPCR technology was not available for the detection of SARS-CoV-2 during the early stage of the pandemic and not until day 7 of the neonate's life.

Serial quantitative antibody measurements showed that the level of IgM was elevated on the day of birth and gradually declined to negative within 28 days of life; the level of IgG declined gradually, but remained at a high level from day 0 to day 50 of life. The sequential dynamic changes in IgM and IgG levels were consistent with those in his mother. Meanwhile, when the result of the one-step RT-ddPCR showed positive on day 7 of life, the antibody titers still showed a decreasing tendency, which ruled out infection after birth (Figure 1B).

Discussion

The significance of IgM detection in intrauterine SARS-CoV-2 infection

In the adult, serological testing has been used as a complementary tool in COVID-19 diagnostics. Both IgM and IgG have been shown to appear at around 13 days post disease onset. Plateau IgM levels have been shown to last for at least 4 weeks and gradually decline, and IgG antibody lasts for a longer time (Long et al., 2020). Since maternal IgM cannot cross the placenta due to its large size, the detection of unequivocal IgM in the neonate suggests an intrauterine infection. Studies have sought to diagnose intrauterine infection of SARS-CoV-2 via elevated specific IgM titers in three neonates after birth (Dong et al., 2020; Zeng et al., 2020a), however other researchers have cast doubt, indicating the potential for false-positive IgM (Kimberlin and Stagno, 2020). In the present report, the consecutive positive SARS-CoV-2-specific IgM results and consistency of the sequential dynamic changes in antibody levels between the neonate and his mother confirmed the accuracy of the antibody testing results, which ruled out the potential for false-positives. The levels of IgM and IgG in the mother and infant with COVID-19 declined gradually over time from the day of birth, which might suggest that neither of the mother and infant with COVID-19 was in the acute phase of infection post-partum.

Possibility of intrauterine infection and favorable prognosis

Although there is no evidence to support intrauterine infection with severe acute respiratory syndrome coronavirus (SARS-CoV) or Middle East respiratory syndrome coronavirus (MERS-CoV), there is evidence supporting intrauterine infection with other coronaviruses (Schwartz and Graham, 2020). Virological findings in the placenta have supported placental infection with SARS-CoV-2 but no intrauterine infection in the fetus, perhaps due to short-term exposure to maternal COVID-19 (Baud et al., 2020). In the case presented here, there was long-term exposure to maternal COVID-19 and the neonate may have become infected by SARS-CoV-2 in utero based on positive virus nucleic acid testing results, unequivocal positive IgM, and slight inflammation of the placenta. The previous research found maternal vascular malperfusion in the placenta of pregnant women with COVID-19 (Shanes et al., 2020). While, the placental histology demonstrated fibrin deposition and inflammatory infiltrates in the placenta infected with SARS-CoV-2 (Baud et al., 2020). Our result was consistent with the latter findings. Although intrauterine infection with SARS-CoV-2 was possible in the case presented herein, the neonate developed normally in utero and had no symptoms of COVID-19.

Conclusions

In conclusion, this report enables us to re-evaluate the significance of IgM detection in intrauterine SARS-CoV-2 infection and presents a favorable prognosis for the infant with a high possibility of intrauterine infection based on placental pathology, virus nucleic acid testing, and sequential antibody titers.

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Ethical approval

The study was approved by the Tongji Hospital Institutional Review Board (approval number TJ-IRB ID20200146). Written informed consent was obtained from the mother.

Conflict of interest

The authors declare no conflicts of interest.

Appendix A. Supplementary data

Supplementary material related to this article can be found, in the online version, at https://doi.org/10.1016/j.ijid.2020.07.063.

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