

Supplemental Online Content

Perl SH, Uzan-Yulzari A, Klainer H, et al. SARS-CoV-2–specific antibodies in breast milk after covid-19 vaccination of breastfeeding women. *JAMA*. doi:10.1001/jama.2021.5782

eMethods.

This supplemental material has been provided by the authors to give readers additional information about their work.

eMethods

Human breastmilk samples were centrifuged at 800g for 10 min at 4°C in order to separate fat content from the cells. Skimmed milk was used for the detection of SARS-CoV-2 antibodies, IgA and IgG, using Enzyme-Linked Immunosorbent Assay (ELISA) kits.

For IgA analysis, samples were first diluted (1:25) in the appropriate sample dilution buffers and next evaluated using a semi-quantitative analysis ELISA kit according to the manufacturers' instructions (ELISA, Kit EUROIMMUN AG, Luebeck, Germany). The microplate wells are coated with recombinant S1 structural protein. The results are evaluated by calculation of a ratio of the extinction of samples over the extinction of the internal calibrator, with a ratio > 0.8 interpreted as positive.

For IgG analysis we used the ROCHE Elecsys Anti-SARS-CoV-2 S quantitative serology assay on the Roche cobas e801 Analyzer according to the manufacturers' instructions. (La Roche Ltd, Basel, Switzerland) A result > 0.8 units/ml based on internal calibration curves is considered positive.