












RESEARCH ARTICLE

REVISED Efficiency of transplacental transfer of respiratory syncytial virus (RSV) specific antibodies among pregnant women in Kenya [version 2; peer review: 2 approved]

Joyce U. Nyiro ¹, Elizabeth Bukusi², Dufton Mwaengo³, Amek Nyaguara ¹, Bryan Nyawanda⁴, Nancy Otieno⁴, Godfrey Bigogo⁴, Nickson Murunga ¹, Marc-Alain Widdowson ^{5,6}, Jennifer R. Verani⁷, Sandra S. Chaves ^{5,7}, Hope Mwangudza ¹, Calleb Odundo¹, James A. Berkley ¹, D James Nokes ^{1,8}, Patrick K. Munywoki ⁵

¹Centre for Geographic Medicine Research-Coast, Kenya Medical Research Institute (KEMRI)-Wellcome Trust Research Programme, Kilifi, Kenya

²Centre for Microbiology Research, Kenya Medical Research Institute (KEMRI), Nairobi, Kenya

³Institute of Tropical and Infectious Diseases, University of Nairobi, Nairobi, Kenya

⁴Centre for Global Health Research, Kenya Medical Research Institute (KEMRI), Kisumu, Kenya

⁵Division of Global Health Protection, CDC-Kenya, U.S Centers for Disease Control and Prevention (CDC), Nairobi, Kenya

⁶Institute of Tropical Medicine, Antwerp, Belgium

⁷Influenza Division, U.S. Centers for Disease Control and Prevention (CDC), Atlanta, USA

⁸School of Life Sciences and Zeeman Institute (SBIDER), University of Warwick, Coventry, UK

v2 First published: 08 Feb 2022, 7:43
<https://doi.org/10.12688/wellcomeopenres.17636.1>

Latest published: 01 Apr 2022, 7:43
<https://doi.org/10.12688/wellcomeopenres.17636.2>




Abstract



Background: Maternal immunisation to boost respiratory syncytial virus (RSV) antibodies in pregnant women, is a strategy being considered to enhance infant protection from severe RSV associated disease. However, little is known about the efficiency of transplacental transfer of RSV-specific antibodies in a setting with a high burden of malaria and HIV, to guide the implementation of such a vaccination program.

Methods: Using a plaque reduction neutralization assay, we screened 400 pairs of cord and maternal serum specimens from pregnant women for RSV-specific antibodies. Participants were pregnant women of two surveillance cohorts: 200 participants from a hospital cohort in Kilifi, Coastal Kenya and 200 participants from a surveillance cohort in Siaya, Western Kenya. Transplacental transfer efficiency was determined by the cord to maternal titre ratio (CMTR). Logistic regression was used to determine independent predictors of impaired transplacental transfer of RSV-specific antibodies.

Results: A total of 800 samples were screened from the 400 participants. At enrollment the median age was 25 years (Interquartile

Open Peer Review**Approval Status**  

	1	2
version 2		
(revision)		
01 Apr 2022		view
		
version 1		
08 Feb 2022	view	view

1. **Kondwani Charles Jambo** , University of Malawi College of Medicine, Blantyre, Malawi
2. **Beate Kampmann** , London School of Hygiene and Tropical Medicine, London, UK
London School of Hygiene and Tropical Medicine, London, UK

range (IQR): 21-31). Overall, transplacental transfer was efficient and did not differ between Kilifi and Siaya cohort (1.02 vs. 1.02; $p=0.946$) but was significantly reduced among HIV-infected mothers compared to HIV-uninfected mothers (mean CMTR: 0.98 vs 1.03; $p=0.015$). Prematurity <33 weeks gestation (Odds ratio [OR]: 0.23, 95% confidence interval [CI] 0.06–0.85; $p=0.028$), low birth weight <2.5 kgs (OR: 0.25, 95% CI: 0.07–0.94; $p=0.041$) and HIV infection (OR: 0.47, 95% CI: 0.23–0.98; $p=0.045$) reduced efficiency of transplacental transfer among these women.

Conclusions: Transplacental transfer of RSV-specific antibodies among pregnant women in Kenya is efficient. A consideration to integrate other preventive interventions with maternal RSV vaccination targeting infants born premature (<33 weeks gestation), with low birth weight <2.5 kgs, or HIV-infected mothers is likely to improve vaccine outcomes in this setting.

Keywords

Pregnant women; transplacental transfer efficiency; Respiratory Syncytial Virus; Neutralising antibody, Maternal vaccine, Effectiveness

Any reports and responses or comments on the article can be found at the end of the article.



This article is included in the [KEMRI | Wellcome Trust gateway](#).

Corresponding author: Joyce U. Nyiro (jnyiro@kemri-wellcome.org)

Author roles: **Nyiro JU:** Conceptualization, Funding Acquisition, Methodology, Project Administration, Visualization, Writing – Original Draft Preparation; **Bukusi E:** Methodology, Writing – Review & Editing; **Mwaengo D:** Methodology, Writing – Review & Editing; **Nyaguara A:** Methodology, Writing – Review & Editing; **Nyawanda B:** Methodology, Writing – Review & Editing; **Otieno N:** Methodology, Writing – Review & Editing; **Bigogo G:** Methodology, Writing – Review & Editing; **Murunga N:** Visualization, Writing – Review & Editing; **Widdowson MA:** Methodology, Writing – Review & Editing; **Verani JR:** Methodology, Writing – Review & Editing; **Chaves SS:** Methodology, Writing – Review & Editing; **Mwangudza H:** Methodology, Writing – Review & Editing; **Odundo C:** Investigation, Writing – Review & Editing; **Berkley JA:** Funding Acquisition, Methodology, Writing – Review & Editing; **Nokes DJ:** Funding Acquisition, Methodology, Writing – Review & Editing; **Munywoki PK:** Methodology, Supervision, Writing – Review & Editing

Competing interests: No competing interests were disclosed.

Grant information: This work was supported by Pfizer (Grant # 52863073_CRG), CDC (Grant# 5U01GH002133-02) and Wellcome (Grant # 093804, 102975 and 077092).

The funders had no role in study design, data collection and analysis, decision to publish, or preparation of the manuscript.

Copyright: © 2022 Nyiro JU *et al.* This is an open access article distributed under the terms of the [Creative Commons Attribution License](#), which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

How to cite this article: Nyiro JU, Bukusi E, Mwaengo D *et al.* **Efficiency of transplacental transfer of respiratory syncytial virus (RSV) specific antibodies among pregnant women in Kenya [version 2; peer review: 2 approved]** Wellcome Open Research 2022, 7:43 <https://doi.org/10.12688/wellcomeopenres.17636.2>

First published: 08 Feb 2022, 7:43 <https://doi.org/10.12688/wellcomeopenres.17636.1>

REVISED Amendments from Version 1

The new manuscript version has been revised based on reviewer comments. Addition of text has been made to Introduction and Methods sections to rephrase statements which initially were not explicitly explained.

In the 'Introduction' section, a paragraph and a reference were added to provide evidence of impaired transplacental transfer of antibodies among malnourished women. Two other references were added to support definition of efficiency of transplacental transfer and analysis of efficiency of transplacental transfer of RSV antibodies using cord to maternal antibody titre ratio.

In 'Methods' section, a paragraph was added to explain how sampling was done from the two cohorts of pregnant women. The plaque reduction neutralisation test procedure was added and described in detail. A sentence on how logistic regression model was conducted was added in statistical analysis section.

In the 'Results' section, Table 2 and Figure 2 were removed from the main manuscript and deposited as supplementary files at Havard Dataverse. Table 3 was edited and changed to Table 2 while Table 4 was edited and changed to Table 3 in version two(V2) of the manuscript.

In the 'Discussion' section, paragraph 3 which described levels of RSV-specific antibodies and protection against RSV disease was deleted.

A total of seven references were added to the revised manuscript.

Any further responses from the reviewers can be found at the end of the article

Abbreviations

RSV	Respiratory syncytial virus
KHDSS	Kilifi Health and Demographic Surveillance System
KEMRI	Kenya Medical Research Institute
KWTRP	KEMRI Wellcome Trust Research Programme
KCH	Kilifi County Hospital
LMICs	Low- and Middle -Income Countries
SD	Standard deviation
95% CI	95% Confidence interval
CMTR	Cord to Maternal Transfer Ratio

Introduction

Globally, respiratory syncytial virus (RSV) is a significant cause of acute lower respiratory tract infection (LRTI) among infants leading to hospital admissions and in-hospital deaths, with 99% of these deaths occurring in developing countries¹. In sub-Saharan Africa and Asia, RSV has been observed to be responsible for about 40% of all hospital admissions with severe or very severe pneumonia among infants under 1 year². Severe RSV-associated LRTI is most common among infants under six months of age^{3,4}, resulting in about 32% of hospitalised infants in the rural coast of Kenya, during epidemics³.

Maternal immunisation is currently being considered as a strategy to protect infants from severe RSV-associated disease because of the lack of licenced RSV vaccines targeting infants^{5,6}. Additionally, the frequency of cases and the peak hospitalization occurs at 2 months of age which is less than the timing for first scheduled infant immunisations and during this neonatal period, the infant may not mount strong immune response against RSV infection due to immature immune system and/or interference from maternal antibodies⁷⁻¹¹. Efforts to advance maternal immunisation for RSV have shown promise and several candidate maternal RSV vaccines are in the late stages of clinical trials^{6,12,13}. However, despite the advancement in development of maternal RSV vaccines, the success of this program will depend on how efficiently vaccine-induced RSV-specific antibodies can be transferred in utero to the infant.

Previous studies have shown that transplacental transfer of RSV-specific antibody to the infant is usually efficient with a cord blood to maternal blood antibody titre ratio of ≥ 1 ¹⁴⁻¹⁶. This is because IgG antibody is actively transported across the syncytiotrophoblast cells rather than a passive diffusion process which is why transplacental transfer is expected to be more than one¹⁷. Transplacental transfer efficiency has been defined as either normal or impaired in other studies^{16,18}. Where the cord to maternal titre ratio (CMTR) is one or greater than one, this is considered as a normal or efficient transfer, while a CMTR of less than one is considered as an impaired transfer¹⁶.

Transplacental transfer of IgG antibodies begins during the 28th week of gestation which is coincident with the timing for expression of Fc gamma RII (FcγII) receptor responsible for the materno-foetal transfer of antibodies¹⁹. Thus, infants born preterm, shortly after or before initiation of transplacental transfer of antibodies may be less likely to benefit from a maternal RSV vaccine program. In general, placental malaria, hypergammaglobinaemia (total IgG >15g/L), HIV infection and possible illness episodes or infection occurring during the third trimester of pregnancy have been known to influence the level of antibodies transferred to the infant²⁰⁻²². Moreover, passive transfer of Haemophilus influenzae type b (Hib) vaccine antibodies was found to be 14% lower in malnourished pregnant group compared to control group among Brazilian women, suggesting less efficient transplacental transfer of antibody among malnourished women²³. However, there is limited data on the efficiency of transplacental transfer of RSV-specific antibodies in settings where the maternal population experiences comorbidities such as malaria, HIV and undernutrition as well as premature deliveries, which could negatively impact the effectiveness of a maternal RSV vaccine.

In this study, we describe the efficiency of transplacental transfer of RSV-specific antibodies measured as CMTR among pregnant women in Kenya using cord-maternal blood sample pairs collected from pregnant women in the counties of Kilifi (coast region) and Siaya (western region) where population-level prevalence of malaria and HIV, respectively, have been documented and we explore factors that could

influence transplacental transfer of RSV-specific antibodies to support the successful implementation of a maternal RSV vaccine program in Kenya.

Methods

Study sites and population

We utilized existing data and serum samples from two independent cohorts of pregnant women in Kilifi and Siaya County, Kenya. One cohort was a hospital-based surveillance investigating risk factors for severe morbidity and mortality in mothers and their infants in Kilifi, coastal Kenya, and the other, a cohort in Siaya County, Western Kenya, for surveillance of influenza disease among pregnant women and their infants.

The surveillance to investigate risk factors for severe morbidity and mortality in mothers and their infants was set up by KEMRI-Wellcome Trust Research Programme (KWTRP) at the maternity ward of Kilifi County Hospital (KCH) and the Kilifi Health and Demographic Surveillance System (KHDSS) area in Coastal Kenya in 2011²⁴. This surveillance was designed to observe 4600 births with approximately 2300 being residents of KHDSS. All mothers presenting for delivery at the maternity ward of KCH were invited to enroll. Routine clinical data were collected using a standardized questionnaire at admission to the maternity department and following delivery. During this surveillance, consent was sought from pregnant women presenting at KCH maternity ward to collect cord and maternal blood samples after delivery. These samples were securely stored at -80°C at KEMRI-Wellcome Trust laboratories in Kilifi for molecular and serological testing for viral and bacterial pathogens. The KIPMAT surveillance enrolled 4047 pregnant women in 2018 and 2019 who had a cord and maternal blood samples collected at delivery and stored for future use.

From the Western part of Kenya, in Siaya County, a cohort of pregnant women was set up through a collaboration between KEMRI-Centre for Global Health Research and the U.S. Centers for Disease Control and Prevention (CDC) Kenya in 2015. This surveillance included pregnant women recruited either from their homes or when they visited for antenatal care at Bondo sub-County or Siaya County Referral Hospital. Participants were enrolled at gestational age <20 weeks. These pregnant women were followed up weekly through a phone call or home visit to record any occurrence of influenza-like illness episodes. Blood samples were collected at enrolment and a maternal and cord blood at birth. If a pregnant woman was identified with cough or fever during follow up, a respiratory specimen was collected and screened for influenza virus type A and B and for RSV using molecular methods²⁵. All participants were requested to deliver their children in the hospital where birth outcomes were recorded; thereafter, both the baby and the mother were followed up weekly for up to six months post-delivery to assess infection from respiratory viruses by testing nasal and throat swabs from symptomatic cases by RT-PCR.

During the RSV RT-PCR procedure, nucleic acids were extracted from 100 μl of the combination of nasopharyngeal and oropharyngeal specimens using the MagMAXTM Viral RNA Isolation

Kit on the Kingfisher mL platform (Life Technologies, New York). A 5 μl of the Nucleic acid extract was then tested for RSV in a 1-step real-time reverse-transcription polymerase chain reaction (rRT-PCR) assay, using the AgPath-ID One-step RT-PCR kit (Applied Biosystems, Foster City, California) using CDC's primers and fluorescent-labelled hydrolysis probes. The assay was considered positive for RSV when exponential fluorescence curves crossed the assigned threshold at a cycle threshold value of <37.0 .

The influenza surveillance had initially proposed to recruit 2250 pregnant women in a period of 3–5 years from 2015, based on assumptions that, each participant was to be followed up for a period of six months during pregnancy and 15% of participants were expected to have at least one influenza-like illness (ILI) episode. The study changed to a surveillance in 2018 and continued to enrol participants beyond the proposed sample size. From 2018 to 2019, the influenza surveillance had recruited 1458 pregnant women from whom, 795 participants had a cord and maternal blood samples at delivery, and these were used as a sampling frame for this study.

For Kilifi cohort, 200 women were randomly selected from the cohort registers based on the availability of meta-data and paired cord and maternal blood samples for births (including preterm births) that occurred in 2018 and 2019. For Siaya cohort, where we had additional meta data, we selected all women ($n=106$) with lab-confirmed RSV infection, HIV, malaria, or anaemia, and randomly selected the other 94 women with available blood (maternal and cord) sample. A total of 400 participants were selected (200 from each region). The sample size was estimated using Edgar C. Fieller methods of calculating confidence intervals for the ratio of two means. This sample size method used CMTRs of 1.03 (0.88–1.19) observed in women in rural Nepal¹⁸. Assuming that CMTR of RSV-specific antibodies among women in Kenya were similar to those of Nepalese women and both the cord and maternal antibody levels followed a Gaussian distribution, a sample size of 200 mother-infant pair was sufficient to detect a CMTR of 1.03 with a 95% confidence interval of 1.01–1.06.

Laboratory procedures to identify RSV specific-antibodies

All blood samples were screened for RSV specific antibodies using an inhouse plaque reduction neutralization titre (PRNT) assay^{26,27} at KWTRP laboratories, Kilifi, Kenya. The PRNT procedure determines the concentration of functional antibodies from a human serum sample (or antibody preparation) required to induce 50% neutralization of a known titration of RSV virus using the Spearman Karber method²⁸. The assay has two stages, the neutralization step and a detection step. In this assay, mother-cord pairs of blood samples were assayed in one plate without use of complement sera. In the neutralization step, each serum sample was repeatedly diluted 2-fold over ten consecutive dilutions and mixed with an equal volume of 50 plaque forming units (pfu) of RSV A2 virus. The virus-serum mixture (50 μl per well) was dispensed over a confluent monolayer of Hep2 cells in a 96 well culture plate, incubated at 37°C for 48 hours.

In the detection step, fixation of cells was done by addition of 100µl/well of fixation reagent (30% methanol+70% acetone). This was followed by addition of a primary antibody (RSV F protein mouse monoclonal-BIO-RAD, Catalogue# MCA490) solution diluted 1:500 in PBS with 2 hours incubation at 37°C, and an addition of a 100µl/well of an anti-mouse HRP-conjugated secondary antibody (170-5047 Immun-Star Goat Anti-Mouse (GAM)- IgG (H/L) polyclonal antibody HRP-BIORAD). Micro RSV plaques were stained brown by 100µl/well detection reagent consisting of 16 µl of hydrogen peroxide and 0.6ml of 3-amino-9-ethylcarbazole 3.3mg/ml solution and counted with an ELISPOT reader.

Plaque counts generated by the ELISpot reader were copied and pasted onto an excel analysis template containing Spearman-Kärber formula to generate the neutralizing antibody titre for each sample. The PRNT titre was calculated as a neutralizing dose (ND50) value as follows: $\log_{10}ND50 = m - \Delta (\Sigma p - 0.5)$. Where m was the log₁₀ dilution of the highest dilution of serum (i.e. log₁₀ (1/10,240) = -4.01) Δ was the constant interval between dilutions expressed as log₁₀ (i.e. log₁₀ (2) = 0.3010). The reciprocal was considered as the final antibody titre. This assay procedure has been described elsewhere in detail²⁷.

An RSV group A human reference standard (RSV IS 16/284)²⁹ obtained from National Institute for Biological Standards and Control (NIBSC), Potters Bar, UK, and an inhouse pooled adult sera were incorporated into each assay run to check for antibody deterioration, standardization of sample titres and quality control.

Ethical considerations

Written informed consent to collect samples and data for storage and use in other studies was obtained from all participants through the parent studies, i.e., the influenza cohort surveillance (SERU #2880; CDC IRB number 6709) and the surveillance for risk factors cohort (SERU #1778). Ethical approval to screen samples for RSV-specific antibodies and use of data from the parent studies for this study was granted by the KEMRI Scientific and Ethical Review Unit Committee (SERU #3716). All methods were carried out in accordance with relevant guidelines and regulations.

Statistical analyses

Separate analysis was done for each cohort and with combined data from both cohorts. All PRNT titres were log normalised (log base2) before analysis. The efficiency of transplacental transfer of RSV-specific antibodies was calculated for each mother-infant pair of blood samples. A CMTR (i.e., PRNT titre cord/PRNT titre maternal blood) of ≥ 1 was considered normal or efficient, CMTR < 1 but ≥ 0.8 as moderately impaired and < 0.8 as severely impaired or poor. Duration of transplacental transfer was calculated as gestational age at delivery minus 28; where 28th week was estimated as the gestational age when a transplacental transfer of IgG antibodies begins during pregnancy. For this analysis, preterm birth (PTB) was defined as baby born alive before 37 weeks of pregnancy are completed and very early PTB as baby born < 33 weeks of gestation.

The difference in CMTR, cord or maternal RSV PRNT titres between HIV-infected versus HIV-uninfected mothers and RSV-infected infants vs. uninfected infants were analysed using a two-sample paired t-test. The Chi-square test was used to compare characteristics of women between Kilifi and Siaya cohort; and was also applied to determine the association between maternal/infant characteristics (HIV infection, malaria infection, RSV infection, anaemia, education level, occupation, gestational age at delivery and birth weight) and efficiency of transplacental transfer of RSV-specific antibodies. Logistic regression adjusted for each variable category (HIV infection, malaria infection, gestational age at delivery, gravida, birth-weight and RSV infection during pregnancy) was used to determine independent predictors of an impaired transplacental transfer of RSV specific antibodies. In the multivariable logistic regression model, transplacental transfer efficiency was used as a binary outcome (normal [CTMR ≥ 1] vs impaired [CMTR < 1] transfer). All data analysis was conducted using STATA version 15.0 (Stata Corp, College Station, Texas). However, a R Statistical software version 4.1.1 which is on open-access, can perform the equivalent analysis. To replicate the same analysis in R, we advise the user to import the CSV version of the data and follow the steps provided in the STATA do-file.

Results

Characteristics of study participants

A total of 800 cord and maternal blood samples from 400 participants selected from the two cohorts were screened for RSV-specific neutralizing antibodies³⁰. The median age of the women at enrollment was 25 years (Interquartile range (IQR): 21–31 years). About 95% of these women reported being married, 6% had no formal education, 57% were housewives and 21% had experienced more than six live births (Table 1).

The overall mean (standard deviation [SD]) birth weight (from both cohorts combined) of infants was 3.03 kgs (0.56), and 55 (14%) of the infants were born with low birth weight < 2.5 kilograms. The mean (SD) gestational age at delivery was 38.3 weeks (2.62). There were 11 infants out of 200 infants born from women sampled from the Siaya cohort who got RSV infection under 6 months of age. Additionally, among women from the Siaya cohort, 37 (19%) were HIV infected, 52 (26%) had malaria infection, 5 (3%) had RSV infection and 12 (6%) had severe anaemia during pregnancy. These additional data were not available for women from the Kilifi cohort.

Analysis of the difference in characteristics of participants from the two cohorts showed these women were significantly different in most characteristics (Table 1). Compared to the Siaya sample, Kilifi pregnant women had more premature births with gestational age < 33 weeks (5% vs. 2%; $P=0.022$), more babies were born with low birthweight < 2.5 kgs (21.5% vs. 6%; $P<0.001$), more women had lower than the secondary level of formal education (73.5% vs. 55%; $P<0.001$), more were younger, i.e.19 years and below (16% vs. 2%; $P<0.001$), and more women were housewives (68% vs. 46%; $P<0.001$). However, these women were similar in proportion

Table 1. Characteristics of study participants from Kilifi and Siaya.

Characteristic	Kilifi (n)	%	Siaya (n)	%	Total (n)	%	P* value
	200	50	200	50	400	100	
Maternal age							
15-19	31	15.50	4	2.00	35	8.75	
20-24	65	32.50	81	40.50	146	36.50	
25-29	43	21.50	60	30.00	103	25.75	<0.001
30-34	30	15.00	32	16.00	62	15.50	
35-39	16	8.00	19	9.50	35	8.75	
40-44	15	7.50	2	1.00	17	4.25	
45-49	0	0.00	2	1.00	2	0.50	
Marital status							
Married	191	95.50	190	95.00	381	95.25	
Single	9	4.50	10	5.00	19	4.75	0.814
Gestational age at delivery							
<33 weeks	10	5.00	3	1.50	13	3.25	
33-37 weeks	59	29.50	44	22.00	103	25.75	0.022
38-42 weeks	131	65.50	153	76.50	284	71.00	
Education level							
None	23	11.50	2	1.00	25	6.25	
Primary	124	62.00	108	54.00	232	58.00	<0.001
Secondary	37	18.50	74	37.00	111	27.75	
Tertiary-College/University	16	8.00	16	8.00	32	8.00	
Gravida							
1-2	112	56.00	41	20.50	153	38.25	
3-5	65	32.50	127	63.50	192	48.00	<0.001
6-9	18	9.00	31	15.50	49	12.25	
10-15	5	2.50	1	0.50	6	1.50	
Occupation							
Farmer	1	0.50	13	6.50	14	3.50	
Business woman	23	11.50	59	29.50	82	20.50	
House wife	136	68.00	92	46.00	228	57.00	<0.001
Salaried worker	16	8.00	18	9.00	34	8.50	
Other	24	12.00	18	9.00	42	10.50	
Number of ANC visits							
1	9	4.52	146	73.00	155	38.85	
2	22	11.06	45	22.50	67	16.79	
3	45	22.61	7	3.50	52	13.03	<0.000
4	57	28.64	2	1.00	59	14.79	
5	38	19.10	0	0.00	38	9.52	
6	28	14.07	0	0.00	28	7.02	
Sex of child							
Female	94	47.00	96	48.00	190	47.50	0.841
Birthweight							
Underweight (<2.5kgs)	43	21.50	12	6.00	55	13.75	<0.000
Transplacental transfer Efficiency							
Impaired	77	38.50	89	44.50	166	41.50	0.223

(*P-Chi squared value)

of those married (95.5% vs. 95%; $P=0.814$) and female sex of the infant (47% vs. 48%; $P=0.841$).

Distribution of Cord and Maternal RSV-specific antibodies among pregnant women in Kenya

Overall, the mean cord PRNT titres from both cohorts was $10.69 \log_2$ PRNT (SD: 1.17), with a median titre of $10.85 \log_2$ PRNT (IQR 10.00–11.59), while the mean maternal \log_2 PRNT RSV antibodies was $10.53 \log_2$ PRNT (SD:1.19), with a median of $10.62 \log_2$ PRNT (IQR 9.92–11.40). The mean titres of cord RSV-specific antibodies from the Kilifi cohort was $10.64 \log_2$ PRNT (SD:1.25), with a median titre of $10.91 \log_2$ PRNT (IQR 9.92–11.62). In the Siaya cohort, the mean cord RSV antibody titre was $10.74 \log_2$ PRNT (SD:1.08), and median $10.79 \log_2$ PRNT (IQR 10.09–11.55) respectively (Figure 1). Both mean cord (10.64 vs. 10.74; $p=0.374$) and mean maternal (10.47 vs. 10.59; $p=0.319$) \log_2 PRNT titres of RSV-specific antibodies between Kilifi and Siaya cohorts were not significantly different.

For the Siaya cohort, we found no difference in the mean \log_2 PRNT titres in cord blood for infants with RSV infection compared to infants without RSV infection in the first 6 months of life ($10.50 \log_2$ PRNT (SD: 0.99) vs. 10.74 (SD: 1.07) respectively, $p=0.186$). The mean (SD) cord PRNT titres transferred to infants of 37 (19%) HIV-infected mothers of 10.41 (SD: 1.14) \log_2 PRNT was significantly lower ($p=0.039$) than that of infants from 163 HIV-uninfected mothers 10.81 (SD: 1.05) \log_2 PRNT. The cord \log_2 PRNT RSV antibody levels of infants from mothers with and without

severe anaemia (10.87 vs. 10.73 ; $p=0.202$) and mothers with and without malaria (10.90 vs. 10.68 ; $p=0.666$) were not significantly different.

Efficiency of Transplacental Transfer of RSV-specific antibodies

The transplacental transfer of RSV-specific antibodies among these women was efficient with a mean CMTR of 1.02 (SD=0.09) and median 1.01 (IQR 0.97–1.06). The mean CMTR values were similar for pregnant women from Kilifi and Siaya (1.02 vs. 1.02; $p=0.946$; $t=0.067$).

The transplacental transfer of RSV specific antibodies for women from Kilifi and Siaya cohort was severely impaired in 1 (0.5%) and 6 (3.0%), moderately impaired in 76 (38.0%) and 83 (41.5%), and normal in 123 (61.5%) and 111 (55.5%), respectively. The overall proportion of women from the two cohorts with impaired transplacental transfer was 41.5% (166/400; 77 Kilifi vs. 89 Siaya).

Analysis of the trend in efficiency of transplacental transfer and characteristics of these women (Supplementary File 1³⁰), showed a significantly lower CMTR value among women who were HIV-infected (mean CMTR: 0.98 vs. 1.03; $p=0.015$) and women who reported their occupation as farming (mean CMTR: 0.96 vs.1.02; $p=0.012$). The CMTR value among women who got RSV infection during pregnancy (mean CMTR: 0.98 vs. 1.02; $p=0.416$) and that of mothers whom infants got RSV disease under 6 months of age (1.01 vs. 1.02; $p=0.489$) was not different.

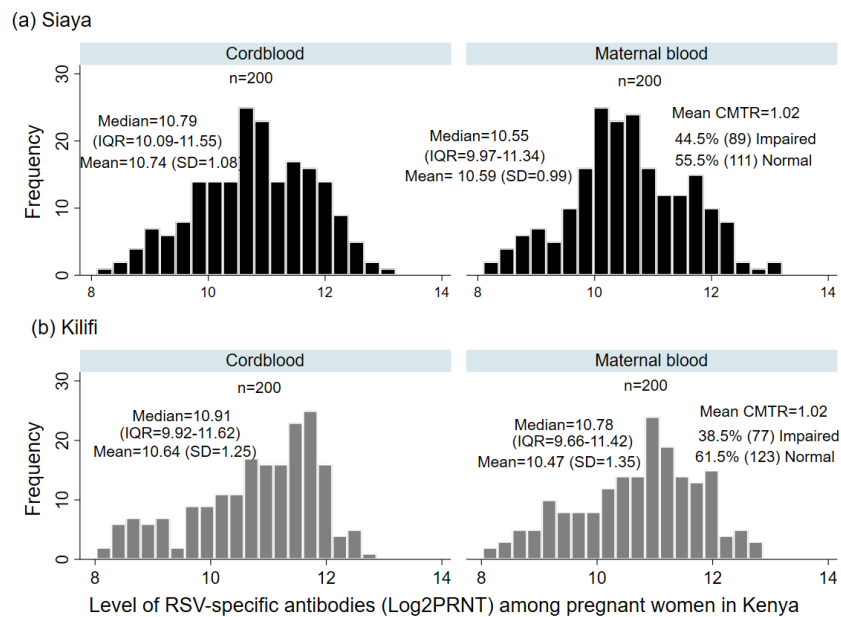


Figure 1. Frequency distribution of cord and maternal RSV specific antibodies (\log_2 transformed PRNT titres) at birth from 400 women from Kilifi and Siaya, Kenya. The mean (standard deviation), median (Inter quartile range) and the mean cord to maternal transfer ratio for each study cohort is shown.

Illness episodes during pregnancy and transplacental transfer of RSV-specific antibodies

Assessment of illness episodes from the 200 women sampled from the Siaya cohort showed 120 (60%) pregnant women had sick outpatient visits captured during weekly follow-ups and 6 of them required hospitalisation. Data on illness episodes during pregnancy among the Kilifi women was not available for this study because the surveillance was not designed to follow up participants prior to delivery.

The most common complaints about the outpatient visits among Siaya women were cough 67 (55.8%), abdominal pain 67 (55.8%), other acute respiratory illness (runny nose, shortness in breathing and chest pain while breathing 35 (29.2%)), joint pain 27 (22.5%), vomiting or diarrhoea 22 (18.3%), urinary tract infection 18 (15.0%), fever 17 (14.2%) and sore throat 12 (10.0%). One participant had premature labour. Experiencing cough episodes during pregnancy was found to be associated with impaired transplacental transfer of RSV-specific antibodies (67.3% vs. 32.7%; $\text{Chi}^2 \text{ p}=0.027$).

Multiple illness episodes during the third trimester of pregnancy occurred in 79/120 (66%) of the sick participants. There were none of the following illness episodes reported during pregnancy in this sample of women: gestational diabetes, hypertension or pre-eclampsia. No illness episode during the third trimester

of pregnancy was found to be associated with the efficiency of transplacental transfer of RSV-specific antibodies.

Gestational age at delivery and transplacental transfer of RSV-specific antibodies

The effect of gestational age in influencing transplacental transfer of RSV antibodies is demonstrated in a scatterplot of CMTR by duration in weeks of transplacental transfer (Supplementary File 2³⁰). The graph shows the level of CMTR is less than 1 or impaired within 4 weeks after onset of transplacental transfer (onset period estimated at 28th week of gestation); CMTR gradually increases above one in most participants in the next 8 weeks and starts to decline to low levels 12 weeks after onset of transplacental transfer.

Factors influencing transplacental transfer of RSV-specific antibodies among pregnant women in Kenya

Among pregnant women in Kilifi, gestational age at delivery of <33 weeks was found to be significantly associated with reduced transplacental transfer of RSV-specific antibodies ($\text{p}=0.034$). In a univariate logistic analysis, transplacental transfer was likely to be increased 5.8 times more in births occurring between 34–37 weeks compared to births in less than 33 weeks of gestation (Odds ratio (OR): 5.8, 95% confidence interval (CI) 1.33–24.95) (Table 2).

Table 2. Predictors of an Impaired transplacental transfer among pregnant women from Kilifi cohort (univariate analysis).

Characteristic	Category	Efficiency of transplacental transfer (Kilifi)				Chi2		Odds ratio		P value
		Normal		Impaired		P value	OR	Odds Ratio (95% CI)		
		n(123)	%	n(77)	%			LCL UCL		
Gestational age delivery										
	<33 weeks	3	2.4	7	9.1		Ref			
	34–37 weeks	42	34.2	17	22.1	0.034	5.8	1.33	24.95	0.019
	38–42 weeks	78	63.4	53	68.8		3.4	0.84	13.88	0.083
Birthweight										
	Underweight (<2.5kgs)	25	20.3	18	23.4					
	Normal	98	79.7	59	76.6	0.609				
Maternal age(yrs)										
	16–19	18	14.6	13	16.9					
	20–29	65	52.9	43	55.8	0.589				
	30–39	32	26.0	14	18.8					
	40–49	8	6.5	7	9.1					
Gravida										
	1–2	72	58.54	40	51.0					
	3–5	41	33.33	24	31.2	0.305				
	6–9	8	6.5	10	13.0					
	10–15	2	1.63	3	3.9					

In the Siaya cohort, transplacental transfer of RSV-specific antibodies was found to be significantly impaired among women with; gravida of more than 6 (OR: 0.56, 95% CI:0.35–0.91; $p=0.02$), occupation as farming (OR: 0.13, 95% CI:0.03–0.60; $p=0.009$), HIV infection (OR: 0.47, 95% CI:0.23–0.98; $p=0.045$) and infants with low birth weight <2.5 kilograms (kgs) (OR: 0.25, 95% CI:0.065–0.94; $p=0.041$). In a multivariate analysis including gravida, HIV infection,

occupation and birthweight to the model for Siaya women, only low birth weight <2.5 kgs was strongly associated with reduced efficiency of transplacental transfer of RSV specific antibodies (OR: 0.21, 95% CI:0.05–0.85; $p=0.029$). Malaria infection was significantly associated with increased transplacental transfer of RSV-specific antibodies (OR: 1.95, 95% CI:1.01–3.78; $p=0.048$) (Table 3). The majority of pregnancies (76%) among Siaya women were delivered at

Table 3. Predictors of an Impaired transplacental transfer among pregnant women from Siaya cohort (univariate analysis).

Characteristic	Category	Transplacental transfer efficiency (Siaya)				Chi2		Odds ratio							
		Normal		Impaired		P value	OR	Odds Ratio (95%CI)		P value					
		n(111)	%	n(89)	%			LCL	UCL						
Maternal age (yrs)	15-19	2	1.82	1	1.12	0.068									
	20-29	83	74.77	58	65.17										
	30-39	25	22.52	26	29.21										
	40-49	0	0.00	4	4.49										
Gestational age at delivery	<33 weeks	0	0.00	2	2.25	0.186									
	34-37 weeks	22	19.82	22	24.72										
	38-42 weeks	88	80.18	65	73.03										
Birthweight	Underweight (<2.5kgs)	3	2.70	9	10.11	0.028	4.05	1.06	15.40	0.041					
	Normal	108	97.30	80	89.89										
Gravida	1-2	27	24.32	14	15.73	0.049	Ref	0.67	0.33	1.42	0.301				
	3-5	72	64.86	55	61.80										
	6-9	12	10.81	20	22.47							0.31	0.12	0.82	0.018
Occupation	Farmer	2	1.80	11	12.36	0.031	Ref	6.55	1.37	31.21	0.018				
	Business woman	37	33.33	22	24.72										
	Housewife	50	45.05	42	47.19										
	Salaried worker	10	9.01	8	8.99							6.87	1.17	40.38	0.033
	Other	12	10.81	6	6.74							11.00	1.82	66.37	0.009
HIV status	Negative	96	86.49	67	75.28	0.043	Ref	0.48	0.23	0.98	0.045				
	Positive	15	13.51	22	24.72										
Sick_Cough	No	36	52.94	17	32.69	0.027	Ref	2.30	1.09	4.90	0.028				
	Yes	32	47.06	35	67.31										
Malaria	No	76	68.47	72	80.90	0.046	Ref	1.95	1.00	3.78	0.048				
	Yes	35	31.53	17	19.10										

term, i.e., ≥ 37 weeks and there was no significant association found between efficiency of transplacental transfer and gestational age at delivery ($p=0.186$).

In a multivariate logistic analysis of combined data from the Kilifi and Siaya cohorts, occupation as a farmer (OR: 0.16, 95% CI:0.03–0.73; $p=0.018$), gravida >6 (OR: 0.70, 95% CI:0.52–0.94; $p=0.023$) and gestational age at delivery <33 weeks (OR: 0.22, 95% CI:0.06–0.84; $p=0.027$) were significantly associated with reduced transplacental transfer of RSV-specific antibodies. Adjusting for the study site did not have any effect on these factors.

Discussion

In this study, we found Kenyan women from the two geographical regions of Siaya and Kilifi differing in many characteristics, but these differences did not affect the mean levels of RSV-specific antibodies transferred to infants or the overall efficiency of transplacental transfer. We also found multiple factors, including gestational age less than 33 weeks, having had multiple pregnancies and farming as an occupation to be associated with reduced transplacental transfer of RSV specific antibodies among pregnant women from the two cohorts.

The concentration of RSV antibody transferred to infants by HIV-infected mothers were significantly reduced. Similarly, the trend of cord to maternal antibody titre ratio showed a decrease with HIV infection. These findings are in line with previous studies³¹ which together raise concerns involving the effectiveness of a maternal RSV vaccine introduction to low-and middle-income countries (LMICs), which is thought might be negatively impacted by the existing comorbidities. Ongoing clinical trials (NCT04424316; NCT04605159)⁶ of maternal RSV vaccines are not taking into account HIV-diverse populations, or populations with high malaria prevalence. Therefore, investigating differences in transplacental transfer in these populations could be important in validating vaccine response in the future.

In this study, gestational age at delivery <33 weeks showed reduced transplacental transfer of RSV-specific antibodies. Gestational age has been known to influence the transplacental transfer of IgG antibodies in the Gambia²⁰ and in Sri Lanka³² where materno-foetal transfer of RSV-specific antibodies was impaired in premature babies. Furthermore, in estimating the duration of transplacental transfer by gestational age using the 28th week of pregnancy as the onset for transplacental transfer, we found babies born shortly (<4 weeks) after the beginning of this transfer had an impaired transplacental transfer. Similarly, babies born more than 3 months after onset of transplacental transfer showed decreased CMTR which was as a result of antibody decay³³. Studies have also shown that, before the 26th week of gestation, IgG transfer is blocked by a barrier of cytotrophoblasts under the syncytiotrophoblast layer³⁴ and Fc gamma RII (FcγII) receptors responsible for mediation of materno-foetal transfer of antibodies are not well expressed¹⁹. Our results, therefore, demonstrate the phenomenon of accumulation of antibody concentration among infants

with the time of transfer, waning of RSV-specific antibodies occurring with wild type RSV infection and confirm the influence of gestational age on timing for vaccination, likely to be observed during the implementation of a maternal RSV vaccine in this setting.

We also found having had more than 6 pregnancies, HIV infection, and low birth weight <2.5 kgs was associated with impaired transplacental transfer of RSV-specific antibodies. These results are similar to findings in the Gambia, where low birthweight <2.5 kgs was found to influence the transplacental transfer of RSV-specific antibodies²⁰. The role of HIV infection in impairing transplacental transfer of RSV antibodies has been found in studies conducted in Botswana³⁵ and Malawi³⁶. HIV-infected pregnant women have shown reduced immunogenicity to vaccines and this is thought to be related to immune activation leading to the production of inflammatory cytokines at the materno-foetal interface^{37,38}. Women with multiple births or pregnancies are usually older and due to repeated exposure to RSV infection, they are thought to have an accumulation of higher levels of antibodies which causes saturation of Fc transport receptors³⁹ leading to reduced transfer and thereby low levels of antibodies observed in neonates.

Women diagnosed with malaria during pregnancy had an efficient transplacental transfer in this study contrary to what has been observed in other studies in Papua New Guinea and Malawi^{36,40} where malaria was associated with a decrease in the transplacental transfer of IgG antibodies of 81%. The process of materno-foetal transfer of pathogen specific antibodies is likely to vary between different populations due to impairment caused by saturation of these receptors with infection-related hypergammaglobinemia³⁷. We would therefore like to argue that, since antimalarial prophylaxis uptake is mandatory for all pregnant women in Kenya during antenatal visits, this might have played an important role in reducing malaria related hypergammaglobinemia. Thus, 67% of the women diagnosed with malaria infection were found to have a normal transplacental transfer of RSV-specific antibodies. However, further screening of samples for evidence of placental malaria is warranted among this sample of women.

This study has some limitations. First, we did not screen for total immunoglobulin G levels and thus are not able to confirm any infection related hypergammaglobulinemia to the impaired placental transfer of RSV antibody seen in HIV-infected women, women with illness episodes during pregnancy and those diagnosed with malaria in this study. Second, by the time this analysis was conducted, results for placental malaria among women from Siaya were not yet available, we could not, therefore, ascertain the positive effect of transplacental transfer in the presence of malaria infection. In addition, we could not get data for HIV antiviral therapy, adherence, or viral load among these women, although they were all under comprehensive care programme. We also used data from cohorts not necessarily designed for this study outcome. For instance, Kilifi cohort women did not have data on follow ups during pregnancy. The sample size was small

for women with premature births, HIV infection, RSV infection, leading to wider confidence intervals and not so strong positive effect on predictors. However, we have provided important baseline data on the efficiency of transplacental transfer of RSV-specific antibodies in a setting where the maternal population experiences a high prevalence of malaria and HIV infections, and we have outlined some of the factors which would require mitigation or use of alternative prevention strategies during the introduction of a maternal RSV vaccine program for optimal vaccine outcome among infants.

Conclusions

Transplacental transfer of RSV-specific antibodies among pregnant women in Kenya is efficient. Maternal characteristics differed between women from the two different geographical regions, but this did not have a significant effect on the overall transplacental transfer of RSV antibodies. Maternal immunisation in the third trimester of pregnancy as a strategy to prevent infants from severe RSV disease should be considered with other interventions which will help protect infants born prematurely < 33 weeks gestation, infants with low birth weight and infants from HIV infected mothers.

Data availability

The data is stored under restricted access and available from the authors upon request through our Data Governance Committee (dgc@kemri-wellcome.org).

The dataset used and analysis scripts generated for this manuscript are available at Harvard Dataverse: Replication Data for: Efficiency of transplacental transfer of respiratory syncytial virus (RSV) specific antibodies among pregnant women in Kenya, <https://doi.org/10.7910/DVN/XOKFFK>³⁰.

This project contains the following underlying data:

- JNyro_RSV_Antibody_Efficiency_Data_Codebook.pdf
- JNyro_RSV_Antibody_Efficiency_Data_Readme.txt
- RSV_antibody_efficiency_analysis_script_04042021.do
- RSV_antibody_Efficiency_KIPMAT_MATFLU_dataset_04062021-1.tab

- RSV_antibody_Efficiency_KIPMAT_MATFLU_dataset_04062021.tab
- Supplementary File 1: Transplacental transfer of RSV specific antibodies provided as cord to maternal transfer ratio (CMTR) with characteristics of women from Kilifi and Siaya, Kenya
- Supplementary File 2: **A scatter plot for cord to maternal transfer ratio by duration of transplacental transfer among pregnant women from Kilifi and Siaya, Kenya.** The line for efficiency in each cohort is shown.

Data are available under the terms of the [Creative Commons Attribution 4.0 International license](https://creativecommons.org/licenses/by/4.0/) (CC-BY 4.0).

Disclaimer

The findings and conclusions in this report are those of the author(s) and do not necessarily represent the official position of the Centers for Disease Control and Prevention. Names of specific vendors, manufacturers, or products are included for public health and informational purposes; inclusion does not imply endorsement of the vendors, manufacturers, or products by the Centers for Disease Control and Prevention or the US Department of Health and Human Services.

Acknowledgements

We thank all pregnant women from Siaya and Kilifi for availing themselves and willingly provided data which answered the questions in this study. We are very grateful to the field study teams both CGHR-Kisumu and KWTRP-Kilifi who collected data from the surveillance cohorts. We appreciate the funding support from CDC (Grant# 5U01GH002133-02) for the influenza surveillance cohort and that of Wellcome Trust for surveillance of factors for mortality and morbidity in mothers and infants (Grant # 093804 and 077092). James Berkley was supported by MRC/DFiD/Wellcome Trust Joint Global Health Trials scheme grant# MR/M007367/1. This work was made possible in part by the funding support from Pfizer (Grant #52863073-CRG) and James Nokes' Wellcome Trust senior investigator award (Grant # 102975). This paper is published with the permission of the Director of KEMRI.

References

1. Nair H, Nokes DJ, Gessner BD, *et al.*: **Global burden of acute lower respiratory infections due to respiratory syncytial virus in young children: a systematic review and meta-analysis.** *Lancet.* 2010; **375**(9725): 1545–1555. [PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
2. Pneumonia Etiology Research for Child Health (PERCH) Study Group: **Causes of severe pneumonia requiring hospital admission in children without HIV infection from Africa and Asia: the PERCH multi-country case-control study.** *Lancet.* 2019; **394**(10200): 757–779. [PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
3. Nokes DJ, Ngama M, Bett A, *et al.*: **Incidence and severity of respiratory syncytial virus pneumonia in rural Kenyan children identified through hospital surveillance.** *Clin Infect Dis.* 2009; **49**(9): 1341–1349. [PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
4. Shi T, McAllister DA, O'Brien KL, *et al.*: **Global, regional, and national disease burden estimates of acute lower respiratory infections due to respiratory syncytial virus in young children in 2015: a systematic review and modelling study.** *Lancet.* 2017; **390**(10098): 946–958. [PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)

5. Englund J, Glezen WP, Piedra PA: **Maternal immunization against viral disease.** *Vaccine.* 1998; **16**(14-15): 1456-1463.
[PubMed Abstract](#) | [Publisher Full Text](#)
6. Madhi SA, Polack FP, Piedra PA, et al.: **Respiratory Syncytial Virus Vaccination during Pregnancy and Effects in Infants.** *N Engl J Med.* 2020; **383**(5): 426-439.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
7. Glezen WP, Paredes A, Allison JE, et al.: **Risk of respiratory syncytial virus infection for infants from low-income families in relationship to age, sex, ethnic group, and maternal antibody level.** *J Pediatr.* 1981; **98**(5): 708-715.
[PubMed Abstract](#) | [Publisher Full Text](#)
8. Roca A, Abacassamo F, Loscertales MP, et al.: **Prevalence of respiratory syncytial virus IgG antibodies in infants living in a rural area of Mozambique.** *J Med Virol.* 2002; **67**(4): 616-623.
[PubMed Abstract](#) | [Publisher Full Text](#)
9. Sande CJ, Cane PA, Nokes DJ: **The association between age and the development of respiratory syncytial virus neutralising antibody responses following natural infection in infants.** *Vaccine.* 2014; **32**(37): 4726-4729.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
10. Esposito S, Scarselli E, Lelii M, et al.: **Antibody response to respiratory syncytial virus infection in children < 18 months old.** *Hum Vaccin Immunother.* 2016; **12**(7): 1700-1706.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
11. Jounai N, Yoshioka M, Tozuka M, et al.: **Age-Specific Profiles of Antibody Responses against Respiratory Syncytial Virus Infection.** *EBioMedicine.* 2017; **16**: 124-135.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
12. Higgins D, Trujillo C, Keech C: **Advances in RSV vaccine research and development - A global agenda.** *Vaccine.* 2016; **34**(26): 2870-2875.
[PubMed Abstract](#) | [Publisher Full Text](#)
13. PATH: **RSV vaccine and mAb snapshot.** 2020; (updated 26 March 2020).
[Reference Source](#)
14. Suara RO, Piedra PA, Glezen WP, et al.: **Prevalence of neutralizing antibody to respiratory syncytial virus in sera from mothers and newborns residing in the Gambia and in The United States.** *Clin Diagn Lab Immunol.* 1996; **3**(4): 477-479.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
15. Cox MJ, Azevedo RS, Cane PA, et al.: **Seroepidemiological study of respiratory syncytial virus in Sao Paulo state, Brazil.** *J Med Virol.* 1998; **55**(3): 234-239.
[PubMed Abstract](#) | [Publisher Full Text](#)
16. Homaira N, Binks M, Walker G, et al.: **Transplacental transfer of RSV antibody in Australian First Nations infants.** *J Med Virol.* 2022; **94**(2): 782-786.
[PubMed Abstract](#) | [Publisher Full Text](#)
17. Palmeira P, Quinello C, Silveira-Lessa AL, et al.: **IgG placental transfer in healthy and pathological pregnancies.** *Clin Dev Immunol.* 2012; **2012**: 985646.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
18. Chu HY, Tielsch J, Katz J, et al.: **Transplacental transfer of maternal respiratory syncytial virus (RSV) antibody and protection against RSV disease in infants in rural Nepal.** *J Clin Virol.* 2017; **95**: 90-95.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
19. Kameda T, Koyama M, Matsuzaki N, et al.: **Localization of three subtypes of Fc gamma receptors in human placenta by immunohistochemical analysis.** *Placenta.* 1991; **12**(1): 15-26.
[PubMed Abstract](#) | [Publisher Full Text](#)
20. Okoko JB, Wesumperuma HL, Hart CA: **The influence of prematurity and low birthweight on transplacental antibody transfer in a rural West African population.** *Trop Med Int Health.* 2001; **6**(7): 529-534.
[PubMed Abstract](#) | [Publisher Full Text](#)
21. Okoko BJ, Wesumperuma LH, Ota MO, et al.: **The influence of placental malaria infection and maternal hypergammaglobulinemia on transplacental transfer of antibodies and IgG subclasses in a rural West African population.** *J Infect Dis.* 2001; **184**(5): 627-632.
[PubMed Abstract](#) | [Publisher Full Text](#)
22. Hartter HK, Oyedele OI, Dietz K, et al.: **Placental transfer and decay of maternally acquired antimeasles antibodies in Nigerian children.** *Pediatr Infect Dis J.* 2000; **19**(7): 635-641.
[PubMed Abstract](#) | [Publisher Full Text](#)
23. Cavalcante RS, Kopelman BI, Costa-Carvalho BT: **Placental transfer of Haemophilus influenzae type b antibodies in malnourished pregnant women.** *Braz J Infect Dis.* 2008; **12**(1): 47-51.
[PubMed Abstract](#) | [Publisher Full Text](#)
24. Seale AC, Barsosio HC, Koeh AC, et al.: **Embedding surveillance into clinical care to detect serious adverse events in pregnancy.** *Vaccine.* 2015; **33**(47): 6466-6468.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
25. Nyawanda BO, Otieno NA, Otieno MO, et al.: **The impact of maternal HIV infection on the burden of respiratory syncytial virus among pregnant women and their infants, western Kenya.** *J Infect Dis.* 2020; **jiaa490**.
[PubMed Abstract](#) | [Publisher Full Text](#)
26. Sande CJ, Mutunga MN, Okiro EA, et al.: **Kinetics of the neutralizing antibody response to respiratory syncytial virus infections in a birth cohort.** *J Med Virol.* 2013; **85**(11): 2020-2025.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
27. Nyiro JU, Kiyuka PK, Mutunga MN, et al.: **Agreement between ELISA and plaque reduction neutralisation assay in Detection of respiratory syncytial virus specific antibodies in a birth Cohort from Kilifi, coastal Kenya [version 1; peer review: 2 approved, 2 approved with reservations].** *Wellcome Open Res.* 2019; **4**: 33.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
28. Sande CJ, Mutunga MN, Medley GF, et al.: **Group- and genotype-specific neutralizing antibody responses against respiratory syncytial virus in infants and young children with severe pneumonia.** *J Infect Dis.* 2013; **207**(3): 489-492.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
29. McDonald JU, Rigsby P, Dougall T, et al.: **Establishment of the first WHO International Standard for antiserum to Respiratory Syncytial Virus: Report of an international collaborative study.** *Vaccine.* 2018; **36**(50): 7641-7649.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
30. Nyiro JU, Bukusi E, Mwaengo D, et al.: **Replication Data for: Efficiency of transplacental transfer of respiratory syncytial virus (RSV) specific antibodies among pregnant women in Kenya.** Harvard Dataverse, V1, UNF6: aOIBDpFDzoTNDQaRw/DH0w== [fileUNF]. 2021.
<http://www.doi.org/10.7910/DVN/XOKFFK>
31. Alonso S, Vidal M, Ruiz-Olalla G, et al.: **Reduced Placental Transfer of Antibodies Against a Wide Range of Microbial and Vaccine Antigens in HIV-Infected Women in Mozambique.** *Front Immunol.* 2021; **12**: 614246.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
32. Wesumperuma HL, Perera AJ, Pharoah PO, et al.: **The influence of prematurity and low birthweight on transplacental antibody transfer in Sri Lanka.** *Ann Trop Med Parasitol.* 1999; **93**(2): 169-177.
[PubMed Abstract](#) | [Publisher Full Text](#)
33. Swamy GK, Garcia-Putnam R: **Maternal immunization to benefit the mother, fetus, and infant.** *Obstet Gynecol Clin North Am.* 2014; **41**(4): 521-534.
[PubMed Abstract](#) | [Publisher Full Text](#)
34. Kristoffersen EK: **Placental Fc receptors and the transfer of maternal IgG.** *Transfus Med Rev.* 2000; **14**(3): 234-243.
[PubMed Abstract](#) | [Publisher Full Text](#)
35. Patel SM, Jallow S, Boiditswe S, et al.: **Placental Transfer of Respiratory Syncytial Virus Antibody Among HIV-Exposed, Uninfected Infants.** *J Pediatric Infect Dis Soc.* 2020; **9**(3): 349-356.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
36. de Moraes-Pinto MI, Verhoeff F, Chimsuku L, et al.: **Placental antibody transfer: influence of maternal HIV infection and placental malaria.** *Arch Dis Child Fetal Neonatal Ed.* 1998; **79**(3): F202-205.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
37. Wilcox CR, Holder B, Jones CE: **Factors Affecting the FcRn-Mediated Transplacental Transfer of Antibodies and Implications for Vaccination in Pregnancy.** *Front Immunol.* 2017; **8**: 1294.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
38. Abu-Raya B, Smolen KK, Willems F, et al.: **Transfer of Maternal Antimicrobial Immunity to HIV-Exposed Uninfected Newborns.** *Front Immunol.* 2016; **7**: 338.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
39. Clements T, Rice TF, Vamvakas G, et al.: **Update on Transplacental Transfer of IgG Subclasses: Impact of Maternal and Fetal Factors.** *Front Immunol.* 2020; **11**: 1920.
[PubMed Abstract](#) | [Publisher Full Text](#) | [Free Full Text](#)
40. Atwell JE, Thumar B, Formica MA, et al.: **Hypergammaglobulinemia and Impaired Transplacental Transfer of Respiratory Syncytial Virus Antibody in Papua New Guinea.** *Pediatr Infect Dis J.* 2019; **38**(9): e199-e202.
[PubMed Abstract](#) | [Publisher Full Text](#)

Open Peer Review

Current Peer Review Status:  

Version 2

Reviewer Report 01 April 2022

<https://doi.org/10.21956/wellcomeopenres.19709.r49637>

© 2022 Kampmann B. This is an open access peer review report distributed under the terms of the [Creative Commons Attribution License](#), which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.



Beate Kampmann 

¹ The Vaccine Centre, Faculty of Infectious and Tropical Diseases, London School of Hygiene and Tropical Medicine, London, UK

² MRC Unit The Gambia, London School of Hygiene and Tropical Medicine, London, UK

I am satisfied with the changes made in this manuscript and it can be approved from my end.

Competing Interests: No competing interests were disclosed.

Reviewer Expertise: Maternal immunisation, Vaccinology, Infant immunity

I confirm that I have read this submission and believe that I have an appropriate level of expertise to confirm that it is of an acceptable scientific standard.

Version 1

Reviewer Report 01 March 2022

<https://doi.org/10.21956/wellcomeopenres.19508.r48687>

© 2022 Kampmann B. This is an open access peer review report distributed under the terms of the [Creative Commons Attribution License](#), which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.



Beate Kampmann 

¹ The Vaccine Centre, Faculty of Infectious and Tropical Diseases, London School of Hygiene and Tropical Medicine, London, UK

² MRC Unit The Gambia, London School of Hygiene and Tropical Medicine, London, UK

The paper by Joyce Nyiro *et al.* summarises the results of a study measuring the transfer of anti-RSV antibody via the placenta in two Kenyan communities.

The samples used stem from two unrelated cohort studies and measure neutralising RSV antibody in paired cord and maternal banked serum samples from a total of 400 mother/infant pairs, using an in-house plaque reduction neutralisation assay conducted at the KEMRI Wellcome Trust Research Programme laboratories in Kenya.

The authors found that overall the antibody transfer was “efficient” with no significant differences between the 2 cohorts. However, HIV status and gestational age of the infant were strong predictors for reduced transplacental transfer ratios.

The subject is important given the efforts to develop and implement an RSV vaccine targeted at pregnant women to prevent RSV infection in young infants. RSV is a known cause of significant morbidity and mortality in early life and the LEMRI team has a longstanding interest in this research area. To study transplacental antibody is therefore timely, although vaccine-induced antibody might give different results-and potentially much higher levels- than the antibody measured here which is due to natural infection.

Conceptually, I have some questions regarding the wider interpretation of the results, as the title talks about efficiency which somewhat raises the expectation that protection from RSV disease is also addressed in this manuscript, which it is not. The results are also not interpreted in the context of potential protective efficacy or indeed should be discussed in correlation to what might be expected from the vaccine-induced antibody.

I have made some more specific comments going through the various sections of the paper:

Introduction:

- The statement that neonates cannot mount protective immune responses is incorrect.
- Undernutrition is mentioned as a contributor to poor vaccine responses but not referenced- where is this evidence? Also this isn't addressed in the manuscript.
- The rationale for using the 2 different cohorts is not provided- maybe opportunistic?
- The paper talks a lot about efficiency but a definition is not provided -more than 1.0 transfer ratio I guess is what has been defined-needs clarification.

Methods:

- Whilst there is more than enough description of the 2 cohorts- in fact a lot of this info is not relevant to the study aims reported here and could be moved to supplementary materials- there is a very rudimentary description of the laboratory methods.
- The sample selection is also not very clearly described as -although apparently randomly selected samples-, there is probably targeted selection for HIV+ve mothers and premature infants to address two key factors that are likely to influence the transplacental transfer ratios? Please make sure the sample selection process is very clear and state if you enriched for these subgroups as they might not have been randomly distributed across the studies.

- There is description of the RSV diagnostics which was not outlined as a research question - which might be that people wanted to ask how/if recent natural infection impacts on measurable antibody- if this was an aim it should be clearly stated. The comparison between antibody acquired from past infection and recent infection should then also be shown.
- A similar issue arises with mentioning of the infants- was an aim of the study to assess if infants who were infected with RSV had lower titres than those not? The sample size is really small to address this issue and it should be clearly stated if this was an exploratory angle or a main question.
- Both of these questions above are of interest but there is no power calculation to really address them in the current study design and it all, therefore, reads like thrown in for good measure-(or part of a doctoral thesis chapter??)

Results:

- The data provided are generally reassuring but their display varies between talking about PRNT titres and then suddenly switch to odds ratios- why not stick with transfer ratios which are more accessible, especially since there is next to no information on the measurements in this neutralisation assay.
- A lot of parameters are being analysed and a multi-variant analysis would be the best way to combine the read outs- HIV and preterm birth stand out-and these results align with results from other studies measuring transplacental transfer ratios for other either naturally occurring or vaccine induced transplacental antibody.
- Preterm birth and low birth weight are obviously related variables.
- Definitions for severely impaired and impaired were not introduced in the Methods section and don't appear to be referenced- where does the definition come from and what is the clinical relevance of 0.96 versus 1.02? This needs to be discussed or at least acknowledged how we would know if/why it might make a difference- the discussion lacks depth in this area.
- Figure 2 could be better explained and the tables have far too many demographic details that do not matter- a lot of this could be moved to the supplementaries if at all of interest- the key variables that might impact on transfer are the only ones that need to be mentioned in my view- again, too much distracting detail that might come from a thesis?

The **discussion** finally touches on the differences between naturally occurring and vaccine-induced antibody but it is very short in this context- no examples of vaccine-induced antibody levels, although some of this data is publically accessible, no comparison with Palivizumab-equivalent antibody titres, no discussion about antibody not being the only correlate of protection against RSV.

What the paper does well is to show that transplacental antibody is measurable, the transfer ratio is above 1, hence there must be active transport via nFcG receptor (not discussed!), and that the

levels can be impacted upon by HIV infection and gestational age of the infant-and this is important information given the vaccines are under advanced development, but a lot of the added detail is not positioned to address additional aspects which have however also not been clearly formulated- natural versus vaccine antibody, recent infection vs past infection in the women, any evidence of protection in the infants, and evidence/assumptions for lack of protection in infants-in context of assumed correlates of protection.

Is the work clearly and accurately presented and does it cite the current literature?

Partly

Is the study design appropriate and is the work technically sound?

Yes

Are sufficient details of methods and analysis provided to allow replication by others?

No

If applicable, is the statistical analysis and its interpretation appropriate?

Partly

Are all the source data underlying the results available to ensure full reproducibility?

Yes

Are the conclusions drawn adequately supported by the results?

Partly

Competing Interests: No competing interests were disclosed.

Reviewer Expertise: Maternal immunisation, Vaccinology, infant immunity

I confirm that I have read this submission and believe that I have an appropriate level of expertise to state that I do not consider it to be of an acceptable scientific standard, for reasons outlined above.

Author Response 25 Mar 2022

Joyce Nyiro, Kenya Medical Research Institute (KEMRI)-Wellcome Trust Research Programme, Kilifi, Kenya

Thank you for the useful comments. We have addressed each of the comments below comprehensively and revised the manuscript accordingly.

All our responses have been provided in italic font.

Regarding the comment about the title of the manuscript, it is important to note that efficiency in transplacental transfer of antibodies is not the same as efficacy or effectiveness (those terms indicating protection). Our manuscript did not intend to assess antibody protection but to address the efficiency of transplacental transfer.

Introduction:

Comment:

- The statement that neonates cannot mount protective immune responses is incorrect.

Response:

Thank you for picking this out. The wording has been changed to bring out the intended message. We have deleted the word “cannot” from the sentence and replaced it with the word “may not”, as a less definitive statement. However, it is well documented that very young infants may not generate a robust immune response after exposure compared to older children and adults. This has been shown to be the case with other vaccines like measles:

<https://www.ncbi.nlm.nih.gov/pmc/articles/PMC4135535/> and

<https://www.frontiersin.org/articles/10.3389/fimmu.2020.595297/full>.

In the case of RSV, infants during the neonatal period may not mount a strong immune response following infection or vaccination. It is likely the response is obscured by the presence of maternal antibodies which are decaying or might reflect an immature immune system. This phenomenon has been observed in a number of studies including a study conducted in Kilifi, Kenya by Sande et al., 2014, a study among Italian infants by Esposito et al., 2016 and in Japan in a study by Jounai et al., 2017.

Please see these changes in the introduction section, paragraph 2 of the revised manuscript.

Comment:

- Undernutrition is mentioned as a contributor to poor vaccine responses but not referenced-where is this evidence? Also this isn't addressed in the manuscript.

Response:

Thank you for the insight and we agree that this statement might not have come out clearly within the manuscript. Our mention of undernutrition in the introduction section of this manuscript was to highlight it as one of the factors which have been found to impair the transplacental transfer of antibodies. Malnutrition has been found to reduce the passive transfer of IgG antibodies from the mother to infant which reduces the efficacy of vaccines in pregnancy intended to prevent infant infection. The mechanism of malnutrition in reducing the transplacental transfer of antibodies has been related to differences in placental size, morphology, and vascular development.

Evidence of reduced transplacental transfer in malnourished pregnant women has been shown with Haemophilus influenzae type b (Hib) vaccine in Brazil where the vertical transmission rate was 14% lower in the malnourished pregnant group (Cavalcante, et al., 2008). This reference has been added in the introduction section and paragraph 4 of the manuscript revised accordingly.

Comment:

- The rationale for using the 2 different cohorts is not provided- maybe opportunistic?

Response:

Thank you for this comment. To an extent, the selection was opportunistic as they were the only

cohorts available to provide the required samples and data. In this study, we used already collected serum samples from the two existing cohorts, hence the selection of the study sites preceded the current analyses. The KIPMAT cohort was specifically set up to do this kind of analysis in relation to maternal vaccines including Group B Streptococcus (GBS) and RSV, while the Influenza surveillance cohort was set up for analysis of maternal influenza vaccine trials.

The choice of two cohorts instead of one cohort was purposive, in that it enabled a comparison between pregnant women from two diverse geographical regions in Kenya which fit with the main objective of the study. Additionally, one cohort was from a region known for high rates of HIV infection and the other from a region endemic for malaria which were some of the factors of interest in the transplacental transfer of antibodies. The methods section has been updated to explicitly state the rationale.

Comment:

- The paper talks a lot about efficiency but a definition is not provided -more than 1.0 transfer ratio I guess is what has been defined-needs clarification.

Response:

We appreciate the concern raised. The definition for efficiency was provided in the statistical analysis section of this manuscript and was defined as cord to maternal RSV antibody titre ratio (CMTR) at birth. Where CMTR was one or greater than one, this was considered as normal or an efficient transfer; if CMTR was less than one, the transplacental transfer was considered as impaired. IgG antibody is actively transported across the syncytiotrophoblast cells rather than a passive diffusion process which is why we expect it to be more than 1 (Palmeira et al., 2012).

Since this comment has been raised multiple times, we have considered the inclusion of this definition in the introduction section of the revised manuscript for more clarity.

Methods:

Comment:

- Whilst there is more than enough description of the 2 cohorts- in fact a lot of this info is not relevant to the study aims reported here and could be moved to supplementary materials- there is a very rudimentary description of the laboratory methods.

Response:

Thank you for this suggestion. We added the description of the two cohorts to provide the context of the current study and show collected metadata. We thought it was necessary to retain this information in the main manuscript following a request made by other reviewers during the process for seeking clearance of the manuscript for publication by CDC.

Concerning the laboratory methods, we have now provided a detailed description of the plaque reduction neutralization assay, in addition to referencing previously published procedures (Nyiro et al., 2019). This addition has been made at the laboratory procedures section in methods of the revised manuscript in paragraphs 1 to 3.

Comment:

- The sample selection is also not very clearly described as -although apparently randomly selected samples-, there is probably targeted selection for HIV+ve mothers and premature infants to address two key factors that are likely to influence the

transplacental transfer ratios? Please make sure the sample selection process is very clear and state if you enriched for these subgroups as they might not have been randomly distributed across the studies.

Response:

Thank you for this comment and we have now clearly described the sample selection. For the Siaya cohort, we ensured adequate representation of women with HIV, malaria, known RSV infection or no recorded risk factor. In each stratum, all or a random selection of the women was conducted. This is because pregnant women with HIV, malaria and known RSV infection from the database were very few. For the Kilifi cohort, we used the random selection approach.

The description of the sample selection process has been updated in the Methods section of the revised manuscript.

Comment:

- There is description of the RSV diagnostics which was not outlined as a research question -which might be that people wanted to ask how/if recent natural infection impacts on measurable antibody- if this was an aim it should be clearly stated. The comparison between antibody acquired from past infection and recent infection should then also be shown.

Response:

You are right, the question on how/if recent natural infection impacts on measurable antibodies was not part of the aim of this study. Our initial submission, before the editor's comments, didn't include the detailed description of the PCR methodology used in testing for RSV antigen from pregnant women who presented at the hospital for care with acute respiratory infection. However, we felt it important to include this description after a request by the journal. We have therefore retained this description in the main manuscript because removing it might conflict with the journal requirements.

Comment:

- A similar issue arises with mentioning of the infants- was an aim of the study to assess if infants who were infected with RSV had lower titres than those not? The sample size is really small to address this issue and it should be clearly stated if this was an exploratory angle or a main question.

Response:

Thank you for this comment. This section has now been deleted from the manuscript due to a similar comment by the first reviewer. Again, this was not part of the objectives of this study but an exploratory analysis.

Comment:

- Both of these questions above are of interest but there is no power calculation to really address then in the current study design and it all, therefore, reads like thrown in for good measure-(or part of a doctoral thesis chapter??)

Response:

Thank you for this comment. We agree we didn't include power calculation in this manuscript because we didn't find its utility. However, we had conducted power calculation in the design of the study to assess the degree of reduction of transplacental transfer by any of the covariates. The power calculation applied reduced transplacental transfer by malaria and

hypergammaglobulinaemia for RSV specific antibodies of 58% and 90% respectively which was observed in the Gambia (Okoko et al., 2001). We had estimated a sample of 160 pregnant women with past RSV infection in our study to have 82.8% power to detect a 12.5% reduction in transplacental transfer of RSV specific antibodies by any of the covariates/factors such as anaemia, malaria, HIV status, maternal age and parity. However, due to the small sample size for women with the different characteristics of HIV infection, anaemia, and other illnesses, the study only focused on the sample size estimation for the primary objective which was to determine the cord to maternal titre ratio as described in this manuscript, and using this data explored the effect of covariates. The lack of adequate sample size for each group was addressed in the limitation section of this manuscript.

Results:**Comment:**

The data provided are generally reassuring but their display varies between talking about PRNT titres and then suddenly switch to odds ratios- why not stick with transfer ratios which are more accessible, especially since there is next to no information on the measurements in this neutralisation assay.

Response:

Thank you for this comment. To clarify further, the results in this manuscript also present the distribution of PRNT titres from maternal and cord blood samples (Figure 1). These PRNT titres have been used to calculate the cord to maternal RSV antibody titre ratios (CMTR) which were used to define the efficiency of the RSV antibody transplacental transfer. The transplacental transfer efficiency was then used as a binary outcome (normal vs impaired transfer) in a multivariable logistic regression model to determine the effect of covariates. These details have been described in the methods, statistical analysis section and also shown in the results section of this manuscript.

Comment:

- A lot of parameters are being analysed and a multi-variant analysis would be the best way to combine the read outs- HIV and preterm birth stand out-and these results align with results from other studies measuring transplacental transfer ratios for other either naturally occurring or vaccine induced transplacental antibody.

Response:

Thank you and we agree with this comment. This is what we did in our analysis. These details are shown in the statistical analysis section and the data analysis script provided with the replication dataset of this manuscript.

Comment:

- Preterm birth and low birth weight are obviously related variables.

Response:

We agree with this statement. However, whereas preterm infants born before 37 weeks of gestation normally have low birth weight, an infant can also have low birth weight even if they are born at full term (>37weeks).

Comment:

- Definitions for severely impaired and impaired were not introduced in the Methods section and don't appear to be referenced- where does the definition come from and what is the clinical relevance of 0.96 versus 1.02? This needs to be discussed or at least acknowledged how we would know if/why it might make a difference- the discussion lacks depth in this area.

Response:

Thank you for this comment. All definitions of transplacental efficiency were provided in the statistical analysis (methods) section of this manuscript. The efficient transplacental transfer was defined as $CMTR > 1$ and an impaired transplacental transfer as $CMTR < 1$. A $CMTR$ of 0.96 is less than one meaning transplacental transfer is impaired while a $CMTR$ of 1.02 is greater than one meaning transplacental transfer is normal/ efficient. This definition and analysis have already been used by the following studies: Australia (Nusrat, H. et al., 2022), in Papua New Guinea (Atwell, J.E. et al., 2016) and in Nepal (Chu, H. et al., 2017)

Comment:

- Figure 2 could be better explained and the tables have far too many demographic details that do not matter- a lot of this could be moved to the supplementaries if at all of interest- the key variables that might impact on transfer are the only ones that need to be mentioned in my view- again, too much distracting detail that might come from a thesis?

Response:

This has been well noted and Figure 2 has been removed from the main manuscript and is now available together with the replication dataset of this manuscript as Supplementary File 2. The tables have been revised as suggested and Table 2 has also been moved to the replication dataset as Supplementary File 1.

Comment:

The **discussion** finally touches on the differences between naturally occurring and vaccine-induced antibody but it is very short in this context- no examples of vaccine-induced antibody levels, although some of this data is publically accessible, no comparison with Palivizumab-equivalent antibody titres, no discussion about antibody not being the only correlate of protection against RSV.

What the paper does well is to show that transplacental antibody is measurable, the transfer ratio is above 1, hence there must be active transport via nFcG receptor (not discussed!), and that the levels can be impacted upon by HIV infection and gestational age of the infant- and this is important information given the vaccines are under advanced development, but a lot of the added detail is not positioned to address additional aspects which have however also not been clearly formulated- natural versus vaccine antibody, recent infection vs past infection in the women, any evidence of protection in the infants, and evidence/assumptions for lack of protection in infants- in context of assumed correlates of protection.

Response:

Thank you for the useful comments and input into this manuscript. We have considered your

concerns and have done the following changes in the manuscript based on your suggestions and where we thought the data available was not sufficient to address some of those aspects.

1. *The aspect of natural versus vaccine antibody,*

We agree this would have been worth discussing in this manuscript since the nature and quality of natural antibody is different from that of vaccine antibody. However, this was not part of the study objectives and was not supported by the available data. Our manuscript only focused on the transplacental transfer of IgG antibodies and the factors likely to impact this transfer. The section in the discussion which had highlighted this aspect has been deleted.

2. *Recent infection versus past infection*

We acknowledge that this aspect didn't come out very clearly in the earlier version of the manuscript. This was not part of the study objectives. However, methodologies for testing of recent infections using PCR were only provided in the manuscript to describe the procedure used to identify women with RSV related acute respiratory infection who presented for care at the hospital. In this study, we did not measure antibodies due to recent infection and that's why it is not discussed in this manuscript. Since inclusion of the PCR laboratory procedure was a request by the journal editor, we have retained this section in the main manuscript.

3. *Evidence of protection in infants and evidence/assumptions for lack of protection in infants*

We agree we didn't explicitly explain this aspect in the manuscript. This is because, it was not an objective of the study, and the data could not support this analysis. Paragraph 3 in the discussion which had mentioned this aspect has been deleted from this manuscript.

Since these aspects are of high interest, we would like to mention that, there are other studies conducted in Kilifi, Kenya which previously sought to address some of these questions. These include Sande et al., 2014 (Recent infection versus past infection) and Nyiro et al., 2016 (evidence of protection by maternal antibody from natural RSV infection).

Competing Interests: No competing interests were disclosed.

Reviewer Report 15 February 2022

<https://doi.org/10.21956/wellcomeopenres.19508.r48576>

© 2022 Jambo K. This is an open access peer review report distributed under the terms of the [Creative Commons Attribution License](#), which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.



Kondwani Charles Jambo 

Malawi-Liverpool-Wellcome Trust Clinical Research Programme, University of Malawi College of Medicine, Blantyre, Malawi

The authors conduct a serological cohort study in Kenya where they study the transplacental transfer of RSV-specific antibodies. They report the efficient transplacental transfer of RSV-specific

antibodies among the cohort. They observe that prematurity (<33 weeks gestation), HIV infection, and low birth weight as key factors associated with reduced efficiency of transplacental transfer. They conclude that there is an efficient transplacental transfer of RSV-specific antibodies among pregnant women in Kenya and recommend the integration of other preventive interventions with maternal RSV vaccination targeting infants born premature, low birth weight, and HIV-infected mothers.

This is a timely study in the field as there is a lot of traction on maternal RSV vaccination. The study identifies key groups that may not maximally benefit from maternal RSV vaccination due to inefficient transplacental transfer of RSV-specific antibodies from mother to child. It has important implications on future RSV vaccination policy as it suggests that extra interventions may be needed for children born to HIV-infected mothers, low birth weight babies, and infants born premature.

Overall, this study is well-conducted with most of the conclusions supported by the presented data. However, I recommend that the authors remove paragraph 3 from the discussion section as the concluding statement is not supported by their data.

Is the work clearly and accurately presented and does it cite the current literature?

Yes

Is the study design appropriate and is the work technically sound?

Yes

Are sufficient details of methods and analysis provided to allow replication by others?

Yes

If applicable, is the statistical analysis and its interpretation appropriate?

Yes

Are all the source data underlying the results available to ensure full reproducibility?

Yes

Are the conclusions drawn adequately supported by the results?

Yes

Competing Interests: No competing interests were disclosed.

Reviewer Expertise: Immunology

I confirm that I have read this submission and believe that I have an appropriate level of expertise to confirm that it is of an acceptable scientific standard.

Author Response 24 Mar 2022

Joyce Nyiro, Kenya Medical Research Institute (KEMRI)-Wellcome Trust Research Programme, Kilifi, Kenya

Thank you for your comments and positive feedback on our manuscript. The referred paragraph 3 in the manuscript's discussion section has now been deleted.

Competing Interests: No competing interests were disclosed.
