Supplementary Appendix

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

Supplement to: The RTS,S Clinical Trials Partnership. A phase 3 trial of RTS,S/AS01 malaria vaccine in African infants. N Engl J Med 2012;367;2284-95. DOI: 10.1056/NEJMoa1208394

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Supplement to: A Phase 3 Trial of RTS,S/AS01 Malaria Vaccine in African Infants.

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1. AUTHORS AND AFFILIATIONS

Submitted by the RTS,S Clinical Trials Partnership.

2. SUPPLEMENTARY METHODS

This supplement describes general information on study population and study conduct with a particular emphasis on the 6-12 weeks age category.

2.1. Ethical considerations

This phase III, double-blind (observer-blind), randomized, controlled multi-center study is currently being undertaken in 11 centers across sub-Saharan Africa. The study design and rationale for selection of endpoints have been described previously.[1] The study is being conducted in accordance with the current Declaration of Helsinki, International Committee on Harmonization Good Clinical Practice guidelines[2] and with the local rules and regulations of each country. The study is monitored by the sponsor, GlaxoSmithKline (GSK) Biologicals SA (GSK monitors or outsourced monitors from Quintiles (Quintiles, Centurion, South Africa) contracted by GSK Biologicals SA), and overseen by a formally constituted Independent Data Monitoring Committee (IDMC), that reviewed, among other information, unblinded comprehensive safety data every three months to authorize trial continuation. The IDMC conferred before the initiation of the study and has had three-monthly teleconferences and one annual meeting thereafter. A Local Safety Monitor, who was an experienced clinician not taking part in the study, was available at each study center to support the clinical investigators and to act as a link between the investigators and the IDMC. The study protocol and amendments, consent forms, and

other information that required pre-approval were reviewed and approved by a national, regional, or research center ethics committee (EC) or institutional review board (IRB) as per local requirements. A list of all EC/IRBs is provided in supplementary table 1a.

2.2. Roles of investigators and sponsor

The study is sponsored by GSK Biologicals SA, the vaccine developer and manufacturer, and funded by both GSK Biologicals SA and the PATH Malaria Vaccine Initiative (MVI). The data generated by the trial are subject to a confidentiality agreement between the sponsor and investigators, which allows the investigators full access to the study data at the end of the study and includes an obligation to permit publication without excessive delay.

2.3. Study centers and affiliated partners

The study is being conducted in 11 centers located in seven countries in sub-Saharan Africa and involves collaboration with partner institutions. The study sites represent the range of malaria transmission seen across sub-Saharan Africa (Supplementary figure 1). The list of study centers and affiliated partners is provided in supplementary table 1b.

2.4. Screening and informed consent

Two groups of children were eligible for inclusion in the trial. One group was comprised of infants who were 6-12 weeks of age (inclusive) at the time of first vaccination and who had not previously received a dose of vaccine against diphtheria, tetanus, pertussis or *Haemophilus influenzae* type b and the other group was comprised of children 5 to 17 months of age (inclusive) at the time of first vaccination. Subjects should not have

received any vaccine within the 7 days preceding the first dose of study vaccine. Screening procedures included a review of the child's medical history, a physical examination and a blood test for assessment of hemoglobin concentration. The main exclusion criteria were: moderate or severe illness at the time of enrolment, a major congenital defect, malnutrition requiring hospitalization, severe anemia - defined as a hemoglobin concentration < 5.0 g/dL or a hemoglobin concentration < 8.0 g/dL associated with clinical signs of heart failure or severe respiratory distress, or a past history of a neurological disorder or atypical febrile seizure. A past history of a simple febrile seizure was not an exclusion criterion. Children with active HIV disease of Stage III or Stage IV severity, as defined by the World Health Organization, at the time of screening were excluded.[3] A previous history of active Stage III or Stage IV HIV disease was not an exclusion criterion.

There was no routine testing for HIV in this trial. HIV positive cases were reported on the general medical history taken at screening or identified by morbidity surveillance during the trial. The decision to report a new HIV infection was on the investigators judgment whether it met the criteria for a serious adverse event. Likewise, it was at the investigators discretion to perform antibody or PCR confirmatory testing. Voluntary counseling and testing, highly active anti-retroviral therapy (HAART) and prevention of mother to child transmission (PMCT) were available at all study centers according to national policies.

Prior to enrolment, study teams conducted a series of information activities. Study teams held discussion meetings with the administrative leaders and/or community leaders. They described the outline of the proposed study, paying particular attention to study

procedures including screening of children, immunization, blood collection, follow-up and their associated risks.

Following the community meetings, and a positive recommendation from community leaders, the parent(s)/guardian(s) of children in the eligible age groups were approached. The need for a vaccine against malaria was discussed and the objectives of the study were explained. The study procedures were carefully described including immunization and blood collection. Parent(s)/guardian(s) interested in enrolling their child into the study were invited to the screening visit.

The site investigator or his/her designate described the protocol to the parent(s)/guardian(s) of potential participating children face to face or the informed consent information was presented to groups at an initial information session. Information was provided in both oral and written form in a language fully comprehensible to the child's family. Each child's family had the opportunity to inquire about details of the study and ask any questions individually in a private place. Formal informed consent was obtained from each child's parent(s) or guardian(s) prior to the performance of any study-specific procedures. Literate parent(s)/guardians willing to let their child enter into the study were asked to sign and date the informed consent form (ICF). If the parents or guardians were illiterate, the study and the ICF were explained point by point in the presence of an impartial witness. The impartial witness could be a friend or family member accompanying the parents or any other literate person independent from the study team. Parent(s)/guardian(s) confirmed their consent for their child to take part in the study by marking the ICF with their thumbprint and the impartial witness personally signed and dated the ICF.

2.5. Randomization and blinding

After verification of eligibility criteria, and prior to the first vaccination, a unique treatment number was assigned to each participating child. Participating children from each age-category were randomized into one of three study groups according to a 1:1:1 ratio (R3R, R3C or C3C) using a randomization algorithm with SAS version 9.1 (Supplementary figure 2). Randomization was stratified for age-category using center as a minimization factor, ensuring balanced treatment allocation within each study center. All children's parent(s)/guardian(s) were provided with a study identification card with a photo of their child, the child's name and a unique subject number. All data were collected using remote data entry (RDE) and electronic case report forms (eCRF).

Data were collected in a double-blinded (observer-blind) manner; the vaccinated children and their parent(s)/guardian(s) as well as those responsible for the evaluation of study endpoints were unaware of whether RTS,S/AS01 or control vaccine had been administered to a particular child. The vaccines used in this study were of different appearance. The content of the syringe was, therefore, masked with an opaque tape to ensure that parent(s)/guardian(s) were blinded. The only study staff who knew of the vaccine assignment were those responsible for preparation and administration of vaccines; these staff played no other role in the study except screening or collection of biologic specimens.

2.6. Contribution to the per-protocol analyses

To be included in the per-protocol analysis of efficacy, participants must have received three doses of RTS,S/AS01 or control vaccine and three doses of co-administered vaccine

(DTPwHepB/Hib and OPV) according to protocol procedures within specified intervals and contributed to the time at risk in the follow-up period starting 14 days post Dose 3.

To be included in the per-protocol analysis of immunogenicity, participants must have received three doses of RTS,S/AS01 or control vaccine and three doses of co-administered vaccine (DTPwHepB/Hib and OPV) according to protocol procedures. Subject should also have followed protocol defined intervals for vaccinations and blood sampling schedules. Subjects with protocol deviations in terms of administration of concomitant vaccinations, screening procedures or subjects unblinded by safety department or investigators were excluded from the per-protocol analysis of immunogenicity.

2.7. Additional analysis of efficacy against severe malaria

As specified in the per-protocol endpoint, severe malaria was analyzed when 250 subjects across both age categories had been diagnosed with a case of severe malaria, these results were published previously.[4] In addition, it was decided during the development of the statistical analysis plan, prior to the performance of any statistical analysis, to add an analysis of efficacy against severe malaria in the same population evaluated for the co-primary endpoint, i.e the first 6000 (approximately) subjects enrolled in each age category followed up for 12 months post Dose 3. The corresponding results were reported in 2011[4] for the children enrolled in the 5-17 months age category and are reported now for the infants in the 6-12 weeks age category.

2.8. Study vaccines

Each child received three doses of either the candidate malaria vaccine RTS,S/AS01 or the control vaccine; in the 6-12 weeks age group the control vaccine was a Meningococcal C conjugate vaccine Menjugate™ (Novartis) (Supplementary figure 2). RTS,S/AS01 or control vaccine were administered into the left anterolateral thigh. The choice of control vaccines was guided by the principles of benefit to the control group without compromising the evaluation of clinical study endpoints. Infants enrolled in the 6-12 weeks age category received the RTS,S/AS01 or control vaccine in co-administration with the DTPwHepB/Hib pentavalent vaccine (Tritanrix™ HepB/Hib, GSK Vaccines) administered into the right anterolateral thigh and an oral polio vaccine containing serotypes 1, 2 and 3 (Polio Sabin™, GSK Vaccines).

The RTS,S/AS01 candidate vaccine has been developed and manufactured by GSK Vaccines and is designed to protect against *P. falciparum* malaria. Manufacturing and quality control are performed in line with current Good Manufacturing Practices. No quality issues in the vaccines used in this trial were recorded. “RTS,S” comprises the carboxyl terminal portion (amino acids 207 to 395) of the circumsporozoite protein from the NF54 strain of *P. falciparum* fused to the hepatitis B surface antigen, co-expressed in yeast with non-fused hepatitis B surface antigen."AS01" describes the Adjuvant System comprising liposomes, MPL (3-O-desacyl-4’-monophosphoryl lipid A) and QS-21 (a triterpene glycoside purified from the bark of *Quillaja saponaria*). Each dose of reconstituted RTS,S/AS01 (0.5 mL) contains approximately 25 μg of antigen, 25 μg of MPL and 25 μg of QS-21 with liposomes.[5]

GSK Vaccines' candidate malaria vaccine has been formulated with the Adjuvant Systems AS02 (oil-in-water emulsion with MPL and QS-21) or AS01 (liposome with MPL and QS-21). RTS,S/AS01 was selected as the best formulation for this phase III trial following a number of phase II trials with both formulations. In all RTS,S/AS02, and RTS,S/AS01 head-to-head comparisons, AS01 proved to be more immunogenic, with recipients of RTS,S/AS01 achieving higher peak anti-CS antibody responses than recipients of AS02.[6,7,8,9]

One dose (0.5 mL) of Novartis's Meningococcal C conjugate vaccine contains 10 μg *Neisseria meningitidis* (strain C11) Group C oligosaccharide conjugated to 12.5-25 μg *Corynebacterium diptheriae* CRM_{197} protein adsorbed on aluminum hydroxide (1.0 mg). The excipients of the reconstituted vaccine include mannitol, sodium phosphate monobasic monohydrate, sodium phosphate dibasic heptahydrate, sodium chloride and water for injections.

GSK Vaccines' DTPwHepB/Hib vaccine is prepared by reconstitution of the Hiberix™ pellet with the Tritanrix™ HepB suspension. Each 0.5 mL dose contains not less than 30 IU of adsorbed diphtheria toxoid, not less than 60 IU of adsorbed tetanus toxoid, not less than 4 IU of whole cell pertussis, 10 μg of recombinant hepatitis B antigen (HBsAg) protein and 10 μg of purified capsular polyribosyl ribitol phosphate (PRP) covalently bound to approximately 30 μg tetanus toxoid. Tritanrix™ HepB also contains 2-phenoxyethanol, polysorbate 20, sodium chloride, thiomersal and water for injection. Hiberix™ also contains lactose.

The oral polio vaccine obtained from GSK Vaccines is a stabilized suspension of types 1, 2 and 3 live attenuated polioviruses (Sabin strains): Type 1 (strain LSc, 2ab), Type 2

(strain P 712 ch, 2ab), Type 3 (strain Leon 12a, 1b). The excipients comprise magnesium chloride, L-arginine, polysorbate 80, neomycin sulphate (residual), polymyxin B sulphate (residual) and purified water.

Children were observed closely for at least 30 minutes after vaccination, with appropriate medical treatment and equipment readily available in case of an anaphylactic reaction. A study clinician accredited in pediatric resuscitation was available at all vaccination sessions.

2.9. Bednets and indoor residual spraying (IRS)

The research team ensured that insecticide treated bednet use was optimized in each study population: in two centers (Kilifi, Kenya and Bagamoyo, Tanzania) this was achieved through close collaboration with the National Malaria Control Programs. In the other centers, impregnated bednets were distributed by the study teams to all children who underwent screening, regardless of whether they were eligible for the trial.

Data were collected on malaria control measures used by the participants' families during the period of surveillance. Bednet usage and indoor residual spraying (IRS) were documented 12 months after the third vaccine dose had been given. Children's parents were asked if their house had been sprayed with a residual insecticide and, if so, when this was done. Then they were asked if their child sleeps under a bednet. During a home visit, a field worker inspected the child's bednet and the integrity of the net was recorded as follows: 1: no bednet; 2: impregnated bednet with no hole large enough to admit three fingers; 3: impregnated bednet with at least one hole large enough to admit three fingers;

4: untreated bednet with no hole large enough to admit three fingers; 5: untreated bednet with at least one hole large enough to admit three fingers.

2.10. Safety assessments

During the study, investigators or their designates were responsible for documenting and reporting events meeting the criteria and definition of an adverse event (AE) or serious adverse event (SAE). Parents/guardians of children participating in the study were requested to contact study personnel immediately if their child showed any signs or symptoms they perceived as serious.

An adverse event was defined as any untoward medical occurrence in a child participating in the clinical trial, temporally associated with vaccination whether or not considered related to the vaccine. An AE could, therefore, be any unfavorable and unintended sign (including an abnormal laboratory finding), symptom, or disease (new or exacerbated) temporally associated with vaccination.

For the purpose of this study, a serious adverse event was defined as any untoward medical occurrence that resulted in death, was life-threatening, required hospitalization or prolongation of existing hospitalization, resulted in disability/incapacity, or a seizure within 30 days of vaccination. Abnormal laboratory findings that were judged by the assessing clinician to be clinically significant were recorded as SAEs if they met the criteria for SAE as defined above.

Seizures occurring within 30 days of vaccination and immune-mediated disorders were reported as SAEs in order to ensure availability of full case narrative descriptions.[1] Data on seizures occurring within 7 days following vaccination were collected and analyzed

according to the Brighton Collaboration guidelines[10] and have been published previously.[4]

In previous phase II RTS,S/AS studies, an imbalance was observed in the incidence of rash.[11] In this trial, the occurrence of rashes and mucocutaneous lesions that occurred within 30 days of vaccination in the first 200 subjects enrolled at each center in the 6-12 weeks age category was reported as an AE or SAE. Medical documentation of the events was reported. Rashes and mucocutaneous lesions that met the criteria for an SAE were reported in all subjects throughout the study period. The analysis of rashes and mucocutaneous diseases was based on the Brighton Collaboration Guidelines.[12] The Brighton Collaboration recommends that the principle analyses are presented on those lesions of diagnostic certainty 1-3. However in the field the classification was open to different interpretations and not consistently applied. For that reason the analyses were based on all mucous or cutaneous lesions reported as unsolicited events in the 30 days post vaccination.

Because pediatric auto-immune diseases are rare and may be underestimated in sub-Saharan Africa, training material on pediatric auto-immune disease presentation and diagnosis was provided by the study sponsor. A specific, standardized clinical data collection questionnaire was generated. Collaborations with reference laboratories in South Africa were initiated so that serum samples or histopathologic specimens could be sent to South Africa for analyses not locally available.

In the first set of results published for this trial, meningitis was reported as a SAE more frequently in the RTS,S/AS01 group than in the control group.[4] All the information available on cases of meningitis reported as a SAE were reviewed in detail by two

experienced investigators. The IDMC also reviewed unblinded safety reports containing specific sections on seizures and meningitis. The IDMC recommended study continuation and will continue to review specific meningitis report.

All solicited AEs were reported for 7 days (day of vaccination and 6 subsequent days) following each vaccine dose for the first 200 infants enrolled at each center. Local AEs solicited were: pain at injection site; swelling at injection site and redness at injection site. Solicited general AEs were: drowsiness, fever; irritability/fussiness and loss of appetite. Intensity of AEs was assessed as described in supplementary table 4.

All unsolicited AEs were reported for 30 days following each vaccine dose for the first 200 infants enrolled at each center.

SAEs were collected for all participating children throughout the study period, from the time of parental consent. At every visit/contact, information was sought on the occurrence of AEs/SAEs. SAEs were identified by surveillance at health facilities in the study area and through monthly home visits with the participating children. All AEs that were observed directly or that were observed by a clinical collaborator, those that were identified through surveillance at health facilities in the study area or those reported by the child's parent/guardian spontaneously or in response to a direct question were evaluated.

Assessments were made of the maximum intensity of all unsolicited AEs and SAEs during the period of the event. The assessment was based on the attending clinician's medical judgment. A grade was assigned to all adverse events as follows; grade 1 (mild): an AE which is easily tolerated by the child, causing minimal discomfort and not interfering with everyday activities; grade 2 (moderate): an AE which is sufficiently

discomforting to interfere with normal everyday activities and grade 3 (severe): an AE which prevents normal, everyday activities.

SAEs were coded according to the MedDRA (Medical Dictionary for Drug Regulatory Activities). Non-malaria SAEs were defined as those which excluded the MedDRA terms "*Plasmodium falciparum* infection", "Malaria" and "Cerebral malaria".

Verbal autopsies were carried out on all children who died outside a health facility to ascribe the cause of death using a questionnaire based on the International Network for the Demographic Evaluation of Populations and Their Health in Developing Countries (INDEPTH) standard questionnaire, adapted to be locally appropriate.[13] To support the timely reporting of SAEs, diagnoses were made according to the usual processes of each center.

2.11. Surveillance for clinical and severe malaria episodes

During the informed consent process, parents were asked to bring their child to a study health facility as soon as possible if their child fell sick during the trial. All participating children who presented to a health facility in the study area were evaluated as potential cases of malaria using a standardized algorithm. All parents were asked whether the child had a fever within the previous 24 hours and all children had their temperature measured. A blood sample was taken for testing for malaria parasites in all children who had a history of fever during the prior 24 hours or who had a measured axillary temperature ≥ 37.5°C at the time of presentation.

Children who needed inpatient care were provided transport to a hospital participating in the trial. All participating children who presented for admission were evaluated as

potential cases of severe malaria following a predefined algorithm (Supplementary table 3). Detection and management of severe malaria have been described in detail by Vekemans et al.[14] During any hospitalization, the child's course was monitored to capture the signs and blood parameters indicative of progression to severe malaria. If a child's condition deteriorated following admission then additional investigations were performed.

Treatment of malaria was performed in accordance with national guidelines. In nine of the 11 study centers, the first line treatment for uncomplicated malaria was a 6-dose regimen of artemether-lumefantrine whilst in two, both in Ghana, it was artesunate-amodiaquine. Children who required inpatient care were admitted to the hospital and received treatment with intravenous quinine, according to national guidelines.

2.12. Chest Radiographs

Chest radiographs were performed as part of the standardized evaluation of study participants brought to a healthcare facility with tachypnea, lower chest wall indrawing, abnormally deep breathing or if a study clinician considered this to be an appropriate investigation.[14]

A digital radiography system was provided to each study center to facilitate radiological assessment of study participants. The radiographers and the physicians who read the images for the trial endpoints received standardized technical training by the manufacturer of the radiography equipment and training on interpretation of chest radiograph images by expert radiologists and physicists. To ensure a robust and verifiable radiograph data base, quality control systems that included local on-site training,

development of quality manuals, quality control checks, on-site radiology committees and external audits were implemented. Digital images were anonymized and sent to a central repository at GSK Vaccines via a satellite internet connection.

For the purpose of endpoints assessment, to ensure accurate diagnosis of pneumonia, a process developed by WHO[15] was followed. Each radiograph was read independently by a clinician attached to the center where the radiograph was taken, and by an external radiologist. GSK Vaccines reviewed all readings made by the centers and by the external radiologists and any images with discordant readings were sent to another panel of radiologists for a final reading. The reporting of pneumonia as a SAE was made based on clinicians' judgment and independent of this protocol-specific assessment.

Clinicians and external radiologists were trained in chest radiograph interpretation according to WHO guidelines.[15]

2.13. Anthropometry

Length/height, weight and mid-upper arm circumference were measured at screening and one month after the third dose of vaccine. The methodologies used for anthropometry were adapted from Cogill.[16]

2.14. Laboratory analysis

Development of standardized laboratory methods and quality control processes for this trial have been described fully in a separate publication[17] and are summarized briefly here.

- **Hematology and biochemistry**

Automated biochemical and hematologic methods were used. All biochemistry automated analyzers were initially enrolled with International External Quality Assessment (EQA) but later switched to the program run by the Royal College of Pathologists of Australia, because the latter was more appropriate for the study requirements at the time. All hematology automated analyzers were enrolled in EQA. Each laboratory had to demonstrate method qualification for biochemistry and hematology, including analysis of repeatability, reproducibility, linearity, QC stability and accuracy between main and back-up analyzers. Data were sent to GSK Vaccines for analysis and feedback was provided to laboratories.

Daily internal QC was performed at each center, and external quality control was performed monthly for biochemistry and hematology samples.

- **Microbiology**

Standard microbiology methods for blood and cerebrospinal fluid (CSF) culture were followed using automated Bactec™ incubators and pediatric bottles (Bactec BD Diagnostic Systems, USA). Positive cultures were sub-cultured using standard methods.[18, 19] For the purpose of trial analysis, as opposed to clinical care, results were classified by standardized case definitions based on an established methodology.[20] A blood culture was considered positive if a definite pathogen was isolated (e.g. *Streptococcus pneumoniae, S. agalactiae, S. pyogenes, Haemophilus influenzae*, Salmonella species) or if a bacterium that could be either a pathogen or a contaminant was isolated within 48 hours of incubation (e.g. *Escherichia coli*, *Klebsiella pneumoniae*, *Staphylococcus aureus*, *Enterococcus faecalis*). A blood culture was considered to be

contaminated if a known contaminant was isolated or if a bacterium that could be either a pathogen or a contaminant was isolated after 48 hours of incubation.[20]

CSF was examined by Gram stain and a white cell count was performed using a hemocytometer. Direct agglutination methods using commercial kits (Remel Wellcogen Bacterial Meningitis Antigen Latex Kit or BIO-RAD Pastorex Meningitis Kit) were used for early detection of specific organisms like *S. pneumoniae,* group B streptococci*, H. influenzae* type b*, E. coli* and *Neisseria meningitidis.* In parallel, CSF was inoculated directly onto recommended culture media and in the same bottles used for blood culture in automated incubators to allow for bacterial growth, identification and antimicrobial sensitivity testing using the disk diffusion method.

For the assessment of protocol endpoints, bacterial meningitis was defined as the presence of a CSF white cell count of $\geq 50 \times 10^6$/L, a positive CSF culture of compatible organisms or a positive CSF latex agglutination test for either *H. influenzae* type b (Hib), *N. meningitidis* or *S. pneumoniae*.[14, 21] The reporting of meningitis cases as SAE was independent of this definition. SAE diagnoses were made by the study clinicians, using clinical judgment, based on clinical and laboratory evidence available.

Microbiology quality assessment included evaluation of microscopy, culture, identification and antimicrobial susceptibility testing. Each laboratory received six samples (with at least two meningeal and two enteric organisms) three times per year, and the criteria of acceptability were defined by the National Institute of Communicable Disease (NICD, South Africa). Internal quality control was performed using American Type Culture Collection (ATCC) control strains for species identification every week or when a new batch of reagent was received or when discordant results were obtained. The

contamination rate of the clinical specimens was evaluated monthly by internal assessment. Continuous assessment allowed re-training programs for both clinical and laboratory staff and more intense quality evaluation when there was a high contamination rate.

- ***P. falciparum* counts by blood smear**

All slides were read independently by two trained microscopists. A third independent microscopist read the slide if there were any of the following discrepancies between the first two readings: (1) a positive reading by one microscopist and a negative reading by the other; (2) both microscopists recorded a parasitemia >400 parasites/μL but the higher count divided by the lower count was >2; (3) at least one microscopist recorded a parasitemia ≤400 parasites/μL but the higher reading was more than 10 times the lower reading.

If the initial two readings gave concordant results, the final parasite density was considered to be the geometric mean of the two readings. If the readings were discordant, then the following principles were applied: (1) where one reading was positive and the other negative, the majority decision obtained following the reading by the third microscopist was adopted – and, when the slide was considered positive, the parasite density was recorded as the geometric mean of the two positive results; (2) when all three readings were positive, the final result was the geometric mean of the two closest readings (in log scale). As a quality measure, agreement between the two microscopists was calculated by means of the Kappa statistic.

Internal QC was performed on one negative and one positive slide for each batch of stain. The External QA process for slide reading comprised species identification and parasite

quantification. Three assessments per year were carried out, including 20 samples per microscopist. Microscopists who were below the level defined as competent were considered to be 'in training' and were not allowed to read study slides until they were retrained and re-assessed.

2.15. Immunological assessment

Anti-circumsporozoite (anti-CS) antibody titers were measured in the first 200 infants enrolled at each study center in the 6-12 weeks age category.

Antibodies specific for the circumsporozoite protein tandem repeat epitope were assessed by a standard, validated ELISA with plates adsorbed with the recombinant antigen R32LR that contains the sequence [NVDP(NANP)15] 2LR as described prevously.[22] Briefly, R32LR protein was coated onto a 96-well polystyrene plate. Serial dilutions of serum were added to the 96-well plate and, after incubation, the plates were washed and Horseradish Peroxidase conjugated polyclonal rabbit anti-human IgG was added. After a final washing step, a color reaction was developed with 3, 3',5,5' tetramethylbenzidine and the plates were read in an ELISA reader. Antibody concentrations were calculated from a standard curve with the software SoftMax® Pro (using a four parameters equation) and expressed as EU/mL. Anti-CS antibodies were tested at the CEVAC Laboratory, University of Ghent, Belgium. The cut-off for the anti-CS ELISA was 0.5 EU/mL. Serum samples with a titer below the cut-off value were given a value of 0.25 EU/mL.

2.16. Data collection, data management and statistical analysis

At each study center, data were remotely entered on electronic case report forms and transferred to GlaxoSmithKline for data management. External monitors reviewed medical records, sample storage, and laboratory procedures to ensure data integrity.

In order to preserve the blinding of the ongoing trial, all data cleaning processes were blinded to study group and analyses were conducted by an external statistician, Catherine Dettori (4Clinics), who performed the analyses using SAS Drug Development (SDD) version 3.5 on SunOS/5.10. (SAS Institute Inc., Cary, NC, USA) based on a cleaned dataset and quality controlled programs provided by GSK Vaccines.

2.17. Major protocol deviations

When reporting the first results from this study, defaults in bednet distribution at screening and study vaccine exposure to temperatures outside recommended ranges were reported in detail.[4] These deviations did not pertain to participants enrolled in the 6-12 weeks age category. There was no protocol deviation estimated to potentially impact the integrity of the results presented here when considering the 6-12 weeks age category.

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Omari Abdul, Salim Abdulla, Béatrice Peggy Abossolo,George Adjei, Samuel Adjei, Tsiri Agbenyega, Selidji Todagbe Agnandji, Alex Agyekum, Saumu Ahmed, Pedro Aide, Ali Mohammed Ali, Pauline Akoo, Pedro Alonso, Daniel Ansong, Isaac Asante, Kwaku Poku Asante, Norbert Awino, John Bawa, Philip Bejon, Owusu Boahen, Harry Owusu Boateng, Daniel Chandramohan, Roma Chilengi, Cornelia Conzelmann, David Dosoo, Chris Drakeley, José Francisco Fernandes, Arnaud Flamen, Samwel Gesase, Jesse Gitaka, Stacey Gondi, Robert Tinga Guiguemdé, Ali Said Hamad, Mary J. Hamel, Saadou Issifou, Erik Jongert, Omar Juma, Bérenger Kaboré, Portia Kamthunzi, Simon Kariuki, Christine Kerubo, Peter Gottfried Kremsner, Hassan Kilavo, Evans Kwara, Miguel Lanaspa, Trudie Lang, Kayla F. Laserson, Bertrand Lell, Martha Lemnge, Edwin Liheluka, John Lusingu, Eusébio Macete, Francisca Machera, Coline Mahende, Charity Maingi, Anangisye Malabeja, Sofia Mandjate, Kevin Marsh, Francis Martinson, Rose Minja, Benjamin Mordmüller, Maxmillian Mpina, Ali Mtoro, Vincent Muturi-Kioi,

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John Aponte, Marc Lievens, and Ali Mohamed Ali developed the analysis plan of the data;

John Aponte and Marc Lievens vouch for the data and analysis;

Salim Abdulla, Tsiri Agbenyega, Selidji Agnandji Todagbe, Pedro Aide, Daniel Ansong, John Aponte, Philip Bejon, Joe Cohen, Umberto D'Alessandro, Samwel Gesase, Brian Greenwood, Yolanda Guerra, Mary J. Hamel, Tinto Halidou, Irving Hoffman, Erik Jongert, Simon Kariuki, David Kaslow, Peter Gottfried Kremsner, Didier Lapierre, Amanda Leach, Didier Leboulleux, Bertrand Lell, Marc Lievens, John Lusingu, Francis Martinson, Patricia Njuguna, Bernhards Ogutu, Aurélie Olivier, Lucas Otieno, Walter Otieno, Afiya Radford, Barbara Savarese, Jahit Sacarlal, David Schellenberg, Laurence Slutsker, Marcel Tanner, and Johan Vekemans interpreted the results;

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All authors decided to publish the paper, reviewed manuscript drafts, and approved the final version of the manuscript.

6. CONFLICTS OF INTEREST

The trial was sponsored by GlaxoSmithKline Biologicals SA (GSK), the vaccine developer and manufacturer, and funded by both GSK Biologicals SA and the PATH Malaria Vaccine Initiative (MVI). All centers declare receiving a grant from MVI for running the trial. Author travel and accommodation related to this trial were financed by MVI. GlaxoSmithKline Biologicals received a grant from MVI to run the trial. MVI received a grant from the Bill and Melinda Gates Foundation to run this trial and to compensate MVI authors for trial-related travel. Other conflicts of interest are disclosed below and in forms available with the full text of this article at NEJM.org.

Pedro Aide declares having received a grant from the GlaxoSmithKline group of companies as a contribution to post-graduate training, John Aponte has received for the center consultancy fees from the GlaxoSmithKline group of companies for membership of the DSMB Board for a pneumococcal vaccine. Jahit Sacarlal has received (for the center) some GlaxoSmithKline group of companies'consultancy fees for other studies. Marcel Tanner is a board member of the Optimus Foundation, and his institution is reimbursed for his activities on the Scientific advisory board of the Novartis institute for tropical diseases. He also has received for his institution other grants from MVI, and travel reimbursements from MVI and Sanaria corp. Patricia Njuguna, Philip Bejon, and Gabriel Mwambingu declare that their institution has received grants from MVI for other malaria studies. Kevin Marsh declared his center receiving a grant from EDCTP for other Phase II malaria vaccine trials. Jon Ben Woods declares his employer, the Walter Reed

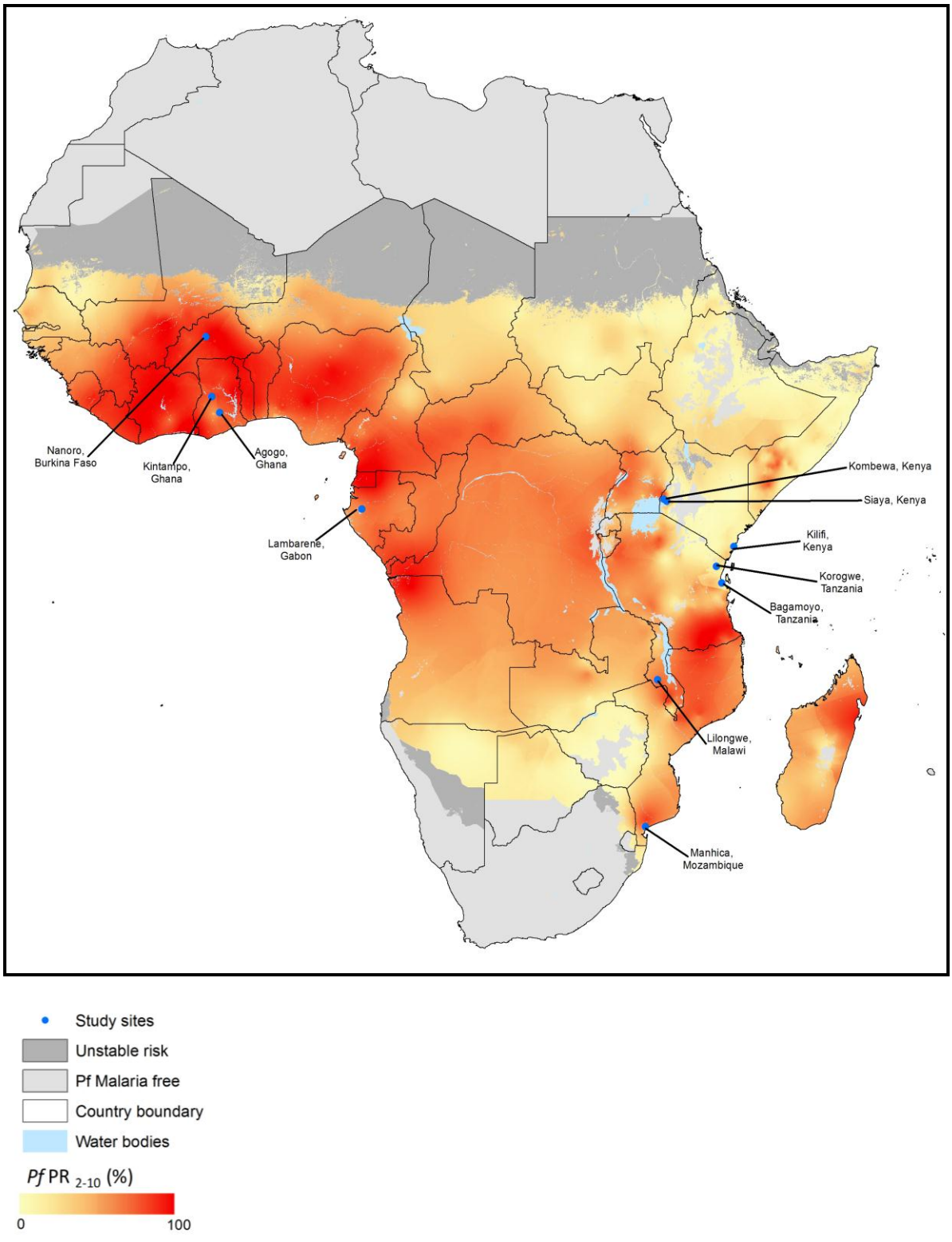
Army Institute of Research, receiving grants from the GlaxoSmithKline group of companies. Brian Greenwood declares that the London School of Hygiene and Tropical Medicine, at which he is employed, receives a grant from the GlaxoSmithKline group of companies for work on a pneumococcal protein vaccine. John Lusingu declares having received consultancy fees from the Task Force on Immunization in the WHO-AFRO region outside the submitted work, and having other grants from the GlaxoSmithKline group of companies and MVI. Umberto D'Alessandro declares his institution receiving consultancy and speaker fees from Sigma Tau and Novartis.

All GSK Vaccines authors are, or were at the time of the study, employed by the GlaxoSmithKline group of companies. Joe Cohen now works as an independent consultant for GSK Vaccines. W. Ripley Ballou, Joe Cohen, Didier Lapierre, Amanda Leach, Marc Lievens, Opokua Ofori-Anyinam and Johan Vekemans have shares/stock options in the GlaxoSmithKline group of companies. Joe Cohen and W. Ripley Ballou declare that they are named inventors on patents for which the rights have been assigned to GlaxoSmithKline group of companies.

Terrell Carter, David Kaslow, Didier Leboulleux, Christian Loucq, Afiya Radford, Barbara Savarese, Marla Sillman, and Preeti Vansadia are, or were at the time of the study, employees at PATH-MVI. David Schellenberg is employed by the London School of Hygiene and Tropical Medicine, and his consultancy activities for MVI are funded as a grant to the LSHTM by MVI. Christian Loucq holds shares in GlaxoSmithKline group of companies. David Kaslow holds stock or stock options from Merck & Co, Inc.

7. SUPPLEMENTARY TABLES AND FIGURES

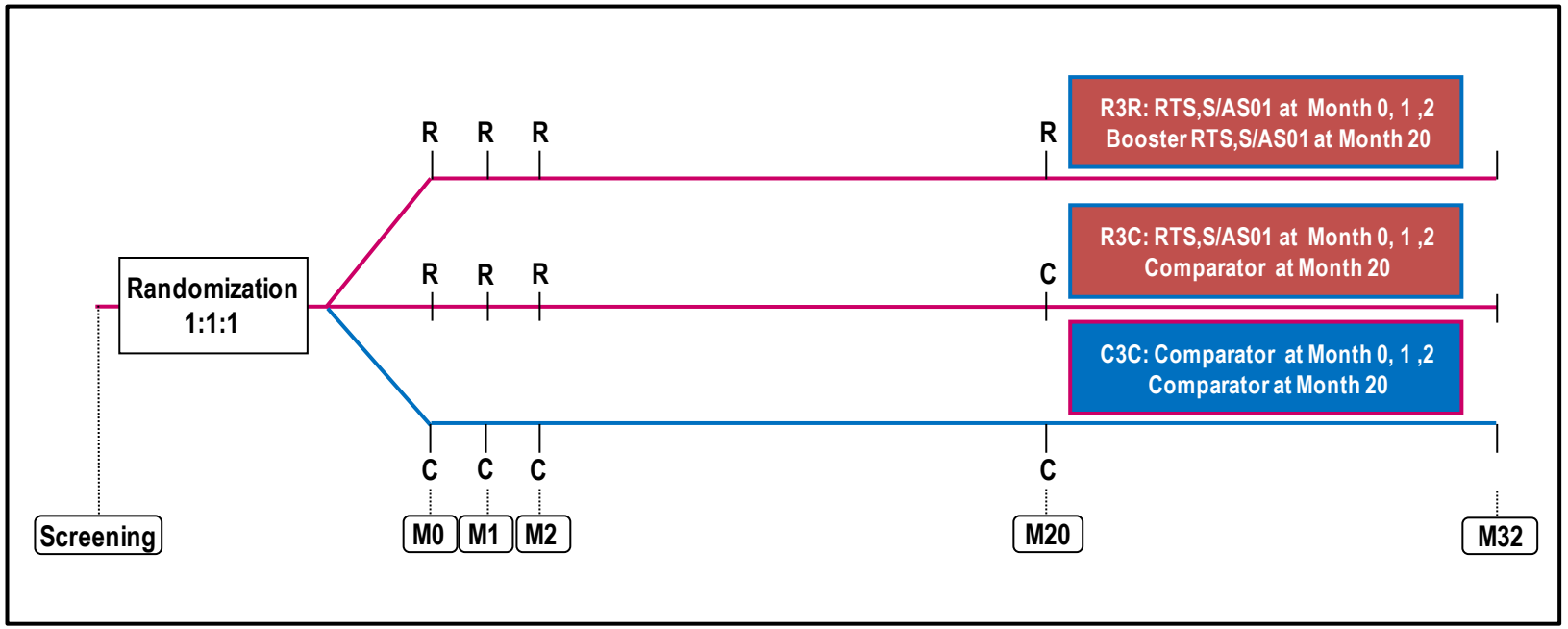
Figure S1. Study centers and malaria endemicity



Adapted from Hay et al, 2009.[23]

The location of each participating center has been added to this previously published map showing the spatial distribution of *P. falciparum* malaria endemicity. The data are the model-based geostatistical point estimates of the annual mean *P. falciparum* parasite rate age-standardized for 2-10 years for 2007 within the stable spatial limits of *P. falciparum* malaria transmission, displayed as a continuum of yellow to red from 0%–100% (see map legend). The rest of the land area was defined as unstable risk (medium grey areas) or no risk (light grey). Nanoro, Burkina Faso has highly seasonal malaria transmission.

Figure S2. Study design



M = Study Month

Figure S3. Consort diagram of the first 200 infants enrolled in each center at 6-12 weeks of age

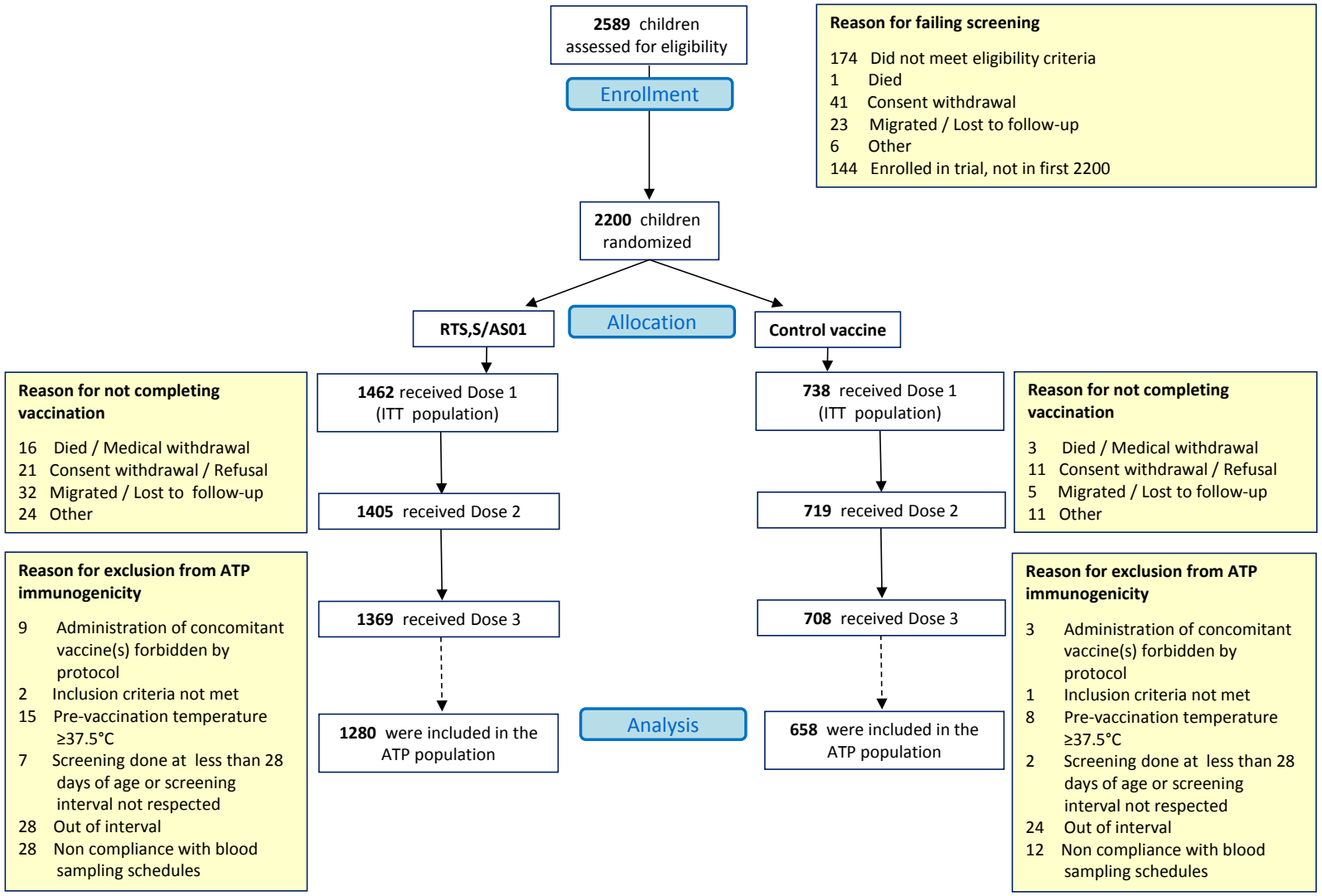
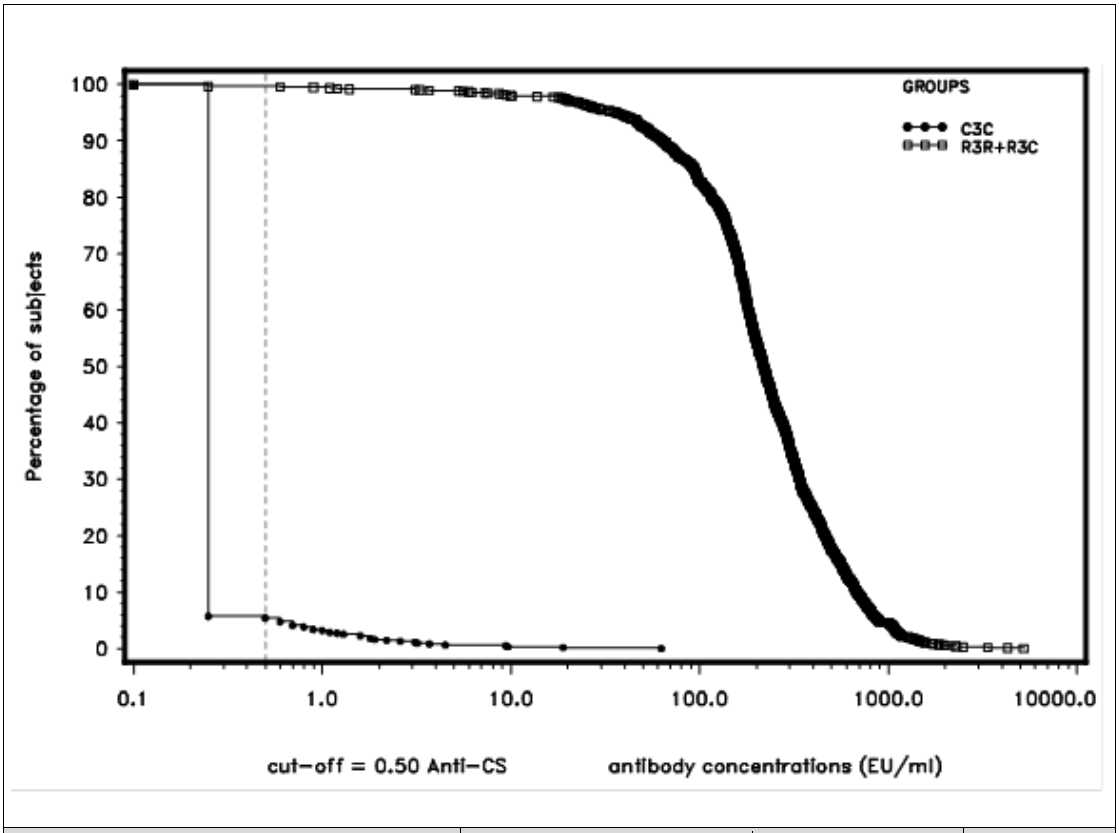


Figure S4. Reverse cumulative distribution curve for anti-CS antibodies (1 month post dose-3), seropositivity and GMTs (Per-protocol population)



Anti-CS			Seropositivity (≥ 0.5 EU/mL)				GMT				
					95% CI			95% CI			
Group	Timing	N	n	%	LL	UL	value	LL	UL	Min	Max
RTS,S/AS01	Screening	1235	424	34.3	31.7	37.1	0.4	0.4	0.4	<0.5	45.0
	1 month post dose-3	1223	1219	99.7	99.2	99.9	209	197	222	<0.5	5210
MenC vaccine	Screening	627	221	35.2	31.5	39.1	0.4	0.4	0.5	<0.5	6.2
	1 month post dose-3	627	36	5.7	4.1	7.9	0.3	0.3	0.3	<0.5	62.8

MenC = Meningococcal serogroup C conjugate vaccine

GMT = geometric mean antibody titer calculated on all infants

N = number of infants with available results. Immunogenicity data were analyzed only in the first 200 infants enrolled at each site.

n/% = number/percentage of infants with titer equal to or above the specified value

95% CI = 95% confidence interval; LL = Lower Limit, UL = Upper Limit

Min/Max = Minimum/Maximum

Table S1a. List of Ethics Committees and Review Boards

Study Centers	Ethics Review Body
Institut de Recherche en Science de la Santé, Nanoro, Burkina Faso	Western Institutional Review Board (WIRB)
	Comité d'Ethique Institutionnel du Centre Muraz (Institutional Ethics Committee of Muraz Center)
	Comite d'Ethique pour la Recherche en Santé (Ethics Committee for Health Research)
Albert Schweitzer Hospital, Lambarene, Gabon	Western Institutional Review Board (WIRB)
	Comité d'Ethique Régional Indépendant de Lambaréné (CERIL) (Independent Regional Ethics Committee of Lambaréné)
	Comité National d'Ethique pour la Recherche (National Ethics Committee for Research The Board)
School of Medical Sciences, Kumasi (Agogo), Ghana	Western Institutional Review Board (WIRB)
	Ghana Health Service (GHS) Ethical Review Committee (ERC) Research and Development Division
	Committee on Human Research Publication and Ethics (CHRPE)
Kintampo Health Research Center, Kintampo, Ghana	Western Institutional Review Board (WIRB)
	Kintampo Health Research Centre (KHRC) Institutional Ethics Committee (IEC)
	London School of Hygiene and Tropical Medicine Ethic Committee
	Ghana Health Service (GHS) Ethical Review Committee (ERC) Research and Development Division
KEMRI - Walter Reed Project, Kombewa, Kenya	Western Institutional Review Board (WIRB)
	Kenya Medical Research Institute (KEMRI) National Ethics Review Committee
	Walter Reed Army Institute of Research (WRAIR) IRB
KEMRI - Wellcome Trust Research Program, Kilifi, Kenya	Western Institutional Review Board (WIRB)
	Kenya Medical Research Institute (KEMRI) National Ethics Review Committee

KEMRI/CDC Research and Public Health Collaboration, Siaya, Kenya	Western Institutional Review Board (WIRB)
	Kenya Medical Research Institute (KEMRI) National Ethics Review Committee
	Centers for Disease Control and Prevention(CDC) – IRB
University of North Carolina Project, Lilongwe, Malawi	Western Institutional Review Board (WIRB)
	National Health Sciences Research Committee
	Office of Human Research Ethics
Centro de Investigação em Saúde de Manhiça, Manhiça, Mozambique	Western Institutional Review Board (WIRB)
	Comitè Etic Investigació Clinica (Hospital Clinic (Barcelona University) Ethics Committee)
	Comité Nacional de Bioética para a Saúde (National Bioethical Health Committee, Mozambique)
Ifakara Health Institute, Bagamoyo, Tanzania	Western Institutional Review Board (WIRB)
	National Institute for Medical Research (NIMR)
	Ethikkommission beider Basel (EKBB) (Ethics Committee of the Swiss Tropical Institute)
	Ifakara Health Institute - IRB
National Institute for Medical Research, Korogwe, Tanzania	Western Institutional Review Board (WIRB)
	London School of Hygiene and Tropical Medicine
	Tanzania Medical Research Coordinating Committee (MRCC) operating within National Institute for Medical Research (NIMR)
	The Danish National Committee on Biomedical Research Ethics

Table S1b. Investigational centers and affiliated partners

Country	Investigational center	Abbreviated name	Affiliated partner
Burkina Faso	Institut de Recherche en Science de la Santé	Nanoro	Prince Leopold Institute of Tropical Medicine, Belgium
Gabon	Albert Schweitzer Hospital, Medical Research Unit	Lambaréné	University of Tübingen, Germany
Ghana	Kwame Nkrumah University of Science and Technology, School of Medical Sciences, Kumasi	Agogo	
Ghana	Kintampo Health Research Centre	Kintampo	London School of Hygiene and Tropical Medicine, UK
Kenya	KEMRI - Wellcome Trust Research Program	Kilifi	University of Oxford, UK
Kenya	KEMRI - Walter Reed Project	Kombewa	Walter Reed Army Institute of Research, USA
Kenya	KEMRI/CDC Research and Public Health Collaboration	Siaya	US Centers for Disease Control and Prevention, USA
Malawi	University of North Carolina Project	Lilongwe	University of North Carolina at Chapel Hill, USA
Mozambique	Centro de Investigação em Saúde de Manhiça	Manhica	Barcelona Centre for International Health Research (CRESIB), Hospital Clinic - Universitat de Barcelona
Tanzania	Ifakara Health Institute	Bagamoyo	Swiss Tropical and Public Health Institute, Switzerland
Tanzania	National Institute for Medical Research	Korogwe	London School of Hygiene and Tropical Medicine, UK Center for Medical parasitology at University of Copenhagen and

			Copenhagen University Hospital, Denmark Kilimanjaro Christian Medical Centre, Tanzania

Table S2. Case definitions of severe malaria

Primary definition	*P. falciparum* > 5000 parasites per µL	**AND** with one or more marker of disease severity: • Prostration • Respiratory distress • Blantyre score ≤ 2 • Seizures 2 or more • Hypoglycemia < 2.2 mmol/L • Acidosis BE ≤-10.0 mmol/L • Lactate ≥ 5.0 mmol/L • Anemia < 5.0 g/dL **AND** without diagnosis of a co-morbidity: • Radiographically proven pneumonia • Meningitis on CSF examination • Positive blood culture • Gastroenteritis with dehydration
Secondary definition with co-morbidity	*P. falciparum* > 5000 parasites per µL	**AND** with one or more marker of disease severity

Prostration: in an acutely sick child, the inability to perform previously-acquired motor function: in a child previously able to stand, inability to stand; in a child previously able to sit, inability to sit and in a very young child, inability to suck.

Respiratory distress: lower chest wall indrawing or abnormally deep breathing.

2 or more seizures: occurring in the total time period including 24 hours prior to admission, the emergency room and the hospitalization.

Radiographically proven pneumonia: a consolidation or pleural effusion defined per protocol on a chest x-ray taken within 72 hours of admission.

Meningitis on CSF examination: WC ≥ 50 x10 ^{6}/L or positive culture of compatible organism or latex agglutination positive for Hib, pneumococci or meningococci.

Gastroenteritis with dehydration: history of 3 or more loose or watery stools in previous 24 hours and an observed watery stool with decreased skin turgor (> 2 seconds for skin to return following skin pinch).

Positive blood culture: defined per protocol on a blood culture taken within 72 hours of admission.

Table S3. Algorithm for the evaluation of a hospital admission as a potential case of severe malaria

For all acute hospital admissions (i.e. except planned admissions for medical investigation/care or elective surgery and trauma admissions), a blood sample was taken for evaluation of:
Malaria parasite density
Blood culture
Hemoglobin
Blood glucose, lactate and base excess
Lumbar Puncture was indicated by the presence of:
Seizure except simple febrile seizure (defined as associated with fever, lasts for 5 minutes or less, generalized as opposed to focal, not followed by transient or persistent neurological abnormalities, occurring in a child ≥ 6 months of age, with full recovery within 1 hour)
Blantyre Coma Score < 5 (children ≤ 9 months of age < 4 [in association with best motor response of 1])[24]
Prostration in child < 3 year of age
Meningism/stiff neck/bulging fontanelle
Clinician's judgment
Chest X-ray (CXR) was indicated by the presence of:
Tachypnea (≥50 breaths per minute in a child < 1 year and ≥ 40 breaths per minute in a child ≥ 1 year)[25]
Lower chest wall indrawing
Abnormally deep breathing
Clinician's judgment

Table S4. Grading of solicited adverse events (AE)

Adverse Event	Intensity grade	Parameter
Pain at injection site	0	Absent
	1	Minor reaction to touch
	2	Cries/protests on touch
	3	Cries when limb is moved/spontaneously painful
Swelling at injection site	0	Absent
	1	<5 mm
	2	5-20 mm
	3	>20 mm
Redness at injection site	0	Absent
	1	<5 mm
	2	5-20 mm
	3	>20 mm
Fever	0	<37.5°C
	1	37.5-38°C
	2	>38-39°C
	3	>39°C
Irritability/Fussiness	0	Behavior as usual
	1	Crying more than usual/ no effect on normal activity
	2	Crying more than usual/ interferes with normal activity
	3	Crying that cannot be comforted/ prevents normal activity
Drowsiness	0	Behavior as usual
	1	Drowsiness easily tolerated
	2	Drowsiness that interferes with normal activity
	3	Drowsiness that prevents normal activity
Loss of appetite	0	Appetite as usual
	1	Eating less than usual/ no effect on normal activity
	2	Eating less than usual/ interferes with normal activity
	3	Not eating at all

Table S5. Baseline characteristics of infants aged 6-12 weeks at enrollment (ITT population)

		RTS,S/AS01 N = 4358	MenC vaccine N = 2179
Age in weeks at first dose	Mean ± SD	7.1 ± 1.4	7.1 ± 1.4
Male Gender	n (%)	2236 (51.3)	1081 (49.6)
Distance outpatient [km]	Mean ± SD	4.2 ± 4.3	4.1 ± 4.0
Distance inpatient [km]	Mean ± SD	13.7 ± 11.3	13.3 ± 10.8
Height for age z-score	Mean ± SD	-1.3 ± 1.3	-1.2 ± 1.3
Weight for age z-score	Mean ± SD	-0.5 ± 1.1	-0.5 ± 1.1
Hemoglobin [g/dL]	Mean ± SD	11.1 ± 1.6	11.1 ± 1.7
Moderate anemia [Hb <8g/dL]	n (%)	67 (1.5)	29 (1.3)

MenC = Meningococcal C conjugate vaccine

N = number of infants

n = number of infants in a given category

SD = Standard deviation

Table S6. Malaria episodes by study center (Per-protocol population)

Site	Number of infants enrolled by center included in the per-protocol population	All clinical malaria episodes (meeting the primary case definition)	All severe malaria episodes (meeting the primary case definition)
Agogo	638	405	12
Bagamoyo	746	71	3
Kilifi	287	3	0
Kintampo	299	350	16
Kombewa	583	538	20
Korogwe	565	21	0
Lambarene	209	17	0
Lilongwe	759	172	4
Manihça	569	37	2
Nanoro	666	1141	14
Siaya	682	1172	42
Total	**6003**	**3927**	**113**

Table S7. Malaria prevention in infants aged 6-12 weeks at 14 months post dose-1 (ITT population)

		RTS,S/AS01 N = 4358		MenC vaccine N = 2179	
Characteristics	**Categories**	**n**	**%**	**n**	**%**
Bednet use, measured at Month 14 post dose-1	ITN no holes	1922	51.3	917	48.9
	ITN holes	1302	34.8	682	36.4
	Untreated w/o holes	78	2.1	36	1.9
	Untreated with holes	68	1.8	33	1.8
	No bednet	376	10.0	208	11.1
	Missing	612	-	303	-
Indoor residual spraying*	N	3573	95.2	1819	96.7
	Y	179	4.8	63	3.3
	Missing	606	-	297	-

MenC = Meningococcal C conjugate vaccine

N = number of infants

n = number of infants in a given category

% = n / Number of infants with available results x 100

ITN no holes = insecticide treated bednet with no hole large enough to admit three fingers

ITN holes = insecticide treated bednet with at least one hole large enough to admit three fingers

Untreated w/o holes = untreated bednet with no hole large enough to admit three fingers

Untreated with holes = untreated bednet with at least one hole large enough to admit three fingers

All categories of bednets use were compared between the two study groups (RTS,S/AS01 and MenC vaccine) overall and by site and all *p*-values were above 0.05 (Fisher exact test).

*IRS coverage was low and was conducted as a public health intervention in 4 out of 11 study centers: Manhiça (IRS coverage = 29.9%), Lambarene (IRS coverage = 16.9%), Kintampo (IRS coverage = 5.2%) and Bagamoyo (IRS coverage = 2.4%). IRS coverage was higher overall in the RTS,S/AS01 group when compared with the control group (*p*-value = 0.0122) (Fisher exact test), but the difference in IRS coverage by site was non-significant except in Manhiça (*p*=0.001). Manhiça contributed few clinical or severe malaria cases to the analysis, and therefore this finding likely did not influence the overall VE estimates presented.

Table S8. Vaccine efficacy: model selection using Cox regression with time dependent covariates for clinical malaria (primary case definition) (Per-protocol population)

Model	-2 log likelihood	Parameters	Akaike's criterion	Schwarz Bayesian criterion[§]
No time-varying covariates	22288.5	1	22290.5	22296.0
group*time^{-2}	22283.4	2	22287.4	22298.5
group*time^{-1}	22278.5	2	22282.5	22293.6
group*time$^{-0.5}$	22271.9	2	22275.9	22287.0
group*log(time)	22264.5	2	22268.5	22279.6
group*time	**22259.8**	**2**	**22263.8**	**22274.8**
group*time$^{0.5}$	22260.7	2	22264.7	22275.8
group*time^{2}	22261.2	2	22265.2	22276.3
Piecewise Cox regression[§§]	22263.0	3	22269.0	22285.6

[§]Lowest Swartz Bayesian Criterion (SBC) corresponds to best model fit among pre-specified models evaluated. Best fit is shown in bold.

[§§]Piecewise Cox regression splitting the time at risk in three periods, allowing in each one a third of the cases

Per protocol, the primary analysis of vaccine efficacy against clinical malaria is based on a Cox regression model. These models assume hazards are proportional throughout the follow up period. It is, therefore, good practice to present, alongside the overall effect, an indication whether or not there is evidence that the observed effect varies with time. A variation over time may represent waning efficacy, differential changes between vaccinees and controls as time continues due to acquisition of natural immunity, treatment effects, or susceptibility, or the lack of important covariates in the model. In order to check the proportionality of hazards, a number of models that allowed the effect to vary over time were pre-specified and were evaluated to determine whether any of

those models fitted the data better than the model without a time-varying component. As indicated in the table above, among the pre-specified models, the model that allowed the observed group effect to vary with a linear function of time has the lowest SBC value, and this provides evidence that the observed effect on first or only malaria episodes is not constant over time.

Table S9. Percentage of infants aged 6-12 weeks at enrollment reporting a serious adverse event during 14 months post dose-1 by MedDRA Preferred Term (ITT population)

	RTS,S/AS01 N = 4358				MenC vaccine N = 2179			
			95% CI				95% CI	
	n	%	LL	UL	n	%	LL	UL
At least one symptom	782	17.9	16.8	19.1	419	19.2	17.6	20.9
At least one symptom excluding Malaria	760	17.4	16.3	18.6	407	18.7	17.1	20.4
Anaemia	90	2.1	1.7	2.5	58	2.7	2.0	3.4
SAE by Preferred Term								
Haemolytic anaemia	1	0.0	0.0	0.1	1	0.0	0.0	0.3
Lymphadenitis	*1*				*1*			
Pericardial effusion	*1*				*1*			
Cerebral palsy	*1*				*1*			
Congenital megacolon	*1*				*1*			
Fallot's tetralogy	*1*				*1*			
Sickle cell anaemia	4	0.1	0.0	0.2	3	0.1	0.0	0.4
Sickle cell anaemia with crisis	4	0.1	0.0	0.2	2	0.1	0.0	0.3
Trisomy 21	*1*				*1*			
Urethral valves	*1*				*1*			
Conjunctivitis	*1*				*1*			
Constipation	*1*				*1*			
Enteritis	11	0.3	0.1	0.5	12	0.6	0.3	1.0
Gastritis	2	0.0	0.0	0.2	2	0.1	0.0	0.3
Inguinal hernia	*1*				*1*			
Intestinal obstruction	*1*				*1*			

	RTS,S/AS01 N = 4358				MenC vaccine N = 2179			
			95% CI				95% CI	
	n	%	LL	UL	n	%	LL	UL
Intussusception	*1*				*1*			
Rectal prolapse	*1*				*1*			
Stomatitis	*1*				*1*			
Death	1	0.0	0.0	0.1	1	0.0	0.0	0.3
Drowning	*1*				*1*			
Hypothermia	*1*				*1*			
Injection site reaction	*1*				*1*			
Pyrexia	15	0.3	0.2	0.6	11	0.5	0.3	0.9
Hepatitis	*1*				*1*			
Anaphylactic reaction	*1*				*1*			
Immune reconstitution syndrome	*1*				*1*			
Abscess	7	0.2	0.1	0.3	2	0.1	0.0	0.3
Abscess limb	*1*				*1*			
Abscess neck	*1*				*1*			
Amoebiasis	*1*				*1*			
Arthritis bacterial	*3*				*3*			
Bacterial infection	*1*				*1*			
Bronchiolitis	28	0.6	0.4	0.9	21	1.0	0.6	1.5
Bronchitis	11	0.3	0.1	0.5	3	0.1	0.0	0.4
Bronchopneumonia	35	0.8	0.6	1.1	20	0.9	0.6	1.4
Bullous impetigo	*1*				*1*			
Burn infection	*1*				*1*			
Cellulitis	6	0.1	0.1	0.3	2	0.1	0.0	0.3
Central nervous system viral infection	*1*				*1*			

	RTS,S/AS01 N = 4358				MenC vaccine N = 2179			
			95% CI				95% CI	
	n	%	LL	UL	n	%	LL	UL
Cerebral malaria	2	0.0	0.0	0.2	1	0.0	0.0	0.3
Conjunctivitis bacterial	*2*				*2*			
Dysentery	6	0.1	0.1	0.3	6	0.3	0.1	0.6
Encephalitis viral	*1*				*1*			
Escherichia sepsis	2	0.0	0.0	0.2	2	0.1	0.0	0.3
Exanthema subitum	*1*				*1*			
Febrile infection	*1*				*1*			
Gastroenteritis	260	6.0	5.3	6.7	139	6.4	5.4	7.5
Gastroenteritis salmonella	1	0.0	0.0	0.1	2	0.1	0.0	0.3
Giardiasis	*1*				*1*			
Helminthic infection	*1*				*1*			
Hiv infection	27	0.6	0.4	0.9	9	0.4	0.2	0.8
Hiv infection who clinical stage iii	*2*				*2*			
Impetigo	*3*				*3*			
Injection site abscess	*1*				*1*			
Listeria sepsis	*1*				*1*			
Lobar pneumonia	11	0.3	0.1	0.5	6	0.3	0.1	0.6
Lower respiratory tract infection	4	0.1	0.0	0.2	2	0.1	0.0	0.3
Ludwig angina	*1*				*1*			
Malaria	184	4.2	3.6	4.9	115	5.3	4.4	6.3
Mastoiditis	*1*				*1*			
Measles	20	0.5	0.3	0.7	7	0.3	0.1	0.7
Meningitis	3	0.1	0.0	0.2	1	0.0	0.0	0.3
Meningitis pneumococcal	3	0.1	0.0	0.2	1	0.0	0.0	0.3

	RTS,S/AS01 N = 4358				MenC vaccine N = 2179			
			95% CI				95% CI	
	n	%	LL	UL	n	%	LL	UL
Meningitis salmonella	*3*				*3*			
Moraxella infection	*1*				*1*			
Oral candidiasis	3	0.1	0.0	0.2	1	0.0	0.0	0.3
Oropharyngeal candidiasis	*1*				*1*			
Osteomyelitis	*2*				*2*			
Otitis externa	*2*				*2*			
Otitis media	14	0.3	0.2	0.5	4	0.2	0.1	0.5
Otitis media acute	3	0.1	0.0	0.2	1	0.0	0.0	0.3
Periorbital cellulitis	*1*				*1*			
Pneumococcal sepsis	6	0.1	0.1	0.3	2	0.1	0.0	0.3
Pneumocystis jiroveci pneumonia	*5*				*5*			
Pneumonia	302	6.9	6.2	7.7	152	7.0	5.9	8.1
Pneumonia primary atypical	*1*				*1*			
Pneumonia viral	*1*				*1*			
Pulmonary tuberculosis	9	0.2	0.1	0.4	1	0.0	0.0	0.3
Rubella	*1*				*1*			
Salmonella sepsis	26	0.6	0.4	0.9	16	0.7	0.4	1.2
Sepsis	26	0.6	0.4	0.9	10	0.5	0.2	0.8
Septic shock	*1*				*1*			
Staphylococcal sepsis	7	0.2	0.1	0.3	1	0.0	0.0	0.3
Staphylococcal skin infection	*1*				*1*			
Subcutaneous abscess	3	0.1	0.0	0.2	3	0.1	0.0	0.4
Tonsillitis	*1*				*1*			
Tuberculosis	3	0.1	0.0	0.2	3	0.1	0.0	0.4

	RTS,S/AS01 N = 4358				MenC vaccine N = 2179			
			95% CI				95% CI	
	n	%	LL	UL	n	%	LL	UL
Upper respiratory tract infection	36	0.8	0.6	1.1	19	0.9	0.5	1.4
Urinary tract infection	16	0.4	0.2	0.6	10	0.5	0.2	0.8
Vaginal infection	*1*				*1*			
Varicella	1	0.0	0.0	0.1	1	0.0	0.0	0.3
Viral infection	*1*				*1*			
Burns second degree	3	0.1	0.0	0.2	1	0.0	0.0	0.3
Clavicle fracture	*1*				*1*			
Femur fracture	*1*				*1*			
Head injury	*3*				*3*			
Herbal toxicity	1	0.0	0.0	0.1	3	0.1	0.0	0.4
Human bite	*1*				*1*			
Pneumonitis chemical	*1*				*1*			
Soft tissue injury	*1*				*1*			
Thermal burn	12	0.3	0.1	0.5	2	0.1	0.0	0.3
Wrist fracture	*1*				*1*			
Failure to thrive	1	0.0	0.0	0.1	1	0.0	0.0	0.3
Hypoglycaemia	4	0.1	0.0	0.2	2	0.1	0.0	0.3
Kwashiorkor	2	0.0	0.0	0.2	1	0.0	0.0	0.3
Malnutrition	29	0.7	0.4	1.0	7	0.3	0.1	0.7
Marasmus	6	0.1	0.1	0.3	5	0.2	0.1	0.5
Dactylitis	1	0.0	0.0	0.1	1	0.0	0.0	0.3
Rickets	*1*				*1*			
Torticollis	*1*				*1*			
Inflammatory pseudotumour	*1*				*1*			

	RTS,S/AS01 N = 4358				MenC vaccine N = 2179			
			95% CI				95% CI	
	n	%	LL	UL	n	%	LL	UL
Cerebellar ataxia	*1*				*1*			
Convulsion	41	0.9	0.7	1.3	19	0.9	0.5	1.4
Encephalitis	*1*				*1*			
Encephalomalacia	*1*				*1*			
Encephalopathy	*1*				*1*			
Epilepsy	*1*				*1*			
Febrile convulsion	82	1.9	1.5	2.3	46	2.1	1.5	2.8
Loss of consciousness	*1*				*1*			
Meningism	*1*				*1*			
Metabolic encephalopathy	*1*				*1*			
Myoclonus	*1*				*1*			
Hydronephrosis	*1*				*1*			
Urinary retention	*1*				*1*			
Asthma	4	0.1	0.0	0.2	3	0.1	0.0	0.4
Bronchial hyperreactivity	*1*				*1*			
Bronchospasm	2	0.0	0.0	0.2	1	0.0	0.0	0.3
Pneumonia aspiration	3	0.1	0.0	0.2	4	0.2	0.1	0.5
Pneumonitis	*1*				*1*			
Respiratory arrest	*1*				*1*			
Rash	*1*				*1*			
Urticaria	*1*				*1*			
Hypovolaemic shock	*1*				*1*			
Shock	3	0.1	0.0	0.2	3	0.1	0.0	0.4

MenC = Meningococcal C conjugate vaccine

At least one symptom = at least one symptom experienced, regardless of the MedDRA Preferred Term

At least one symptom excluding Malaria = at least one symptom experienced (regardless of the MedDRA Preferred Term), excluding Malaria, P. falciparum infection, and Cerebral malaria.

N = number of infants with at least one administered dose

n/% = number/percentage of infants reporting the symptom at least once

95% CI = exact 95% confidence interval; LL = Lower Limit, UL = Upper Limit

Tabulations that present single or multiple SAEs in one study group (RTS,S/AS01 or comparator) are presented in both study groups as *n*, indicating that there are n events in one of the study groups, to preserve the blind of the study.

Table S10. Percentage of infants aged 6-12 weeks at enrollment reporting a fatal serious adverse event during 14 months post dose-1 by MedDRA Preferred Term (ITT population)

	RTS,S/AS01 N = 4358				MenC vaccine N = 2179			
			95% CI				95% CI	
Overview	**n**	**%**	**LL**	**UL**	**n**	**%**	**LL**	**UL**
At least one symptom	66	1.5	1.2	1.9	28	1.3	0.9	1.9
Fatal SAE by Preferred Term								
Anaemia	6	0.1	0.1	0.3	2	0.1	0.0	0.3
Haemolytic anemia	*1*				*1*			
Congenital megacolon	*1*				*1*			
Fallot's tetralogy	*1*				*1*			
Enteritis	1	0.0	0.0	0.1	2	0.1	0.0	0.3
Intussusception	*1*				*1*			
Death	1	0.0	0.0	0.1	1	0.0	0.0	0.3
Drowning	*1*				*1*			
Pyrexia	1	0.0	0.0	0.1	2	0.1	0.0	0.3
Bronchitis	*1*				*1*			
Bronchopneumonia	5	0.1	0.0	0.3	2	0.1	0.0	0.3
Burn infection	*1*				*1*			
Cerebral malaria	*1*				*1*			
Dysentery	*1*				*1*			
Encephalitis viral	*1*				*1*			
Febrile infection	*1*				*1*			
Gastroenteritis	15	0.3	0.2	0.6	10	0.5	0.2	0.8
Hiv infection	9	0.2	0.1	0.4	2	0.1	0.0	0.3
Hiv infection WHO clinical stage 3	*1*				*1*			
Lobar pneumonia	*1*				*1*			

Malaria	3	0.1	0.0	0.2	2	0.1	0.0	0.3
Meningitis pneumococcal	2	0.0	0.0	0.2	1	0.0	0.0	0.3
Pneumococcal sepsis	2	0.0	0.0	0.2	2	0.1	0.0	0.3
Pneumocystis jiroveci pneumonia	*3*				*3*			
Pneumonia	21	0.5	0.3	0.7	7	0.3	0.1	0.7
Pneumonia primary atypical	*1*				*1*			
Salmonella sepsis	1	0.0	0.0	0.1	2	0.1	0.0	0.3
Sepsis	8	0.2	0.1	0.4	5	0.2	0.1	0.5
Septic shock	*1*				*1*			
Tuberculosis	*1*				*1*			
Urinary tract infection	*1*				*1*			
Head injury	*2*				*2*			
Herbal toxicity	*1*				*1*			
Thermal burn	*1*				*1*			
Hypoglycemia	*1*				*1*			
Kwashiorkor	*1*				*1*			
Malnutrition	1	0.0	0.0	0.1	1	0.0	0.0	0.3
Marasmus	3	0.1	0.0	0.2	2	0.1	0.0	0.3
Convulsion	5	0.1	0.0	0.3	4	0.2	0.1	0.5
Encephalitis	*1*				*1*			
Febrile convulsion	1	0.0	0.0	0.1	1	0.0	0.0	0.3
Loss of consciousness	*1*				*1*			
Pneumonia aspiration	*2*				*2*			

MenC = Meningococcal C conjugate vaccine

At least one symptom = at least one symptom experienced, regardless of the MedDRA Preferred Term

N = number of infants with at least one administered dose

n/% = number/percentage of infants reporting the symptom at least once

95% CI = exact 95% confidence interval; LL = Lower Limit, UL = Upper Limit

Tabulations that present single or multiple SAEs in one study group (RTS,S/AS01 or comparator) are presented in both study groups as *n*, indicating that there are n events in one of the study groups, to preserve the blind of the study.

None of the fatal SAE was related to vaccination.

Table S11a. Percentage of infants aged 6-12 weeks at enrollment reporting unsolicited adverse events (AE) within the 30-day post-vaccination period (ITT population)

	RTS,S/AS01 N = 1462				MenC vaccine N = 738			
			95% CI				95% CI	
Overview	**n**	**%**	**LL**	**UL**	**n**	**%**	**LL**	**UL**
At least one AE	1161	79.4	77.2	81.5	600	81.3	78.3	84.1
AE with an incidence ≥ 5% by Preferred Term								
Upper respiratory tract infection	584	39.9	37.4	42.5	312	42.3	38.7	45.9
Gastroenteritis	220	15.0	13.3	17.0	131	17.8	15.1	20.7
Pyrexia	251	17.2	15.3	19.2	112	15.2	12.7	18.0
Rhinitis	148	10.1	8.6	11.8	75	10.2	8.1	12.6
Enteritis	131	9.0	7.5	10.5	72	9.8	7.7	12.1
Malaria	137	9.4	7.9	11.0	70	9.5	7.5	11.8
Conjunctivitis	118	8.1	6.7	9.6	63	8.5	6.6	10.8
Nasopharyngitis	76	5.2	4.1	6.5	55	7.5	5.7	9.6
Pneumonia	86	5.9	4.7	7.2	39	5.3	3.8	7.2

MenC = Meningococcal C conjugate vaccine

At least one AE = at least one AE experienced, regardless of the MedDRA Preferred Term

N = number of infants with at least one administered dose. AEs during the first 30 days post-vaccination were recorded and analyzed only in the first 200 infants enrolled at each center.

n/% = number/percentage of infants reporting the symptom at least once

95% CI = exact 95% confidence interval; LL = Lower Limit, UL = Upper Limit

Table S11b. Percentage of infants aged 6-12 weeks at enrollment reporting an unsolicited adverse event (AE) related or leading to withdrawal from further vaccination within the 30-day post vaccination period classified by MedDRA Preferred Term (ITT population)

	RTS,S/AS01 N = 4358				MenC vaccine N = 2179			
			95% CI				95% CI	
Overview	**n**	**%**	**LL**	**UL**	**n**	**%**	**LL**	**UL**
At least one symptom	578	13.3	12.3	14.3	231	10.6	9.3	12.0
Related AE by Preferred Term								
Neutrophilia	*1*				*1*			
Enteritis	*1*				*1*			
Crying	*1*				*1*			
Injection site erythema	19	0.4	0.3	0.7	7	0.3	0.1	0.7
Injection site induration	33	0.8	0.5	1.1	5	0.2	0.1	0.5
Injection site inflammation	*3*				*3*			
Injection site irritation	*1*				*1*			
Injection site pain	21	0.5	0.3	0.7	5	0.2	0.1	0.5
Injection site reaction	*1*				*1*			
Injection site swelling	49	1.1	0.8	1.5	29	1.3	0.9	1.9
Irritability	16	0.4	0.2	0.6	7	0.3	0.1	0.7
Pyrexia	490	11.2	10.3	12.2	194	8.9	7.7	10.2
Anaphylactic reaction	*1*				*1*			
Hiv infection	*1*				*1*			
Hiv infection who clinical stage iii	*1*				*1*			
Injection site abscess	*2*				*2*			
Injection site cellulitis	*2*				*2*			
Malaria	*1*				*1*			
Decreased appetite	*2*				*2*			
Febrile convulsion	*1*				*1*			

Dermatitis allergic	*1*				*1*			
Rash	*1*				*1*			
Rash maculo-papular	*1*				*1*			

MenC = Meningococcal C conjugate vaccine

At least one symptom = at least one symptom experienced, regardless of the MedDRA Preferred Term

N = number of subjects with at least one administered dose. AEs related or leading to withdrawal occurring during the first 30 days post-vaccination were recorded and analyzed only in all infants enrolled in this trial.

n/% = number/percentage of subjects reporting the symptom at least once

95% CI= exact 95% confidence interval; LL = Lower Limit, UL = Upper Limit

Tabulations that present single or multiple AEs in one study group (RTS,S/AS01 or comparator) are presented in both study groups as *n*, indicating that there are n events in one of the study groups, to preserve the blind of the study.

Table S12. Incidence of solicited general adverse events in infants aged 6-12 weeks at enrollment reported during the 7-day post-vaccination period following each dose and overall (ITT population)

		RTS,S/AS01					MenC vaccine				
					95 % CI					95 % CI	
Symptom	**Type**	**N**	**n**	**%**	**LL**	**UL**	**N**	**n**	**%**	**LL**	**UL**
Dose 1											
Drowsiness	All	1462	164	11.2	9.6	12.9	738	65	8.8	6.9	11.1
	Grade 3	1462	1	0.1	0.0	0.4	738	1	0.1	0.0	0.8
	Related	1462	88	6.0	4.9	7.4	738	26	3.5	2.3	5.1
	Grade 3 Related	1462	1	0.1	0.0	0.4	738	0	0.0	0.0	0.5
Irritability	All	1462	370	25.3	23.1	27.6	738	157	21.3	18.4	24.4
	Grade 3	1462	10	0.7	0.3	1.3	738	3	0.4	0.1	1.2
	Related	1462	226	15.5	13.6	17.4	738	84	11.4	9.2	13.9
	Grade 3 Related	1462	6	0.4	0.2	0.9	738	3	0.4	0.1	1.2
Loss of appetite	All	1462	124	8.5	7.1	10.0	738	52	7.0	5.3	9.1
	Grade 3	1462	2	0.1	0.0	0.5	738	0	0.0	0.0	0.5
	Related	1462	67	4.6	3.6	5.8	738	24	3.3	2.1	4.8
	Grade 3 Related	1462	2	0.1	0.0	0.5	738	0	0.0	0.0	0.5
Temperature	All	1462	459	31.4	29.0	33.8	738	192	26.0	22.9	29.3
(Axillary) (°C)	>39.0	1462	5	0.3	0.1	0.8	738	2	0.3	0.0	1.0
	Related	1462	326	22.3	20.2	24.5	738	127	17.2	14.6	20.1
	>39.0 Related	1462	1	0.1	0.0	0.4	738	1	0.1	0.0	0.8
Dose 2											
Drowsiness	All	1412	135	9.6	8.1	11.2	721	55	7.6	5.8	9.8
	Grade 3	1412	0	0.0	0.0	0.3	721	0	0.0	0.0	0.5
	Related	1412	74	5.2	4.1	6.5	721	15	2.1	1.2	3.4
	Grade 3 Related	1412	0	0.0	0.0	0.3	721	0	0.0	0.0	0.5
Irritability	All	1412	289	20.5	18.4	22.7	721	123	17.1	14.4	20.0

		RTS,S/AS01					MenC vaccine				
					95 % CI					95 % CI	
Symptom	**Type**	**N**	**n**	**%**	**LL**	**UL**	**N**	**n**	**%**	**LL**	**UL**
	Grade 3	1412	7	0.5	0.2	1.0	721	0	0.0	0.0	0.5
	Related	1412	175	12.4	10.7	14.2	721	57	7.9	6.0	10.1
	Grade 3 Related	1412	5	0.4	0.1	0.8	721	0	0.0	0.0	0.5
Loss of appetite	All	1412	105	7.4	6.1	8.9	721	43	6.0	4.3	7.9
	Grade 3	1412	0	0.0	0.0	0.3	721	0	0.0	0.0	0.5
	Related	1412	60	4.2	3.3	5.4	721	8	1.1	0.5	2.2
	Grade 3 Related	1412	0	0.0	0.0	0.3	721	0	0.0	0.0	0.5
Temperature	All	1412	411	29.1	26.7	31.6	721	154	21.4	18.4	24.5
(Axillary) (°C)	>39.0	1412	9	0.6	0.3	1.2	721	6	0.8	0.3	1.8
	Related	1412	278	19.7	17.6	21.9	721	89	12.3	10.0	15.0
	>39.0 Related	1412	5	0.4	0.1	0.8	721	3	0.4	0.1	1.2
Dose 3											
Drowsiness	All	1378	124	9.0	7.5	10.6	710	44	6.2	4.5	8.2
	Grade 3	1378	1	0.1	0.0	0.4	710	1	0.1	0.0	0.8
	Related	1378	49	3.6	2.6	4.7	710	19	2.7	1.6	4.1
	Grade 3 Related	1378	0	0.0	0.0	0.3	710	0	0.0	0.0	0.5
Irritability	All	1378	287	20.8	18.7	23.1	710	104	14.6	12.1	17.5
	Grade 3	1378	3	0.2	0.0	0.6	710	2	0.3	0.0	1.0
	Related	1378	144	10.4	8.9	12.2	710	54	7.6	5.8	9.8
	Grade 3 Related	1378	2	0.1	0.0	0.5	710	0	0.0	0.0	0.5
Loss of appetite	All	1378	106	7.7	6.3	9.2	710	45	6.3	4.7	8.4
	Grade 3	1378	0	0.0	0.0	0.3	710	1	0.1	0.0	0.8
	Related	1378	44	3.2	2.3	4.3	710	20	2.8	1.7	4.3
	Grade 3 Related	1378	0	0.0	0.0	0.3	710	0	0.0	0.0	0.5
Temperature	All	1378	429	31.1	28.7	33.7	710	111	15.6	13.0	18.5

		RTS,S/AS01					MenC vaccine				
					95 % CI					95 % CI	
Symptom	**Type**	**N**	**n**	**%**	**LL**	**UL**	**N**	**n**	**%**	**LL**	**UL**
(Axillary) (°C)	>39.0	1378	13	0.9	0.5	1.6	710	3	0.4	0.1	1.2
	Related	1378	280	20.3	18.2	22.5	710	57	8.0	6.1	10.3
	>39.0 Related	1378	11	0.8	0.4	1.4	710	3	0.4	0.1	1.2
Overall/dose											
Drowsiness	All	4252	423	9.9	9.1	10.9	2169	164	7.6	6.5	8.8
	Grade 3	4252	2	0.0	0.0	0.2	2169	2	0.1	0.0	0.3
	Related	4252	211	5.0	4.3	5.7	2169	60	2.8	2.1	3.5
	Grade 3 Related	4252	1	0.0	0.0	0.1	2169	0	0.0	0.0	0.2
Irritability	All	4252	946	22.2	21.0	23.5	2169	384	17.7	16.1	19.4
	Grade 3	4252	20	0.5	0.3	0.7	2169	5	0.2	0.1	0.5
	Related	4252	545	12.8	11.8	13.9	2169	195	9.0	7.8	10.3
	Grade 3 Related	4252	13	0.3	0.2	0.5	2169	3	0.1	0.0	0.4
Loss of appetite	All	4252	335	7.9	7.1	8.7	2169	140	6.5	5.5	7.6
	Grade 3	4252	2	0.0	0.0	0.2	2169	1	0.0	0.0	0.3
	Related	4252	171	4.0	3.5	4.7	2169	52	2.4	1.8	3.1
	Grade 3 Related	4252	2	0.0	0.0	0.2	2169	0	0.0	0.0	0.2
Temperature	All	4252	1299	30.6	29.2	32.0	2169	457	21.1	19.4	22.8
(Axillary) (°C)	>39.0	4252	27	0.6	0.4	0.9	2169	11	0.5	0.3	0.9
	Related	4252	884	20.8	19.6	22.0	2169	273	12.6	11.2	14.1
	>39.0 Related	4252	17	0.4	0.2	0.6	2169	7	0.3	0.1	0.7
Overall/subject											
Drowsiness	All	1462	285	19.5	17.5	21.6	738	121	16.4	13.8	19.3
	Grade 3	1462	2	0.1	0.0	0.5	738	2	0.3	0.0	1.0
	Related	1462	144	9.8	8.4	11.5	738	51	6.9	5.2	9.0
	Grade 3 Related	1462	1	0.1	0.0	0.4	738	0	0.0	0.0	0.5

		RTS,S/AS01					MenC vaccine				
				95 % CI					95 % CI		
Symptom	**Type**	**N**	**n**	**%**	**LL**	**UL**	**N**	**n**	**%**	**LL**	**UL**
Irritability	All	1462	574	39.3	36.7	41.8	738	244	33.1	29.7	36.6
	Grade 3	1462	17	1.2	0.7	1.9	738	5	0.7	0.2	1.6
	Related	1462	363	24.8	22.6	27.1	738	139	18.8	16.1	21.8
	Grade 3 Related	1462	11	0.8	0.4	1.3	738	3	0.4	0.1	1.2
Loss of appetite	All	1462	243	16.6	14.7	18.6	738	104	14.1	11.7	16.8
	Grade 3	1462	2	0.1	0.0	0.5	738	1	0.1	0.0	0.8
	Related	1462	128	8.8	7.4	10.3	738	41	5.6	4.0	7.5
	Grade 3 Related	1462	2	0.1	0.0	0.5	738	0	0.0	0.0	0.5
Temperature	All	1462	839	57.4	54.8	59.9	738	331	44.9	41.2	48.5
(Axillary) (°C)	>39.0	1462	26	1.8	1.2	2.6	738	11	1.5	0.7	2.7
	Related	1462	598	40.9	38.4	43.5	738	209	28.3	25.1	31.7
	>39.0 Related	1462	16	1.1	0.6	1.8	738	7	0.9	0.4	1.9

MenC = Meningococcal C conjugate vaccine

N= number of administered doses. AEs during the first 30 days post-vaccination were recorded and analyzed only in the first 200 infants enrolled at each center.

n/%= number/percentage of doses followed by at least one type of symptom

95%CI = Exact 95% confidence interval; LL = lower limit, UL = upper limit

Table S13. Incidence of solicited local adverse events in infants aged 6-12 weeks at enrollment reported during the 7-day post-vaccination period following each dose and overall (ITT population)

			RTS,S/AS01					MenC vaccine				
						95 % CI					95 % CI	
Symptom	**Product**	**Type**	**N**	**n**	**%**	**LL**	**UL**	**N**	**n**	**%**	**LL**	**UL**
Dose 1												
Pain	Meningococcal C conjugate vaccine	All						738	110	14.9	12.4	17.7
		Grade 3						738	1	0.1	0.0	0.8
	RTS,S/AS01	All	1462	278	19.0	17.0	21.1					
		Grade 3	1462	3	0.2	0.0	0.6					
	DTPwHepB/Hib	All	1462	395	27.0	24.8	29.4	738	201	27.2	24.1	30.6
		Grade 3	1462	9	0.6	0.3	1.2	738	7	0.9	0.4	1.9
Redness	Meningococcal C conjugate vaccine	All						738	46	6.2	4.6	8.2
		>20.0 mm						738	0	0.0	0.0	0.5
	RTS,S/AS01	All	1462	100	6.8	5.6	8.3					
		>20.0 mm	1462	0	0.0	0.0	0.3					
	DTPwHepB/Hib	All	1462	144	9.8	8.4	11.5	738	78	10.6	8.4	13.0
		>20.0 mm	1462	3	0.2	0.0	0.6	738	3	0.4	0.1	1.2
Swelling	Meningococcal C conjugate vaccine	All						738	39	5.3	3.8	7.2
		>20.0 mm						738	2	0.3	0.0	1.0
	RTS,S/AS01	All	1462	89	6.1	4.9	7.4					
		>20.0 mm	1462	3	0.2	0.0	0.6					
	DTPwHepB/Hib	All	1462	209	14.3	12.5	16.2	738	120	16.3	13.7	19.1
		>20.0 mm	1462	26	1.8	1.2	2.6	738	28	3.8	2.5	5.4
Dose 2												
Pain	Meningococcal C conjugate vaccine	All						719	103	14.3	11.8	17.1
		Grade 3						719	0	0.0	0.0	0.5
	RTS,S/AS01	All	1405	236	16.8	14.9	18.9					

			RTS,S/AS01					MenC vaccine				
						95 % CI					95 % CI	
Symptom	**Product**	**Type**	**N**	**n**	**%**	**LL**	**UL**	**N**	**n**	**%**	**LL**	**UL**
		Grade 3	1405	3	0.2	0.0	0.6					
	DTPwHepB/Hib	All	1411	356	25.2	23.0	27.6	721	165	22.9	19.9	26.1
		Grade 3	1411	3	0.2	0.0	0.6	721	3	0.4	0.1	1.2
Redness	Meningococcal C conjugate vaccine	All						719	57	7.9	6.1	10.1
		>20.0 mm						719	0	0.0	0.0	0.5
	RTS,S/AS01	All	1405	71	5.1	4.0	6.3					
		>20.0 mm	1405	0	0.0	0.0	0.3					
	DTPwHepB/Hib	All	1411	111	7.9	6.5	9.4	721	78	10.8	8.6	13.3
		>20.0 mm	1411	3	0.2	0.0	0.6	721	1	0.1	0.0	0.8
Swelling	Meningococcal C conjugate vaccine	All						719	44	6.1	4.5	8.1
		>20.0 mm						719	0	0.0	0.0	0.5
	RTS,S/AS01	All	1405	85	6.0	4.9	7.4					
		>20.0 mm	1405	2	0.1	0.0	0.5					
	DTPwHepB/Hib	All	1411	220	15.6	13.7	17.6	721	121	16.8	14.1	19.7
		>20.0 mm	1411	27	1.9	1.3	2.8	721	17	2.4	1.4	3.7
Dose 3												
Pain	Meningococcal C conjugate vaccine	All						708	82	11.6	9.3	14.2
		Grade 3						708	0	0.0	0.0	0.5
	RTS,S/AS01	All	1370	220	16.1	14.2	18.1					
		Grade 3	1370	5	0.4	0.1	0.8					
	DTPwHepB/Hib	All	1378	327	23.7	21.5	26.1	710	138	19.4	16.6	22.5
		Grade 3	1378	6	0.4	0.2	0.9	710	2	0.3	0.0	1.0
Redness	Meningococcal C conjugate vaccine	All						708	34	4.8	3.3	6.6
		>20.0 mm						708	0	0.0	0.0	0.5
	RTS,S/AS01	All	1370	73	5.3	4.2	6.7					

			RTS,S/AS01					MenC vaccine				
						95 % CI					95 % CI	
Symptom	**Product**	**Type**	**N**	**n**	**%**	**LL**	**UL**	**N**	**n**	**%**	**LL**	**UL**
		>20.0 mm	1370	1	0.1	0.0	0.4					
	DTPwHepB/Hib	All	1378	96	7.0	5.7	8.4	710	52	7.3	5.5	9.5
		>20.0 mm	1378	0	0.0	0.0	0.3	710	1	0.1	0.0	0.8
Swelling	Meningococcal C conjugate vaccine	All						708	41	5.8	4.2	7.8
		>20.0 mm						708	0	0.0	0.0	0.5
	RTS,S/AS01	All	1370	81	5.9	4.7	7.3					
		>20.0 mm	1370	1	0.1	0.0	0.4					
	DTPwHepB/Hib	All	1378	172	12.5	10.8	14.3	710	102	14.4	11.9	17.2
		>20.0 mm	1378	9	0.7	0.3	1.2	710	12	1.7	0.9	2.9
		Overall/dose										
Pain	Meningococcal C conjugate vaccine	All						2165	295	13.6	12.2	15.1
		Grade 3						2165	1	0.0	0.0	0.3
	RTS,S/AS01	All	4237	734	17.3	16.2	18.5					
		Grade 3	4237	11	0.3	0.1	0.5					
	DTPwHepB/Hib	All	4251	1078	25.4	24.1	26.7	2169	504	23.2	21.5	25.1
		Grade 3	4251	18	0.4	0.3	0.7	2169	12	0.6	0.3	1.0
Redness	Meningococcal C conjugate vaccine	All						2165	137	6.3	5.3	7.4
		>20.0 mm						2165	0	0.0	0.0	0.2
	RTS,S/AS01	All	4237	244	5.8	5.1	6.5					
		>20.0 mm	4237	1	0.0	0.0	0.1					
	DTPwHepB/Hib	All	4251	351	8.3	7.4	9.1	2169	208	9.6	8.4	10.9
		>20.0 mm	4251	6	0.1	0.1	0.3	2169	5	0.2	0.1	0.5
Swelling	Meningococcal C conjugate vaccine	All						2165	124	5.7	4.8	6.8
		>20.0 mm						2165	2	0.1	0.0	0.3
	RTS,S/AS01	All	4237	255	6.0	5.3	6.8					

			RTS,S/AS01					MenC vaccine				
						95 % CI					95 % CI	
Symptom	**Product**	**Type**	**N**	**n**	**%**	**LL**	**UL**	**N**	**n**	**%**	**LL**	**UL**
		>20.0 mm	4237	6	0.1	0.1	0.3					
	DTPwHepB/Hib	All	4251	601	14.1	13.1	15.2	2169	343	15.8	14.3	17.4
		>20.0 mm	4251	62	1.5	1.1	1.9	2169	57	2.6	2.0	3.4

For each dose:

N= number of infants with at least one administered dose

n/%= number/percentage of infants reporting the symptom at least once

For Overall/dose:

N= number of administered doses

n/%= number/percentage of doses followed by at least one type of symptom

95%CI = Exact 95% confidence interval; LL = lower limit, UL = upper limit

Table S14. Percentage of infants with at least one cutaneous and/or mucosal change within the 30-day post-vaccination period (ITT population)

		RTS,S/AS01 N = 1389				MenC N = 704				Total N = 2093			
				95% CI				95% CI				95% CI	
Characteristics	**Categories**	**n**	**%**	**LL**	**UL**	**n**	**%**	**LL**	**UL**	**n**	**%**	**LL**	**UL**
Cutaneous and/or mucosal change	At least one	414	29.8	27.4	32.3	207	29.4	26.1	32.9	621	29.7	27.7	31.7
Cutaneous and mucosal change	At least one	33	2.4	1.6	3.3	22	3.1	2.0	4.7	55	2.6	2.0	3.4
Cutaneous only change	At least one	262	18.9	16.8	21.0	133	18.9	16.1	22.0	395	18.9	17.2	20.6
Mucosal only change	At least one	159	11.4	9.8	13.2	79	11.2	9.0	13.8	238	11.4	10.0	12.8
Cutaneous change focused on the nappy/diaper area	At least one	29	2.1	1.4	3.0	20	2.8	1.7	4.4	49	2.3	1.7	3.1

MenC = Meningococcal C conjugate vaccine

N = number of infants for which data on cutaneous and/or mucosal change were collected

n = number of infants for which data on cutaneous and/or mucosal change were collected in a given category

% = n/Number of infants for which data on cutaneous and/or mucosal change were collected with available results x 100

95%CI = Exact 95% confidence interval; LL = lower limit, UL = upper limit

Table S15. Clinical features of infants with severe malaria aged 6-12 weeks at enrollment (Per-protocol population)

		All episodes primary case definition N = 113	
Frequency of markers of disease severity		**n**	**%**
Prostration		21	18.6
Respiratory distress		11	9.7
Blantyre Coma Score ≤2		8	7.1
Seizures 2 or more		42	37.2
Hypoglycemia <2.2 mmol/L		3	2.7
Acidosis BE ≤-10 mmol/L		46	40.7
Lactate ≥5 mmol/L		36	31.9
Anemia <5 g/dL		32	28.3
Number of disease markers	1	67	59.3
	2	28	24.8
	3	7	6.2
	4	5	4.4
	5	2	1.8
	6	3	2.7
	7	1	0.9

N = number of events meeting the primary case definition

n = number of events meeting the primary case definition in a given category

% = n / Number of events meeting the primary case definition with available results

Table S16. Frequency of co-morbidities in infants 6-12 weeks of age at enrollment with severe malaria (Per-protocol population)

	All episodes, secondary case definition N = 125	
	n	%
No co-morbidity detected and algorithm completed	113	90.4
Pneumonia	2	1.6
Meningitis	1	0.8
Bacteremia*	5	4.0
Gastroenteritis	0	-
Co-morbidity not excluded (Algorithm not completed)	5	4.0

N = number of events meeting the secondary case definition

n = number of events meeting the case definition in a given category

% = n / Number of events meeting the secondary case definition with available results

* Salmonella 3 (2.4%), other pathogens 2 (1.6%)

The total percentage exceeds 100% because a child could have more than one co-morbidity during a severe malaria episode.

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