Results Out of 284 participants data for 245 were analysed (Group 1: 27; Group 2: 107; Group 3: 111). Majority were aged 25–29 years and over 60% had primary/lesser education. There were 39 (Group 1: 5; Group 2: 16; Group 3: 18) VFs with a total VF incidence of 8.12 [95% CI (5.96, 11.17)] per 1000 Person months of observation (PMOs). Group 2 had the lowest VF incidence. Baseline CD4 <349 cells/mm³ and initiation/use of TDF/3TC/EFV were associated with virologic failure (VF).

Conclusion Women at risk of VF based on the identified risk factors should be identified and targeted with appropriate intervention. Further studies are needed to verify and understand the mechanisms of association between VF and TDF/3TC/EFV which is a WHO-recommended first-line ART regimen.

PO 8596 ENHANCING LABORATORY DIAGNOSIS OF MYCOBACTERIUM TUBERCULOSIS IN SAMPLES FROM CHILDREN IN THE GAMBIA

1Abigail Ayorinde*, 1Edward G Coker, 1Alieu Mendy, 1Fatoumatta Cole, 1Abdou K Sillah, 1Francis S Mendy, 1Ishukahpu Egere, 1,2Beate Kampmann, 1,3Lucia B Tientcheu.

1Medical Research Unit The Gambia at London School of Hygiene and Tropical Medicine, The Gambia; 2Imperial College, London, UK; 3Department of Biochemistry, Faculty of Science University of Yaoundé 1, Yaounde, Cameroon

Background Routine laboratory diagnostic methods for M. Tuberculosis complex (MTBC) in induced sputum samples such as smear microscopy, GeneXpert and liquid Mycobacteria growth indicator tube (MGIT) culture are often negative due to the paucibacillary nature of childhood tuberculosis. We hypothesise that prolonged incubation beyond routine culture time could potentially improve MTBC detection in specimens.

Methods Out of over 1000 induced sputum samples collected during our childhood TB contact tracing research programme, we randomly selected 102 MTBC-negative MGIT cultures that had either been reported as contaminated (n=35) or negative (n=67) and further incubated these at 37°C for the duration of one month. Ziehl-Neelsen microscopy, MPT64 Antigen secretion and GeneXpert tests were repeated on all samples to detect MTBC. Bacterial DNA was extracted by CTAB method and genotyped using spoligotyping analysis.

Results Of the 1160 routinely collected induced-sputum samples, 12 (1%) were smear-positives; 41 (3.5%) Xpert-positives and 51 (4.4%) MGIT culture MTBC-positives. The remaining MGIT cultures were flagged as contaminated 393 (33.9%) or MTBC-negative 644 (55.5%). After prolonged incubation and retesting of the randomly selected ones, 26/102 (25.5%) were now microscopy-positives, 2/55 (3.63%) were GeneXpert-positives, 8/102 (7.8%) MPT64-Antigen-positive, and 38/102 (37.2%) had readable spoligotyping patterns. The predominant lineages were Mtb-Euro-American 16 (42.1%), Mtb-Indo-Oceanic 11 (28.9%) and M. africanum West African type-2 8 (21%).

Conclusion Prolonged incubation of routinely MTBC-negative induced-sputum cultures yielded positive results upon retesting, highlighting the low sensitivity of routine diagnosis tools on pauci-bacillary paediatric samples. Spoligotyping was more sensitive to detect MTBC compared to GeneXpert. However, prolonged incubation will cause diagnostic delays and thus better strategies are needed to improve timely childhood TB diagnosis.
Pfhrp-2 gene deletion is implicated in limiting RDT sensitivity. Studies evaluating Pfhrp-2 and Pfhrp-3 deletion and the amino acid sequence diversity has not been investigated in Nigeria. We therefore hypothesised that malaria parasites in Nigeria are lacking Pfhrp-2/Pfhrp-3 genes with variable amino acid repeats sequences.

Methods The study was part of a prospective cohort study evaluating RDTs performance. We pooled 66 samples comprising false negatives (n=31) and true positives (n=35) to elucidate Pfhrp-2/Pfhrp-3 gene deletion, RDT cross-reactivity with Pfhrp-3 antigen and amino acid sequence diversity. The 18SrRNA, msp1, msp2 and glurp genes were amplified to establish active Plasmodium falciparum infection and the exon-2 regions of Pfhrp-2 and Pfhrp-3 genes were amplified to determine the presence or absence of Pfhrp-2 and Pfhrp-3 genes. Isolates with conserved Pfhrp-2/Pfhrp-3 were sequenced.

Results All 66 samples were positive for 18SrRNA, msp1, msp2 and glurp, indicating active P. falciparum infection. However, 16.7% and 6.0% of the samples were lacking Pfhrp-2 and Pfhrp-3 genes. Of the false negative samples, 25.8% and 12.9% has Pfhrp-2 and Pfhrp-3 deletions. Three Pfhrp-3 conserved antigens cross reacted to give RDT positive results. An extensive diversity in the amino acid sequence was observed.

Conclusion Plasmodium falciparum parasites in Nigeria lack Pfhrp-2 and Pfhrp-3 genes. However, the proportion of deletions is low compared to reports from other malaria-endemic regions. In addition, a high amino acid tandem repeat was observed. A combination of pLDH and Pfhrp-2 based RDTs is recommended for accurate malaria diagnosis.