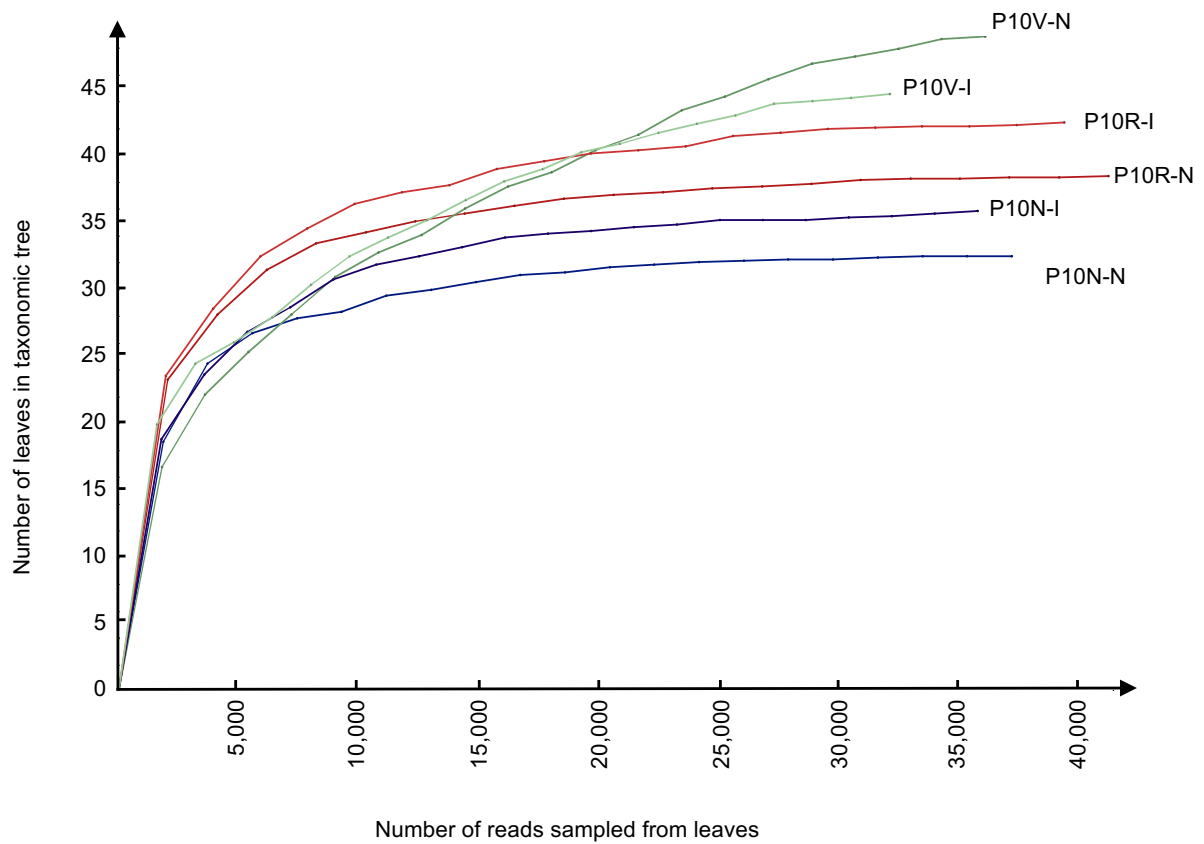


**Supplementary Figure 1. Timeline diagrams for healthy preterms (P106, P116, P103).**

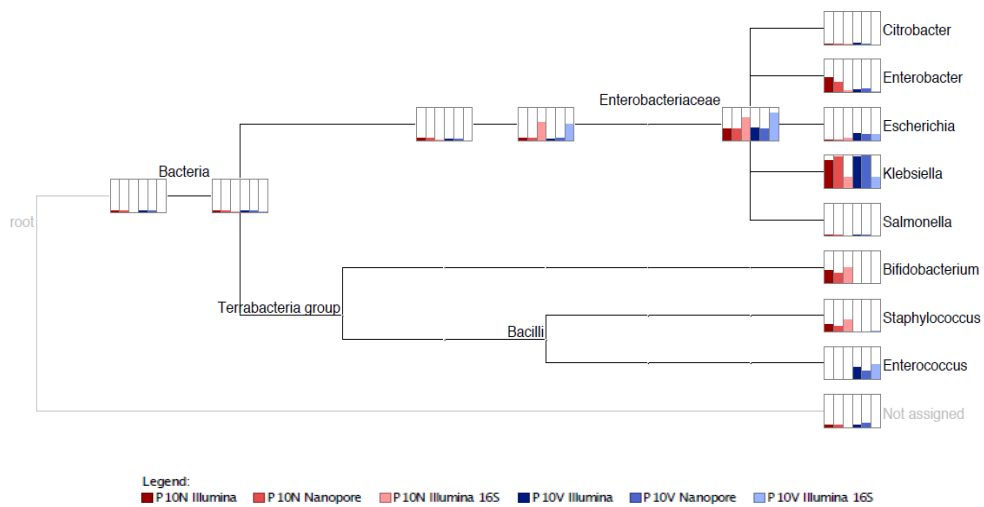
Timeline diagrams indicating time points of faecal sample collection, duration of antibiotic, probiotic treatment, and relevant clinical observations. The timeline diagrams are divided in weeks and dots represent days within the scale. Blue dots indicate days of probiotic treatment, red dots antibiotic treatment, and black squares time points for sample collection. (a) timeline diagram for preterm P106, (b) timeline diagram for preterm P116 and (c) timeline diagram for preterm P103.





**Supplementary Figure 3. Rarefaction curves comparing MinION and Illumina HiSeq 2500 for preterm P10.**

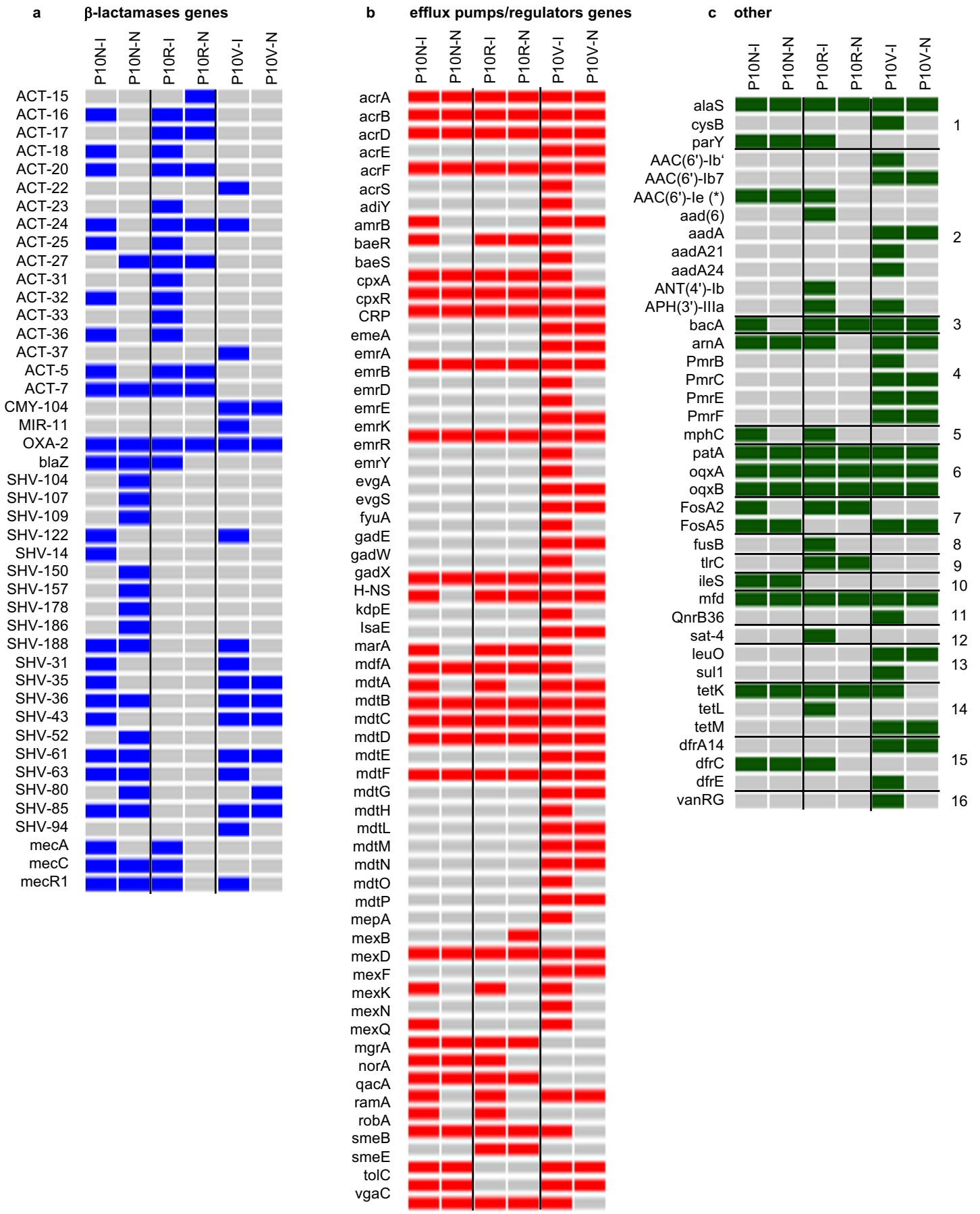
Rarefaction curves representing number of species (leaves) detected in the taxonomic tree vs number of reads sampled. Three samples from preterm P10 were analysed: samples (P10N-N, P10R-N and P10V-N) were sequenced with MinION technology, while samples (P10N-I, P10R-I and P10V-I) were sequenced with Illumina technology.



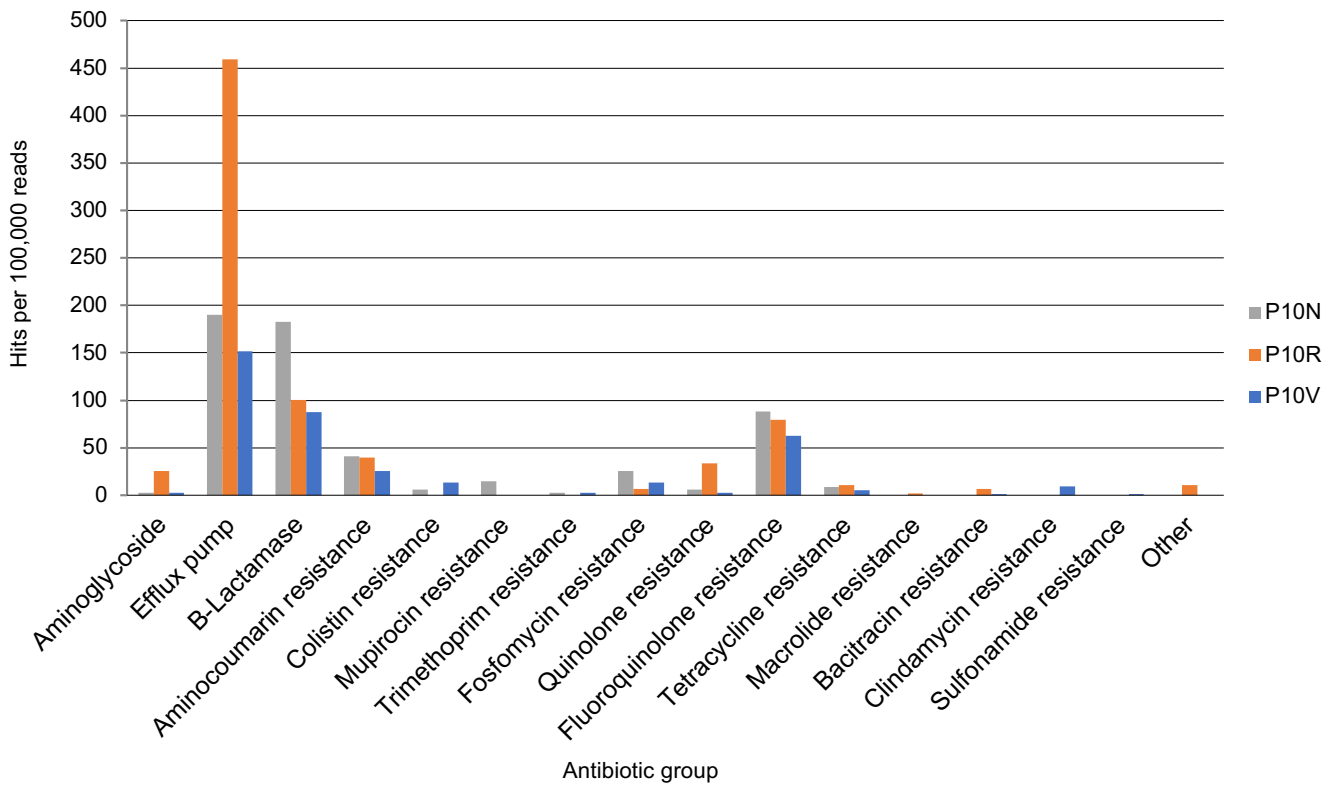
**Supplementary Figure 4. Megan taxonomic tree comparing assignments obtained by Illumina HiSeq 2500 WGS, ONT MinION, and Illumina 16S rRNA gene sequencing.**

Megan taxonomic tree showing bacteria taxa identified and their corresponding abundances. The height of the bars represents the number of reads assigned for each species taxa. Samples highlighted in red belong to sample P10N sequenced using Illumina HiSeq 2500 WGS, ONT MinION, and Illumina 16S rRNA gene sequencing. Samples highlighted in blue belong to sample P10V sequenced using Illumina HiSeq 2500 WGS, ONT MinION, and Illumina 16S rRNA gene sequencing.

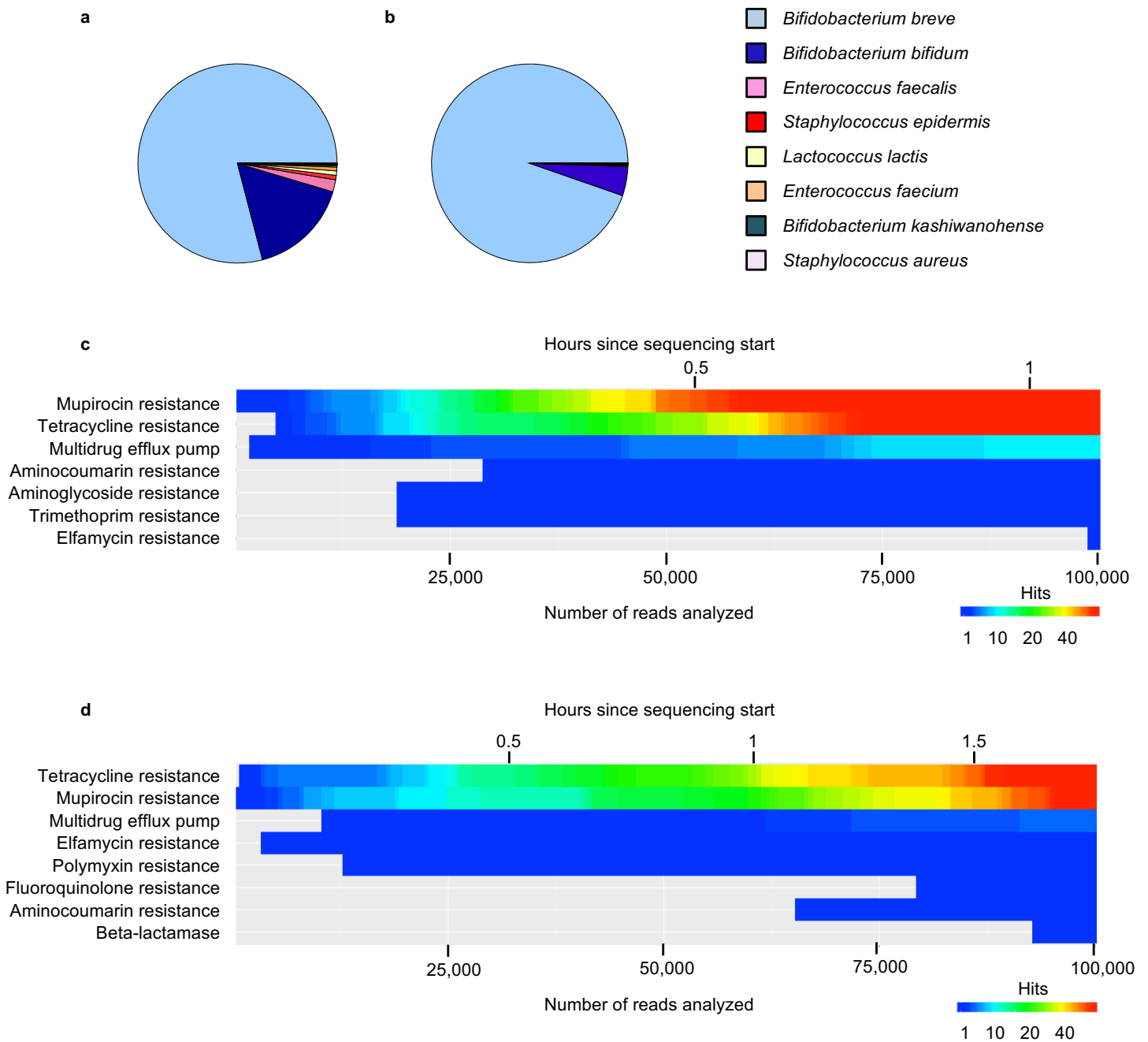




**Supplementary Figure 5. Heat map displaying presence or absence of AMR hits found in preterm P10 using MinION and Illumina HiSeq 2500.** Three samples of preterm P10 were analysed and sequenced using MinION (P10N-I, P10R-I and P10V-I), and Illumina technology (P10N-N, P10R-N and P10V-N). (a) Heat map representing  $\beta$ -lactamases hits found in samples P10N, P10R and P10V. (b) Heat map highlighting efflux pumps inhibitors or regulators genes. (c) Heat map showing (1) aminocoumarin resistance genes (2) aminoglycosides resistance (3) bacitracin resistance (4) colistin resistance (5) erythromycin resistance (6) fluoroquinolone resistance (7) fosfomycin resistance (8) fusidic acid resistance (9) macrolide resistance (10) mucopirocin resistance (11) quinolone resistance (12) streptothricin resistance (13) sulphonamide resistance (14) tetracycline resistance (15) trimethoprim resistance (16) vancomycin resistance.



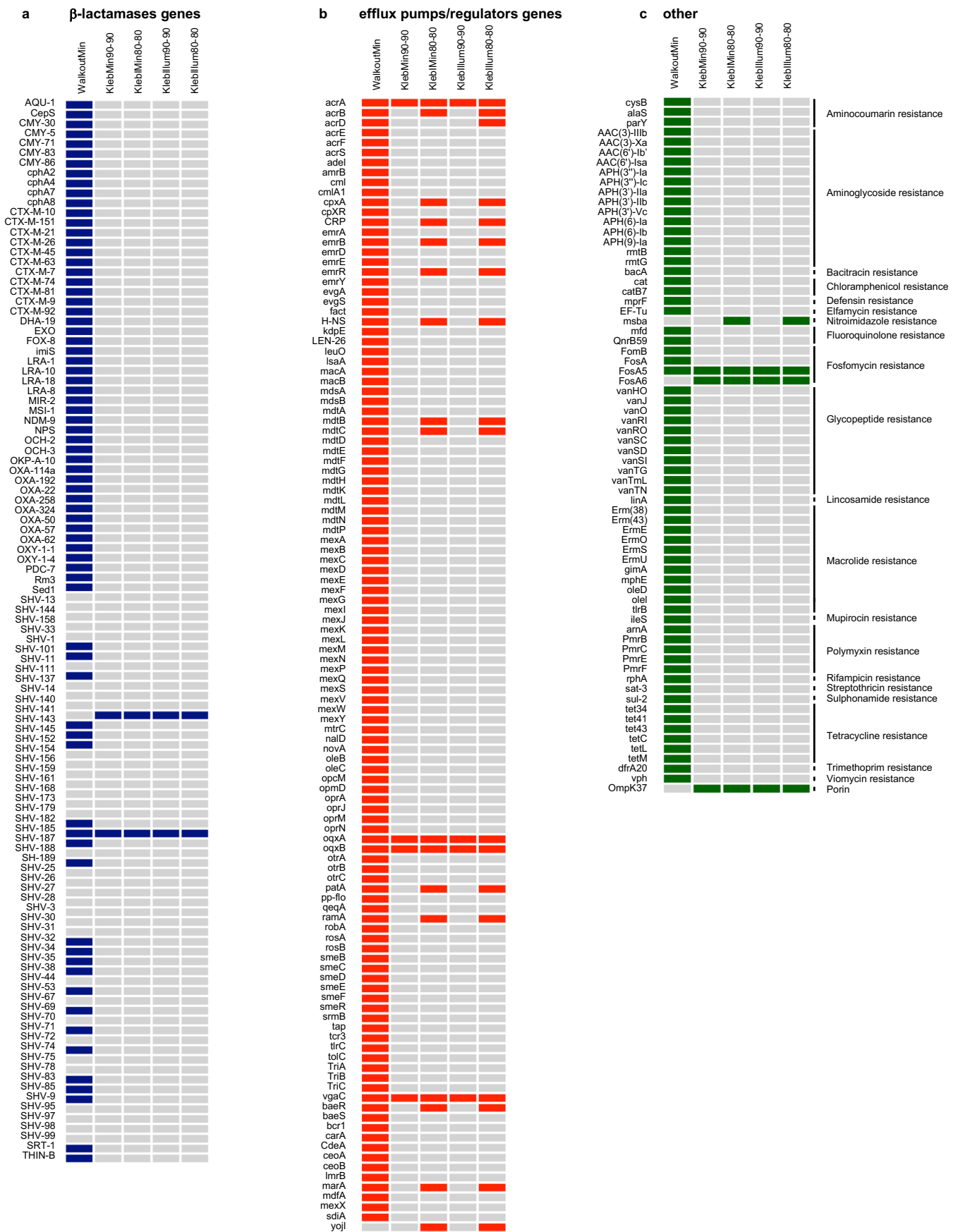
**Supplementary Figure 6. Comparison of AMR hits found in preterm P10.** Height of bars indicates number of CARD database hits per 100,000 reads to each of 16 antibiotic resistance groups for sample P10N (grey, 13 days after birth), P10R (orange, 28 days after birth), P10V (blue, 64 days after birth).



**Supplementary Figure 7. Rapid diagnostic of healthy preterms P106 and P116 receiving probiotic supplementation.**

(a), (b) Taxonomic profiles obtained using MinION technology for preterms P106 and P116, respectively. Figure legend comprises the 8 most abundant taxonomic taxa obtained. Further information on all the bacteria taxa and the number of reads obtained can be found in **Supplementary Table 3**.

(c), (d) Heat maps displaying number of CARD database hits detected among the most common groups of antibiotic resistance genes found in preterms P106 and P116. Top and lower panel indicate the hours since sequencing started and the number of reads analyzed, respectively within this timeframe. Further information on all the AMR genes obtained can be found in **Supplementary Table 5**



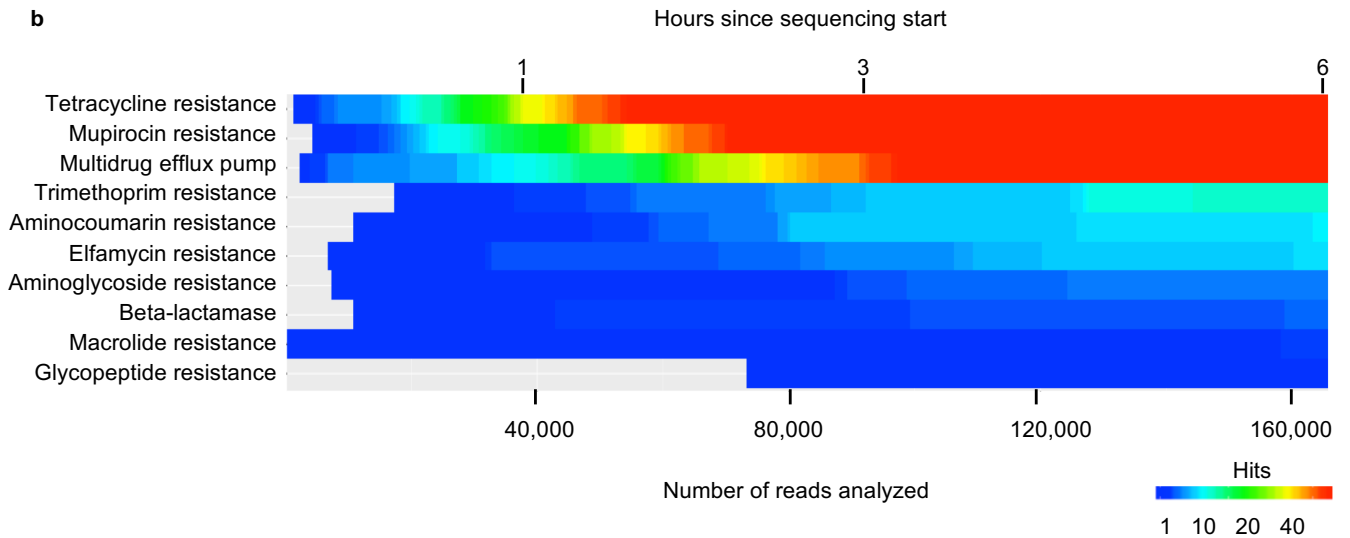
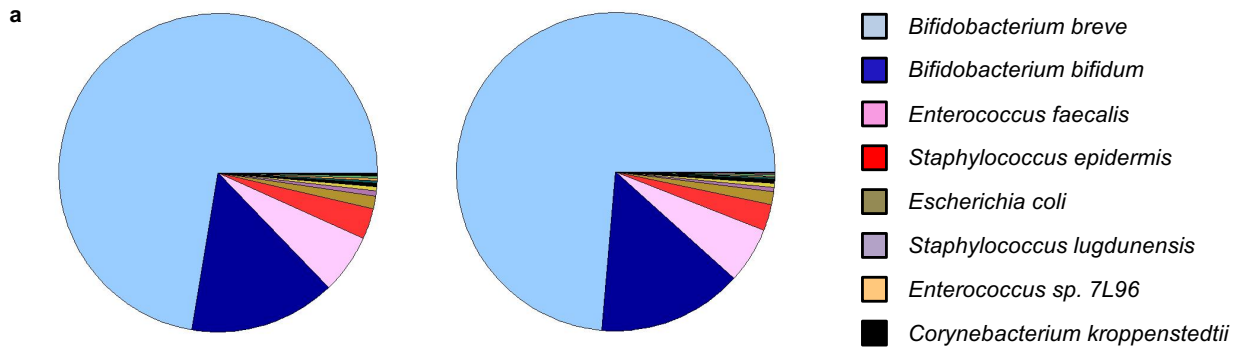
**Supplementary Figure 8. AMR genes associated to *Klebsiella pneumoniae* from a metagenomic sample compared to those found in a *Klebsiella pneumoniae* isolate from the same sample.** Heat maps displaying AMR genes associated to *Klebsiella pneumoniae* from faecal sample P8E and AMR genes detected from a *Klebsiella pneumoniae* isolate from the same sample. Sequencing of the metagenomic sample was performed for only 6h using MiniION and NanoOK RT tool (highlighted as 'Walkout'). Sequencing of the *Klebsiella pneumoniae* isolate was performed using both MiniION (marked in the heat map as 'KlebMiniON') and Illumina HiSeq (highlighted as 'KlebIllu'). Two different filtering criteria were used to analyse the data from the *Klebsiella pneumoniae* isolate: i) a 'strict' one; identity 90% and coverage 90% (marked as 'KlebMini90-90' and 'KlebIllu90-90') and ii) a 'relax' one; identity 80% and coverage 80% (marked as 'KlebMini80-80' and 'KlebIllu80-80'). AMR genes detected were grouped according to antibiotic mode of actions: (a) Heat map comparing *Klebsiella* specific β-lactamases (b) Heat map comparing *Klebsiella* specific efflux pumps or regulator genes (c) Heat map showing other *Klebsiella* specific AMR genes.

a

<b>Antibiotic</b>	<b>MIC mg/L</b>	<b>Eucast (mg/L)</b>
Gentamicin	3.12	2
Benzylpenicillim	780	ND
Amoxicillin	3900	>512
Metronidazole	1250	ND
Vancomycin	1562	ND
Meropenem	6.25	0.125
Cefotaxime	0.19	0.25

**Supplementary Figure 9. Broth microdilution test for *Klebsiella pneumoniae* isolate from baby P8 (sample P8E).**

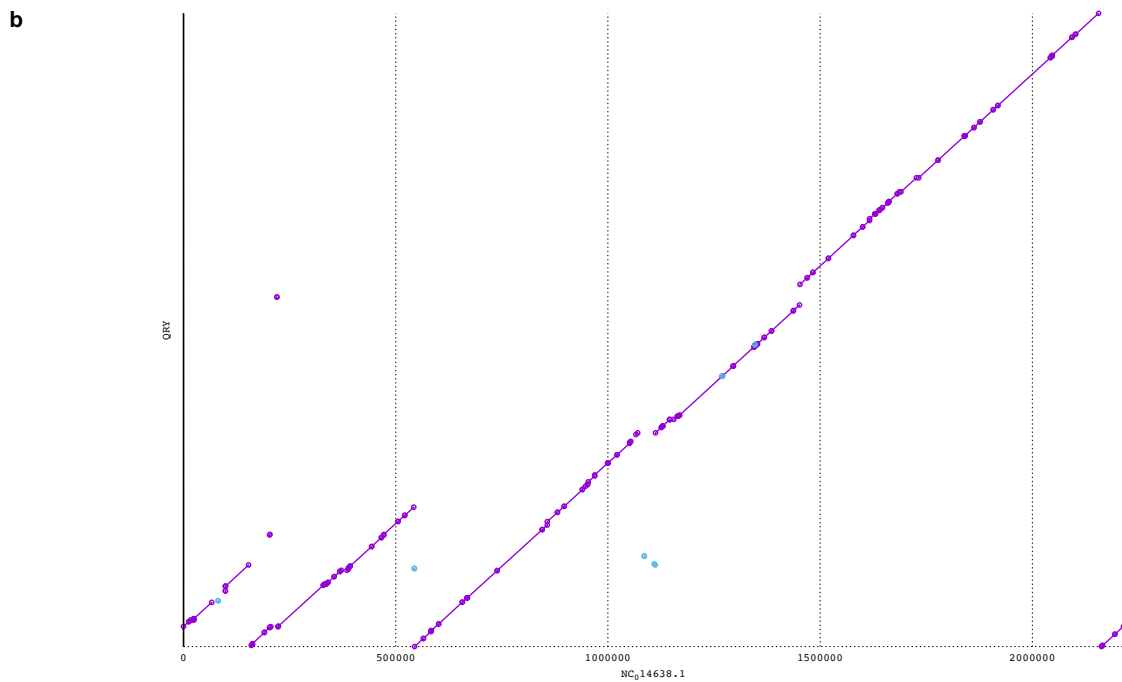
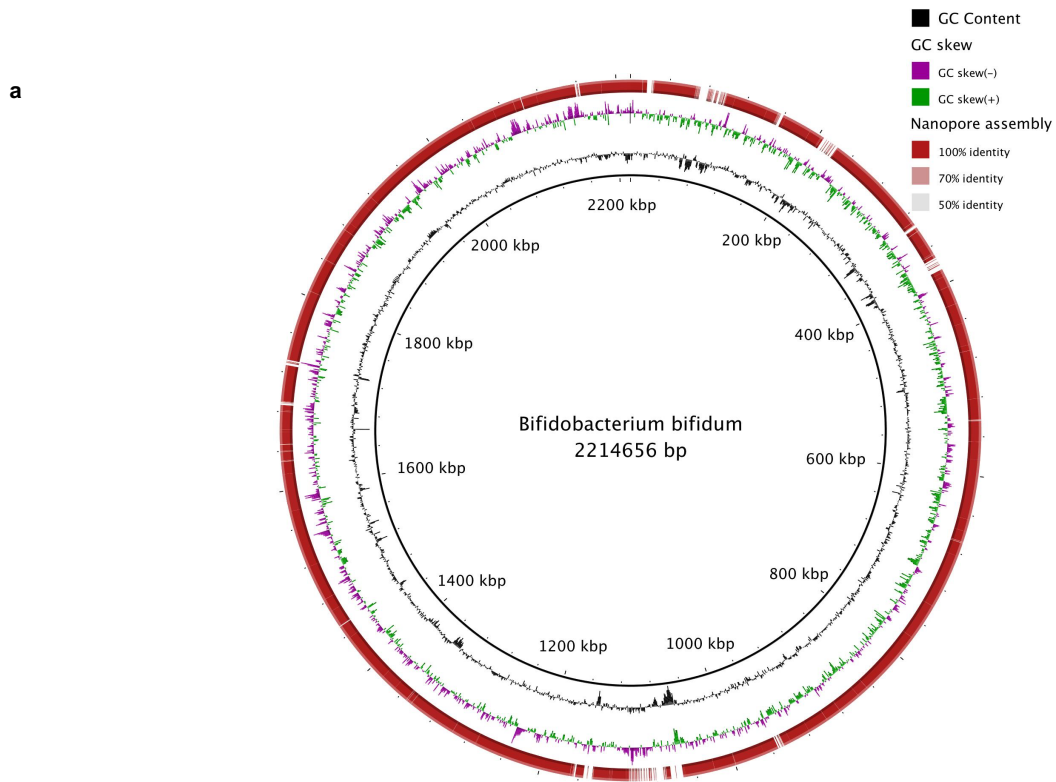
(a) Minimal Inhibitory Concentrations (MIC) for *Klebsiella pneumoniae* isolate from sample P8E. Antibiotics used are indicated in the table. Table includes breakpoint values from European Committee on Antimicrobial Susceptibility Testing (Eucast) as a reference. ND = no data available.



**Supplementary Figure 10. Rapid diagnostic of healthy preterm P103 using SQK-RAD004.**

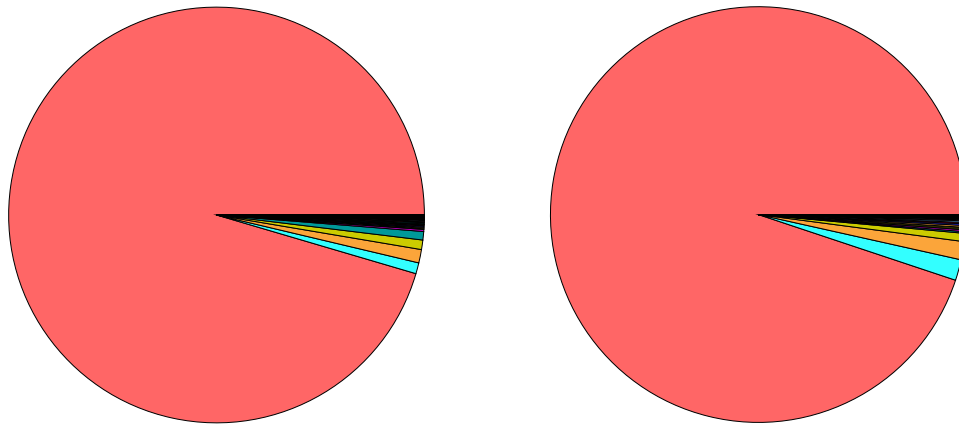
(a) Taxonomic profiles detected for preterm P103. Figure legend comprises the 8 most abundant taxonomic taxa obtained. Further information on all the bacteria taxa and the number of reads obtained can be found in **Supplementary Table 3**.

(b) Heat map displaying number of CARD database hits detected among the most common groups of antibiotic resistance genes found in preterm P103. Top and lower panel indicate the hours since sequencing started and the number of reads analyzed, respectively within this timeframe. Further information on all the AMR genes obtained can be found in **Supplementary Table 5**.



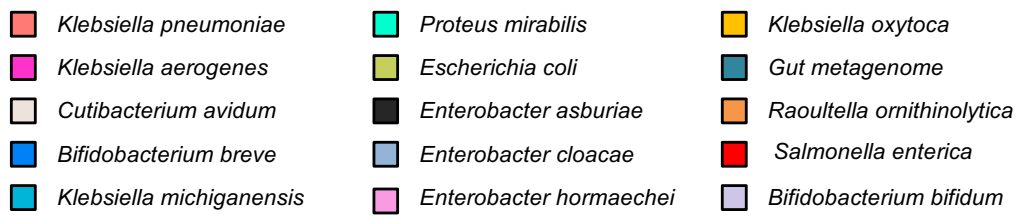
**Supplementary Figure 11. Comparison between MinION assembly of *Bifidobacterium bifidum* and the reference strain *Bifidobacterium bifidum* PRL2010**

(a) BRIG plot showing BLAST alignment identity between *Bifidobacterium bifidum* assembled out of sample P103M and the reference strain NC\_014638.1. Also shown is reference GC content and skew. (b) Dot plots showing similarity between assembly and reference.



Illumina HiSeq 2500

MinION Nanopore



**Supplementary Figure 12. Comparison of taxonomic profiles using Illumina HiSeq 2500 and MinION technology for preterm P8.**

Taxonomic profiles obtained using Illumina HiSeq 2500 and MinION technology for preterm P8. Figure legend comprises the 15 most abundant taxonomic taxa obtained.