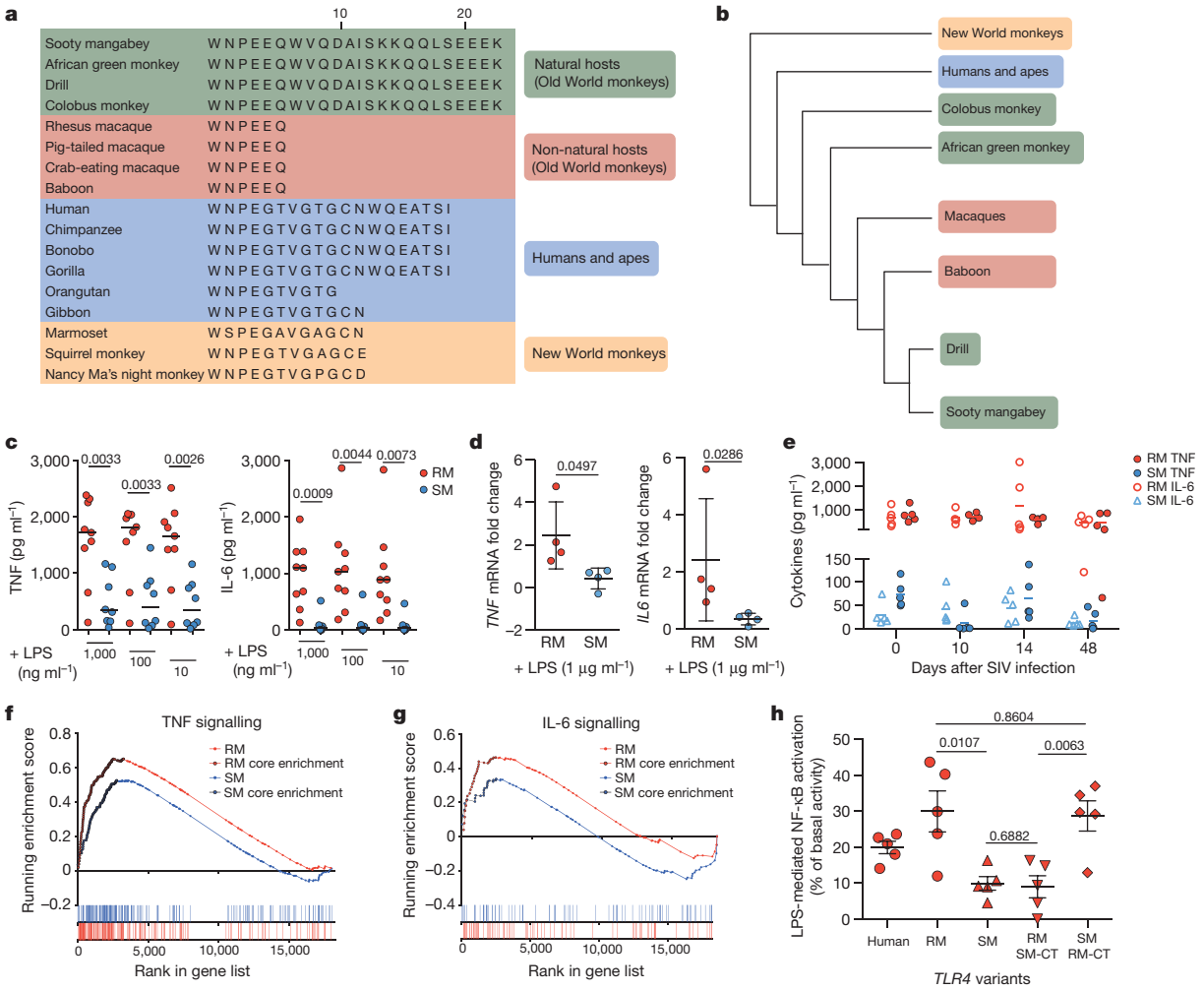

Figure 3 | The TLR-4 C terminus is distinctive in natural SIV hosts. **a**, Alignment of C-terminal TLR-4 protein sequences from different primate species (starting at human TLR-4 amino acid position 821). **b**, Primate phylogenetic tree with colour-coding according to the TLR-4 C terminus as indicated in **a**. Phylogeny appears as in ref. 14. **c**, Cytokine release from blood of rhesus macaques ($n = 9$ biologically independent samples) and sooty mangabeys ($n = 8$ biologically independent samples) after LPS stimulation as measured by cytometric bead array. **d**, mRNA expression in whole blood after LPS stimulation quantified by quantitative PCR (qPCR). $n = 4$ biologically independent samples for each species. **e**, TNF and IL-6 cytokine release from blood of rhesus macaques and sooty mangabeys over the course of SIV infection. $n = 5$ biologically independent samples for each species. Data are mean ± s.d. (**c**–**e**), unpaired two-sided Student's *t*-test, *P* values are indicated (**c**, **d**). **f**, Gene set enrichment analysis of LPS-stimulated monocytes of rhesus macaques and sooty mangabeys using the TNF signalling via NF-κB hallmark gene set. **g**, GSEA of LPS-stimulated monocytes of rhesus macaques and sooty mangabey using the IL6 JAK–STAT3 hallmark gene set. **h**, NF-κB response to LPS of primate *TLR4* variants in transfected HEK293T cells. NF-κB firefly-luciferase signals were normalized to *Gaussia* luciferase signals, and the relative increase in NF-κB activity compared to unstimulated controls (100%) was calculated. Data are mean ± s.e.m. of $n = 5$ independent experiments performed in triplicate transfections are shown. Unpaired two-sided Student's *t*-test, *P* values are indicated. For source data of the animal studies, see Supplementary Table 1. RM SM-CT, *Mm*TLR-4 with the C terminus of *Ca*TLR-4; SM RM-CT, *Ca*TLR-4 with the C terminus of *Mm*TLR-4.

of *TLR4* in cells from *C. atys* and *M. mulatta*, nor did the expression of any factors in the TLR-4–MyD88–TRIF signalling axis correlate with TNF and IL-6 production (Extended Data Fig. 8d and Extended Data Table 3). To more broadly characterize the effect of attenuated TLR-4 signalling in *C. atys*, we performed comparative RNA-seq profiling of LPS-treated monocytes, and found lower production of Ca*TNF* and Ca*IL6* mRNA (Extended Data Fig. 8e). Moreover, using gene set enrichment analysis (GSEA), we observed that induction of pro-inflammatory genes was broadly and significantly reduced in cells from *C. atys* (Fig. 3f, g and Extended Data Fig. 9). Overall, these results indicate that LPS stimulation of blood cells from *C. atys* results in a blunted production of pro-inflammatory cytokines. To establish a link between the C-terminal *TLR4* sequence difference and the responsiveness to LPS, we analysed the TLR-4 orthologues of humans, *C. atys* and *M. mulatta* in an NF-κB reporter assay. We observed a significantly attenuated NF-κB response to LPS of *C. atys* TLR-4 (*Ca*TLR-4) compared to *M. mulatta* TLR-4 (*Mm*TLR-4). Using chimaeric constructs encoding Mm*TLR4* with the C terminus of Ca*TLR4* or Ca*TLR4* with the C terminus of *Mm*TLR4, we confirmed that the TLR4 C terminus is responsible for this phenotypic difference (Fig. 3h). This demonstrates a sequence–function relationship of the TLR4 C terminus and suggests a novel mechanism contributing to the lower immune activation of SIV-infected sooty mangabeys.

Over the past decade the genomes of more than 25 nonhuman primate species have been sequenced, assembled and annotated[18]. This knowledge has improved our understanding of primate evolution, biology and general physiology, which has informed human biology and medicine. Here, we report a high-coverage, high-contiguity whole-genome sequence for *C. atys*, a natural SIV host. Comparative genomic analyses of natural and non-natural SIV hosts provide candidate genes that potentially influence susceptibility to AIDS in SIV-infected hosts. We have previously used trancriptomics to