Supplementary figure legends

**Supplementary Figure 1 -** Sequential gating strategy to determine the human antibody-secreting cell (ASC) phenotype. PBMCs were cultivated with DENV4 TVP particles in the MOI of 10 for 7 days. Cells were harvested, and labelled for surface staining antigens with fluorochrome-conjugated antibodies. A) The representative FACS data showing an enrichment for ASCs: 1) extended lymphocyte including blasting cells; 2–3) singlets; 4) CD20− cells; 5) CD38hi CD27hi cells.

**Supplementary Figure 2 -** Dengue virus triggers the IgM secretion upon culture with PBMC-containing B cells. A total of 3.3 x 104 cultured PBMCs were harvested at day 7 of culture and added per well of ELISPOT plates to enumerate IgM-secreting cells in each culture condition.

**Supplementary Figure 3 -** IgG secretion detected in the supernatants of PBMC-DENV cultures did not react against DENV1-4 EDIII recombinant proteins or viable DENV1-4 particles. Reactivity of supernatant samples harvested at day 7 of cultures were tested through ELISA in different dilutions: a) 1:30 - human IgG; b) 1:40 - EDIII recombinant proteins from DENV1-4 serotypes; c) 1:40 - viable DENV particles from DENV1-4 serotypes. NT - not tested.

**Supplementary Figure 4 -** Small IgG reactivity of plasma samples from blood donors of this study against distinct DENV targets through ELISA. Plasma samples were tested in different dilutions: a) 1:100 - NS1 recombinant proteins from all 4 serotypes; b) 1:80 - EDIII recombinant proteins from DENV1-4 serotypes; c) 1:80 - DENV viable particles from DENV1-4 serotypes.

**Supplementary Figure 5 -** Differential gene expression profile (fold change) between PBMC cultures stimulated with DENV4 TVP, Mitogens, or Mock supernatants for genes related to cellular signalling and B cell differentiation into ASCs. Targeted genes were: a) BAFF; b) IL-10; c) SYK; d) SRC. Data are represented as boxes and whiskers (10-90% percentile). Dashed lines represents the gene expression within the Mock culture condition.

**Supplementary Figure 6 -** Activation of the tryptophan metabolism through either serotonine (a) and kynurenine pathways (b-e) in the in vitro model of DENV-mediated ASC differentiation in comparison to mitogen-stimulated culture. Different PBMC cultures derived from 19 healthy donors had their supernatants obtained at day seven of the culture for the measurement of the following metabolites: a) 5-hydroxyindoleacetic acid; b) Anthranilic acid; c) Kynureninic acid; d) Quinolinic acid; e) Nicotinic acid. Dashed lines represent the standard amount of the metabolite found in the culture medium kept under the same conditions, but without cells.

Supplementary table legends

Supplementary Table 1 - Fluorochrome-conjugated antibodies used to estimate the percentage and absolute number of antibody-secreting cells within the cell cultures.

| Human antigen | Clone | Fluorophore | Supplier  |
| --- | --- | --- | --- |
| CD20 | 2H7 | FITC | Biolegend |
| CD38 | HI72 | PE-Cy7 | Biolegend |
| CD27 | CD27M-t271 | PE | Biolegend |

Supplementary Table 2 - qRT-PCR primers.

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| Target | Forward primer5’-3’ | Reverse primer5’-3’ | Amplicon size (bp) | Reference |
| *SYK* | AAAGACAAATGGAAAGTTCCTGA | CTTTGTCGATGCGATAGTGC | 104 | (Grammatikos et al., 2013) |
| *IL10* | GGTTGCCAAGCCTTGTCTGA | AGGGAGTTCACATGCGCCT | 101 | (Pfaffl, 2001) |
| *SRC* | AGTGCTGGCGGAAGGAGCCT | ATCCAAGCCGAGAAGCCGGT | 149 | (Laghi & Al, 2001) |
| BAFF(*TNFSF13B)* | AGGCAACTCCAGTCAGAACAGC | TCATCCCCAAAGACATGGACC | 303 | (Schaumann et al., 2007) |
| IDO1 | AGACCACAAGTCACAGCGCC | TTGGCAAGACCTTACGGACAT | 70 | (Opitz et al., 2011) |
| IDO2 | TGCTTCATGCCTTTGATGAG | GAAGGCCTTATGGGAAGGAG | 104 | (Opitz et al., 2011) |
| DENV | GACTAGTGGTTAGAGGAGACC | GTCTCCTCTAACCTCTAGTCCT | 155 | Laboratório de Virologia Molecular, Instituto Carlos Chagas, ICC/Fiocruz-PR, Curitiba, PR - Brazil |

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