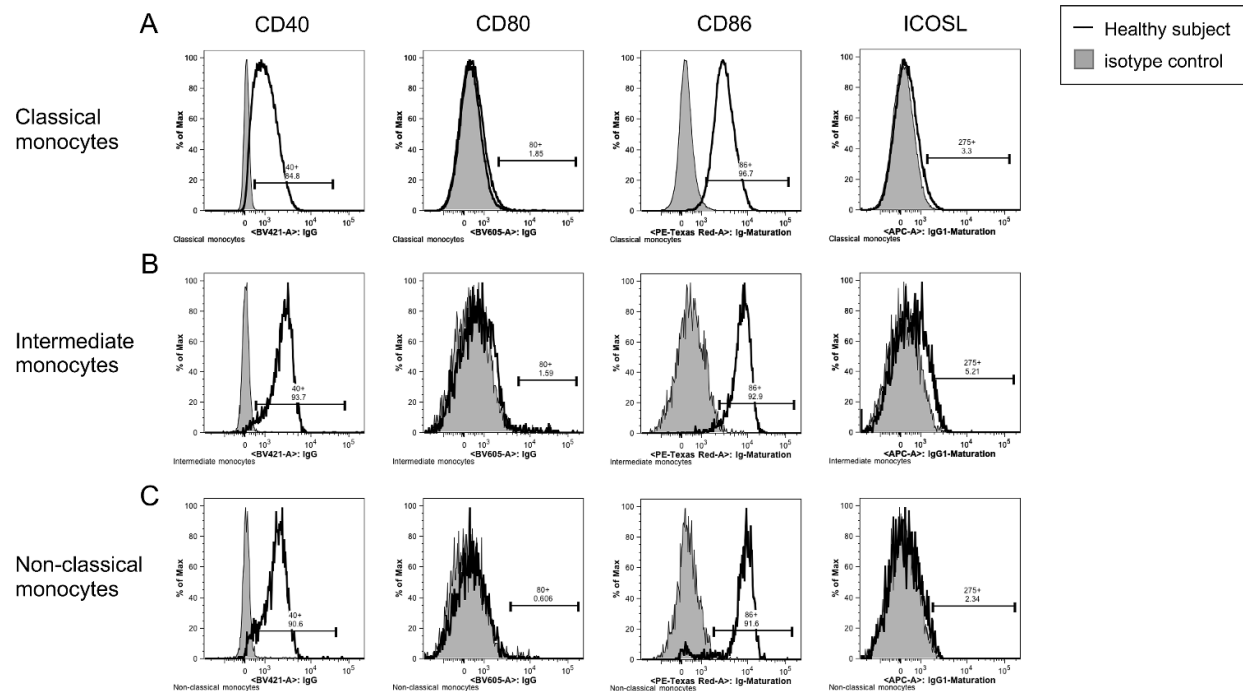
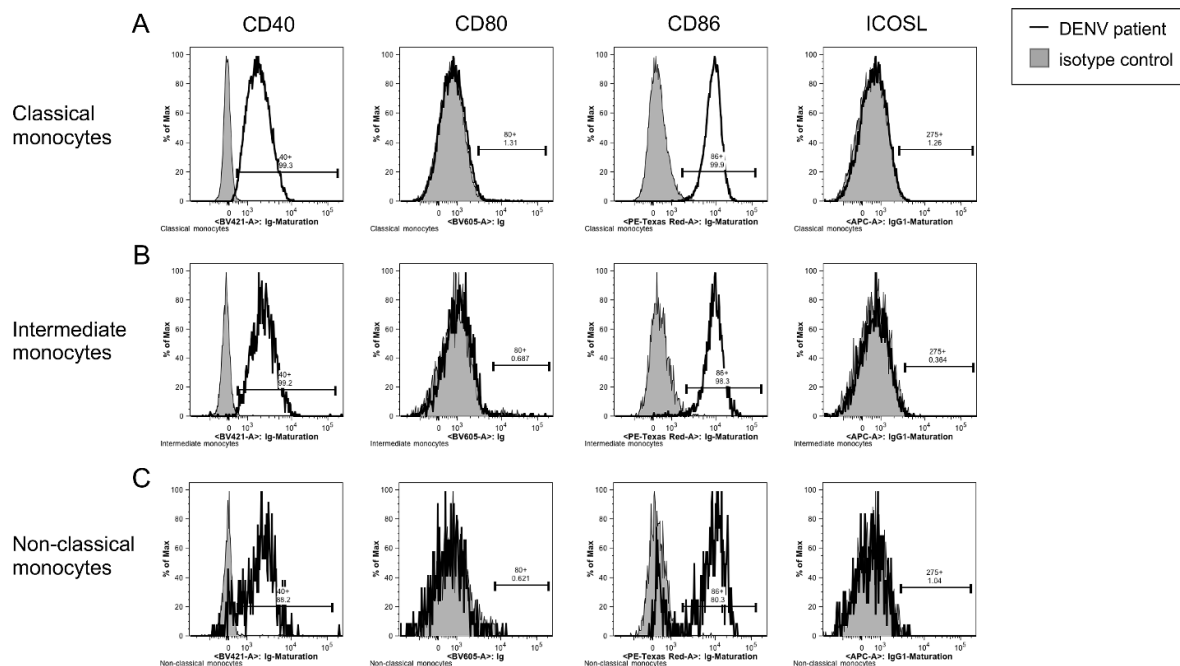


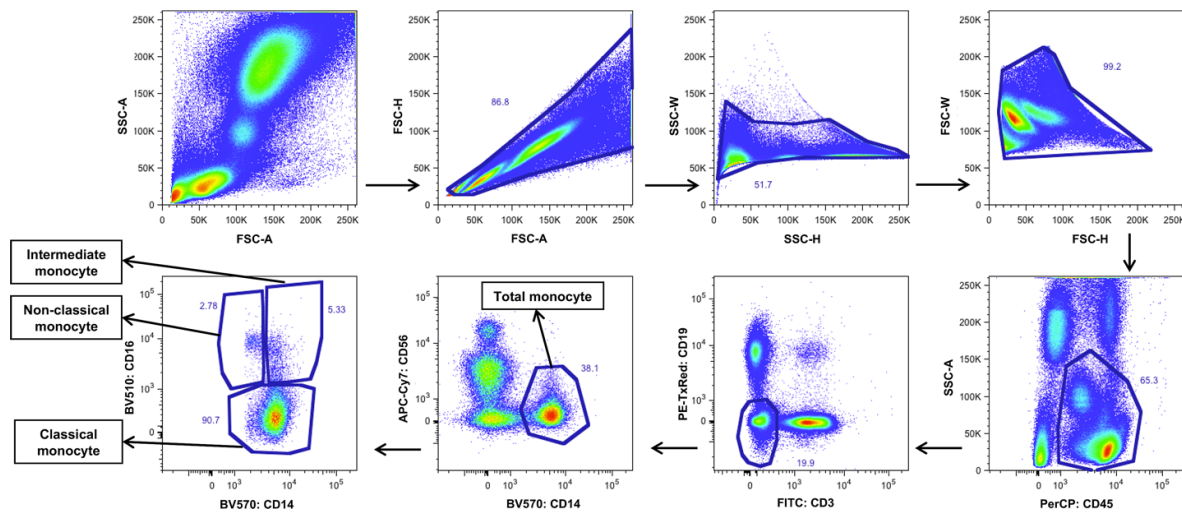
# Supplementary Materials



**Figure S1.** Gating strategy of co-stimulatory molecules expressed on each monocyte subset in healthy subjects. Representative histogram plots display the co-stimulatory molecules CD40, CD80, CD86 and ICOSL expressed by (A) classical monocytes, (B) intermediate monocytes and (C) non-classical monocytes on blood samples from healthy subjects (solid black line) and showing their isotype control (shaded grey).



**Figure S2.** Gating strategy of co-stimulatory molecules expressed on each monocyte subset during acute DENV infection. Representative histogram plots display the co-stimulatory molecules CD40, CD80, CD86 and ICOSL expressed by (A) classical monocytes, (B) intermediate monocytes and (C) non-classical monocytes on blood samples from a representative DENV-infected patient (solid black line) and showing their isotype control (shaded grey).



**Figure S3.** Representative flow cytometric gating strategy of monocytes from peripheral blood of healthy subject. After exclusion of doublets, monocytes were identified on the gated population of CD45<sup>+</sup> cells. The CD45<sup>+</sup>/CD3<sup>−</sup>/CD19<sup>−</sup>/CD56<sup>−</sup>/CD14<sup>+</sup> cells were further analyzed for monocyte subsets based on their levels of expression of CD14 and CD16. Monocyte subsets were identified as classical monocytes (CD14<sup>++</sup>CD16<sup>−</sup>), intermediate monocytes (CD14<sup>++</sup>CD16<sup>+</sup>) and non-classical monocytes (CD14<sup>+</sup>CD16<sup>++</sup>). The gating strategy of CD45<sup>+</sup> cells was adapted from Lertjuthaporn et al. [43].

**Table S1.** Primer sequences for amplification of dengue viral RNA1

| Primer               | Sequence                      | Product Size (bp) |
|----------------------|-------------------------------|-------------------|
| <i>E region</i>      |                               |                   |
| DEUL                 | 5'-TGGCTGGTGCACAGACAATGGTT-3' | 641               |
| DEUR                 | 5'-GCTGTGTCACCCAGAATGGCCAT-3' |                   |
| <i>Type-specific</i> |                               |                   |
| D1L                  | 5'-GGGGCTTCAACATCCCAAGAG-3'   | 504               |
| D1R                  | 5'-GCTTAGTTTCAAAGCTTTTTCAC-3' |                   |
| D2L                  | 5'-ATCCAGATGTCATCAGGAAAC-3'   | 346               |
| D2R                  | 5'-CCGGCTCTACTCCTATGATG-3'    |                   |
| D3L                  | 5'-CAATGTGCTTGAATACCTTTGT-3'  | 196               |
| D3R                  | 5'-GGACAGGCTCCTCCTTCTTG-3'    |                   |
| D4L                  | 5'-GGACAACAGTGGTGAAAGTCA-3'   | 143               |
| D4R                  | 5'-GGTTACACTGTTGGTATTCTCA-3'  |                   |

**Table S2.** Surface staining panels for the phenotypic characterization of monocytes and their co-stimulatory molecules1

| Panel for monocytes | FITC | PE-TxRd | PerCP | APC   | APC-Cy7   | Pacific Blue | BV510 | BV570 | BV605 |
|---------------------|------|---------|-------|-------|-----------|--------------|-------|-------|-------|
| Frequency           | CD3  | CD19    | CD45  | –     | CD56      | –            | CD16  | CD14  | –     |
| Co-stimulatory      | –    | CD86*   | CD45  | CD275 | CD3+19+56 | CD40         | CD16  | CD14  | CD80  |

PE-TxRd, PE-Texas Red; \*CD86-PE/Dazzle594.