**Extended Data**

**Supplementary Text**

## Alterations in cell subtype abundance and gene expression are only partially associated with prior DENV exposure

To determine whether the alterations we detected in cell subtype abundance and gene expression in SD progressors are associated with prior exposure to DENV, we analyzed cell subtype fractions and DEGs in primary vs. secondary DENV-infected patients across disease outcomes (**Supplementary Table 1**). Activated B cells and exhausted CD8+ T cell fractions were expanded in secondary relative to primary dengue patients regardless of disease outcome, suggesting association with prior DENV exposure but not disease severity (**Extended Data Fig. 2c**). In contrast, memory B cell and Treg fractions were expanded only in SDp with secondary infection, confirming that some alterations are unique to secondary SDp. The expansion of proliferating plasmablasts and memory CD4+ T cell fractions was comparable in primary and secondary SDp relative to uncomplicated dengue (D/DWS), thus more likely associated with disease progression rather than prior exposure to DENV. Lastly, reduction of non-classical monocyte and cytotoxic NK cell fractions was observed in all patient categories except for primary uncomplicated dengue (D/DWS), suggesting association with both prior exposure and progression.

Next, we compared the expression of DEGs we identified between SD progressors and D patients in APCs (**Fig. 2a-c**) and effector cells (**Fig. 3a-c**) in primary vs. secondary dengue infection in D and SDp patients. Several interesting observations were made; however, these require further validation given the small number of patients for each disease category: primary (total n=4: D (n=2), SDp (n=2)); secondary (total n=10: D (n=5), and SDp (n=5)).

Genes involved in antigen uptake (*CD163*, *FCGR1A* and *FCGR1B)* were mildly upregulated in secondary dengue infections in APCs (**Extended Data Fig. 4a**), and *IgG* genes and genes involved in antibody secretion were upregulated in secondary dengue infections in plasmablasts (**Extended Data Fig. 5f**). Since similar patterns were observed for these genes in SDp vs. D (**Fig. 2a and Fig. 3f**), these findings highlight association of signatures linked with ADE with prior DENV exposure, as previously reported[7,8].

In contrast, MHC-II genes were upregulated in secondary vs. primary infection in APCs (**Extended Data Fig. 4a**), suggesting that their downregulation in SDp vs. D (**Fig. 2a-c**.) is not associated with prior exposure to DENV, but rather with disease progression. Variable patterns were observed upon comparison of IFN response genes and genes involved in inflammation, migration and adhesion between secondary vs. primary infections (**Extended Data Fig. 4a**) and SDp vs. D (**Fig. 2a-c**), suggesting that these APCs' responses are only

partially associated with DENV exposure. Similarly, whereas genes associated with exhaustion were upregulated in T and NK cells in secondary vs. primary infections as in SDp vs. D, variable expression patterns were observed for genes involved in cell activation (e.g. *IL32* in T cells and *KLRB1* in NK cells) and IFN response (e.g. *IFIT3* in T cells), suggesting only partial association of altered responses observed in these effector cells in SDp with prior DENV exposure (**Extended Data Fig. 5f, Fig. 3a-c**).

This analysis is subject to a substantial limitation due to the relatively modest number of patients in each category. Because of the small n, it is not possible to perform robust statistical analyses after subdividing the patients by both outcome and previous exposure simultaneously. As a consequence, we only stratify patients by either outcome (in the main text) or prior exposure (here). Because prior exposure is itself associated with severe outcome, this simpler stratification is not as clean as an ideal experimental design would warrant. In future studies with a larger sample size, for which sample collection is ongoing, we plan to perform a double subdivision based on both criteria simultaneously in order to obtain more definitive answers.

Taken together, these findings support that prior DENV exposure is associated with pathways involved in ADE, as previously reported, and possibly with effector cell exhaustion. Importantly, these findings reveal that other hallmarks of SD progression, such as downregulation of MHC-II genes, may be only partially or not associated with DENV exposure. Yet, these findings require validation in larger cohorts given the limited number of samples.