



SUPPLEMENTARY FIG. S3. Comparison between the in vitro immunomodulatory effect on T-cell proliferation of MSCs, MSC-derived MVs, and supernatants collected during serial ultracentrifugation. The graph shows the proliferation of healthy donor peripheral blood mononuclear cells (PBMCs) stimulated with PHA, in the presence or in the absence of either MSCs (at two different MSCs:PBMCs ratios 1:2 and 1:10) or MVs, isolated from MSCs with two different procedures (MVs-1 and MVs-2), or intermediate supernatants collected during ultracentrifugation procedures. In detail, as for MVs-1, first supernatant sample was collected after 2,500 g centrifugation, second supernatant sample after 10,000 g centrifugation, and third supernatant sample after 100,000 g ultracentrifugation; whereas, as for MVs-2, first supernatant sample was collected after 1,000 g centrifugation, second supernatant after 2,000 g centrifugation, and third supernatant after 100,000 g ultracentrifugation. Each bar represents the percentage of proliferation of 10^5 PBMCs, calculated by measuring ^3H -thymidine incorporation after 3-day culture. The counts per minute (cpm) values at each cell concentration were normalized to the cpm of PBMCs alone in each experiment. Each bar represents the median \pm SD of 12 experiments (each point being in triplicate).