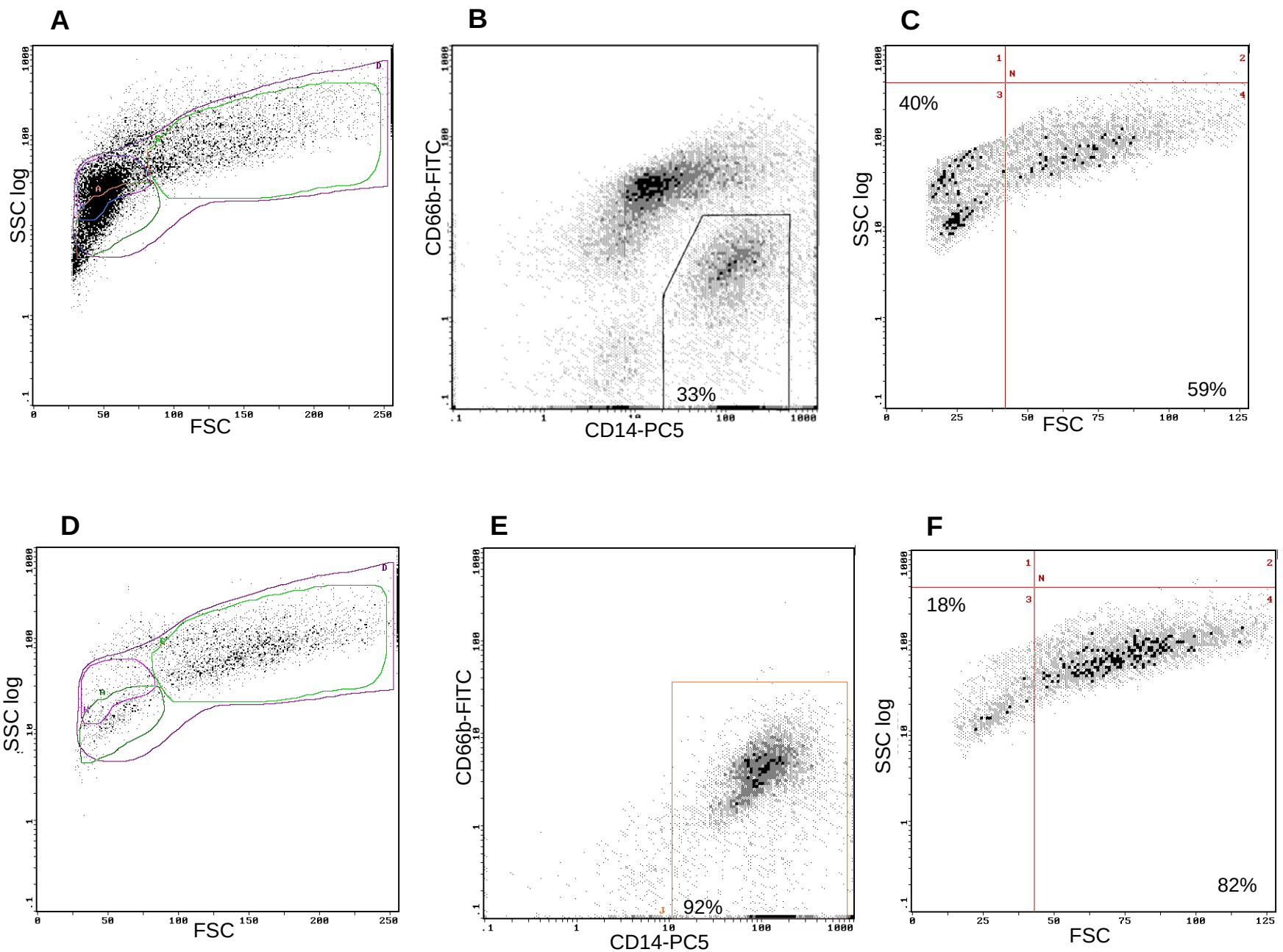
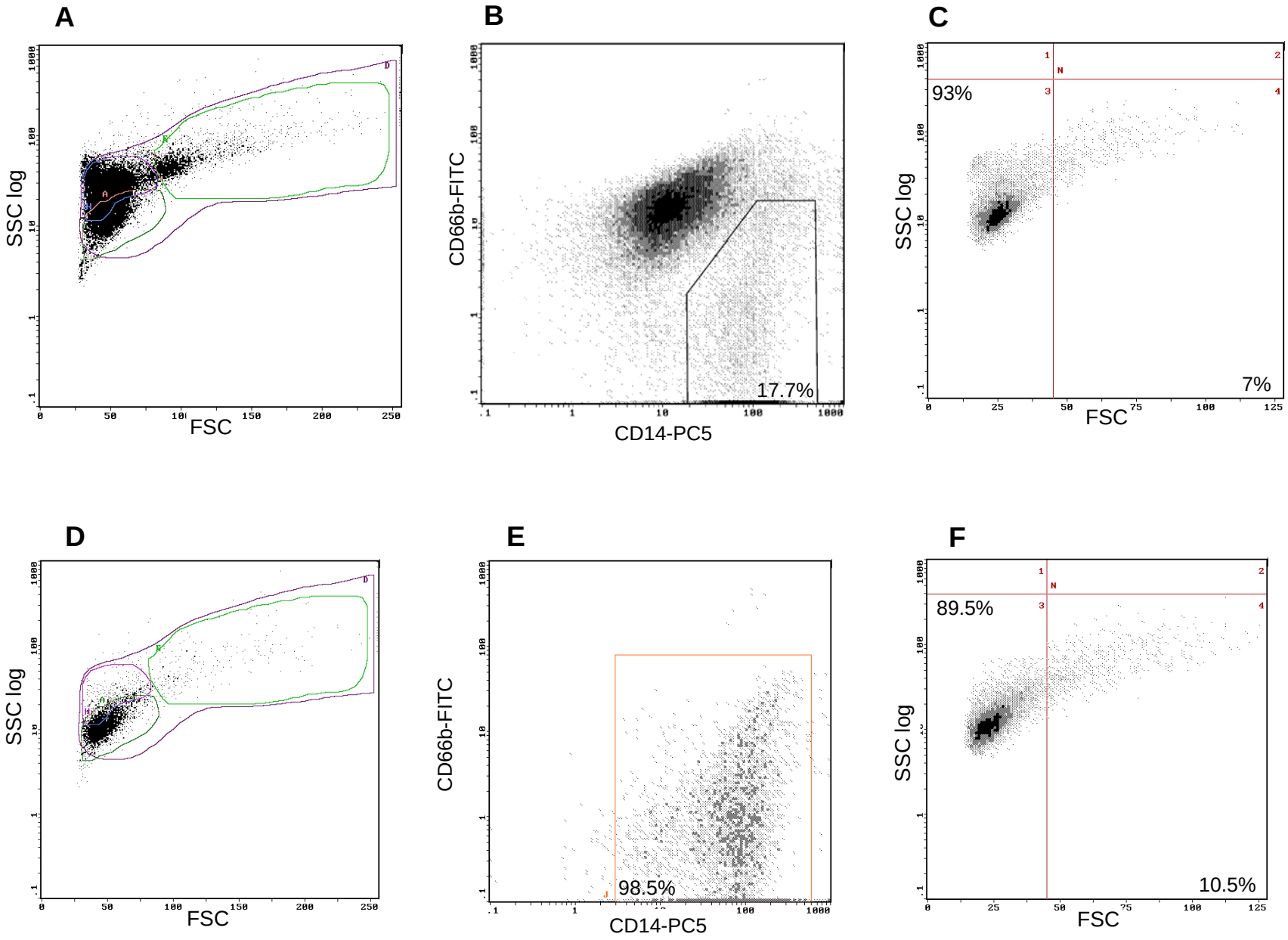


Suppl. Fig. 1: Frankenberger et al.

Frankenberger et al.
Legend to suppl. Fig1
Flow cytometry analysis of sputum macrophages before and after RosetteSep purification in a healthy control donor

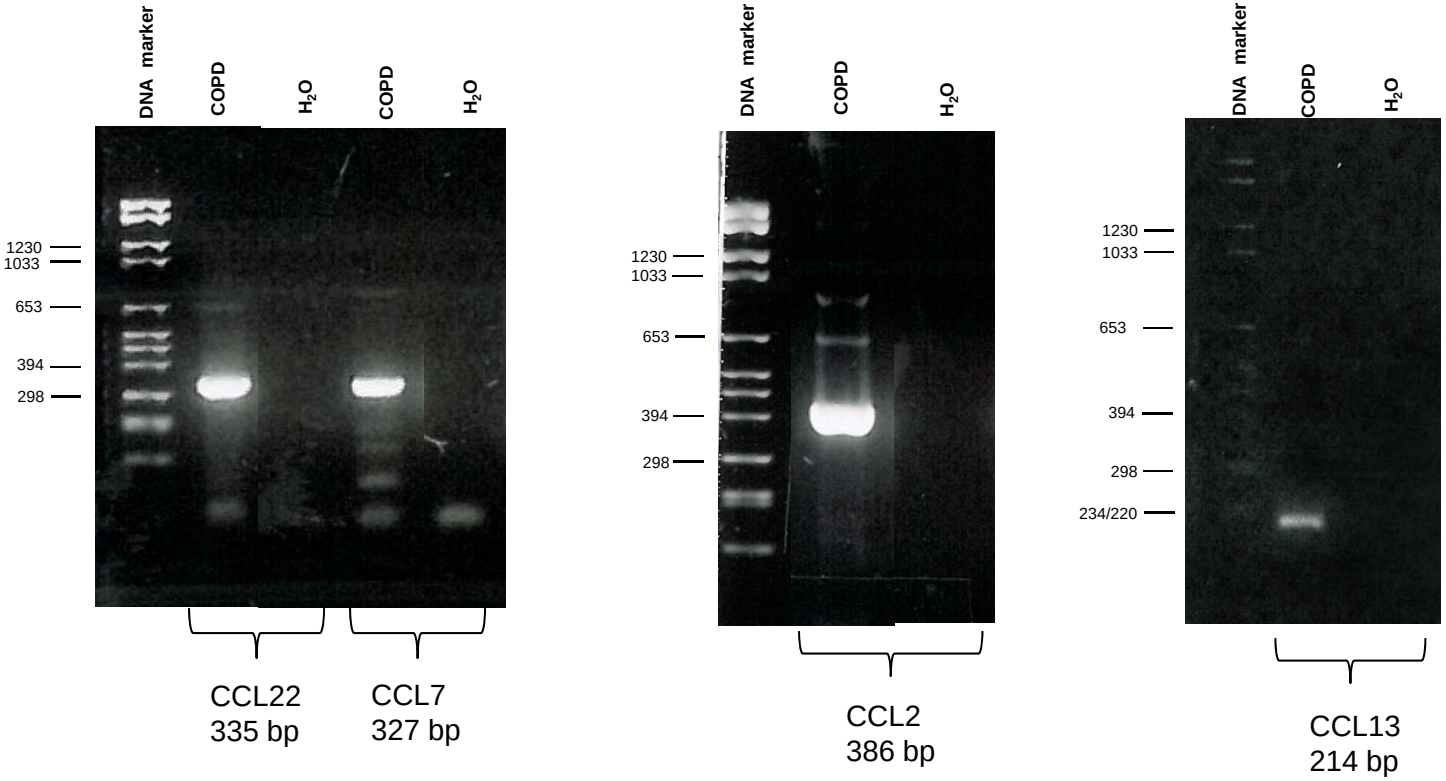
Sputum cells were isolated as described in „Material and methods“. Macrophages were further purified by the RosetteSep technique. An aliquot of cells was taken before and after RosetteSep and stained with CD66b-FITC/CD16b-PE/CD14-PC5 + Propidiumiodide for analysis in the flow cytometer. Dot plots A, B, and C show the analysis before purification and dot plots D, E and F reveal results from purified sputum macrophages. Dot plotsA and D show forward versus side scatter analysis of all cells analysed. Plots B and E reveal two color analysis for CD14 versus CD66b with an analysis gate set on whole sputum macrophages for calculation of macrophage purity (here 33% before RosetteSep in B and 98% after RosetteSep in E). The calculated macrophages were then re-gated on FSC versus SSC to determine the percentage of small macrophages which were 40% without RosetteSep (C)and 18% with RosetteSep in (F).



Suppl. Fig. 2: Frankenberger et al.

Frankenberger et al.
Legend to suppl. Fig2
Flow cytometry analysis of sputum macrophages before and after RosetteSep purification in a COPD patient

Sputum cells were isolated as described in „Material and methods“. Macrophages were further purified by the RosetteSep technique. An aliquot of cells was taken before and after RosetteSep and stained with CD66b-FITC/CD16b-PE/CD14-PC5 + Propidiumiodide for analysis in the flow cytometer. Dot plots A, B, and C show the analysis before purification and dot plots D, E and F reveal results from purified sputum macrophages. Dot plotsA and D show forward versus side scatter analysis of all cells analysed. Plots B and E reveal two color analysis for CD14 versus CD66b with an analysis gate set on whole sputum macrophages for calculation of macrophage purity (here 17.7% before RosetteSep in B and 98.5% after RosetteSep in E). The calculated macrophages were then re-gated on FSC versus SSC to determine the percentage of small and large sputum macrophages (here 89.5% small and 10.5% large macrophages after RosetteSep in F). The calculated macrophages were then re-gated on FSC versus SSC to determine the percentage of small macrophages which were 93% without RosetteSep (C)and 89.5% with (RosetteSep in F).



Suppl. Fig. 3: Frankenberger et al.

Frankenberger et al.
Legend to suppl. Fig3
Agarose gel electrophoresis

Products obtained after LightCycler PCR were separated on a 2% agarose gel to determine the product size. As test samples we used cDNA of different purified sputum macrophages from COPD patients.