

Supplementary data 1: IL-34 and M-CSF differentially activate the M-CSFR. (A) Western blot analysis of tyrosine phosphorylation patterns of M-CSFR and Erk1/2 phosphorylation in THP-1 cells after addition of increasing doses of M-CSF (1-30 ng/mL) for 5 minutes compared to 25 ng/mL of IL-34. (B) Differential effects of M-CSF and IL-34 on the phosphorylation of M-CSFR and Erk1/2 in myelomonocytic U937 cells.

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Supplementary data 2: M-CSF did not bind HEK- and HOS-Mock cells licking the M-CSFR. Increasing concentrations of ¹²⁵I-labelled M-CSF (M-CSF*) were incubated for 60 min at 4°C with 106 cells and bound and unbound fractions were determined as described in « Experimental Procedures ». Specific binding fraction of iodinated cytokines. Curve fitting was performed as described in the « Materials and Methods » section.



Supplementary data 3: IL-34 does not induce detectable signaling pathways in absence of M-CSFR and RPTPB/E. (A-B) HEK Mock and HOS Mock, which did not express neither the MCSF-R nor the RPTP β/ξ , were stimulated with 200 ng/mL of IL-34 for 1, 5, 15, 30 or 60 min. (A) The total tyrosine phosphorylation pattern or (B) the phosphorylations of mTor, and Akt were analyzed by Western blot. β-actin, total mTor and total Akt were used as loading controls.



Supplementary data 4: Syndecan-2 and -4 have no effect on IL-34 induced signaling pathways in contrast to Syndecan-1. HEK M-CSFR were transfected with the different forms of syndecan (1, 2 and 4). Fourty eight hours after transfection, cells were stimulated with 50 ng/mL of IL-34 for 5 min, and M-CSFR tyrosine phosphorylations were assessed by Western blot. β -actin was used as a loading control. NT: Non-transfected cells; Mock: cells transfected with an empty plasmid; Syndecan-1, -2, -4: cells transfected with a plasmid coding for syndecan-1, -2 or -4.



Supplementary data 5: Syndecan-1 expression regulates the levels IL-34-induced M-CSFR phosphorylation. HEK M-CSF expressing differentially the syndecan-1 expression were isolated by cell sorting flow cytometry. Western blot analysis of tyrosine phosphorylation patterns of M-CSFR and Erk1/2 phosphorylation were carried out in the presence of increasing concentrations of IL-34 for 5 minutes.