

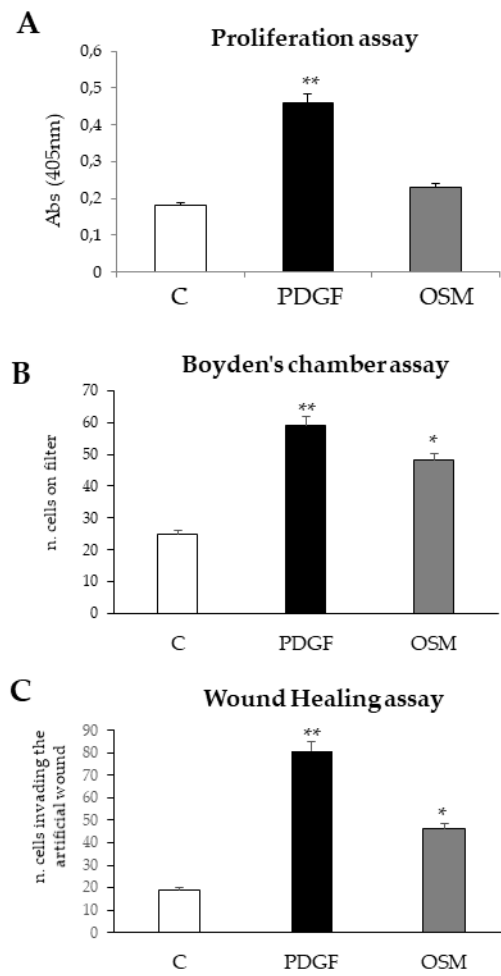


Figure 1. (A–C) Effect of hrOSM on proliferation and migration of HSC/MFs cells. Proliferation assay (A) Boyden's chamber assay (B) and wound healing assay (C) were performed on HSC/MFs exposed to PDGF-BB 10 ng/mL, used as positive control, or to hsOSM 10 ng/mL. Data in bar graphs represent mean $\pm$ SEM ($n = 3$, in triplicate). * $p < 0.05$, ** $p < 0.01$ versus control value.

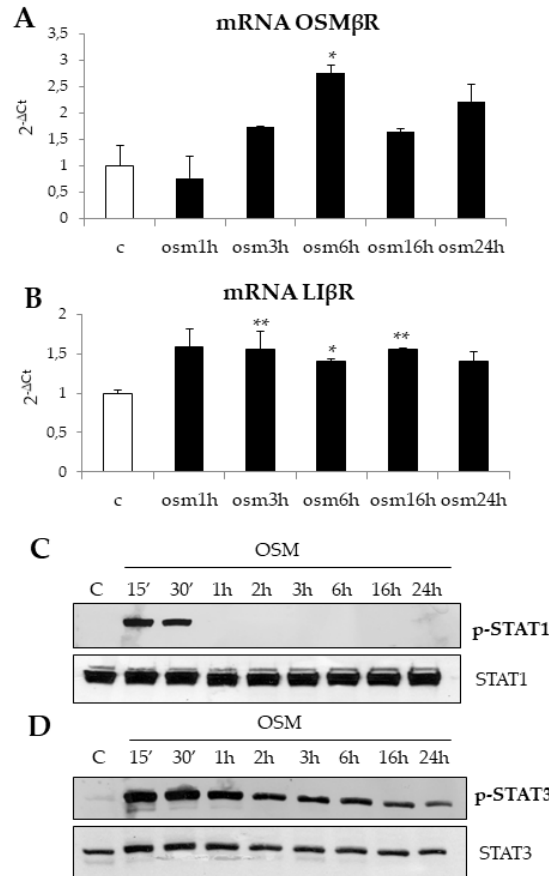


Figure 2. (A–D) Involvement of both OSM-receptors, OSMβR and LIFβR, as well as of STAT1 and STAT3 signal pathways in HSC/MFs in response to hrOSM. Quantitative real time PCR (q-PCR) analysis of OSMβR (A) and LIFβR (B) was performed in HSC/MFs exposed to hrOSM 10 ng/mL up to 24 h. Data are expressed as means ± SEM of three independent experiments. **p* < 0.05, versus control value. Western blotting analysis of phosphorylated STAT1 (C) and STAT3 (D) in HSC/MFs exposed to hrOSM 10 ng/mL (starting from 15 min up to 24 h). Equal loading was confirmed by reprobings the same membrane with un-phosphorylated protein.

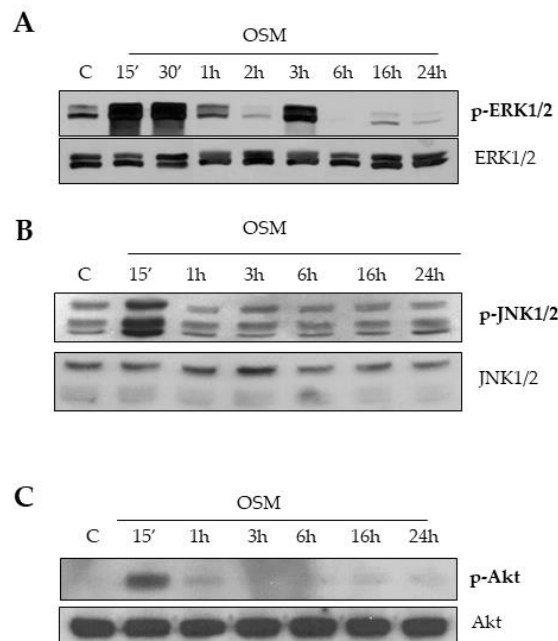


Figure 3. (A–C) HSC/MFs response to hrOSM involved different signal pathways. Western blotting analysis of phosphorylated ERK 1/2 (A), JNK1/2 (B) and c-Akt (C) in HSC/MFs exposed to hrOSM 10 ng/mL (starting from 15 min up to 24 h). Equal loading was confirmed by reprobing the same membrane with the un-phosphorylated protein.

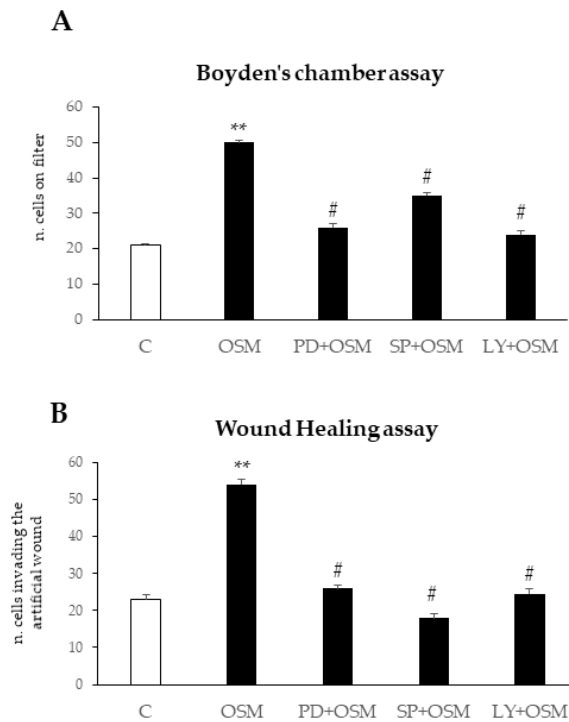


Figure S4. (A,B) Involvement of signal transduction pathways in the modulation of OSM-dependent migration of HSC/MFs. Boyden's chamber assay (A) and wound healing assay (B) were performed on HSC/MFs treated with hrOSM 10 ng/mL or ,where indicated, pre-treated for 30 min with specific pharmacological inhibitors PD98059 (MEK-1 inhibitor), SP600125 (JNK 1/2 inhibitor), LY294002 (Akt inhibitor) and then exposed to hrOSM 10 ng/mL. ** $p < 0.01$ versus control value, # $p < 0.05$ versus OSM value.

Figure S5

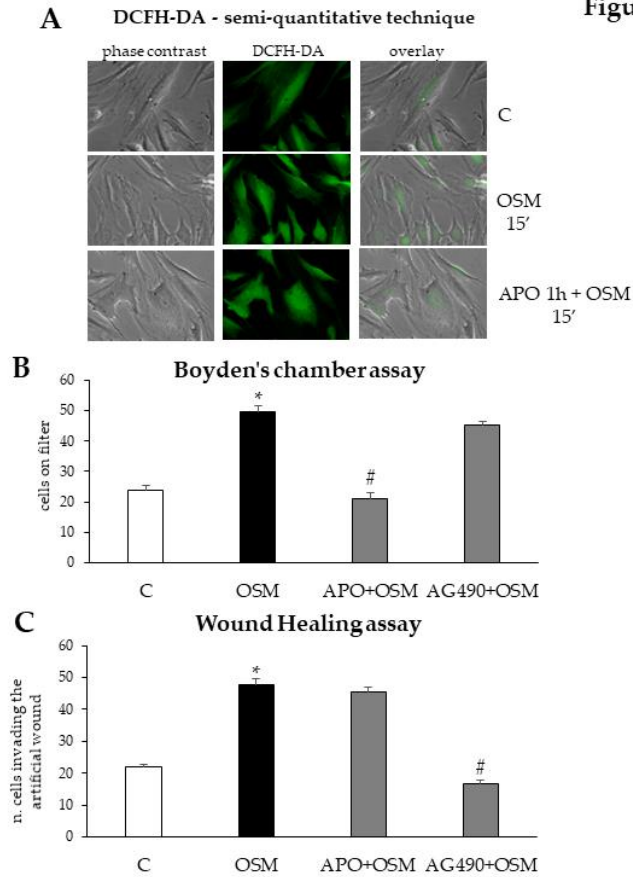


Figure 5. (A–C) hrOSM dependent induction of migration relies on intracellular generation of ROS and STAT1/3 activation. DCFH-DA semi-quantitative analysis was performed to evaluate ROS intracellular generation in HSC/MFs untreated for 15 min with OSM 10 ng/mL or pre-treated for 1 h with apocynin, the pharmacological inhibitor of NADPH oxidase (APO 250 μ M), and then exposed to OSM 10 ng/mL for 1 h (A). Boyden's chamber assay (B) and wound healing assay (C) were performed on HSC/MFs cells treated with hrOSM 10 ng/mL or, where indicated, pre-treated for 1 h with apocynin, the pharmacological inhibitor of the NADPH oxidase (APO 250 μ M), or with the JAK2 inhibitor (AG490 100 μ M) and then exposed to OSM 10 ng/mL. * $p < 0.05$, versus control value, # $p < 0.01$ versus OSM value.

Figure S6

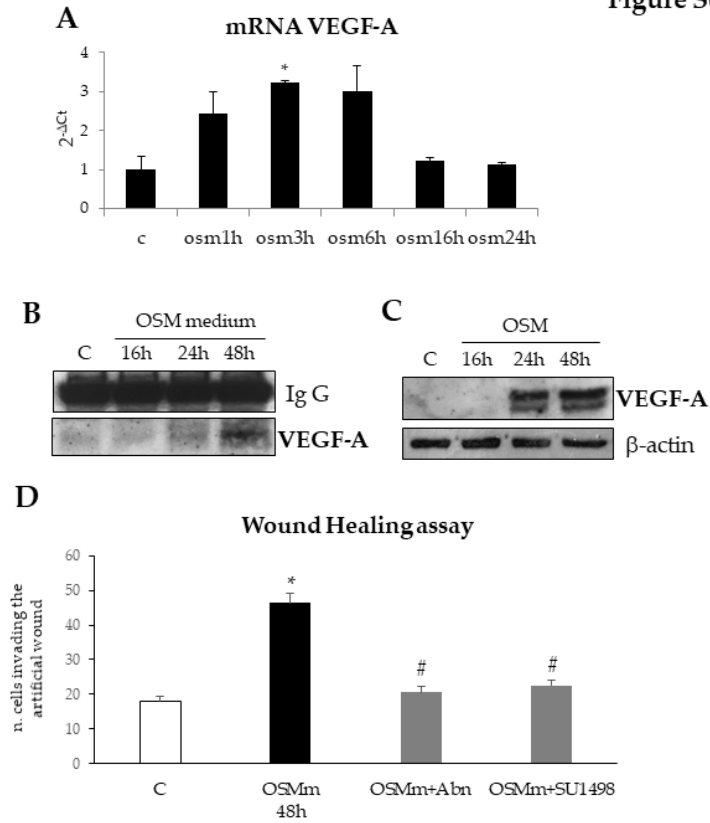


Figure 6. (A–D) involvement of VEGF-A in OSM-dependent non oriented migration. Quantitative real time PCR (q-PCR) analysis of VEGF-A (A) in HSC/MFs exposed to hrOSM 10 ng/mL up to 24 h. Data are expressed as means $\pm$ SEM of three independent experiments. * $p < 0.05$, ** $p < 0.01$ versus control value. Immunoprecipitation analysis of VEGF-A released in the medium of HSC/MFs treated with hrOSM 10 ng/mL from 16 to 48 h (B). Western blotting analysis of VEGF protein levels in HSC/MFs treated with hrOSM 10 ng/mL from 16 to 48 h (C). Equal loading was confirmed by re-probing the same membrane with β -actin. Wound healing assay (D) in HSC/MFs untreated or exposed to medium (OSMm) obtained from HSC/MFs treated with OSM for 48 h or, where indicated, pre-treated with the neutralizing antibody for VEGFR2 or with the pharmacological inhibitor of VEGFR2, SU1498 and then exposed to OSMm. * $p < 0.05$, versus control value, # $p < 0.05$; versus OSMm value.