Supporting Information

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SI Text

SI Materials and Methods. ***Immunoblotting.*** Rat vascular smooth muscle cells (rVSMCs) were starved for 48 h in DMEM media +0.1% BSA and 10 mM Hepes prior to addition of 100 nM (Stromal-derived factor) SDF-1α or interferon-inducible T-cell alpha chemoattractant (ITAC). PhosphoERK immunoblotting were carried out as described (1). Anti-phospho-p44/42 MAPK (1:3,000; Cell Signaling Technology) and anti-MAPK 1/2 (1:10,000; Upstate Biotechnology) were used for detection of phosphoERK and total ERK on immunoblot, respectively.

Thymidine incorporation. rVSMCs plated in 12-well plates (104 cells/well) were rendered quiescent in serum-free medium for 48 hr, then refed with serum-free medium containing 1 μCi/mL of [^{3}H]thymidine without (basal) or with the indicated agonist, and incubated for 48 hr. DNA synthesis was estimated as the incorporation of [^{3}H]thymidine into trichloroacetic acid-insoluble material, in triplicate wells.

1. Ahn S, Shenoy SK, Wei H, Lefkowitz RJ (2004) Differential kinetic and spatial patterns of beta-arrestin and G protein-mediated ERK activation by the angiotensin II receptor. *J Biol Chem* 279(34):35518–35525.

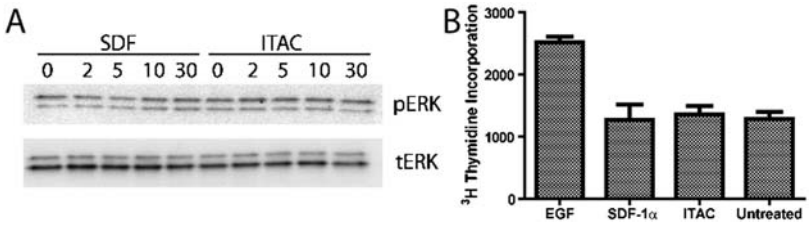

Fig. S1. Absence of pERK activation or mitogenic response to CXCR7 ligands SDF-1a and ITAC in rVSMCs. (*A*) Representative blot (of three independent experiments) of pERK and tERK levels in rVSMCs treated with SDF-1α or ITAC from 0 to 30 minutes. (*B*) Lack of mitogenic effect of either SDF-1α or ITAC on rVSMCs as assayed by [^{3}H]thymidine incorporation for 48 hr after stimulation (positive control of epidermal growth factor). Data shown are mean ± SEM from at least three independent experiments.

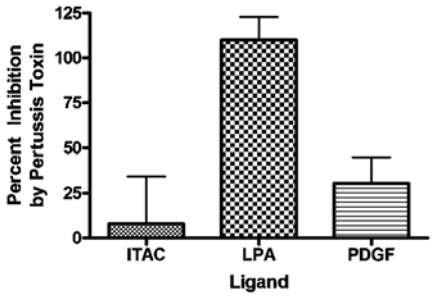

Fig. S2. Migration by rVSMCs in response to ITAC is independent of $G_\alpha i$. rVSMCs were treated with pertussis toxin at a concentration of 100 ng/mL for 16 hr prior to assessing migration by Transwell assay (see *Materials and Methods*). Ligands used were lysophosphatidic acid, whose response is known to be regulated by $G_\alpha i$, PDGF, whose response is known to be independent of G proteins, and ITAC. Shown is the percent inhibition of ligand-induced migration by pertussis toxin from six Transwell assays.