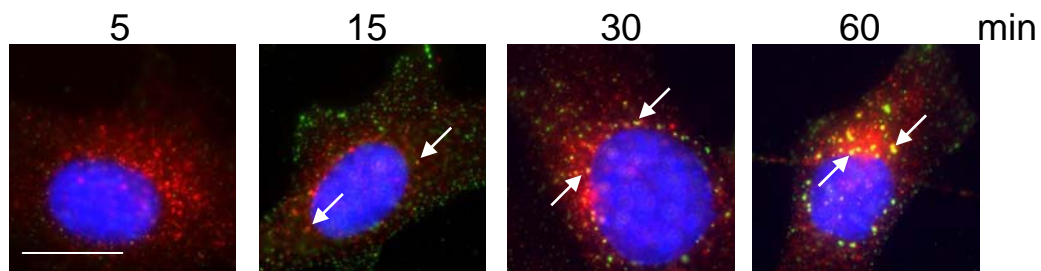
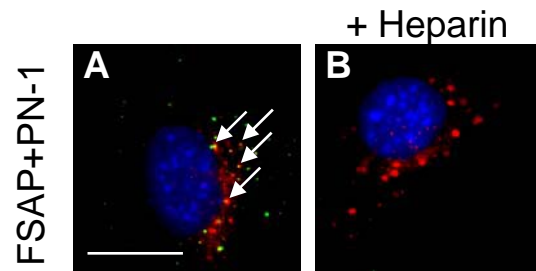


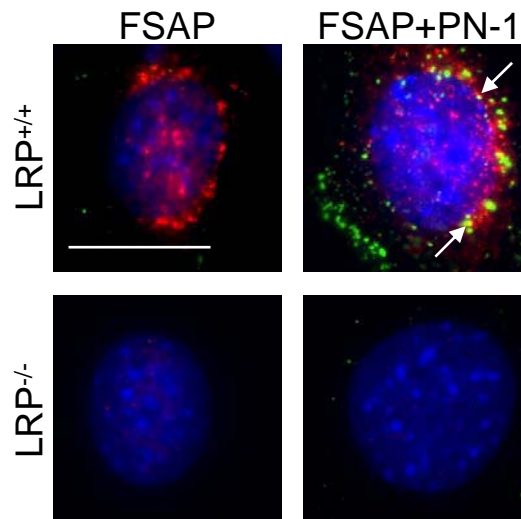
Muhl et al. Online supplemental data 1



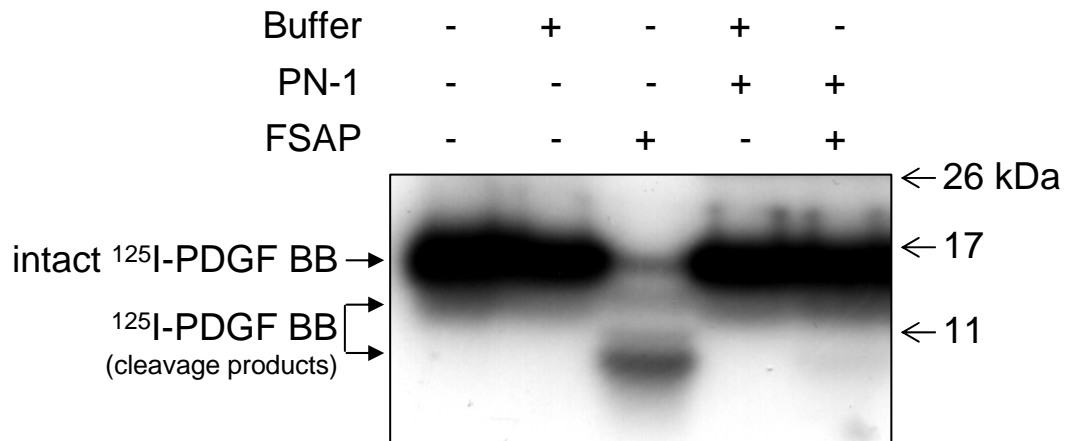
**Supplemental data 1: Time-dependent binding and uptake of FSAP-PN-1 complex in VSMC via LRP.** 1  $\mu\text{g/ml}$  FSAP and 2  $\mu\text{g/ml}$  PN-1 were preincubated for 30 min and the mixture was added to the cells for indicated times at 37°C. FSAP is stained with FITC-labeled secondary antibody indicated in green, LRP is stained with Rhodamine red-X-labeled secondary antibody indicated in red and nuclei are DAPI stained indicated in blue. Yellow color indicates co-localization of FSAP and LRP. White arrows highlight examples of co-localization. Calibration bars indicates 20  $\mu\text{m}$ . FSAP-PN-1 complex internalization proceeds in a time dependent manner starting at 15 min and reaching a maximum at 60 min. Similar results were obtained in 2 separate experiments.



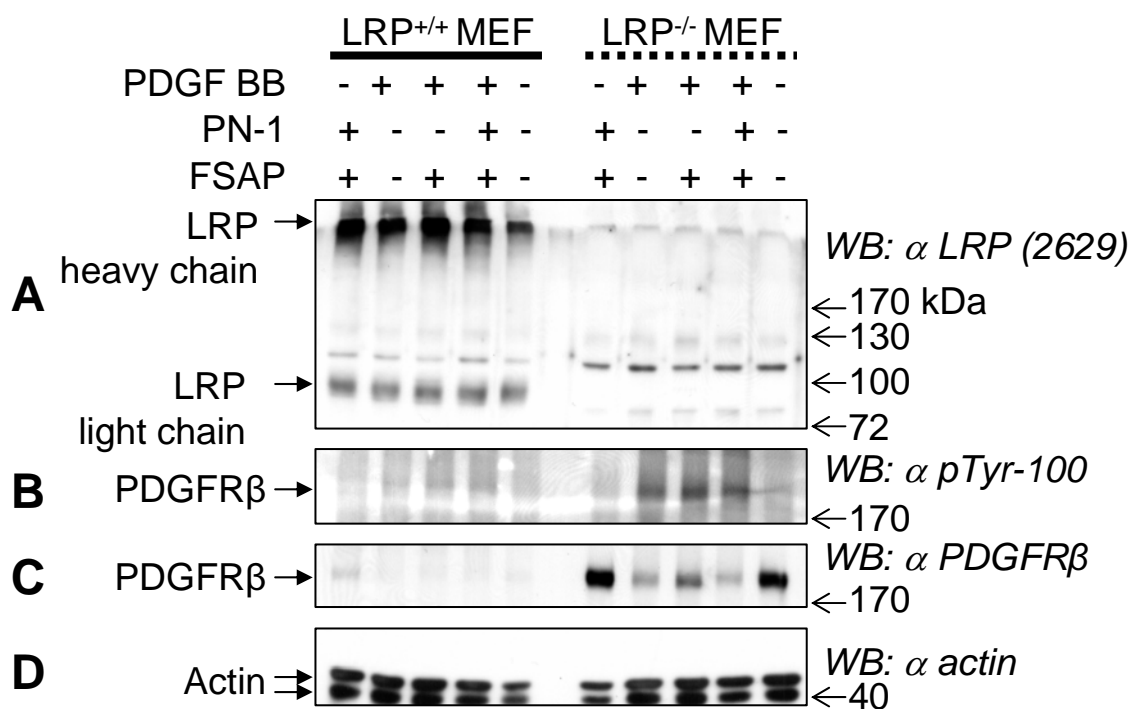
**Supplemental data 2: Heparin-dependent binding and internalization of FSAP-PN-1 complex in VSMC via LRP.** 1  $\mu\text{g/ml}$  FSAP and 2  $\mu\text{g/ml}$  PN-1 were preincubated for 30 min (A) without or (B) with 10  $\mu\text{g/ml}$  Heparin and the mixture was added to the cells for indicated times at 37°C. FSAP is stained with FITC-labeled secondary antibody indicated in green, LRP is stained with Rhodamine red-X-labeled secondary antibody indicated in red and nuclei are DAPI stained indicated in blue. Yellow color indicates co-localization of FSAP and LRP. White arrows highlight examples of co-localization. Calibration bar indicates 20  $\mu\text{m}$ . FSAP-PN-1 complex interaction with LRP and therefore internalization via LRP is blocked by Heparin.



**Supplemental data 3: Internalized FSAP-PN-1 complexes via LRP in mouse embryo fibroblasts (MEF).** 1  $\mu\text{g/ml}$  FSAP and 2  $\mu\text{g/ml}$  PN-1 were preincubated for 30 min and the mixture was added to LRP<sup>+/+</sup> MEF (upper panels) and LRP<sup>-/-</sup> MEF (lower panels) for 45 min. FSAP is stained with FITC-labeled secondary antibody indicated in green, LRP is stained with Rhodamine red-X-labeled secondary antibody, nuclei are stained with DAPI indicated in blue. Yellow color indicates co-localization of FSAP and LRP. White arrows highlight examples of co-localization. Calibration bar indicate 20  $\mu\text{m}$ . The lack of staining of LRP<sup>-/-</sup> cells with the  $\alpha$  **LRP antibody** indicates specificity of staining **with this antibody**.



**Supplemental data 4: <sup>125</sup>I-PDGF BB-binding to VSMC:** <sup>125</sup>I-PDGF BB was incubated with buffer, 1 µg/ml FSAP or FSAP + 2 µg/ml PN-1 for 30 min and then added to VSMC for 60 min on 37°C. After extensively washing the cells the cell associated radioactivity was recovered by lysis and SDS-PAGE was performed under reducing conditions followed by autoradiography. FSAP alone induced cleavage of <sup>125</sup>I-PDGF BB and this was inhibited by PN-1. Intact <sup>125</sup>I-PDGF BB bound to cells but cleaved <sup>125</sup>I-PDGF BB did not.



**Supplemental data 5: Effect of FSAP-PN-1 complex on PDGF-BB mediated stimulation of LRP<sup>+/+</sup> and LRP<sup>-/-</sup> MEF.** 1  $\mu$ g/ml FSAP and 2  $\mu$ g/ml PN-1 were preincubated for 30 min. 20 ng/ml PDGF BB was then preincubated with either buffer, FSAP or FSAP-PN-1 for 60 min and the mixtures were used for stimulating cells for 10 min. MEF were lysed with lysis-buffer and processed for Western Blot analysis. Heavy chain and light chain of LRP is expressed in LRP<sup>+/+</sup> MEF but not in LRP<sup>-/-</sup> MEF (A). Since LRP internalizes PDGF $\beta$ R, more PDGF $\beta$ R is present in LRP<sup>-/-</sup> than LRP<sup>+/+</sup> cell (C). Stimulation of cells with PDGF-BB leads to tyrosine phosphorylation of PDGF $\beta$ R (B) and its internalization and degradation (C). FSAP or FSAP-PN-1 complex do not influence this process. Actin blot shows equal loading of cell extracts in all lanes (D).