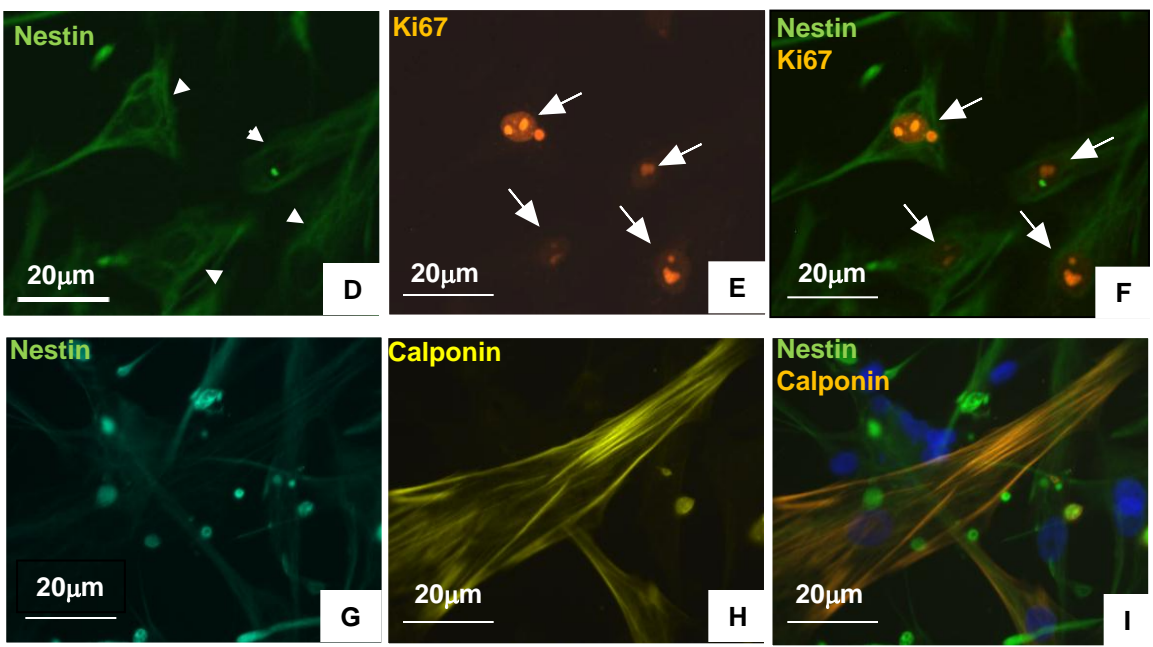
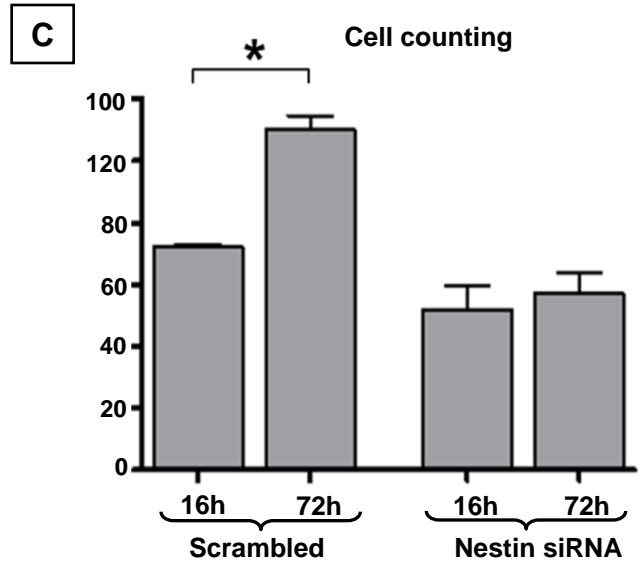
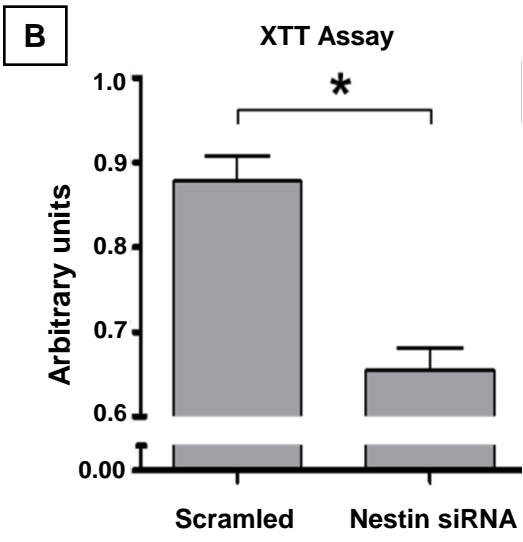
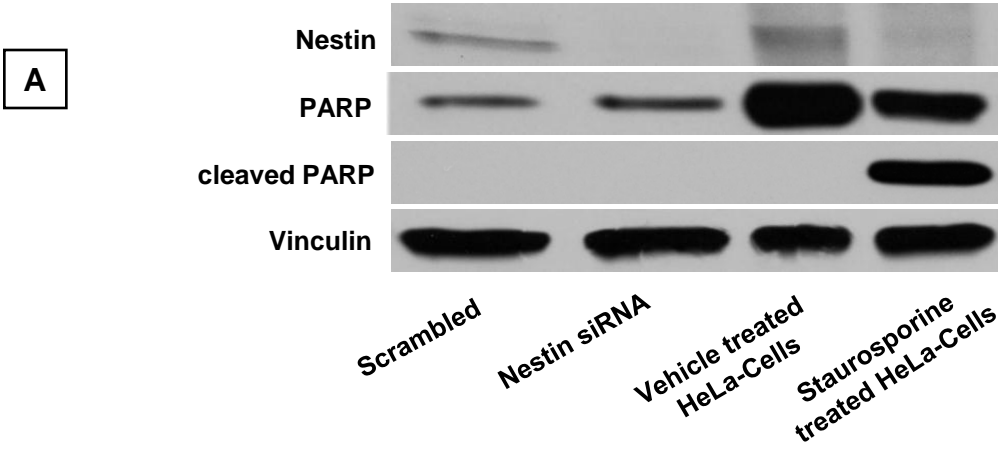


# Supplemental Figure 4



**Supplemental Figure 4. Isolated VSMCs.** Nestin effects on cell number and nestin expression in isolated VSMCs (HPASMCs). **A:** Western blot showing nestin and the apoptosis marker cleaved PARP (89kDa) after treatment of nestin siRNA-treated and control (scrambled siRNA) cells. 80  $\mu$ g total protein. Vehicle-and staurosporine-treated HeLa-cells served as control for apoptosis. Vinculin was used as loading control. **B:** XTT assay suggests a significantly lower cell number in case of nestin siRNA treatment. Bars show mean scores ( $\pm$  SEM) from 3 assessments. \* $P < 0.0001$ . **C:** Cell counting: Cell number only increases in the presencence of nestin (scrambled siRNA) \* $P < 0.01$ . There is no significant increase in cell number in the absence of nestin (nestin siRNA). Cells were counted after 16h (overnight) of siRNA treatment and after 72h of siRNA treatment. **D-I** (Immunostaining of untransfected HPASMCs): **D-F:** The proliferation marker Ki67 (red, arrow) is co-localized with nestin (green, arrowhead). (D: nestin, E: Ki67, F: merge). **G-I:** Different to nestin (green) calponin (red) is present only in a low number of cells displaying a more differentiated phenotype.