

## Supplementary figure labels

### **Fig. S1. Alternative flow patterns**

By the use of dyes, two separate flow patterns in the microfluidic device are visualized. These are achieved by operating different sets of channels and are determining the composition of fluids in the cell cultivation chamber. Heterogeneous flows (a) for localized application of agents are obtained by feeding the upper inlets with different fluids and applying suction to the lower port, using a syringe pump. Perfusion flow (b, I-III are progressive images) for culture maintenance is performed by siphoning, using the distal ports (see text). Flow directions are indicated in the overview image (upper left).

### **Fig. S2. MSC localization is not restricted by BSA boundaries**

Progressive images of iMSC#3 cultivated in serum containing medium on a glass substrate that was coated alternately with FN ( $\Phi$ ) and BSA (\*) by the use of microchannels. Although the cells initially adhered to FN coated regions only (a, 1h after seeding), cells migrated out of these areas (b) and appeared unpatterned the next day (c).

### **Fig. S3. Simultaneous adipogenesis and osteogenesis on patterned PS substrates**

Dark field images showing one conventional (upper panel) and one microfluidic (lower panel) culture of iMSC#3 after 3 weeks in a 50:50 mixture of adipogenic and osteogenic medium. Adipocytes were stained by Oil Red-O (left panel) and subsequent staining of the same culture with Alizarin Red S revealed calcium produced by osteoblasts (right panel). The device top layer was removed for better end-point evaluation of the microfluidic culture, (d, dotted lines indicate former wall position) due to calcium deposition on the PDMS channel surface (being the cause of some blurring in c).