Figure legends

Supplemental Figure 1: Assessment of the "dead core" area

Representative Hoechst stainings of nuclei in hMO sections derived from three different WT NESC lines and cultured under shaking or fluidic conditions. The dashed line encircling the "dead core" was set at the border between high and low nuclei density. Usually, the densely packed core region gets loose and breaks out in the course of the sectioning process leaving a central hole (*). In some cases, remnants of the core region remain in the section (#) separated from the surrounding hMO by a gap.

Supplemental Figure 2: Computational modelling of medium fluidics

Red lines indicate the computed velocity field of the laminar medium flow in a fluidic chamber.

Supplemental Figure 3: Line specific reduction in SOX2⁺ NESC under fluidic culture conditions

(i) Immunofluorescence stainings of representative hMO sections derived from three different WT NESC lines and cultured under shaking or fluidic conditions (merge of SOX2 and Hoechst staining; scale bar = 500μ m). For better comparability of SOX2 quantities, the corresponding SOX2 staining (white) is depicted next to the merge. (ii) Mask-based image quantification (pixel ratios) of SOX2⁺ nuclei in corresponding immunofluorescence stainings (n = 3 per cell line and culture condition; **p<0.01, *p<0.05). (iii) Mean pixel ratio values for SOX2⁺ hMO of three different WT lines under fluidic conditions relative to those under shaking conditions (set to 100%).

Supplemental Figure 4: Line specific increase in LMX1⁺/TH⁺ mDA neurons under fluidic conditions

(i) Flow cytometry analysis (contour plots) of single cells obtained from dissociated MOs from three different WT NESC lines and cultured under shaking or fluidic conditions (blue lines = isotype control, red lines = target staining). Percentages of LMX1⁺/TH⁺ cells are indicated in the upper right corner of the plots. (ii) The bar chart summarizes the percentages of LMX1⁺/TH⁺ cells depicted above. (iii) Percentage of LMX1⁺/TH⁺ cells in hMO under fluidic conditions relative to those ones under shaking conditions (set to 100%).





Supplementary figures:











