SUPPLEMENTAL MATERIAL

Blaser et al., https://doi.org/10.1084/jem.20161616

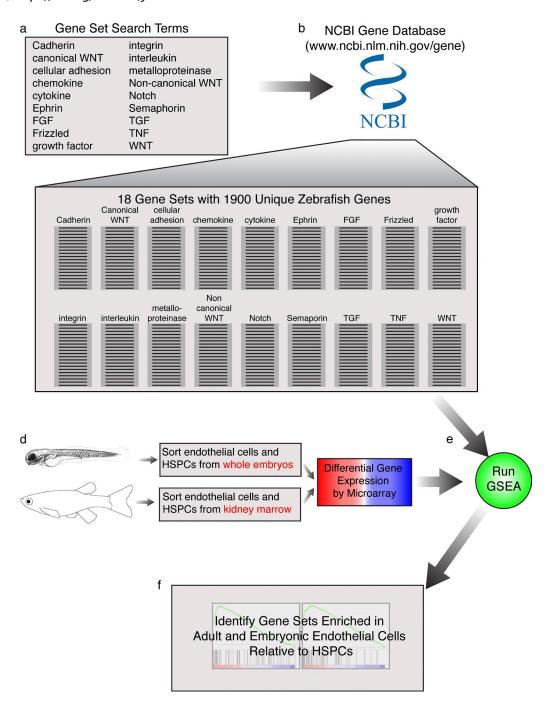


Figure S1. **Schematic for bioinformatic prediction of candidate genes.** (a) 18 terms were used to search the NCBI gene database (b) for zebrafish genes encoding various categories of extracellular and secreted molecules. (c) Gene sets were created from these lists in .gmx format. (d) Differential gene expression data on sorted endothelial cells and HSPCs was generated by microarray analysis (Zebrafish 1.0 ST array; Affymetrix) and converted to .gct format for GSEA. (e) GSEA software v2.2.0 was used for the analysis. The analysis was performed using the Signal2Noise metric and 1,000 permutations. (f) Leading edge gene set and component gene overlap between the adult and embryonic comparisons was identified using Venny 2.0.

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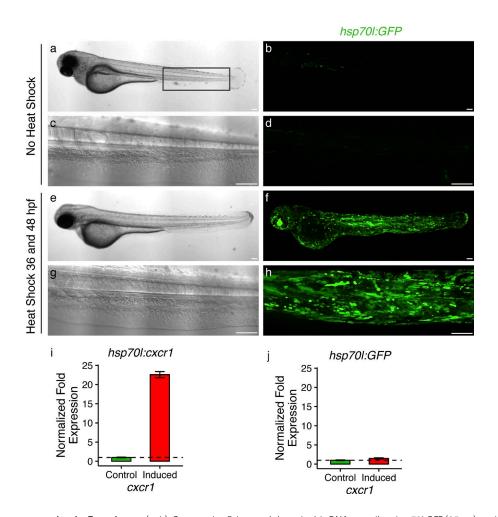
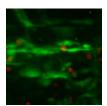


Figure S2. **Transgene expression in F₀ embryos.** (a–h) *Casper* zebrafish were injected with DNA encoding *hsp70l:GFP* (25 pg) at the 1-cell stage. Half of the animals underwent heat shock induction at 36 and 48 hpf (e–h; 40 degrees for 30 min each time). The remainder of the animals was kept at 28 degrees (a–d). All animals were imaged at 72 hpf; representative embryos are shown. Bar = 100 μ m. Box in panel a indicates the CHT as imaged in c, d, g, and h. (I and j) Casper zebrafish were injected with DNA encoding *hsp70l:cxcr1* (i) or *hsp70l:GFP* (j; 25 pg for each construct). Gene expression was induced by heat shock at 36 and 48 hpf (40 degrees, 30 min each time) in one half of each group. Total RNA was extracted at 72 hpf and quantitative PCR was performed. Expression of 18s was used as a reference. Normalized fold change of *cxcr1* expression relative to uninduced controls is plotted. The experiment was repeated twice with similar results.



Video 1. **Time lapse video microscopy of HSPC cuddling by endothelial cells in the CHT.** This movie shows the cuddling event from Fig. 4 f in its entirety.

Tables S1–S6 are available as Excel files. Table S1 shows Zebrafish extracellular and secreted factor gene sets. Table S2 shows GSEA results for the embryonic zebrafish gene expression dataset. Table S3 shows GSEA results for the adult kidney marrow gene expression dataset. Table S4 shows chemokine leading edge gene set for embryonic endothelial cells. Table S5 shows chemokine leading edge gene set for adult kidney marrow endothelial cells. Table S6 shows the intersection of embryonic and adult chemokine leading edge gene sets.