

Supplementary materials

Gene Name	Forward 5'-3'	Reverse 5'-3'
<i>heg1</i> -exon1	GTGGAAACCGCTATCGCCTG	GTCGCAGGACCTAGTTGACTG
<i>heg1</i> -qPCR	CCACTGCCACAGCCGTGGATC	GTCAGATTGAAGATGTTCTG
<i>β-actin</i>	ATGGATGAGGAAATCGCTGCC	CTCCCTGATGTCTGGGTCGTC
<i>myh6</i>	GACATGGCGATGCTGACGTTTC	GATAAGCATTATCTGAGATG
<i>ifabp</i>	CTGAAGATCACCCCTGGAGCAG	GTTGTCCTTGCGTGTGAAAG
<i>cmlc2</i>	GAGCTGGAGTCCATGCTAAC	CATTAGCAGCCTCTTGAACTCA
<i>sox7</i>	GGAGACCCATGAACGCCTT	GCTCGGCTTCCTCCACATAT
<i>flk1</i>	GACCATAAAACAAGTGAGGCAGAAG	CTCCTGGTTTGACAGAGCGATA
<i>scl</i>	GCCAATGGTGAAGTTGTGAGT	CGTCTGCTCTCTACCTGGAT
<i>flt4</i>	ATTACAACTGCGTGCCGTTT	TGTCAACATGGCTCCTCTGT
<i>c-myb</i>	TGAATCATCACGGGTGCCAT	TGTTGTCCCTTCAGCTCGTT
<i>vegfa</i>	ATCCTCCCTCCTGTGAATGC	CCTCAAAGTTGGATCGGTGG

Table S1. List of all primer sequences used for genotypes identify and Quantitative PCR.

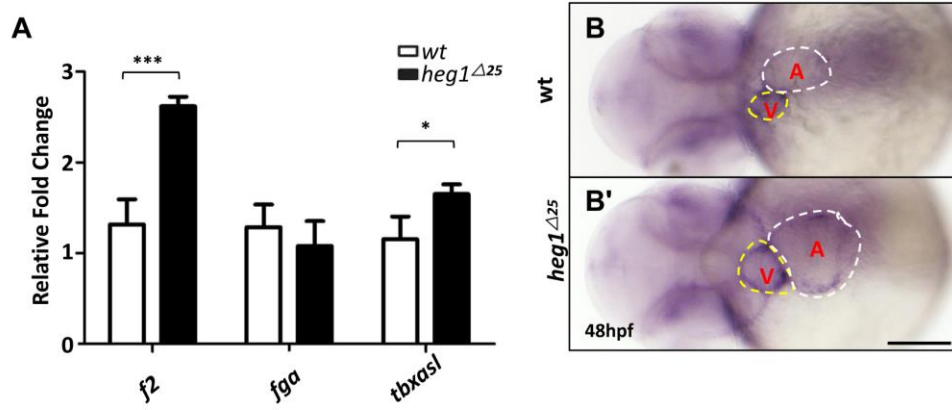


Figure S1. Analysis of *heg1*^{Δ25} mutant embryos. **(A)** The expressions of thrombotic markers, as determined by qRT-PCR, were significantly changed in *heg1*^{Δ25} mutants at 48 hpf. **(B,B')** Representative images of the *heg1*^{Δ25} and wt embryos at 48 hpf stained for the heart marker *bmp4*. Note the enlargement heart in *heg1*^{Δ25} mutants (V: Ventricular, yellow dotted-line boxes; A: atria, white dotted-line boxes, ventral view). Data are represented as mean ± SE from three independent experiments, **p* < 0.05, and ****p* < 0.001 (Student's t-test).

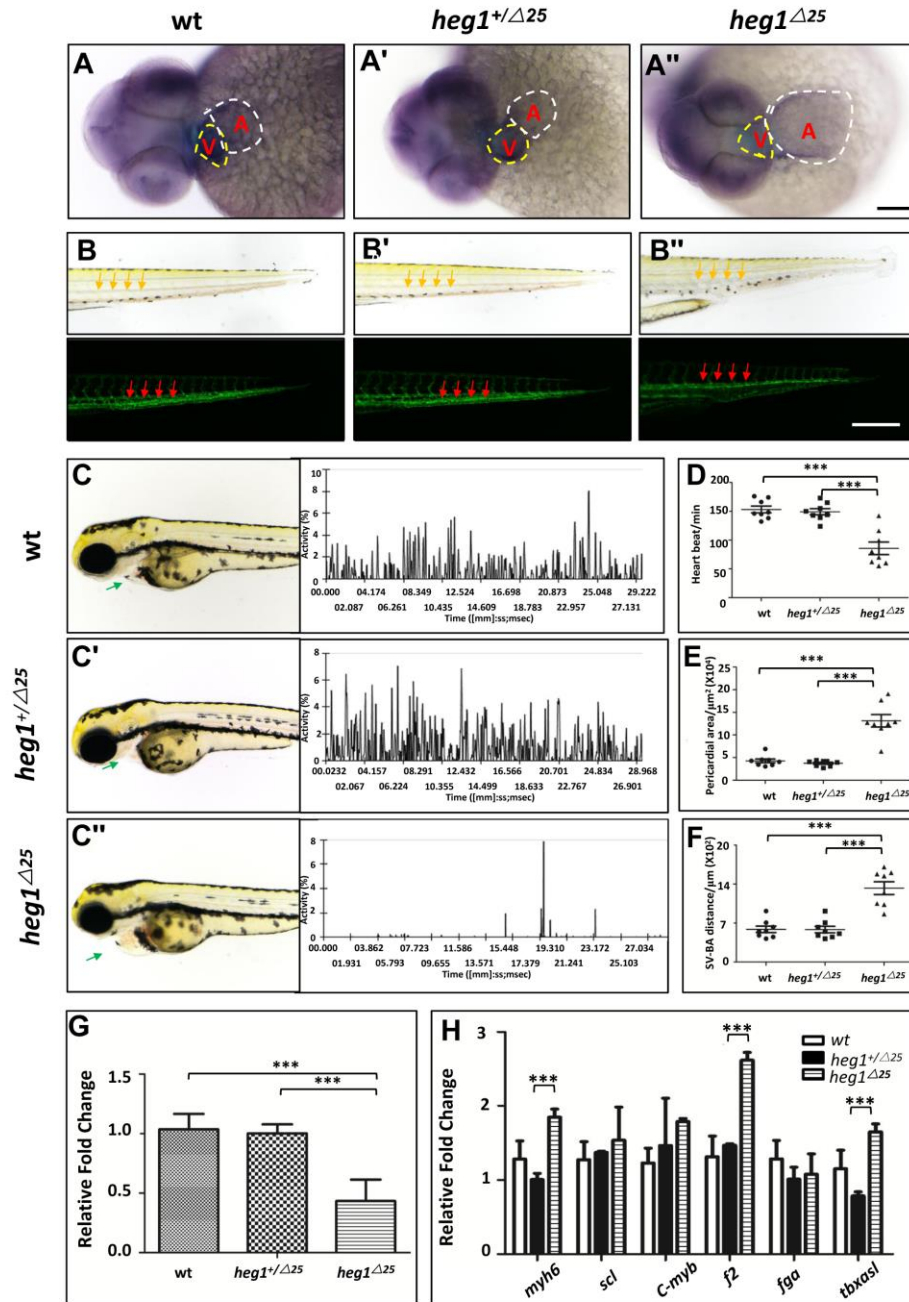


Figure S2. Comparison of wt, *heg1*^{+/Δ25} heterozygous and *heg1*^{Δ25} homozygous mutants. (A,A',A'') Representative images of the wt, *heg1*^{+/Δ25} heterozygous and *heg1*^{Δ25} homozygous mutant embryos at 48 hpf stained for the heart marker *cmlc1*, note the enlargement heart in *heg1*^{Δ25} mutants but not in wt and *heg1*^{+/Δ25} heterozygous embryos (V: Ventricular, yellow dotted-line boxes; A: atria, white dotted-line boxes, ventral view). (B,B',B'') Lateral view of zebrafish larvae at 96 hpf. Representative images of wt, *heg1*^{+/Δ25} heterozygous and *heg1*^{Δ25} homozygous mutants embryos, exhibiting blood congestion (yellow arrows), and dilation of dorsal aorta (DA) lumen (red arrows) in *heg1*^{Δ25} embryos but not in wt and *heg1*^{+/Δ25} heterozygous embryos. (C,C',C'') Lateral view of zebrafish larvae at 72 hpf. Representative images of wt, *heg1*^{+/Δ25}, and *heg1*^{Δ25} embryos. The movement ratio of RBCs based on changes in pixel density of PVC. (D) Heart rate in wt, *heg1*^{+/Δ25}, and *heg1*^{Δ25} zebrafish larvae (n=8 embryos/group). (E) The pericardial area in wt, *heg1*^{+/Δ25}, and

heg1^{Δ25} zebrafish larvae (n=8 embryos/group). (F) The SV-BA distance in wt, *heg1*^{+/^{Δ25}}, and *heg1*^{Δ25} zebrafish larvae (n=8 embryos /group). (G) qRT-PCR confirmation that *heg1* expression was significantly decreased in *heg1*^{Δ25} embryos, no difference between wt and *heg1*^{+/^{Δ25}}, n=30 embryos per group. (H) The expressions of cardiovascular markers, as determined by qRT-PCR at 48 hpf. Data are represented as mean ± SE from three independent experiments, ****p* < 0.001 (Student's t-test).