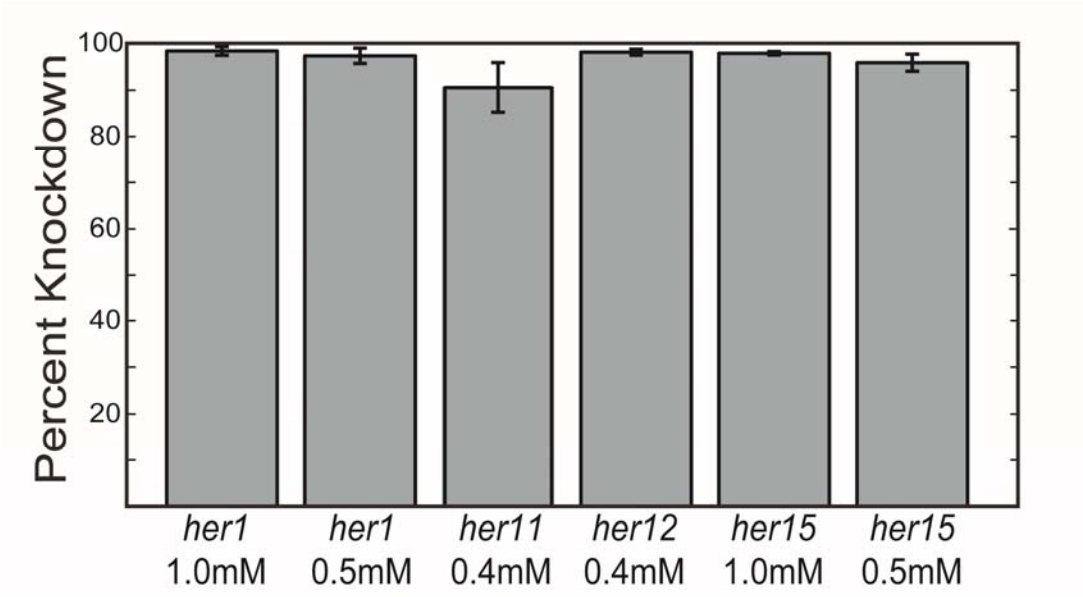

Figure S1 Modified luciferase assay was used to determine knockdown efficiency for all *her/hes* morpholinos. For each measurement, three experimental replicates were performed. Injection of 1.0mM or 0.5mM *her1* morpholino results in 98.3 ± 0.9% or 97.3 ± 1.6% knockdown, respectively. The least efficient knockdown is by *her11* morpholino (90.5 ± 5.3%). *her12* morpholino is 98.1 ±0.7% efficient, and 1.0mM and 0.5mM concentrations of *her15* morpholino are 97.8 ± 0.4% and 95.8 ± 1.8% efficient, respectively.

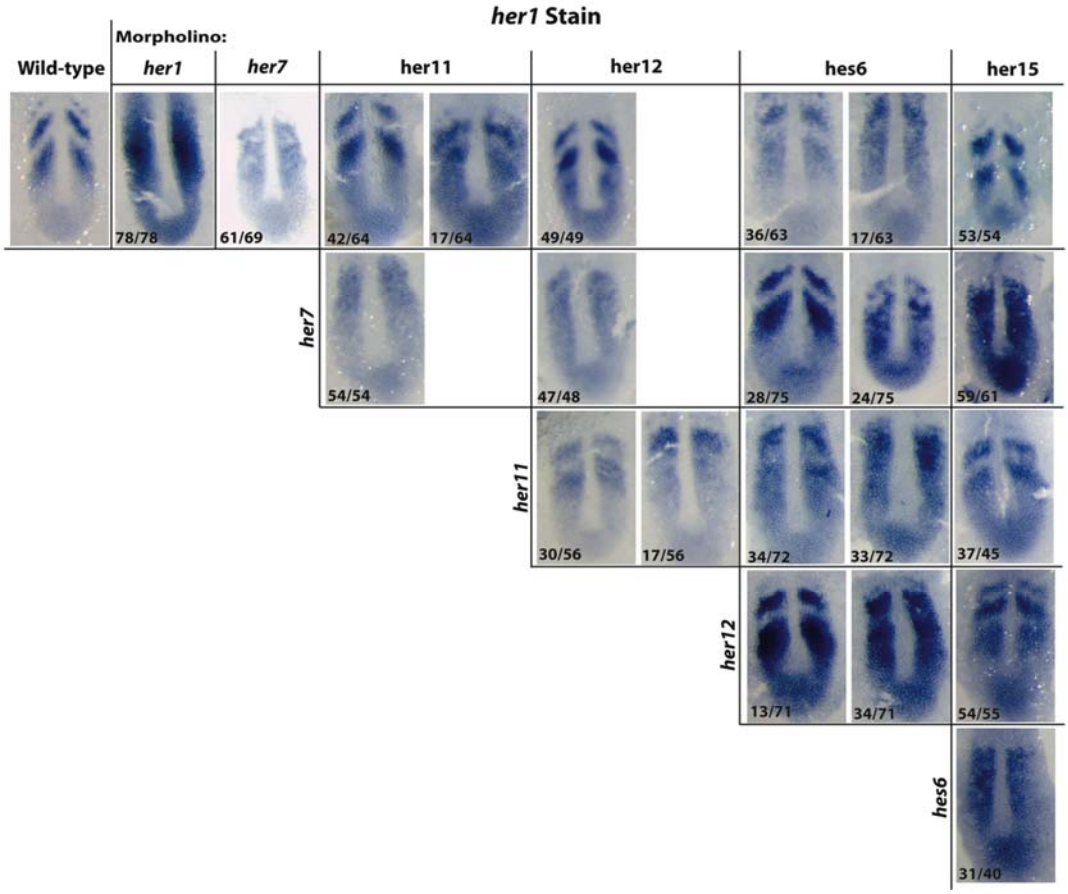

Figure S2 Summary of *her1* patterning defect in single and double *her/hes* knockdowns, excluding combinations with *her1* MO. Injection of *her1* MO stabilizes mRNA and obfuscates *her1* patterning defects. For all knockdowns, at least three experimental replicates were performed. Number of embryos analyzed is given in bottom left of each image.

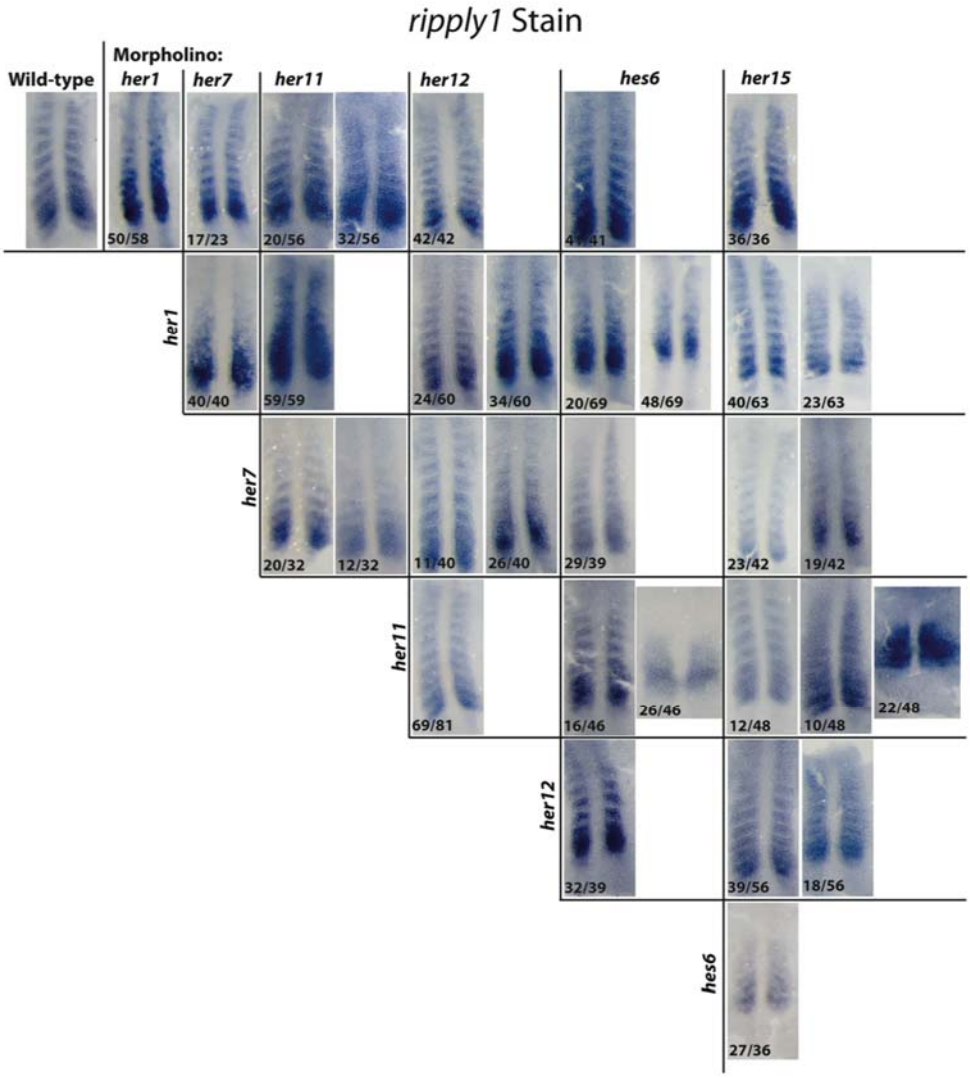

Figure S3 Summary of *ripply1* expression patterns for all *her/hes* knockdowns. For all knockdowns, three experimental replicates were performed. Number of embryos analyzed is given in bottom left of each image.

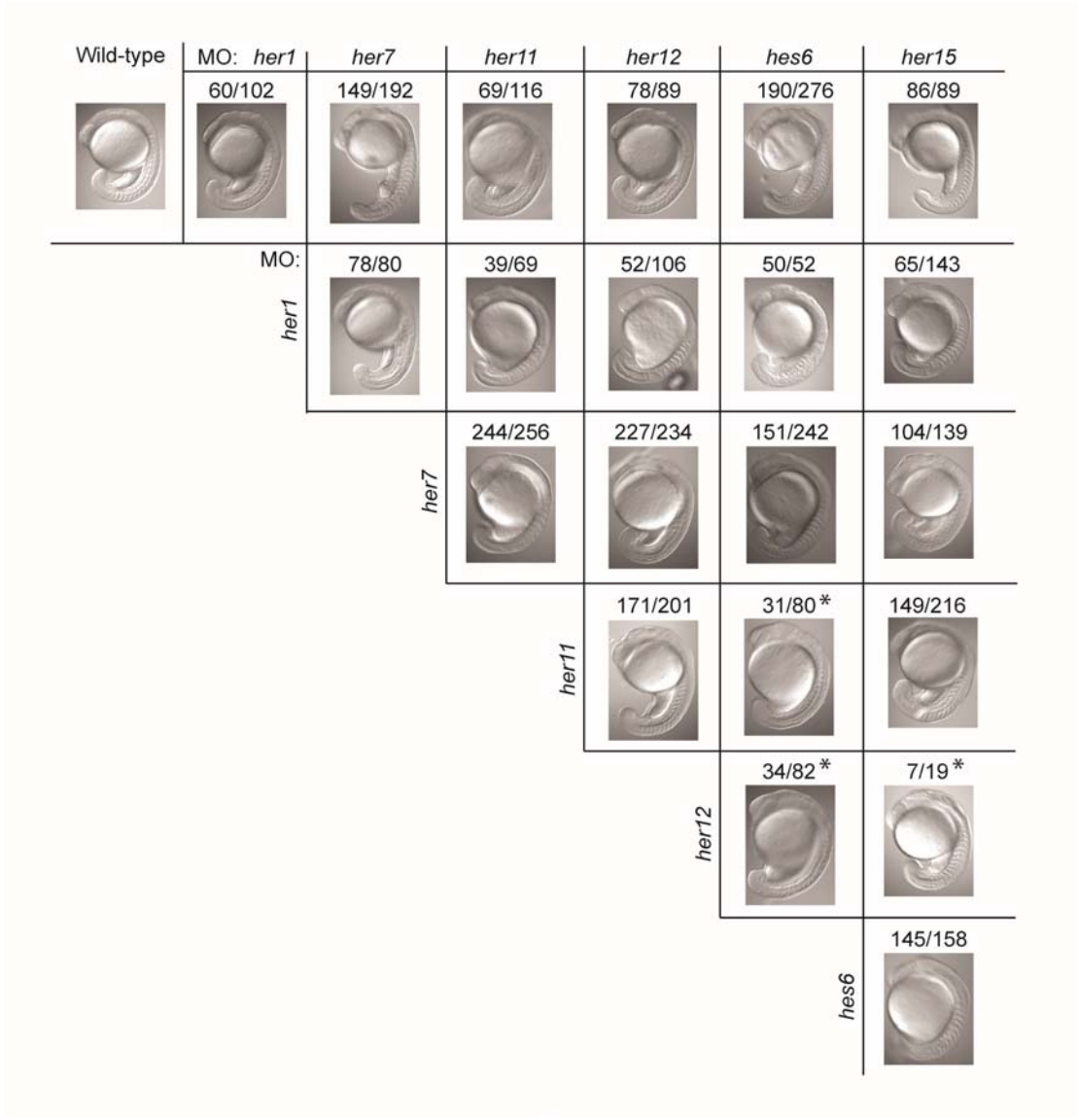

Figure S4 DIC images for all single and pairwise knockdowns. Pictured is the most prevalent, except for conditions marked with an asterisk. Here, a similar proportion of embryos had somite defects as appeared wild type. Shown are embryos exhibiting segment border defects. Number of embryos analyzed is given above each image. Fish with axis defects were excluded from this analysis, and additional details can be found in Table S4. In total, approximately 3000 embryos were analyzed.

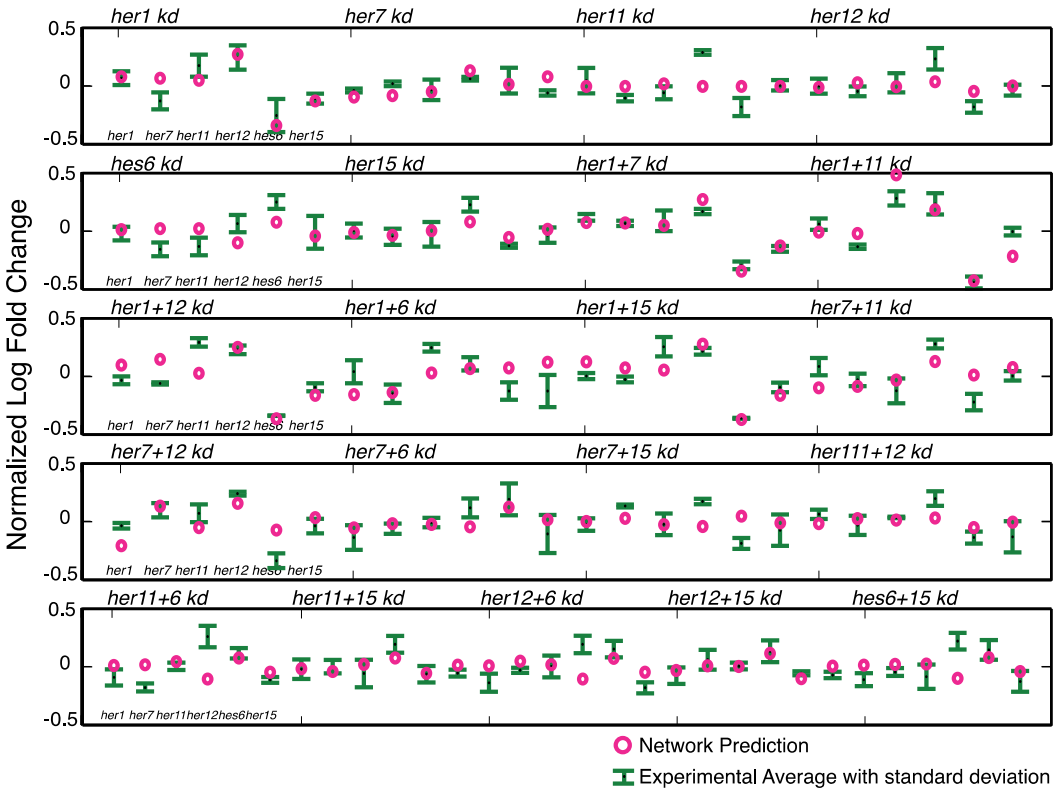

Figure S5 Comparison of qPCR results from all single and double *her/hes* perturbations with fit from predicted GRN. qPCR values are shown as normalized log fold change. For each knockdown condition, results for all six genes: *her1, her7, her11, her12, hes6,* and *her15* are given. Knockdowns are labeled above each graph, and the order of genes shown is constant. For simplicity, each gene is labeled only once per plot. The order is consistent throughout. Experimental results are shown with standard deviation in green. Five replicates were performed for all single knockdowns, and three replicates for all double knockdowns. Computed qPCR values from the predicted GRN are shown as pink circles.

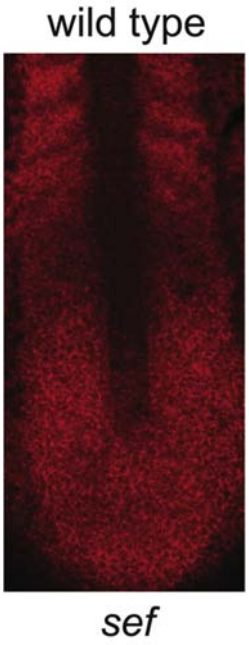

Figure S6 Confocal slice of fluorescent in situ hybridization for *sef* in wild type 8-10-somite stage embryo. A single experiment was performed, with eight embryos imaged by confocal.

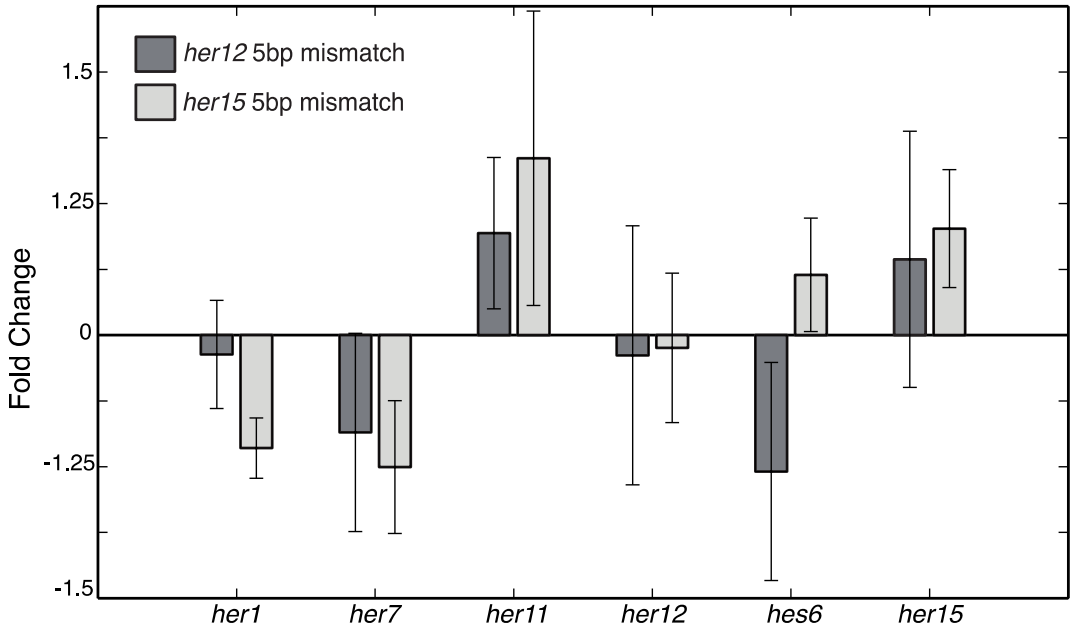

Figure S7 Summary of qPCR data from embryos injected with 0.4mM or 1mM *her12* 5bp-mismatch morpholino or *her15* 5b-mismatch morpholino, respectively. Shown are averages from three replicates with standard error. No response is statistically different from zero.

Table S1 Morpholino and Primer Sequences

A

Morpholino	Sequence	Concentration	Reference
her1 mo1	CGA CTT GCC ATT TTT GGA GTA ACC A	1mM (or 0.5mM)	(HOLLEY *et al.* 2002)
her7 antiATG	CAT TGC ACG TGT ACT CCA ATA GTT G	0.4mM	(GAJEWSKI *et al.* 2003)
her11 ORF	CGT CAT GTT GAA AGT CGG TGT GCT	0.4mM	(SIEGER *et al.* 2004)
her12 ORF2	GTG CCA TGT CTG TGC TCG AAC AGC T	0.4mM	Designed by GeneTools
hes6 MO1	TGC AGT TCA GGA CGC TTG AAT GGG	0.6mM	(KAWAMURA *et al.* 2005)
her15.1 ORF	AGT ATT CAG TCA TAT ATG CAG GAG C	1mM (or 0.5mM)	Designed by GeneTools

B

qPCR Primers	Sequence	Reference
bactin F	CGC GCA GGA GAT GGG AAC C	(KEEGAN *et al.* 2002)
bactin R	CAA CGG AAA CGC TCA TTG C	(KEEGAN *et al.* 2002)
her1 intron F	TGC ATG CCT TTC CAC TCT CCC TAA C	(STULBERG *et al.* 2012)
her1 R	ATG GCA TCT GGG GTC TCC TT	(STULBERG *et al.* 2012)
her7 intron F	TGT CAA CTC TTA TTT TTG TAG CAA CC	(STULBERG *et al.* 2012)
her7 R	CGG GCT GCT TTT TGA AGA CA	(STULBERG *et al.* 2012)
her11 F	TAG ACC TTG CAG TTC AGT ACA TCA A	
her11 intron R	GAC AAG AAA GAA AAA TTA GGC AAA TGA	
her12 F	TTG GCA CAA CAT AAA GCA AGC T	
her12 intron R	TCA AAT TAG TTG AAA TGC TGG AAT G	
hes6 F	GCA ACA CTC ACG ACG AGG AT	(STULBERG *et al.* 2012)
hes6 intron R	AGT TGT GGG AAA CGT CGA CAA	(STULBERG *et al.* 2012)
her15 intron F	ACG TTA GCC AAA CGA CAC AGA	
her15 R	GCT GCT CGA TGC AGT TGT TG	
sef intron F	TGA GCT CAC AGC CCT TCT CA	(STULBERG *et al.* 2012)
sef R	GCA GAA AAG ATG GCG GAA AG	(STULBERG *et al.* 2012)
axin2 intron F	GCG CGC ACA AAG TAG ACG TA	(STULBERG *et al.* 2012)
axin2 R	CCA GCA GCA AAG CCT TCA GT	(STULBERG *et al.* 2012)

C

Probe Primers	**Sequence**	**Reference**
sef F	TCA CGG AGA CTT GCG CAT TTT T	
sef R	AGC ACA ACA GGT TAT CCC GCA	
ripply1 F	ATG AAT TCT GTG TGC TTT GCC A	(ZHANG *et al.* 2008)
ripply1 R	GTT GAA AGC TGT GAA GTG ACT	(ZHANG *et al.* 2008)

(A) Morpholino, **(B)** qPCR and **(C)** in situ hybridization primer sequences used. Morpholinos and primers that have been previously published are referenced on right. Morpholinos for *her12* and *her15* were designed by GeneTools, and validated using 5bp-mismatch controls (Figure S7). Further, the efficiency of these morpholinos was calculated as ~98% and they recapitulated published morphant phenotypes (SHANKARAN *et al.* 2007).

Table S2 Parameter Values and Ranges

Parameter	Description	Initial Value	Range and Unit	Average Value from parameter search	Average Value for min hes6 oscillations
δm	mRNA degradation	0.31	min^{-1}		
δm6	*hes6* mRNA degradation	0.31	0-1 min^{-1}	0.72	0.65
δP	protein degradation	0.15	min^{-1}		
δP6	Hes6 protein degradation	0.15	0-1 min^{-1}	0.12	0.061
δD	dimer degradation	0.15	min^{-1}		
δD6	Hes6/Hes6 degradation	0.15	0-1 min^{-1}	0.032	0.015
βm	maximum transcription	150	min^{-1}		
βm6	maximum *hes6* transcription	150	0-10,000 min^{-1}	1444	93
βP	translation	18.5	min^{-1}		
βP6	Hes6 translation	18.5	0-100 min^{-1}	18.3	62.8
r	critical concentration	13,900			
s	strength scaling factor	162.7	0-10,000	61.3	64.7
k+	dimer on rate	0.02	min^{-1}		
k-	dimer off rate	1			
k- Hes6/6	Hes6/Hes6 off rate	1	0-10	5.2	3.0
n	relative difference between weak and strong dimers	1.26	1-10	1.31	1.31
τm	transcriptional delay	5	min		
τ	translational delay	1.5	min		

Biochemical parameters used in computational model, their initial values and the average fitted values corresponding to Figure 5B. Transcription, translation and degradation rates are from (CINQUIN 2007) along with *r* and *S* values, which were scaled to account for the different number of repressive dimers in our simulation and the assumption of independent, rather than competitive, repressors.

Files S1-S8

Available for download at http://www.genetics.org/lookup/suppl/doi:10.1534/genetics.114.163642/-/DC1

File S1 qPCR Values. Summary of qPCR values in **(A)** log fold change and **(B)** normalized log fold change (*her/hes* genes only) along with standard error. Three replicates were performed for all double knockdowns, and five replicates for single knockdowns of *her/hes* genes. Three replicates were performed for *sef* and *axin2* measurements. File S1 is available for download as an Excel file (.xlsx).

File S2 Morphological Defects. Summary of morphological defects for all single and pairwise knockdowns. The number of experimental replicates is listed for each knockdown condition. "Posterior Defect" refers to morphants with disrupted segment borders that begin anywhere after somite eight. "Anterior Defect" typically indicates that somites one through four are perturbed, though the *her1+her12* MO condition is slightly stronger, with up to ten anterior somites affected. "Middle" somites are those between somite four and ten. Embryos with "Anterior and Posterior Defects" typically represent an additive phenotype, with the first four somites affected, and borders posterior to somite ten disrupted. File S2 is available for download as an Excel file (.xlsx).

File S3 MOtesterAP.m File S3 is available for download as a Matlab file (.m).

File S4 MOtesterAP_ant.m File S4 is available for download as a Matlab file (.m).

File S5 MOtesterAP_post.m File S5 is available for download as a Matlab file (.m).

File S6 MO_cost_func.m File S6 is available for download as a Matlab file (.m).

File S7 getAvgSpectrum.m File S7 is available for download as a Matlab file (.m).

File S8 JSdivergence.m File S8 is available for download as a Matlab file (.m)

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