

Figure S1 (Related to Figure 1) Expression of innate immune response gene in X. tropicalis and X. laevis RNA-seq datasets. Expression of Tp53inp1, Tp53 and C3ar1 genes in X. tropicalis (A) and X. laevis (B) in transcripts per million from publically available RNA-seq datasets.

9

10

18

20

26

44





Stage 26



C Morpholino Target: control

Stage 34

cytokine-mediated signaling pathway response to interleukin-1 regulation of I-kappaB kinase/NF-kappaB signaling

extracellular matrix disassembly protein complex oligomerization positive regulation of protein deubiquitinatior

esponse to virus esponse to mechanical stimulus

regulated exocytosis leukocyte migration response to extracellular stimulus stress-activated protein kinase signaling cascade

response to oxidative stress leukocyte cell-cell adhesion

positive regulation of reactive oxygen species metabolic process response to inorganic substance

multi-multicellular organism process response to arsenic-containing substance positive regulation of transcription from RNA polymerase II promoter in response to endoplasmic reti





Stage 34



Figure S2 (Related to figure 2 and 3) Gene ontology analysis of differentially expressed genes by various morpholinos in Gentsch et al. datasets. Gene ontology analysis of the control MO and T/T2 morpholino at stage 26 (A,B) and stage 34 (A,B). We used the criteria overlap \geq 3, p-value \leq 0.01 and enrichment \geq 1.5 to define significance. Red asterisk (*) indicates innate immunity related GO terms.



Figure S3 (Related to figure 2 and 3) Gene ontology analysis of differentially expressed genes by various morpholinos in available X. tropicalis datasets. Gene ontology analysis of the Cdx4 MO (A), Cdx1/2/4 MO (B), and Wnt8a MO (C). We used the criteria overlap \geq 3, p-value \leq 0.01 and enrichment \geq 1.5 to define significance.



Figure S4 (Related to figure 2 and 3) Gene ontology analysis of differentially expressed genes by various morpholinos in available Xenopus laevis datasets. Gene ontology analysis of the β -catenin MO at stage 9 and 10 (A,B), Exosc9 MO (C) and Ptbp1 MO (D). We used the criteria overlap \geq 3, p-value \leq 0.01 and enrichment \geq 1.5 to define significance.

Accession	Experimental Morpholino(s)	Control Morpholino	Replicates	Organism	Collection Stage	Collected Tissue	Citation
GSE72657	Wnt8a	Yes	3	X. tropicalis	St 10	Whole embryo	Nakamura et al., 2016
GSE53654	Foxh1	No	1	X. tropicalis	St 10	Whole embryo	Chiu et al., 2014
GSE56169	E2a	Yes	2	X. tropicalis	St 10	Whole embryo	Wills et al., 2015
DRA000516, DRA000517, DRA000518, DRA001093, DRA001094, DRA001095	Lim1+Otx2+Otx5, Gsc	Yes	1	X. tropicalis	St 11	Whole embryo	Yasuoka et al., 2014
GSE86382	Mov10	Yes	2 or 3	X. laevis	St 10	Whole embryo	Skariah et al., 2018
GSE93195	Beta-catenin	No	2 or 3	X. laevis	Stage 9 and 10	Whole embryo	Ding et al., 2017
GSE76995	Tbp + Tlf + Tbp2, Gcn5 (Antisense DNA)	No	2	X. laevis	St 10	Whole embryo	Gazdag et al., 2016
GSE76915	Ascl1	Yes	3	X. laevis	St 10	Whole embryo	Gao et al., 2016
GSE50593	Rfx2	No	2	X. laevis	Stage 20	Isolated ectoderm	Kwon et al., 2014
GSE45786	Tcf21	Yes	1	X. laevis	St 44 - 45	Whole embryo	Tandon et al., 2013
GSE71006	Cdx1, Cdx2, Cdx4, Cdx1/2/4	Yes	3	X. tropicalis	Stage 14	Whole embryo	Marlétaz et al., 2015
GSE48663	Tbxt/Tbxt2	Yes	3	X. tropicalis	Stage 32	Whole embryo	Gentsch et al., 2013
GSE89271	Foxn4	No	2	X. laevis	Stage 18	Isolated ectoderm	Campbell et al., 2016
PRJEB8711	Ptbp1, Exosc9	No	1	X. laevis	Stage 26	Whole embryo	Noiret et al., 2016
PRJNA266550	Tra2b	Yes	3	X. laevis	Stage 14	Whole embryo	Dichmann et al., 2015
GSE96655	Tbxt/Tbxt2	Yes	3	X. tropicalis	Stage 24 and Stage 36	Whole embryo	Gentsch et al., 2018

Table S1 (Related to Figure 1-4) List of RNA-seq datasets that contained a morpholino experiment used in this study.

Tropicalis	Tp53inp1		Тр	53	C3ar1	
	Reported	Measured	Reported	Measured	Reported	Measured
Control MO	0.97	0.84	1.06	0.96	0.97	1.01
Tra2b MO	1.05	1.07	1.37	1.36	1.43	1.22
Cdx2 MO	1.15	1.35	0.84	0.93	1.55	1.57
Cdx1 MO	1.23	1.33	1.06	1.05	2.69	2.5
Cdx4 MO	2.43	2.55	1.38	1.39	3.19	3.64
CdxA MO	2.06	2.06	1.37	1.27	5.27	4.63
Control MO	1.58	1.48	1.62	1.52	4.37	5.37
T/T2 MO	3.92	3.7	2.67	2.37	14.08	14.8
Laevis	Tp53inp1.L		Tp!	53.L	C3ar1.L	
	Reported	Measured	Reported	Measured	Reported	Measured
Ptbp1 MO	0.87	1.15	0.74	0.75	0.79	0.99
Rfx2 MO	1.74	1.54	1.68	1.36	0.9	1.03
Exosc9 MO	1.62	1.6	1.57	1.52	3.59	0.72

Table S2 (Related to Figure 1) Comparison of fold change generated between our analysisand the Gentsch et al. (2018) analysis.