

Supplementary information

Ftx is dispensable for imprinted X-chromosome inactivation in preimplantation mouse embryos

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Supplementary table S1. Mating of *Ftx*-deficient mice (line #77)

Cross (♀×♂)	Male pups		Female pups		Total pups	Mean litter size
+/+ × -/Y	+/Y		+/-		84	8.4

Cross (♀×♂)	Male pups		Female pups		Total pups	Mean litter size
	+/Y	-/Y	+/+	-/+		
+/- × +/Y	20	19	19	12		
	39		31		70	7.8

Cross (♀×♂)	Male pups		Female pups		Total pups	Mean litter size
	+/Y	-/Y	+/-	-/-		
+/- × -/Y	33	36	26	25		
	69		51		120	7.1

Cross (♀×♂)	Male pups	Female pups	Total pups	Mean litter size
	-/Y	-/-		
-/- × -/Y	12	17	29	5.8

Supplementary Table S2. A list of the PCR primer sets used in this research.

	gene name	primer name	sequenece (5' →3')	
Construction of <i>Ftx</i> disruption vector	Short arm	Forward 169	gcctctccaaaggctagaattc	
	Short arm	Reverse 170	caaccactggcatggtgata	
	Long arm	Forward 185	cacggcccagtgttgccaaat	
	Long arm	Reverse 186	tgctaatgggcaaatccaaagcca	
	Short arm	Forward 174	caggaggcctacactacagtaaca	
5' -RACE PCR	<i>Ftx</i>	GSP1 159	gccaaagcacacatcagcttgagttgg	
	<i>Ftx</i>	GSP2 160	aaagttcagtcctatggccaactggctc	
validation of recombination	<i>Ftx</i> short arm	Forward 173	ggagttgggatatagctcacct	
		Reverse 001	gccttctatcgcttcttgacgagttcttc	
	<i>Ftx</i> long arm	Forward 040	tgatgtggaatgtgtgag	
		Reverse 187	gccatgctgaatgtgtgaaggcc	
Genotyping	<i>Ftx</i>	Forward 233	aatagcctgccccaaagagtt	
		Reverse 234	gtcaaaggatcccgttgaa	
		Forward 330	ccgcgccacatgagcacaac	
RT-PCR	<i>Ftx</i>	Forward 211	gcccattcttgctctgtt	
		Reverse 212	tcgtttgtccacatctcttc	
		Forward 541	tatgccacctagcctttctaca	
		Reverse 542	atctctcaaaagcggcataat	
qPCR	beta-actin	Forward 051	aagtgtgacgttgacatccg	
		Reverse 052	gatccacatctgtggaagg	
	<i>Ftx</i>	Forward 207	atctcttgctcctctt	
		Reverse 208	tgtgtccagggtctctgt	
	<i>Xist</i>	Forward 236	cagagtagcaggactgaagag	
		Reverse 237	gctggtctgtctatctgtggg	
polymorphic analysis(RFLP)				Restriction enzyme
<i>Fmr1</i>	Forward 079	Reverse 080	cttaacactcagggcagg	RsaI
		Reverse 080	cttccctgaactctgcatcc	
<i>Fgd1</i>	Forward 084	Reverse 085	tcacacaaagccacctaagc	HhaI
	Reverse 085	attgactgcattgggagtg		
<i>Pdha1</i>	Forward 086	Reverse 087	ttccagcatatgctgacttt	TaqI
	Reverse 087	tgcaaggcatgaagtgata		
<i>Rnf12</i>	Forward 094	Reverse 091	ctggagagtctcagatgatgta	HaeIII
	Reverse 091	ggtcggcacttctgtactgc		
<i>Xist</i> exon1	Forward 105	Reverse 106	ctaaaactcagcccgttcca	SpeI
	Reverse 106	gcaacccagcaatagtc		
<i>Xist</i> exon 7	Forward 107	Reverse 108	gcccaggtcacattatggt	SacI
	Reverse 108	ctccaattctgggctcaag		
<i>Slc16a2</i>	Forward 109	Reverse 110	tgtacggctcacctcattagg	BseRI
	Reverse 110	ggaagtgaagcattgttc		
<i>Ftx</i>	Forward 116	Reverse 117	gccatctgatgcatgttg	HpyCH4 III
	Reverse 117	ggtgtggtctctgtctcc		

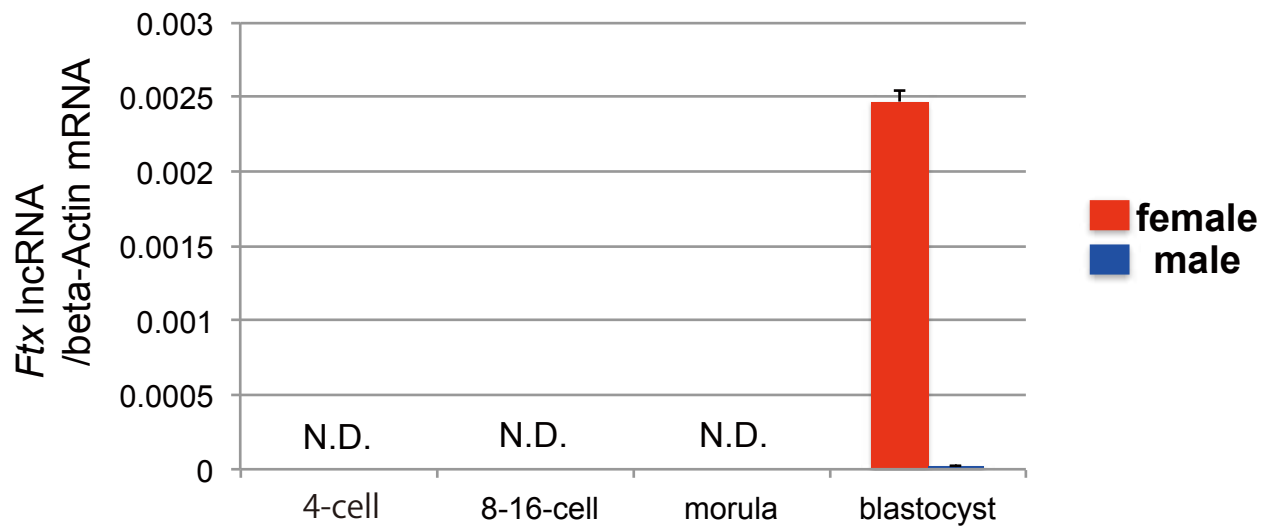


Figure S1. Expression of *Ftx* in males and females before the blastocysts stage

The expression level of *Ftx* was quantified by q-PCR in wild type embryos at preimplantation stages. 4-cell, 8-cell, and morula samples were mixture of both sexes. Only blastocyst sample was separated by sex. N.D.= not detected

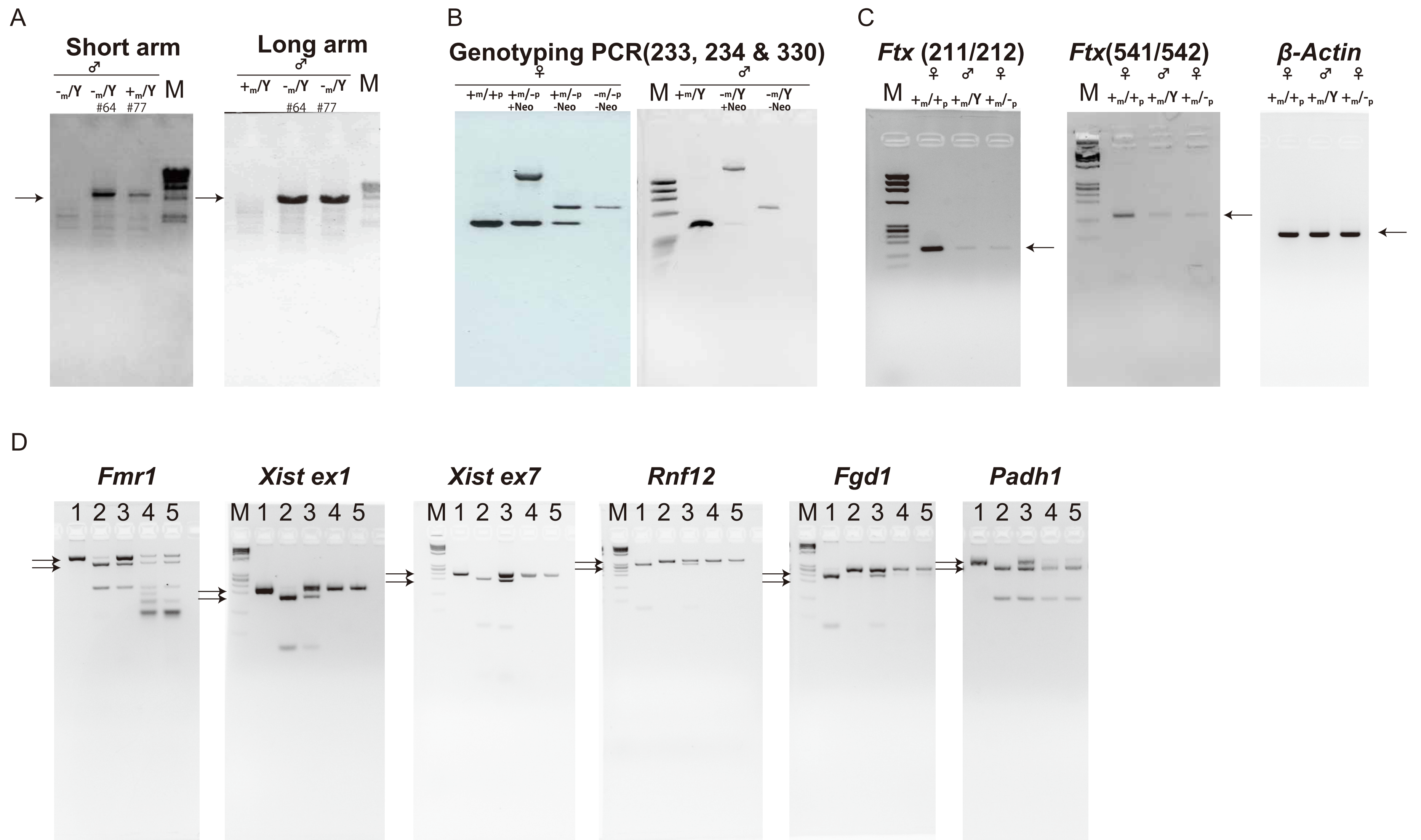


Figure S2. Full-length electrophoretic gels used to analyse the RT-PCR products and genotyping PCR.

Primers used for RT-PCR or genotyping PCR are indicated at the top of gel images. Arrows point to the PCR-amplified specific bands. M; molecular weight marker.

(A) Full-length images of cropped bands shown in Figure2b, (B) Full-length images of cropped bands shown in Figure2c, (C) Full-length images of cropped bands shown in Figure2d.

(D) Full-length images of cropped bands shown in Figure3c. 1: B6 genomic DNA, 2: JF1 genomic DNA, 3: (JF1×B6)F1 genomic DNA, 4: (JF1×B6)F1 RT-PCR products, 5: (JF1×KO)F1 PT-PCR products.