

Supplementary Information

(A)

Pseudogenes with <i>RFWD2</i> origin		Locations	Exon6-Exon2 back-splicing junction	<i>RFWD2</i> exons in pseudogenes	Counts of As in poly(A)	
Putative <i>circRFWD2</i> -derived pseudogenes (42)	High-confidence <i>circRFWD2</i> -derived pseudogenes (33)	Back-splicing junction sequences ≥ 20 bp (7)	chr1:117842919-117843301	exon6(42bp)-exon2(60bp)	6,2,3,4,5	12
			chr1:118040603-118040985	exon6(42bp)-exon2(60bp)	6,2,3,4,5	13
			chr6:130401556-130401938	exon6(42bp)-exon2(60bp)	6,2,3,4,5	12
			chr10:126586694-126586843*	exon6(42bp)-exon2(60bp)	6,2,3	N/A
			chr14:15298521-15298904	exon6(42bp)-exon2(60bp)	6,2,3,4,5	12
			chr15:103804648-103804954	exon6(42bp)-exon2(60bp)	6,2,3,4,5	5
			chrX:65086804-65087178	exon6(35bp)-exon2(60bp)	6,2,3,4,5	13
	Back-splicing junction sequences < 20 bp (26)	chr1:155766132-155766276*	exon6(13bp)-exon2(60bp)	6,2,3	N/A	
		chr2:7631062-7631345	exon6(13bp)-exon2(60bp)	6,2,3,4,5	7	
		chr2:29093836-29094119	exon6(13bp)-exon2(60bp)	6,2,3,4,5	5	
		chr2:34830827-34831109	exon6(13bp)-exon2(60bp)	6,2,3,4,5	7	
		chr3:4504816-4505071	exon6(13bp)-exon2(60bp)	6,2,3,4,5	4	
		chr3:4766084-4766365	exon6(13bp)-exon2(60bp)	6,2,3,4	10	
		chr3:6378332-6378615	exon6(11bp)-exon2(60bp)	6,2,3,4,5	7	
		chr3:155754971-155755247	exon6(13bp)-exon2(60bp)	6,2,3,4,5	6	
		chr4:7763264-7763548	exon6(13bp)-exon2(60bp)	6,2,3,4,5	5	
		chr4:94061360-94061643	exon6(13bp)-exon2(60bp)	6,2,3,4,5	9	
		chr7:28235651-28235934	exon6(13bp)-exon2(60bp)	6,2,3,4,5	6	
		chr8:29417354-29417637	exon6(13bp)-exon2(60bp)	6,2,3,4,5	6	
		chr10:104874215-104874498	exon6(13bp)-exon2(60bp)	6,2,3,4,5	8	
		chr12:94731107-9473390	exon6(13bp)-exon2(60bp)	6,2,3,4,5	6	
		chr14:19263202-19263485	exon6(13bp)-exon2(60bp)	6,2,3,4,5	7	
		chr14:116729593-116729876	exon6(13bp)-exon2(60bp)	6,2,3,4,5	7	
		chr15:3953006-3953272	exon6(13bp)-exon2(60bp)	6,2,3,4,5	4	
		chr15:5714020-5714303	exon6(13bp)-exon2(60bp)	6,2,3,4,5	7	
		chr15:13713972-13714255	exon6(13bp)-exon2(60bp)	6,2,3,4,5	9	
		chr15:14142938-14143221	exon6(13bp)-exon2(60bp)	6,2,3,4,5	7	
		chr18:7844684-7844949	exon6(13bp)-exon2(60bp)	6,2,3,4,5	7	
		chr18:11752288-11752571	exon6(13bp)-exon2(60bp)	6,2,3,4,5	17	
		chrX:75705395-75705678	exon6(13bp)-exon2(60bp)	6,2,3,4,5	7	
		chrX:95467664-95467947	exon6(13bp)-exon2(60bp)	6,2,3,4,5	6	
		chrX:114662183-114662466	exon6(13bp)-exon2(60bp)	6,2,3,4,5	5	
		chrY:1277313-1277596**	exon6(13bp)-exon2(60bp)	6,2,3,4,5	6	
	Low-confidence <i>circRFWD2</i> -derived pseudogenes (9)	chr1:105321124-105321442	No	2,3,4,5	3	
		chr2:22762744-22763077	No	2,3,4,5	3	
		chr2:45329232-45329465*	No	2,3,4	N/A	
		chr2:96133443-96133643	No	2,3,4,5	6	
		chr5:6358836-6359147	No	2,3,4,5	6	
		chr6:14570771-14571041	No	2,3,4,5	6	
		chr10:3303049-3303381	No	2,3,4,5	3	
		chr10:82442603-82442943	No	2,3,4,5	4	
		chr17:57115121-57115447	No	2,3,4,5	3	
Linear <i>RFWD2</i> mRNA-derived pseudogenes (6)	chr3:98846063-98846502	No	19,20	36		
	chr6:41010910-41011386	No	18,19,20	34		
	chr6:120005990-120006346	No	17,18,19,20	7		
	chr6:131431440-131433959	No	1-20	44		
	chr9:32629139-32629479	No	15,16,17	19		
	chrX:103162834-103165519	No	1-20	31		

(B)

Locations in mouse genome	CircRNA exons	Pseudogene exons	Origin genes	Isoforms	Other linear RNA related pseudogenes (and their genomic locations)
chr1:77570670-77570963	8-10	8-10	TASP1	NM_001159640	No
chr1:140047780-140048277	2-3	2-3	ZFP532	NM_207255	No
chr2:21244101-21244441	4-5	4-5	PHF3	ENSMUST00000088310	No
chr2:37671940-37673005	2-3	2-3	ZFP609	NM_172536	No
chr2:131109585-131109796	7-21	20	FUBP1	uc008rsz.1	No
chr4:18193525-18193754	2-5	2	ZFP652	ENSMUST00000107717	No
chr4:131902790-131903312	3-10	7-10	PIK3CB	uc012gyy.1	No
chr5:88423213-88423556	3-7	3-5	SEMA3A	ENSMUST00000095012	No
chr6:58135885-58136125	19-20	19	CNTNAP2	ENSMUST00000114641	No
chr6:58512271-58512511	19-20	19	CNTNAP2	ENSMUST00000114641	No
chr6:76284389-76284927	2-9	2-8	UXS1	ENSMUST00000126008	Yes (exon1-15: chr17:73189666-73191698)
chr6:89200701-89201079	3-12	7-9	BRD1	NM_001033274	No
chr6:119425237-119425605	17-20	17-19	NCOR1	ENSMUST00000101066	No
chr6:146463331-146463607	3-5	3-5	CALM2	NM_007589	Yes (exon1-6: chr6:127281468-127282571)
chr7:14616386-14616789	19-20	20	RANBP2	NM_011240	No
chr7:14756551-14756926	19-20	20	RANBP2	NM_011240	No
chr7:14857028-14857340	19-20	20	RANBP2	NM_011240	No
chr7:15118005-15118384	19-20	20	RANBP2	NM_011240	No
chr7:15369748-15370129	19-20	20	RANBP2	NM_011240	No
chr7:15408517-15408905	19-20	20	RANBP2	NM_011240	No
chr7:15528046-15528427	19-20	20	RANBP2	NM_011240	No
chr7:15566483-15566871	19-20	20	RANBP2	NM_011240	No
chr7:15725924-15726312	19-20	20	RANBP2	NM_011240	No
chr7:15766810-15767202	19-20	20	RANBP2	NM_011240	No
chr7:84804224-84805343	2-5	2-4	PNPLA8	NM_026164	No
chr7:84883579-84884648	2-5	2-4	PNPLA8	NM_026164	No
chr8:112938148-112938513	3-7	4-7	GMDS	ENSMUST00000041859	Yes (exon5-10: chrX:155922915-155923428)
chr8:122683600-122683840	3-9	4-6	ACTR3	ENSMUST00000027579	Yes (exon1-11: chrX:81820383-81822920)
chr9:40929907-40930428	2-4	4	USP10	ENSMUST00000144458	Yes (exon1-14: chr1:45768280-45770200)
chr9:48420165-48420581	3-8	8	NXPE4	uc009phu.2	No
chr9:75061357-75062048	2-7	2-7	API5	ENSMUST00000028617	Yes (exon1-7: chr3:90758659-90759563)
chr9:96810275-96810687	2-4	2-4	D17WSU92E	NM_001044719	No
chr10:60228055-60228859	2	2	HECA	uc007elw.1	No
chr11:45280589-45280955	7-11	8-11	TYW1	ENSMUST00000040213	No
chr11:80423966-80424340	8-10	8-10	DLGAP4	NM_146128	No
chr12:39948444-39949116	2-32	17-20	USP9X	NM_009481	No
chr13:13275132-13275791	3-11	6-11	ROCK1	ENSMUST00000067947	No
chr13:76439327-76439666	8-22	17-19	ACAP2	ENSMUST00000058033	No
chr13:85865048-85865333	5-9	6-9	SFPQ	NM_023603	No
chr14:19035803-19036355	3-8	4-7	GTF2A1	NM_031391	Yes (exon1-9: chr12:82491520-82496623/ chr5:134353719-134356615)/ chr11:8804984-8806936/chrX: 20382438-20383891
chr14:25809631-25809873	3-4	3-4	HMBOX1	uc007uip.1	No
chr14:114492243-114492559	2-5	2	PNPLA8	NM_026164	Yes (exon2-4: chr7:84804225-84805343)
chr16:27843370-27844421	2-10	6-10	ZFP106	ENSMUST00000055241	No
chr16:70747479-70747721	3-5	3-5	TAF1B	uc007nej.2	No
chr17:19422496-19422803	2-17	13-15	SMC2	NM_001301412	No
chr17:55063411-55063913	2-5	2	4933411K20RIK	ENSMUST00000110376	No
chr18:25321983-25322217	24-25	25	SMARCA2	ENSMUST00000025862	No
chrX:53418329-53418967	2-10	5-9	CSNK1G3	NM_146128	No

(C)

Locations in human genome	CircRNA exons	Pseudogene exons	Origin genes	Isoforms	Other linear RNA related pseudogenes
chr1:16314674-16314961	11-15	12	CPD	ENST00000225719	No
chr1:223741976-223742921	2	2	NACC2	NM_144653	No
chr1:238090130-238090890	2-5	2-4	ACTG1	NM_001199954	Yes (exon2-6: chr1:104112105-104114008/ chr1:202842360-202844336/chr20:1141153-1143045/ chrY:19868177-19870002/chrY:20309773-20311598)
chr1:246691881-246692140	11-15	12	CPD	ENST00000225719	No
chr2:159559606-159559995	3-4	3-4	NCOA6	ENST00000374796	No
chr7:39899233-39899892	2	2	LOC650226	NR_029420	No
chr11:21191294-21191683	3	3	KIAA1462	ENST00000375377	Yes (exon2-3: chr21:40897453-40901708)
chr11:34764451-34764745	7-10	8-10	ARHGAP26	ENST00000274498	No
chr11:41681595-41681772	4-7	6	DDB2	ENST00000256996	No
chr13:25091882-25092180	2-8	4	CENPJ	NM_018451	No
chr13:34836998-34837211	4-5	4	KIAA1551	ENST00000312561	No
chr13:77564462-77564724	25-27	25-27	DHX9	NM_001357	Yes (exon1-28: chr13:77558898-77563104)
chr14:52622457-52622894	11-18	12-15	ITGA3	NM_002204	No
chr14:71498397-71498687	4-9	4-6	TTC7B	ENST00000555005	No
chr15:34701760-34702795	4	4	APBA2	ENST00000561069	No
chr15:34847982-34849017	4	4	APBA2	ENST00000561069	No
chr16:87500741-87501671	2	2	NR3C1	NM_000176	No
chr17:26004849-26005113	11-15	12	CPD	ENST00000225719	No
chr17:30330126-30330344	2-3	3	BPTF	ENST00000544778	No
chr17:30423862-30424085	2-3	3	BPTF	ENST00000544778	No
chr19:23437948-23438238	10-26	21-23	IPO5	NM_002271	Yes (exon6-28: chr9:6012290-6015582)
chr20:10869230-10869738	10-11	10	FAT1	NM_005245	No
chrX:118072118-118073108	2-6	2-6	COBL	ENST00000265136	No
chrY:13299925-13300166	6-7	7	AC097374.2	ENST00000582835	No

Table S1. List of circRNA-derived pseudogenes in mouse and human.

(A) List of mouse *RFWD2* related pseudogenes. At least 42 pseudogenes were identified to have the putative *circRFWD2* origin, and other six with a linear *RFWD2* mRNA origin. The sub-classification, genomic locations, non-colinear exon-exon back-splicing junction sequence length, exon composition and counts of “A”s in UCSC poly(A) information are listed. Although most have been annotated, all these *circRFWD2*-derived pseudogenes were not marked with circRNA origin. *, three

circRFWD2-derived pseudogenes were not previously annotated as pseudogenes. **, not identified in all the examined mouse strains, possible due to its location on Y chromosome.

(B) Annotation of low-confidence circRNA-derived pseudogenes in mouse. In total, at least 48 low-confidence circRNA-derived pseudogenes were identified without related non-colinear exon-exon junctions in the mouse reference genome (mm10). The genomic locations, exon compositions in both related circRNAs and pseudogenes, related gene origins and isoforms, and the existence of linear mRNA-derived pseudogenes from the same gene origin is listed. Note that the genomic loci of related linear mRNA-derived pseudogenes (last column) are different from the low-confidence circRNA-derived pseudogenes.

(C) Annotation of low-confidence circRNA-derived pseudogenes in human. In total, at least 24 low-confidence circRNA-derived pseudogenes were identified without related non-colinear exon-exon junctions in the human reference genome (hg19). The genomic locations, exon compositions in both related circRNAs and pseudogenes, related gene origins and isoforms, and the existence of the related linear mRNA-derived pseudogenes is listed. Note that the genomic loci of the related linear mRNA-derived pseudogenes (last column) are different from the low-confidence circRNA-derived pseudogenes.