Table S1: Analyses of dog germ cell purity

	Spermatocytes	Spermatids	Cytoplasm	Others	**Spermatids**	Spermatocytes	Cytoplasm	Others
Dog 4	86	16	2	3	140	2	17	6
Dog5	95	18	4	9	101	1	12	8
Dog6	94	9	1	7	116	1	15	4
Tot.	265 **(79.3%)**	43 (12.9%)	7 (2.1%)	19 (5.7%)	357 **(84.4%)**	4 (0.9%)	44 (10.4%)	18 (4.3%)

Table S2: Relative expression levels of spermatocyte genes in mouse and dog spermatids

Spermatocyte gene	Dog log2 fold change spt/spc	Mouse log2 fold change spt/spc
Sycp1/SYCP1	-0.46	-0.42
Sycp2/SYCP2	-2.88	-1.38
Sycp3/SYCP3	-1.07	-1.11
Average	-1.47	-0.97

Table S3: Relative expression levels of spermatid genes in mouse and dog spermatids

Spermatid gene	Dog log2 fold change spt/spc	Mouse log2 fold change spt/spc
Tnp1/TNP1	3.76	3.54
Tnp2/TNP2	4.02	3.51
Prm1/PRM1	n.a.	3.51
Prm2/PRM2	3.86	3.80
Prm3/PRM3	2.75	3.18
Average	3.60	3.51

Table S4: Comparison between Cufflinks and edgeR results

	# diff. expressed genes	# in common (%)	# sign and >1.5 fold up	# in common (%)	# sign and >1.5 fold down	# in common (%)	# sign and >1.5 fold up X	# in common (%)	#sign and >1.5 fold down X	# in common (%)
Cufflinks	9679	8661 (89)	4632	4323 (94)	4220	4094 (97)	207	197 (95)	126	116 (92)
edgeR	9904	8661 (87)	4828	4323 (90)	4919	4094 (83)	224	197 (88)	160	116 (73)

Table S5: Differentially expressed dog X-linked genes homologous to mouse and/or human multicopy genes

Down in spermatids	**Up in spermatids**
DDX26B	*COL4A6*
MAGEB1	*GLRA2*
MAGEB4	*GLRA4*
MAGED1	*H2BFWT*
MAOB	*MAGEB10*
NGFRAP1	*MAGEB5*
RAB9A	*MAOA*
SHROOM2	*MID2*
SLC9A6	*PHKA2*
SSX5	*PRPS2*
TCEAL4	*RAB9B*
ZNF182	*SCML1*
ZNF185	*SCML2*
ZNF280C	*SPACA5*
	TGIF2LX
	ZCCHC12
	ZNF711
	ZNF81

Table S6: Average FPKM values of gene expression in dog

	Genome	Chr 1	X	PAR
SPERMATOCYTES	27.05	28.31	6.15	2.46
SPERMATIDS	37.92	35.18	22.87	2.71

Table S7: Expression of *SLC25A* variants in spermatocytes and spermatids of mouse man and dog

DOG (RNA-seq)				MOUSE (RNA-seq)			MAN (microarray)		
gene	chr	spc (FPKM)	spt (FPKM)	chr	spc (FPKM)	spt (FPKM)	chr	spc	spt
SLC25A4	16	5.77	2.06	8	6.18	2.81	4	26.44	13.83
SLC25A5	X	4.08	1.77	X	8.50	2.38	X	159.86	297.73
SLC25A6	X(PAR)	47.19	18.11	-	-	-	X(PAR)	113.95	163.43
SLC25A31	19	34.76	20.54	3	34.10	16.26	4	265.19	75.93

Table S8: Molecular and cellular functions associated with X-linked genes upregulated in spermatids of dog and mouse

Category	p-value	Genes
Cell Morphology	9.44E-04-4.66E-02	*PHEX,AKAP4,PAK3,CDK16,TAF7L,MAOA,HMGB3*
DNA Replication, Recombination, and Repair	2.08E-03-3.94E-02	*HUWE1,CUL4B,CETN2,UBE2A,TEX11,ERCC6L*
Cell Signaling	2.37E-03-4.33E-02	*PRKX,TAF1,AKAP4,PDK3,MED12,MED14*
Post-Translational Modification	2.37E-03-3.17E-02	*HUWE1,CDKL5,OGT,PRKX,TAF1,UBE2A,RLIM,UBA1,ASB9,PDK3,MAOA*
Protein Synthesis	2.37E-03-3.56E-02	*CUL4B,PRKX,TAF1,RLIM,EIF1AX,ASB9,PDK3*
Molecular Transport	3.49E-03-1.99E-02	*PHEX,SLC35A2,RPGR,MAOA*
Nucleic Acid Metabolism	3.49E-03-1.99E-02	*CTPS2,SLC35A2,RPGR*
Small Molecule Biochemistry	3.49E-03-4.33E-02	*OGT,CTPS2,SLC35A2,RPGR,SMS,MAOA*
Carbohydrate Metabolism	4.02E-03-2.39E-02	*OGT,SLC35A2*
Cell Death and Survival	4.02E-03-4.98E-02	*HUWE1,OGT,UXT,ATP11C,PAK3,BGN,SH3KBP1,MAOA*
Cell-To-Cell Signaling and Interaction	4.02E-03-4.71E-02	*CDKL5,OGT,OPHN1,PAK3,RLIM,BGN,MAOA*
Cellular Assembly and Organization	4.02E-03-3.94E-02	*OPHN1,PAK3,OFD1,TEX11,MAOA*
Cellular Development	4.02E-03-4.71E-02	*CUL4B,RLIM,BGN,USP9X,CDK16,ZFX,TEX11,TAF7L*
Cellular Function and Maintenance	4.02E-03-1.99E-02	*PHEX,OPHN1,OFD1,MAOA*
Cellular Growth and Proliferation	4.02E-03-4.33E-02	*CUL4B,CETN2,RLIM,BGN*
Drug Metabolism	4.02E-03-2.39E-02	*MAOA*
Gene Expression	4.02E-03-1.72E-02	*MED12,MED14,TXLNG*
Cell Cycle	8.01E-03-3.17E-02	*RLIM,ERCC6L*
Cellular Movement	8.01E-03-4.11E-02	*CDKL5,AKAP4,TAF7L,MAOA*
Energy Production	8.01E-03-1.6E-02	*MAOA*
Lipid Metabolism	1.6E-02-1.6E-02	*SLC35A2*
Protein Degradation	3.06E-02-3.06E-02	*CUL4B,RLIM,ASB9*
Amino Acid Metabolism	3.17E-02-3.17E-02	*SMS*
RNA Post-Transcriptional Modification	4.71E-02-4.71E-02	*CSTF2*

Table S9: X-linked genes upregulated in spermatids of mouse, human, and dog

	Gene	Mouse knockout	Molecular function	Putative function in spermatogenesis
Structural components	*AKAP14*	-	A-kinase-anchoring protein	Ciliary beat frequency [1]
	AKAP4	immotile sperm [2]	A-kinase-anchoring protein	Fibrous sheet formation
	CDK16	malformed sperm , reduced motility [3]	Cyclin dependent kinase	Annulus formation
	OFD1	embryonic lethal [4] Mutations in the human ortholog are associated with Oral–facial–digital type I (OFDI; MIM 311200) syndrome and Simpson-Golabi-Behmel syndrome type 2	Centrosome associated proteins, involved in Cilia formation [5]. Removal through autophagy is essential for proper cilia function [6]	
	CYLC1	-	Cyclicin, basic protein, structural component of cytoskeleton	Component of sperm head cytoskeleton [7]
Ubiquitin pathway	*ASB12*	-	The ankyrin repeat and SOCS box (ASB) family, forms E3 complex with Cul5-Rbx2	Ubiquitylation [8]
	CUL4B	embryonic lethal, epiblast-specific knockout is viable, reproduction not analysed [9]	Component of E3 ligase complex	Ubiquitylation
	UBA1	-	Ubiquitin-activating enzyme	Ubiquitylation
Mitochondrial enzymes	*DUSP21*	-	Mitochondrial membrane phosphatase [10]	
	MAOA	fertile, aggressive behaviour [11]	Monoamine oxidase A degrades serotonin and norepinephrine. Mitochondrial enzyme	
Transcription regulation	*TAF1*	-	Largest subunit of TFIID, transcriptional regulation	
	TAF7L	reduced sperm numbers and quality, fertile [12]	Regulation of transcription program together with TAF1	
	IQSEC2	human mutations cause nonsyndromic intellectual disability [13]	Guanine nucleotide exchange factor for the ADP-ribosylation factor family of small GTPases, involved in membrane trafficking and actin dynamics	
	TBC1D25	-	Rab GTPase-activating protein involved in autophagy [14]	
	CDKL5	neurodevelopmental disorder, fertile [15]	Cyclin-dependent kinase-like	
	CXORF27	-	Huntingtin interacting protein with histone fold [16]	

	GSPT2	-	Polypeptide release factor (translation termination) [17]	
	TEX13A	-	Testis-specific gene [18] possibly involved in mRNA processing [19]	
	PPEF1	fertile, no clear phenotype [20]	Calcium binding phosphatase	
	RIBC1	-	The RIB43A domain with coiled coils 1 (*RIBC1*) gene with unknown function	
	TEX28	-	Testis-specific single exon gene of unknown function [21]	

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