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Author(s)	Murata, Chie; Kuroki, Yoko; Imoto, Issei et al.
Citation	Chromosome research, 24(3), 407-419 https://doi.org/10.1007/s10577-016-9531-y
Issue Date	2016-09
Doc URL	https://hdl.handle.net/2115/67083
Rights	The final publication is available at link.springer.com
Type	journal article
File Information	MS_HUSCAP.pdf

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Ancestral Y-linked genes were maintained by translocation to the X and Y chromosomes fused to an autosomal pair in the Okinawa spiny rat *Tokudaia muenninki*

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Keywords: sex chromosome, evolution, translocation, gene amplification, RNA-seq, BAC

A short running title: Y-linked gene translocations in the Okinawa spiny rat.

Introduction

The mammalian Y chromosome is male-specific and triggers the differentiation of the developing fetus into a male. The Y is one of the smallest chromosomes with the fewest genes, despite previous homology to the X chromosome (Ross et al. 2005). Most present therians (placentas and marsupials) maintain the sex chromosomes formed in the therian ancestor; a core set of Y-linked genes are shared among many eutherian species, while many multiple-copy genes are species- or lineage-specific (Paria et al. 2011; Bellott et al. 2014; Cortez et al. 2014). Models of Y gene loss dynamics revealed that Y chromosomal gene decay proceeds rapidly upon differentiation and then markedly levels off when small, but apparently essential, gene repertoires are defined (Hughes et al. 2012; Bellott et al. 2014; Cortez et al. 2014). In particular, the sex-determining gene *SRY* has persisted over long evolutionary time periods in most therians, with rare examples of *SRY* loss in the *Tokudaia* and *Ellobius* lineages (Just et al. 1995; Soullier et al. 1998; Vogel et al. 1998; Suto et al. 2001). Several long-lived ancestral Y-linked genes were also lost in at least one mammalian lineage during recent evolution in the clade (Bellot et al. 2014). However, other genes have been conserved by compensatory mechanisms, e.g., the rescue of Y-linked gene loss via gene transposition to other chromosomes (Hughes et al. 2015).

This compensatory mechanism was first detected in *Tokudaia* species (Kuroiwa et al. 2010). The Amami and Tokunoshima spiny rats, *Tokudaia osimensis* and *Tokudaia tokunoshimensis*, respectively, have an XO/XO sex chromosome constitution; they lack the entire Y chromosome, except for a small region translocated to the X chromosome (Honda et al. 1977, 1978; Kuroiwa et al. 2010). The proto-Y region on the X chromosome contains at least four Y-linked genes, *EIF2S3Y*, *KDM5D*, *TSPY*, and *ZFY*, but functional *RBMY* and *SRY* have been absent from the genome since the secondary Y chromosome loss (Arakawa et al. 2002; Kuroiwa et al. 2010). Furthermore, *KDM5D* and *EIF2S3Y* are expressed in the gonads of both sexes in *T. osimensis* (Kuroiwa et al. 2010).

The Okinawa spiny rat *Tokudaia muenninki* has the XX/XY sex chromosome

58 constitution and multiple *SRY* copies on the large Y chromosome fused to an autosome
59 (Tsuchiya et al. 1989; Murata et al. 2010, 2012). It has an ancestral Y chromosome region and
60 many *SRY* sequences, but these *SRY* loci may have not retained sex-determining functions
61 (Murata et al. 2010; Kimura et al. 2014). SRY proteins bind to the core region within a
62 testis-specific enhancer of *SOX9* (TESCO) with SF1 to upregulate *SOX9* expression in the
63 undifferentiated gonads of XY embryos of humans and mice (Sekido et al. 2008). TESCO of the
64 genus *Tokudaia* lost enhancer activity due to mutations in its SRY- and SF1-binding sites, and *T.*
65 *muenninki* SRY does not activate the mutant TESCO or mouse TESCO (Kimura et al. 2014).
66 In our previous study, we found that two Y-linked genes, *UTY* and *DDX3Y*, are
67 single-copy genes on the pericentromeric region of Yp, which is the ancestral Y chromosome
68 region in *T. muenninki* (Murata et al. 2012). By contrast, most other Y-linked genes are
69 excessively duplicated, like the *SRY*s. We investigated six apparently multi-copy Y-linked genes,
70 *ZFY*, *UBA1Y*, *EIF2S3Y*, *TSPY*, *USP9Y*, and *RBMY*, and two single-copy genes, *UTY* and
71 *DDX3Y*, to examine whether the functions of multiple-copy genes were compensated by
72 translocation or transposition. We performed RNA-seq using three tissue types to identify
73 transcripts of ancestral Y-linked genes. We then conducted reverse transcription PCR (RT-PCR)
74 and 3′ rapid amplification of cDNA ends (RACE) using six tissue types from an adult male to
75 examine the functional conservation of these Y-linked genes based on expression patterns and
76 the complete coding sequences, and to determine whether mRNA sequence variants existed for
77 each gene. Furthermore, we performed fluorescent *in situ* hybridization (FISH) using cDNA
78 clones of Y-linked genes as probes to display their chromosomal localization. Using quantitative
79 real-time PCR (qPCR) with genomic DNA, we estimated the copy number of each Y-linked
80 gene in the male genome. The six genes were excessively duplicated on the Y chromosome, and
81 several copies of *ZFY* and *UBA1Y* were located on the X chromosome. We then performed
82 next-generation sequencing of five bacterial artificial chromosome (BAC) clones including *UTY*
83 and/or *DDX3Y* to reveal the genomic structure of the two genes as well as adjacent genes in the

ancestral Y euchromatic region. Our results show that functional Y-linked genes are compensated by translocations to other sex chromosomal regions and amplifications in *T. muenninki*.

Materials and Methods

Sample preparation

T. muenninki is endangered (The IUCN Red List of Threatened Species; http://www.iucnredlist.org/, 3/8/2016) and has been protected by the Japanese government as a natural treasure since 1972 (Yamada et al. 2010). With permission from the Agency for Cultural Affairs and the Ministry of the Environment in Japan, all spiny rats were released at their capture sites after a small piece was cut from the tip of their tail. Using the tail tissues, cell cultures and DNA extraction were performed as previously reported (Murata et al. 2010). Total RNA was extracted from the brain, liver, testis, kidney, lung, and spleen of one adult male *T. muenninki* who died of accidental causes in March 2013 using an RNeasy Mini Kit (Qiagen, Hilden, Germany) following the manufacturer's protocol. All experimental procedures conformed to the guidelines for animal experimentation of Tokushima University and all institutional and national guidelines for the care and use of laboratory animals were followed.

RNA-seq and data assembly

The cDNA libraries from the brain, liver, and testis were constructed using the TruSeq RNA Sample Preparation Kit v2 (Illumina, San Diego, CA, USA). Three indexed libraries from each tissue were pooled and sequenced using one lane to obtain 101 nucleotide single-end reads with an Illumina HiSeq 2500 RNA-seq data were deposited in the DDBJ Sequence Read Archive under the accession number DRA004624. *De novo* assembly and reference mapping were performed after removing polyA/T tails and filtering low-quality reads. *De novo* assembly was carried out using Trinity (Grabherr et al. 2011; Haas et al. 2013) with the default settings. BLAST searches using the assembled sequences were performed with Blast+ ver. 2.2.30 (Camacho et al. 2009; ftp://ftp.ncbi.nlm.nih.gov/blast/executables/blast+/LATEST/). The National Center for Biotechnology Information (NCBI) transcript reference sequences (refseq_rna.*.tar.gz) were downloaded via FTP from ftp://ftp.ncbi.nlm.nih.gov/blast/db/. The *de*

113 *novo* assembled sequences matched to Y-linked genes, and these X homologs in the BLAST
114 search were extracted using the 'grep' command. To fill in gaps between the extracted contigs,
115 RNA-seq reads were mapped to RefSeq mRNA sequences from the mouse (refMrna.fa.gz
116 downloaded from http://hgdownload.cse.ucsc.edu/goldenPath/mm10/bigZips/) using BWA 0.5.9
117 with a 15% uniform base error rate (Li and Durbin 2009). The Integrative Genomics Viewer
118 was used to visualize reads mapped to the reference genome (Robinson et al. 2011). Accession
119 numbers of the eight X-linked gene sequences determined in this analysis are listed in Table S1.
120
121 **RT-PCR and 3′ RACE**
122 RT-PCR and 3′ RACE were performed using testis tissue to determine the complete coding
123 sequences of ancestral Y-linked genes by filling in gaps in the *de novo* assembled sequences and
124 to confirm the sequence variation for the transcripts of each Y-linked gene. RT-PCR using
125 multiple tissues from a male was also conducted to determine the expression patterns of the
126 Y-linked genes. The cDNA was synthesized using the SuperScript III First-Strand System for
127 RT-PCR Kit (Thermo Fisher Scientific, MA, USA). The primers for each RT-PCR amplification
128 were designed using the partial transcript sequences determined by *de novo* assembly and the
129 ancestral Y genome sequence determined by BAC sequencing. *ACTB* was used as a positive
130 control, and its primer pair was designed from the common sequence of mouse and rat. The
131 primer sequences are shown in Table S2. Amplification was carried out in a 20 µl mixture
132 containing 0.5 µl of cDNA, 0.3 µM each primer, 0.2 mM each of the four deoxynucleotide
133 triphosphates (dNTPs), 0.25 U of PrimeSTAR GXL DNA Polymerase, and a one-fifth volume
134 of 5× PrimeSTAR GXL Buffer (TaKaRa, Shiga, Japan). The RT-PCR conditions were as
135 follows: 98°C for 2 min; 30 cycles at 98°C for 10 s, 55–60°C for 15 s, and 68°C for 30 s to 4
136 min; and a final extension at 72°C for 7 min. For 3′ RACE, cDNA synthesis was initiated at the
137 poly(A) tail of mRNA using the adapter primer 5′-TGG AAG AAT TCG CGG CCG CAG TTT
138 TTT TTT TTT TTT T-3′ with the PrimeScript II 1st Strand cDNA Synthesis Kit (TaKaRa). The

unknown coding region of *USP9Y* was amplified using the Ex Taq HS System (TaKaRa) with the gene-specific primer 5′-CAC CAG CAC CAC AAG ATA GA-3′ and the universal amplification primer 5′-TGG AAG AAT TCG CGG CCG CAG-3′. PCR products were cloned using the TArget Clone Kit (ToYoBo, Osaka, Japan) after overhanging dA was added to the 3′ ends of the products using A-attachment mix (ToYoBo). Both the PCR products and cDNA clones were sequenced using an ABI 3500xL Genetic Analyzer. The identified Y gene and homologous X sequences were compared with orthologous sequences of the mouse (*Mus musculus*), rat (*Rattus norvegicus*), *T. osimensis*, and *Apodemus sylvaticus* using ClustalX 2.0 and MEGA5.2 (Larkin et al. 2007; Tamura et al. 2008). Furthermore, we performed phylogenetic analyses for each X/Y gene pair including *T. muenninki* and other rodent genes using MEGA5.2 to confirm whether X- and Y-linked genes formed separate clusters. Accession numbers of the Y-linked transcript sequences determined by RT-PCR and 3′ RACE with *de novo* assembled sequences are listed in Table S1. Accession numbers of the X and Y gene sequences in other rodents are also listed in Table S1.

Chromosome preparation and FISH

The preparation of R-banded chromosomes was performed as described by Matsuda et al. (1992), Matsuda and Chapman (1995), and Kobayashi et al. (2008). The cDNA clones of *ZFY* (2,263 bp), *UBA1Y* (1,778 bp), *EIF2S3Y* (1,458 bp), *TSPY* (691 bp), *USP9Y*-2 (3,616 bp), and *RBMY* (1,123 bp) were used as probes. The cDNA clones *ZFX* (2,816 bp) and *UBA1* (2,114 bp) from the brain were also used, and the primer pairs used to detect the two X-linked genes by RT-PCR are shown in Table S2. The FISH analysis using the cDNA clones was performed as described by Kuroiwa et al. (2010). The FISH signals were observed under an Olympus fluorescence microscope BX53, and the images were captured with a DP73 digital camera (Olympus, Tokyo, Japan).

qPCR

The copy number of each amplified ancestral Y-linked gene was quantified using gene-specific primers designed using Primer3 ver. 0.4.0 (http://bioinfo.ut.ee/primer3-0.4.0/). For each qPCR, *DDX3Y* and *UTY*, single-copy genes in the *T. muenninki* male genome, were used as an internal control for copy number. The primer pairs used for this analysis are shown in Table S2. To achieve maximum efficiency of the qPCR, the product size was limited to 90–109 bp.

qPCR was conducted using TaKaRa SYBR *Premix Ex Taq* (Perfect Real Time) in accordance with the manufacturer's protocol, and amplification was detected using the Applied Biosystems 7500 Real-Time PCR System. The qPCR reactions were performed in triplicate in a 10 µl reaction volume in a 96-well plate under the following conditions: 95°C for 30 s, followed by 40 cycles of 95°C for 5 s and 60°C for 34 s. Genomic DNA samples from five *T. muenninki* males were used in the analysis. The presence of a single melting temperature peak, which represented a single amplicon, was used to validate primer specificity, and the slope and R^2 values for serial dilutions of genomic DNA were used to confirm the efficiency of the reaction. To detect potential contamination during plate preparation, nuclease-free water was included in each set of reactions as a negative control.

BAC clone sequencing, data assembly, and sequence comparison

BAC libraries of a single male *T. muenninki* were previously constructed (Murata et al. 2015). Five BAC clone sequences were determined, including *UTY* and/or *DDX3Y* genes (TMB1-066O12, 237P18, 265P11, 324H06, and 389K21), by multiplex sequencing on an Illumina HiSeq 1000 using a 100 bp paired-end protocol, as described in our previous study (Murata et al. 2015). The primers used to screen the BAC libraries were designed within an exon from the transcript sequences of the two genes determined by RT-PCR and sequencing. The primer pairs used for gene amplification were as follows: *UTY*, Primers no. 2 and 3 in Table S2; *DDX3Y*, 5′-GGA AGG ATT CAT TGA CTT TAG TA-3′ and 5′-TCT GTG ATC TGT CTC

191 CAT GA-3′. Amplification was carried out in a 10 µl mixture containing 1.0 µl of library DNA,
192 0.5 µM each primer, 0.2 mM each of the four dNTPs, 0.5 U of BIOTaq HS DNA Polymerase,
193 and a one-tenth volume of 10× ImmoBuffer. PCR was performed under the following
194 conditions: denaturation for 10 min at 95°C, followed by 45 cycles of denaturation for 30 s at
195 95°C, annealing for 30 s at 55°C, and extension for 30 s at 72°C. BAC sequences were *de novo*
196 assembled as previously reported (Murata et al. 2015). Gaps within *de novo* assembled
197 sequences were bridged by PCR and sequencing. Finally 372,407 bp of combined sequence
198 from five BAC clones was determined without gaps (LC066213).
199 To capture genome rearrangements, a dot-plot analysis was performed for the *T.*
200 *muenninki* Y and mouse Y regions. The mouse genome sequence (NC_000087) was obtained
201 from NCBI. A nucleotide-based alignment was generated using NUCmer, which is a mode in
202 MUMmer 3.0, for the large-scale genome comparisons with default settings (minimum length of
203 a maximal exact match: 20, and minimum cluster length: 65; Kurtz et al. 2004). The exon-intron
204 boundaries were defined for the combined sequence of five BAC clones by comparison to the
205 transcript sequences of *T. muenninki* and orthologous genes in the mouse genome. To verify
206 whether the genes located on the ancestral Y genome were transcribed into mRNAs, the
207 predicted mRNA sequences were compared with the actual mRNA sequences of *T. muenninki*
208 using ClustalX 2.0. Furthermore, the sequences surrounding *SRY* with masked repetitive
209 elements were compared between *T. muenninki*, mouse, and rat using ClustalX 2.0. Repetitive
210 elements were identified using the RepeatMasker Web Server
211 (http://www.repeatmasker.org/cgi-bin/WEBRepeatMasker).

Results

Identification of expressed Y-linked genes and their X homologs

Y-linked genes diverge more rapidly than other chromosomal genes among species. Therefore, the *de novo* assembly of RNA-seq data is a useful strategy to find Y-linked genes without a genome sequence, such as in non-laboratory animals. We performed RNA-seq using three tissue types to identify Y-linked genes in *T. muenninki*. We generated 115,946,604, 101,801,400, and 111,401,004 single-end reads from the testis, brain, and liver, respectively. A total of 91–92% of the sequenced reads passed the quality control check and were used for *de novo* Trinity assembly. After the *de novo* assembly of all available reads from the three tissues, we obtained 111,185 components with an N50 of 3,437 bp, defined as the maximum length such that 50% of the total assembled sequences were contained in contigs of at least this length. We extracted the partial transcript sequences that matched eight mouse and rat Y-linked genes (*ZFY*, *UBA1Y*, *EIF2S3Y*, *TSPY*, *UTY*, *DDX3Y*, *USP9Y*, and *RBMY*) by BLAST searches of the *de novo* assembled sequences. We also identified the complete coding sequences of six X-linked genes (*ZFX*, *UBA1X*, *KDM6A*/*UTX*, *DDX3X*, *USP9X*, and *RBMX*) from the assembled sequences, and we determined the sequences of two genes, *EIF2S3X* and *TSPYL2*, by filling in gaps using the read sequences mapped to the mouse RefSeq mRNA sequences. The similarities of the nucleotide and amino acid sequences were 91.8–97.6% and 93.4–99.4%, respectively, between X-linked genes of *T. muenninki* and mouse (Table S3).

Predicted amino acid sequences and expression patterns of Y-linked genes

Using RT-PCR and 3′ RACE with an adult testis tissue sample, we determined the complete coding sequences of seven Y-linked genes, *ZFY*, *EIF2S3Y*, *TSPY*, *UTY*, *DDX3Y*, *USP9Y*, and *RBMY*, but not *UBA1Y* (Fig. S1). The identification of X and Y genes was performed by phylogenetic analysis with each X/Y gene sequences of *T. muenninki* and other rodents, revealing that their Y-linked genes were grouped into the same cluster and formed separate

cluster to the X homologs (Fig. S2). We observed several nucleotide substitution sites within the transcript sequences of *ZFY*, *EIF2S3Y*, *TSPY*, *USP9Y*, and *RBMY*, and no sequence variation within the *UTY* and *DDX3Y* transcripts (Fig. S1). The predicted amino acid sequences of the seven Y-linked genes exhibited 63.3–99.6% similarity between *T. muenninki* and other rodents (Table 1). Some of the *ZFY* transcripts had 1 bp and 7 bp deletions at positions 369 and 494–496, respectively, in *T. muenninki* (Fig. S1a). Therefore, the predicted amino acid sequences of the transcripts without either deletion were compared with the mouse and *T. osimensis* orthologs (Table 1, Fig. S1b). We did not detect sequence variation in *UBA1Y*, but we observed a partial coding sequence caused by an internal stop codon. The internal stop codon was generated by a frameshift 86 bp deletion, which resulted in the deletion of more than 230 amino acid residues (Fig. S1c, d). The predicted amino acid sequences of *EIF2S3Y*, *TSPY*, *UTY*, and *DDX3Y* were highly similar to the orthologs of other rodents without frameshift mutations (Table 1, Fig. S1e–l). In *USP9Y*, we identified a difference between *T. muenninki* and mouse orthologs at the last 22 amino acid residues due to a frameshift mutation (Fig. S1m–n). Based on a sequence comparison of *RBMY* among *T. muenninki*, mouse, and rat, we inferred that the frameshift mutation occurred in the mouse lineage, and sequence similarity was higher for the comparison between *T. muenninki* and the rat than between *T. muenninki* and the mouse (Table 1, Fig. S1o, p).

We examined the expression patterns of the eight Y-linked genes by RT-PCR using six tissues obtained from an adult male. We observed higher *ZFY* expression in the testis and brain than in other tissues, and we detected *UBA1Y* expression in all tissue types, except for the kidney. *EIF2S3Y*, *UTY*, and *DDX3Y* showed ubiquitous expression, *TSPY* and *RBMY* were specifically expressed in the testis, and *USP9Y* was expressed more highly in the testis than in other tissues in *T. muenninki* (Fig. 1).

Chromosomal localization of Y-linked genes

We mapped the *T. muenninki* cDNA clones of six Y-linked genes, *ZFY*, *UBA1Y*, *EIF2S3Y*, *TSPY*, *USP9Y*, and *RBMY*, to chromosomes of a male by direct R-banding FISH. All six genes were located on the long arm of the Y chromosome as multiple-copy genes (Fig. 2). *ZFY* was also distributed on the distal region of Xq as well as Yq, and *ZFX* was located on the middle region of Xq (Fig. 2a–c, Fig. S3a, b). Furthermore, we observed FISH signals of *UBA1Y* along the heterochromatin block of the X chromosome as well as Yq (Fig. 2d–f), and we detected *UBA1* signals in the pericentromeric region of Xq (Fig. S3c, d). *EIF2S3Y* and *USP9Y* copies were mainly located on the pericentromeric and middle region of Yq, respectively (Fig. 2g–l). The other four genes were amplified on the pericentromeric and middle region of Yq (Fig. 2a–f, m–r). We also observed *USP9Y* on the pericentromeric region of Yp (Fig. 2j–l), which was the predicted ancestral Y euchromatic region including two single-copy genes *UTY* and *DDX3Y* (Murata et al. 2012).

Copy numbers of Y-linked genes

We performed qPCR using genomic DNA of five *T. muenninki* males to estimate the copy numbers of the six Y-linked genes (*ZFY*, *UBA1Y*, *EIF2S3Y*, *TSPY*, *USP9Y*, and *RBMY*) that were identified as multiple-copy genes in the FISH analysis (Fig. 2). We also examined the copy number of *SRY*, which was previously demonstrated to have multiple copies (Murata et al. 2010). We used *UTY* and *DDX3Y* as internal control genes with a single copy per male diploid genome. Setting the copy number value of *UTY* (or *DDX3Y*) to 1.0, the relative copy numbers for *DDX3Y* (or *UTY*), *ZFY*, *UBA1Y*, *EIF2S3Y*, *TSPY*, *USP9Y*, *RBMY*, and *SRY* were approximately 1, 63, 66, 10, 61, 34, 50, and 61, respectively (Fig. 3, Table 1).

Genomic structure of ancestral Y genes

We screened five BAC clones including *UTY* and/or *DDX3Y*, which are single-copy genes on Yp, to identify the genomic structure of ancestral Y genes. We determined a 372.4 kb sequence

by next-generation sequencing using the BAC clones. Based on a comparison between *T. muenninki* and the mouse Y chromosome sequence, we identified a large homologous block without apparent inversions (Fig. 4). We obtained the sequences of *SRY*, *EIF2S3Y*, *TSPY*, *UTY*, *DDX3Y*, and *USP9Y* (Fig. 4a, b). We detected the reversed *SRY* sequence upstream of *EIF2S3Y* in *T. muenninki*, although *SRY* was located more than 1 Mb downstream of *USP9Y* in the mouse genome (Fig. 4c). Exons 1–4 and exon 12, including the complete 447 bp coding region, were absent from *EIF2S3Y*, although the upstream and downstream sequences of *EIF2S3Y* were found in the BAC clone sequence of *T. muenninki* (Fig. 4, Fig. S1e). We did not detect exons 1–28 in *USP9Y* because the terminal region of the BAC sequence was within intron 29 of *USP9Y* (Fig. 4a). Additionally, the ancestral *USP9Y* genomic sequence had an internal stop codon within exon 29 (Fig. S1m). The predicted transcript sequences inferred from the genomic sequence of three multiple-copy genes, *EIF2S3Y*, *TSPY*, and *USP9Y*, were different from the actual mRNA sequences (Table 1, Fig. S1e, g, m). For single-copy genes, the *UTY* and *DDX3Y* mRNA sequences were completely consistent with the predicted sequences from the genome, and the exon-intron boundaries for the two genes corresponded to those of mouse orthologs, except for a 3 bp slip of an accepter splicing site in exon 16 of *DDX3Y* (Table 1, Fig. S1i, k and Fig. S4).

Function and structure of the duplicated *SRY*

The *SRY* copy located immediately upstream of *EIF2S3Y* (named *SRY25*) in the euchromatic region was compared with previously reported *SRY* sequences of *T. muenninki*, mouse, and rat in the coding region and the surrounding region, to reveal whether it functions as a sex-determining gene. *SRY25* matches the previously identified *SRY5*, which is a pseudogene with an internal stop codon (Fig. S1q). In the region upstream of *SRY/Sry*, an approximately 3.1 kb sequence in the rat showed more than 80% and 71% similarity with *T. muenninki SRY* sequences segmented by long interspersed element (LINE) insertions (Fig. 5a, Fig. S5a). We did

not detect conservation between mice and the other two species, except for an approximately 0.7 kb region just before the start codon (Fig. 5a, Fig. S5a). Downstream of *SRY/Sry*, we observed more than 6 kb of three conserved segments separated by insertions of several retrotransposable elements between mouse and *T. muenninki*, but the conservation was limited to an approximately 1.3 kb region between rat and the other two species (Fig. 5b, Fig. S5b). In the *SRY* surrounding region, sequence similarity was higher between *T. muenninki* and other rodents than between mouse and rat.

We then identified the distal end of a *SRY* duplication unit to identify factors related to *SRY* amplification in *T. muenninki* by comparing the genomic region surrounding *SRY* between *T. muenninki* and other rodents. We found a long-terminal repeat (LTR) at the distal end of a *SRY* duplication unit, which was adjacent to the region with high similarity to mouse upstream of *Eif2s3y* (Fig. 5b).

328 **Discussion**

329 We determined the complete coding sequences of *ZFY*, *EIF2S3Y*, *TSPY*, *UTY*, *DDX3Y*, *USP9Y*,
330 and *RBMY*, but not *UBA1Y*, by RNA-seq, RT-PCR, and 3′ RACE (Fig. S1). We observed higher
331 sequence similarity between *T. muenninki* and mouse than between mouse and rat for the
332 single-copy genes *UTY* and *DDX3Y*, but slightly lower similarity between *T. muenninki* and
333 mouse/*Apodemus sylvaticus* (the Wood mouse) than between mouse/*A. sylvaticus* and rat for the
334 multiple-copy genes *EIF2S3Y*, *TSPY*, *USP9Y*, and *RBMY* (Table 1). Based on molecular
335 phylogenetic studies, the *Tokudaia* lineage (sister to *Apodemus*) is more closely related to *Mus*
336 than *Rattus* (Rowe et al. 2008), suggesting that the rate of evolution of the latter four Y-linked
337 genes accelerated in *T. muenninki* relative to other rodents owing to the lineage-specific gene
338 amplification. In the mouse and rat, *ZFY*, *UBA1Y*, *USP9Y*, *TSPY*, and *RBMY* are predominantly
339 expressed in testes, whereas *EIF2S3Y*, *UTY*, and *DDX3Y* are broadly expressed in male tissues
340 (Bellott et al. 2014; GEO Profiles in National Center for Biotechnology Information). Similar
341 expression patterns are observed in *T. muenninki* and other rodents for the Y-linked genes,
342 except for *UBA1Y* (Fig. 1). Our results indicated that the *T. muenninki* genome retained the
343 seven functional Y-linked genes. We also observed several nucleotide substitution sites within
344 the transcript sequences of *ZFY*, *EIF2S3Y*, *TSPY*, *USP9Y*, and *RBMY* (Fig. S1), indicating that
345 mRNAs of each gene are transcribed from multiple gene regions with different sequences.
346 Additionally, some of the *ZFY* transcripts had deletions within the coding region in *T. muenninki*,
347 suggesting that both functional and pseudogene sequences of *ZFY* are expressed in *T. muenninki*
348 testes (Fig. S1a).

349 We observed excessive amplification of the ancestral Y-linked genes on the Y chromosome
350 in *T. muenninki* (Fig. 2, Fig. 3). Based on a FISH analysis, multiple copies of six genes (*ZFY*,
351 *UBA1Y*, *EIF2S3Y*, *TSPY*, *USP9Y*, and *RBMY*) were mainly localized to the long arm of the Y
352 chromosome. Our previous study demonstrates that the long arm of the Y chromosomes is
353 entirely heterochromatic (Murata et al 2010). *EIF2S3Y* and *USP9Y* copies were located on the

354 pericentromeric and middle region of Yq, respectively (Fig. 2g–l), while the other four genes
355 and *SRY* were on the pericentromeric and middle regions (Fig. 2a–f, m–r, Murata et al. 2010).
356 Consistent with the FISH results, which showed that *EIF2S3Y* and *USP9Y* copies were not
357 located on the middle and pericentromeric regions, respectively, we detected fewer copies of
358 these two genes than the other multiple-copy genes located in both regions by qPCR (Fig. 3).
359 These results indicated that the Y segment that includes the seven Y-linked genes translocated
360 from Yp to Yq, and the segment was subsequently amplified in the middle and pericentromeric
361 regions of Yq after the loss of *EIF2S3Y* and *USP9Y*, respectively. Y-linked gene amplification is
362 common during mammalian evolution; for example, it has been reported for *ZFY*, *RBMY*, *TSPY*,
363 and *SRY* (Bianchi et al. 1993; Chai et al. 1998; Verkaar et al. 2004; Turner et al. 2007). However,
364 this is the first report of the massive amplification of these seven Y-linked genes in a
365 mammalian species. We also found that the Y-linked genes were translocated to the X
366 chromosome at least two times in *T. muenninki*. *ZFY* and *UBA1Y* were distributed on the distal
367 region of Xq and the centromeric heterochromatin of the X chromosome, respectively (Fig. 2a–
368 f). In the *T. muenninki* X chromosome, the long and short arms consist of the ancestral X
369 chromosome and a fused autosome, respectively (Murata et al. 2012). Dobigny et al. (2004)
370 suggested that constitutive heterochromatin between the X chromosome and the autosome fused
371 to the X prevented the spread of facultative heterochromatinization of the inactive X to an
372 autosome. Therefore, *UBA1Y* would be translocated to the centromeric heterochromatin of the X
373 chromosome and would then be involved in this amplification related to the autosomal fusion in
374 *T. muenninki*.
375 The order and direction of the five Y-linked genes determined by BAC clone
376 sequencing of *T. muenninki* revealed that the genome structure of the ancestral Y euchromatic
377 region was highly conserved between *T. muenninki* and mouse, except for *SRY* (Fig. 4),
378 suggesting that we identified the ancestral genomic sequences of three multiple-copy genes
379 (*EIF2S3Y*, *TSPY*, and *USP9Y*) in the Y euchromatic region of *T. muenninki*. We observed the

loss of five exons in *EIF2S3Y* and the presence of an internal stop codon in *USP9Y* (Fig. 4, Fig. S1). The deletion in *EIF2S3Y* and a missense mutation in *USP9Y* might occur in each original sequence after duplication. The predicted transcript sequences inferred from the genomic sequence of *TSPY* as well as *EIF2S3Y* and *USP9Y* were different from the actual transcript sequences (Fig. S1), suggesting that the amplified copies of these genes were functional, unlike the original sequences. Thus, our results provide evidence for compensatory effects of the amplified gene copies.

We showed that the *SRY* surrounding region was highly conserved between *T. muenninki* and other rodents, although *SRY*25 was a nonfunctional pseudogene with the internal stop codon (Fig. S1q). There is a possibility that *SRY* could be duplicated on the Y chromosome to compensate its function like as other Y-linked genes. However, it is suggested that *SRY* has not retained sex-determining functions (Murata et al. 2010; Kimura et al. 2014).

The 5' and 3' flanking region of *SRY*25 showed high similarity to mouse and rat (Fig. 5), suggesting that the gene was duplicated by not transposition but translocation. The 5' flanking region of *SRY25* was segmented by LINE insertion (Fig. 5a yellow colored), indicating that this LINE insertion occurred after *SRY* duplication. In contrast, a LTR were found at the distal end of a conserved *SRY* unit (Fig 5b, pink colored). Furthermore, the downstream of this LTR showed high homology to the 5' flanking region of mouse *Eif2S3y* (Fig. 5b, black colored). These observations indicated that the LTR was boundary for *SRY* duplication unit and the duplication occurred via these repetitive sequences. Several genetic mechanisms can determine the numbers of repeating units in tandem arrays, including gene amplification by processes such as replication slippage, rolling circle amplification, and unequal exchange (Charlesworth et al. 1994). Because replication slippage and rolling circle amplification involve only a short stretch of DNA at a time, these processes probably play only minor roles in the amplification of very long repeat units (Charlesworth et al. 1994). By contrast, the expansion of repeat units may be strongly associated with unequal exchange; this pattern has been inferred based on a

sequence analysis of satellites consisting of higher-order repeat units (Cabot et al. 1993; Warburton et al. 1993). Furthermore, extensive accumulation of retrotransposable elements has been observed in the Y chromosomes of *Mus* species (Eicher et al. 1989). Actually, unequal sister chromatid exchange mediated by retrotransposable elements could have been involved in *SRY* amplification along the heterochromatic block of the X chromosome in Cabrera's vole, *Microtus cabrerae* (Fernández et al. 2002; Marchal et al. 2008). Gene amplification by unequal exchange mediated by retrotransposable elements has also been described for human olfactory receptor genes (Glusman et al. 2000), the primate γ-globin gene (Fitch et al. 1991), and mouse zinc-finger protein genes (Chen et al. 2002). Therefore, the amplification process for the Y-linked genes of *T. muenninki* can be explained by the same mechanism mediated by retrotransposable elements. In support of this hypothesis, we did not detect an intronless sequence generated by retrotransposition in the six multiple Y-linked genes (*ZFY*, *UBA1Y*, *EIF2S3Y*, *TSPY*, *USP9Y*, and *RBMY*) by PCR using the genomic DNA of *T. muenninki* (data not shown).

Our results revealed that the seven Y-linked genes were translocated to the X and Y chromosomes fused to an autosomal pair and were subsequently amplified in *T. muenninki*. Additionally, we showed that the amplified copies of the Y-linked genes are functional, unlike the original genes in the ancestral Y euchromatic region. The rescue of Y-linked genes via transposition to other chromosomes is widespread among mammals (Hughes et al. 2015). In the common ancestor of the two XO/XO species, *T. osimensis* and *T. tokunoshimensis*, the Y chromosome was lost after a small euchromatic region containing essential Y-linked genes was translocated to the X chromosome and a few Y-linked gene transposed to autosomes (Kuroiwa et al. 2010; Fig. 6). In the lineage of the *T. muenninki*, the Y chromosome was likely rescued from the risk of loss by sex-autosome fusions (Murata et al. 2012; Fig. 6). Nonetheless, the Y-linked genes were specifically translocated to the other region of the Y chromosome in the *T. muenninki* lineage, suggesting that the translocation was necessary to rescue Y-linked genes

from progressive degeneration in the ancestral Y region. As the Y chromosome diverged from the X chromosome, the correction of replication errors by homologous recombination was not possible, leading to the accumulation of deleterious mutations in Y-linked genes (Bachtrog 2013). Therefore, it is hypothesized that becoming multiple copied genes is one strategy to survive on the non-recombining Y (Bellott et al. 2014). Also, gene duplication coupled with gene conversion between duplicates can potentially maintain gene function by counteracting evolutionary Y decay (Rozen et al. 2003; Charlesworth 2003; Noordam and Repping 2006). A combination of analytical and computer simulation methods also showed that gene conversion effectively maintains the functionality of Y-linked duplicates that have already become fixed, although it has little impact on the probability that duplicates become fixed within a population (Connallon and Clark 2010). Therefore, the functions of *T. muenninki* Y-linked genes may have been maintained not only by translocation, but also by amplification on the sex chromosomes fused to an autosomal pair.

445 **Figure Legends**

446 **Figure 1. Expression patterns of eight Y-linked genes in six tissue types from an adult male**
447 **of *Tokudaia muenninki*.**

448 *ACTB* was used as a positive control. N.C.: negative control. Marker: AccuRuler 1 kb DNA
449 RTU Ladder (Maestrogen).

450

451 **Figure 2. FISH mapping of six Y-linked genes on *T. muenninki* male chromosomes.**

452 All six genes, *ZFY* (a–c), *UBA1Y* (d–f), *EIF2S3Y* (g–i), *USP9Y* (j–l), *TSPY* (m–o), and *RBMY*
453 (p–r), were mapped along the long arm of the Y chromosome. The hybridization signals are
454 indicated by arrowheads. Propidium iodide-stained R- (a, b, d, e, g, h, j, k, m, n, p, q) and
455 Hoechst G-banding patterns (c, f, i, l, o, r) are shown. The scale bars represent 10 μm. (s) A
456 summary of the Y-linked gene locations on the sex chromosomes. *: Murata et al. 2012; **:
457 Murata et al. 2010.

458

459 **Figure 3. Quantification of the copy numbers of ancestral Y-linked genes in *T. muenninki*.**

460 The copy number of *UTY* (a) or *DDX3Y* (b) was set to 1.0, and the relative values for the other
461 eight genes were estimated in five males by qPCR. The graph shows the means ± SD.

462

463 **Figure 4. Comparison of the Y chromosome region of mouse (*x*-axis) to the ancestral Y**
464 **chromosome region of *T. muenninki* (*y*-axis).**

465 (a) Dot-plot alignments of a 391 kb region of mouse (NC_000087: 960,000–1,350,695) to a
466 372.4 kb region of *T. muenninki* (LC066213: 1–372,407). (b) Dot-plot alignments of an 8.8 kb
467 region of mouse (NC_000087: 2,655,600–2,664,400) to a 15.9 kb region of *T. muenninki*
468 (LC066213: 14,001–29,875). These alignments were generated using a mummerplot script and
469 gnuplot. Aligned segments are represented as dots or lines in the NUCmer alignment. The
470 positions of the genes on the mouse Y chromosome are shown on the dot-plot alignments. (c)

Comparison of Y gene order between the mouse and *T. muenninki*. *SRY* was located approximately 1.2 Mb downstream of *USP9Y* in the mouse genome.

Figure 5. Sequence conservation of the regions upstream (a) and downstream (b) of *SRY* among *T. muenninki*, mouse, and rat.

Percentages between bars indicate sequence similarity between the segments. Species-specific regions, except for retrotransposable elements, are colored white. TMU: *T. muenninki*.

Figure 6. Evolution of the X and Y chromosomes and genes in genus *Tokudaia*.

The lineage of *T. muenninki* was firstly diverged in genus *Tokudaia* (Murata et al. 2010). Sex-autosome fusion (Murata et al. 2012), Y-to-Y and Y-to-X gene translocations occurred in the lineage. In the common ancestor of *T. osimensis* and *T. tokunoshimensis*, a small euchromatic region of Y chromosome translocated to a distal end of X chromosome, and several Y genes transposed to autosome (Kuroiwa et al. 2010).

485 **Supplementary Figure Legends**

486 **Figure S1. Alignment of the nucleotide and amino acid sequences of nine Y-linked genes**
487 **between *T. muenninki* and other rodents.**

488 The coding region from the mRNA and/or genomic sequences of *ZFY* (a, b), *UBA1Y* (c, d),
489 *EIF2S3Y* (e, f), *TSPY* (g, h), *UTY* (i, j), *DDX3Y* (k, l), *USP9Y* (m, n), *RBMY* (o, p), and *SRY* (q)
490 in *T. muenninki* were compared with orthologous sequences in mouse, rat, *T. osimensis*, and/or *A.*
491 *sylvaticus*. Identical residues are indicated by dots. TMU: *T. muenninki*. The deletion and the
492 internal stop codon are colored in gray (a) and pink (m), respectively. Asterisks show the stop
493 codon sites in the amino acid sequences (b, d, f, h, j, l, n, p). Single nucleotide and amino acid
494 polymorphisms within *T. muenninki* transcripts of each gene are shown in red letters (e–h, m–p).
495 Different nucleotide sites of genomic sequences from the mRNAs of *T. muenninki* are shown in
496 blue letters (e, g, m, o). (q) The start codons and the stop codons are colored in blue. *SRY25* is
497 the genomic sequence observed upstream of *EIF2S3Y* in this study. *SRY1–24* sequences of *T.*
498 *muenninki* are reported by Murata et al. 2010, of which only three (*SRY1–3*) have the conserved
499 coding sequences. The repeat sequence was partially skipped in the mouse (600 bp), and the
500 skipped site is shown by double slashes.

501

502 **Figure S2. Neighbor-joining trees constructed with each X/Y gene sequence of *T.***
503 ***muenninki* and other rodents.**

504 Number above and below branches refer to bootstrap percentages. The values less than 70% are
505 not shown. TMU: *T. muenninki*; MMU: *M.musculus*; RNO: *R. norvegicus*; ASY: *A. sylvaticus*

506

507 **Figure S3. Chromosomal location of two X-linked genes in *T. muenninki*.**

508 A fluorescent *in situ* hybridization (FISH) analysis showed that *ZFX* (a, b) and *UBA1* (c, d) are
509 distributed on the middle and pericentromeric regions of Xq, respectively. The hybridization
510 signals are indicated by arrowheads. R- (a, c) and G-banding (b, d) patterns are shown. The

511 scale bars represent 10 μm.

512

513 **Figure S4. Nucleotide alignment of the accepter splicing site within intron 15 of *DDX3Y***

514 **between *T. muenninki* and mouse.**

515 The intronic sites are colored in gray for each species. Arrows show the boundary between an

516 exon and intron. Asterisks show the identical sites between two sequences.

517

518 **Figure S5. Nucleotide sequence alignment in the conserved upstream (a) and downstream**

519 **(b) regions of *SRY* among *T. muenninki*, mouse, and rat.**

520 Identical residues are indicated by dots. The skipped site is shown by double slashes. TMU: *T.*

521 *muenninki*.

Conflict of Interest

The authors declare that they have no conflict of interest.

Acknowledgments

The authors thank F. Yamada, T. Jogahara, C. Koshimoto, T. Mitani, and K. Nakata for their efforts in capturing the animals and the Fujii Memorial Institute of Medical Sciences, Tokushima University, for support with fluorescence microscopy. This work was supported by JSPS KAKENHI Grant Numbers 23870023 and 26870410.

Authors' Contributions

CM conceived of and designed the study, performed molecular and cytogenetic experiments and all data analysis, and drafted the manuscript. YK conducted RNA-seq. II participated in total RNA and BAC DNA extraction and commented on the manuscript. AK conceived of and designed the study, and participated in drafting the manuscript. All authors read and approved of the final manuscript.

References

Arakawa Y, Nishida-Umehara C, Matsuda Y, Sutou S, Suzuki H (2002) X-chromosomal localization of mammalian Y-linked genes in two XO species of the Ryukyu spiny rat. Cytogenet. Genome Res. 99:303-309

Bachtrog D (2013) Y-chromosome evolution: emerging insights into processes of Y-chromosome degeneration. Nat Rev Genet 14:113-124

Bellott DW, Hughes JF, Skaletsky H, Brown LG, Pyntikova T, Cho TJ, Koutseva N, Zaghlul S, Graves T, Rock S, followed by et al (2014) Mammalian Y chromosomes retain widely expressed dosage-sensitive regulators. Nature 508:494-499

Bianchi NO, Bianchi MS, Bailliet G, de la Chapelle A. (1993) Characterization and sequencing of the sex determining region Y gene *(Sry*) in *Akodon* (Cricetidae) species with sex reversed females. Chromosoma 102:389-395

Cabot EL, Doshi P, Wu ML, Wu CI (1993) Population genetics of tandem repeats in centromeric heterochromatin: unequal crossing over and chromosomal divergence at the *Responder* locus of *Drosophila melanogaster*. Genetics 135:477-487

Camacho C, Coulouris G, Avagyan V, Ma N, Papadopoulos J, Bealer K, Madden TL (2009) BLAST+: architecture and applications. BMC Bioinformatics. 2009 10:421

Chai NN, Zhou H, Hernandez J, Najmabadi H, Bhasin S, Yen PH (1998) Structure and

555 organization of the *RBMY* genes on the human Y chromosome: transposition and
556 amplification of an ancestral autosomal *hnRNPG* gene. Genomics 49:283-289
557 Charlesworth B (2003) The organization and evolution of the human Y chromosome. Genome
558 Biol 4:226
559 Charlesworth B, Sniegowski P, Stephan W (1994) The evolutionary dynamics of repetitive DNA
560 in eukaryotes. Nature 371:215-220
561 Chen HH, Liu TY, Huang CJ, Choo KB (2002) Generation of two homologous and intronless
562 zinc-finger protein genes, *zfp352* and *zfp353*, with different expression patterns by
563 retrotransposition. Genomics 79:18-23
564 Connallon T, Clark AG (2010) Gene duplication, gene conversion and the evolution of the Y
565 chromosome. Genetics 186:277-286
566 Cortez D, Marin R, Toledo-Flores D, Froidevaux L, Liechti A, Waters PD, Grutzner F,
567 Kaessmann H (2014) Origins and functional evolution of Y chromosomes across
568 mammals. Nature 508:488-493
569 Dobigny G, Ozouf-Costaz C, Bonillo C, Volobouev V (2004) Viability of X-autosome
570 translocations in mammals: an epigenomic hypothesis from a rodent case-study.
571 Chromosoma 113:34-41
572 Eicher EM, Hutchison KW, Phillips SJ, Tucker PK, Lee BK (1989) A repeated segment on the

573 mouse Y chromosome is composed of retroviral-related, Y-enriched and Y-specific
574 sequences. Genetics 122:181-92
575 Fernandez R, Barragan MJ, Bullejos M, Marchal JA, Martinez S, Diaz de la Guardia R, Sanchez
576 A (2002) Mapping the *SRY* gene in *Microtus cabrerae*: a vole species with multiple *SRY*
577 copies in males and females. Genome 45:600-603
578 Fitch DH, Bailey WJ, Tagle DA, Goodman M, Sieu L, Slightom JL (1991) Duplication of the
579 gamma-globin gene mediated by L1 long interspersed repetitive elements in an early
580 ancestor of simian primates. Proc Natl Acad Sci U S A 88:7396-7400
581 Glusman G, Sosinsky A, Ben-Asher E, Avidan N, Sonkin D, Bahar A, Rosenthal A, Clifton S,
582 Roe B, Ferraz C, Demaille J, Lancet D (2000) Sequence, structure, and evolution of a
583 complete human olfactory receptor gene cluster. Genomics 63:227-45
584 Grabherr MG, Haas BJ, Yassour M, Levin JZ, Thompson DA, Amit I, Adiconis X, Fan L,
585 Raychowdhury R, Zeng Q followed by et al (2011) Full-length transcriptome assembly
586 from RNA-Seq data without a reference genome. Nat Biotechnol 29:644-652
587 Haas BJ, Papanicolaou A, Yassour M, Grabherr M, Blood PD, Bowden J, Couger MB, Eccles D,
588 Li B, Lieber M followed by et al (2013) *De novo* transcript sequence reconstruction from
589 RNA-seq using the Trinity platform for reference generation and analysis. Nat Protoc
590 8:1494-1512

591 Honda T, Suzuki H, Itoh M (1977) An unusual sex chromosome constitution found in the
592 Amami spinous country-rat, *Tokudaia osimensis osimensis*. Jpn J Genet 52:247-249
593 Honda T, Suzuki H, Itoh M, Hayashi K (1978) Karyotypical differences of the Amami spinous
594 country-rats, *Tokudaia osimensis osimensis* obtained from two neighbouring islands. Jpn.
595 J. Genet. 53:297-299
596 Hughes JF, Skaletsky H, Brown LG, Pyntikova T, Graves T, Fulton RS, Dugan S, Ding Y,
597 Buhay CJ, Kremitzki C, followed by et al (2012) Strict evolutionary conservation
598 followed rapid gene loss on human and rhesus Y chromosomes. Nature 483:82-86
599 Hughes JF, Skaletsky H, Koutseva N, Pyntikova T, Page DC (2015) Sex
600 chromosome-to-autosome transposition events counter Y-chromosome gene loss in
601 mammals. Genome Biol 16:104.
602 Just W, Rau W, Vogel W, Akhverdian M, Fredga K, Graves JA, Lyapunova E (1995) Absence of
603 Sry in species of the vole *Ellobius*. Nat Genet 11:117-118
604 Kimura R, Murata C, Kuroki Y, Kuroiwa A (2014) Mutations in the testis-specific enhancer of
605 *SOX9* in the *SRY* independent sex-determining mechanism in the genus *Tokudaia*. PLoS
606 One 9:e108779
607 Kobayashi T, Yamada F, Hashimoto T, Abe S, Matsuda Y, Kuroiwa A (2008) Centromere
608 repositioning in the X chromosome of XO/XO mammals, Ryukyu spiny rat. Chromosome

609 Res 16:587-593

610 Kuroiwa A, Ishiguchi Y, Yamada F, Abe S, Matsuda Y (2010) The process of a Y-loss event in
611 an XO/XO mammal, the Ryukyu spiny rat. Chromosoma 119:519-526

612 Kurtz S, Phillippy A, Delcher AL, Smoot M, Shumway M, Antonescu C, Salzberg SL (2004)
613 Versatile and open software for comparing large genomes. Genome Biol 5:R12

614 Larkin MA, Blackshields G, Brown NP, Chenna R, McGettigan PA, McWilliam H, Valentin F,
615 Wallace IM, Wilm A, Lopez R, Thompson JD, Gibson TJ, Higgins DG (2007) Clustal W
616 and Clustal X version 2.0. Bioinformatics 23:2947-2948

617 Li H and Durbin R (2009) Fast and accurate short read alignment with Burrows-Wheeler
618 Transform. Bioinformatics 25:1754-60

619 Marchal JA, Acosta MJ, Bullejos M, Diaz de la Guardia R, Sanchez A (2008) Origin and spread
620 of the *SRY* gene on the X and Y chromosomes of the rodent *Microtus cabrerae*: Role of
621 L1 elements. Genomics 91:142-151

622 Matsuda Y, Chapman VM (1995) Application of fluorescence in situ hybridization in genome
623 analysis of the mouse. Electrophoresis 16:261-272

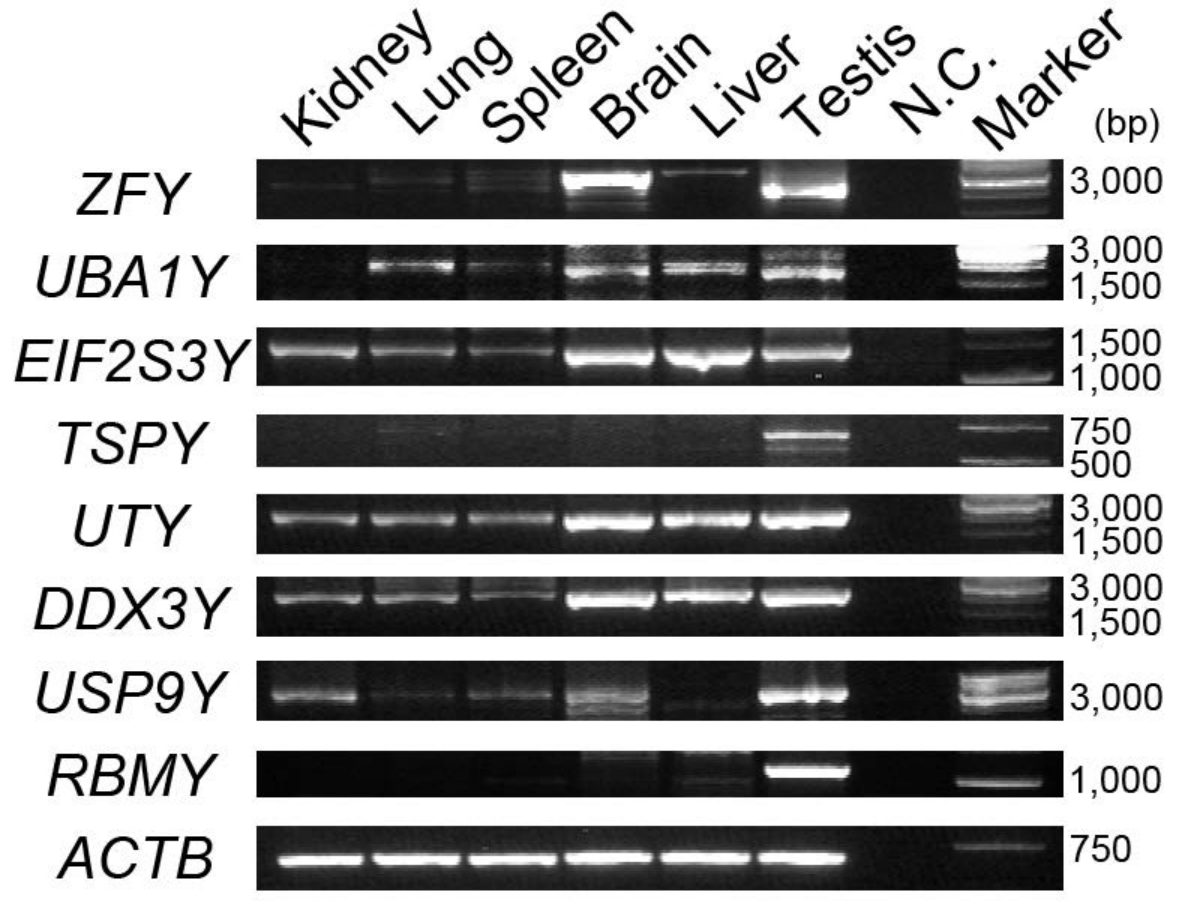
624 Matsuda Y, Harada YN, Natsuume-Sakai S, Lee K, Shiomi T, Chapman VM (1992) Location of
625 the mouse compliment factor H gene (cfh) by FISH analysis and replication R-banding.
626 Cell Genet 61:282-285

627 Murata C, Kuroki Y, Imoto I, Tsukahara M, Ikejiri N, Kuroiwa A (2015) Initiation of
628 recombination suppression and PAR formation during the early stages of neo-sex
629 chromosome differentiation in the Okinawa spiny rat, *Tokudaia muenninki*. BMC Evol
630 Biol 15:234
631 Murata C, Yamada F, Kawauchi N, Matsuda Y, Kuroiwa A (2010) Multiple copies of *SRY* on the
632 large Y chromosome of the Okinawa spiny rat, *Tokudaia muenninki*. Chromosome Res
633 18:623-634
634 Murata C, Yamada F, Kawauchi N, Matsuda Y, Kuroiwa A (2012) The Y chromosome of the
635 Okinawa spiny rat, *Tokudaia muenninki*, was rescued through fusion with an autosome.
636 Chromosome Res 20:111-125
637 Noordam MJ, Repping S (2006) The human Y chromosome: a masculine chromosome. Curr
638 Opin Genet Dev 16:225-232
639 Paria N, Raudsepp T, Pearks Wilkerson AJ, O'Brien PC, Ferguson-Smith MA, Love CC, Arnold
640 C, Rakestraw P, Murphy WJ, Chowdhary BP. (2011) A gene catalogue of the euchromatic
641 male-specific region of the horse Y chromosome: comparison with human and other
642 mammals. PLoS One 6:e21374
643 Robinson JT, Thorvaldsdottir H, Winckler W, Guttman M, Lander ES, Getz G, Mesirov JP
644 (2011) Integrative Genomics Viewer. Nature Biotechnology 29:24-26

645 Ross MT, Grafham DV, Coffey AJ, Scherer S, McLay K, Muzny D, Platzer M, Howell GR,
646 Burrows C, Bird CP, followed by et al (2005) The DNA sequence of the human X
647 chromosome. Nature 434:325-337
648 Rowe KC, Reno ML, Richmond DM, Adkins RM, Steppan SJ (2008) Pliocene colonization and
649 adaptive radiations in Australia and New Guinea (Sahul): multilocus systematics of the
650 old endemic rodents (Muroidea: Murinae). Mol Phylogenet Evol 47:84-101
651 Rozen S, Skaletsky H, Marszalek JD, Minx PJ, Cordum HS, Waterston RH, Wilson RK, Page
652 DC (2003) Abundant gene conversion between arms of palindromes in human and ape Y
653 chromosomes. Nature 423:873-876
654 Sekido R, Lovell-Badge R (2008) Sex determination involves synergistic action of SRY and
655 SF1 on a specific *Sox9* enhancer. Nature 453:930-934
656 Soullier S, Hanni C, Catzeflis F, Berta P, Laudet V (1998) Male sex determination in the spiny
657 rat *Tokudaia osimensis* (Rodentia:Muridae) is not Sry dependent. Mamm Genome
658 9:590-592
659 Sutou S, Mitsui Y, Tsuchiya K (2001) Sex determination without the Y chromosome in two
660 Japanese rodents *Tokudaia osimensis osimensis* and *Tokudaia osimensis* spp. Mamm
661 Genome 12:17-21
662 Tamura K, Peterson D, Peterson N, Stecher G, Nei M, and Kumar S (2008) MEGA5: Molecular

663 Evolutionary Genetics Analysis using Maximum Likelihood, Evolutionary Distance, and
664 Maximum Parsimony Methods. Mol Biol Evol 28:2731-2739
665 Tsuchiya K, Wakana S, Suzuki H, Hattori S, Hayashi Y (1989) "Taxonomic study of *Tokudaia*
666 (Rodentia: Muridae): I. Genetic differentiation. Memoirs Nat Sci Museum, Tokyo
667 22:227-234
668 Turner ME, Martin C, Martins AS, Dunmire J, Farkas J, Ely DL, Milsted A (2007) Genomic and
669 expression analysis of multiple *Sry* loci from a single *Rattus norvegicus* Y chromosome.
670 BMC Genet 8-11
671 Verkaar EL, Zijlstra C, van't Veld EM, Boutaga K, van Boxtel DC, Lenstra JA (2004)
672 Organization and concerted evolution of the ampliconic Y-chromosomal *TSPY* genes
673 from cattle. Genomics 84:468-474
674 Vogel W, Jainta S, Rau W, Geerkens C, Baumstark A, Correa-Cerro LS, Ebenhoch C, Just W
675 (1998) Sex determination in Ellobius lutescens: the story of an enigma. Cytogenet Cell
676 Genet 80:214-221
677 Warburton PE, Waye JS, Willard HF (1993) Nonrandom localization of recombination events in
678 human alpha satellite repeat unit variants: implications for higher-order structural
679 characteristics within centromeric heterochromatin. Mol Cell Biol 13:6520-6529
680 Yamada F, Kawauchi N, Nakata K, Abe S, Kotaka N, Takashima A, Murata C, Kuroiwa A

681 (2010) Rediscovery after thirty years since the last capture of the critically endangered
682 Okinawa spiny rat *Tokudaia muenninki* in the northern part of Okinawa Island. Mammal
683 Study 35:243-255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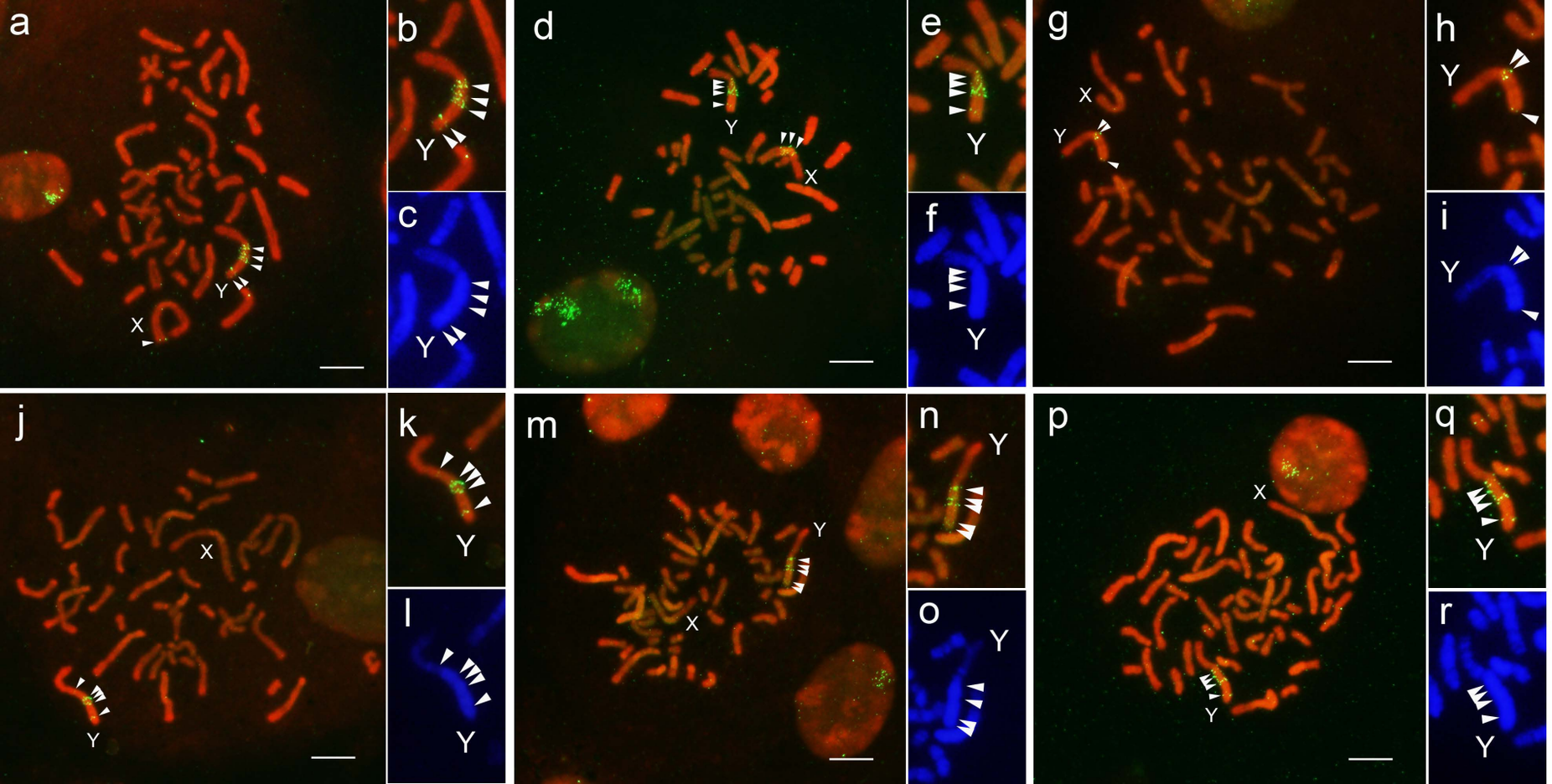

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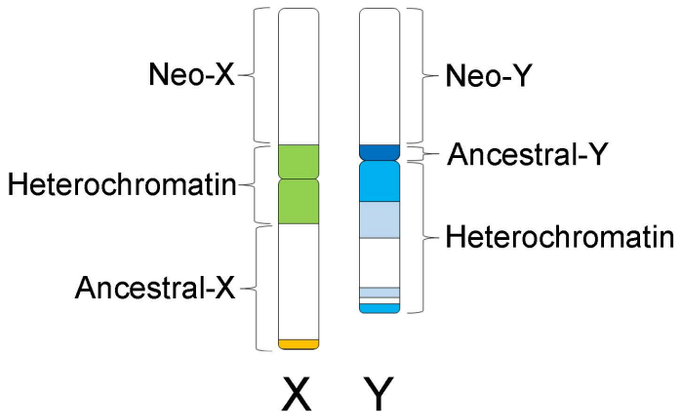

: *UTY**, *DDX3Y**, *USP9Y*

: *ZFY*, *UBA1Y*, *EIF2S3Y*, *TSPY*, *SRY***, *RBMY*

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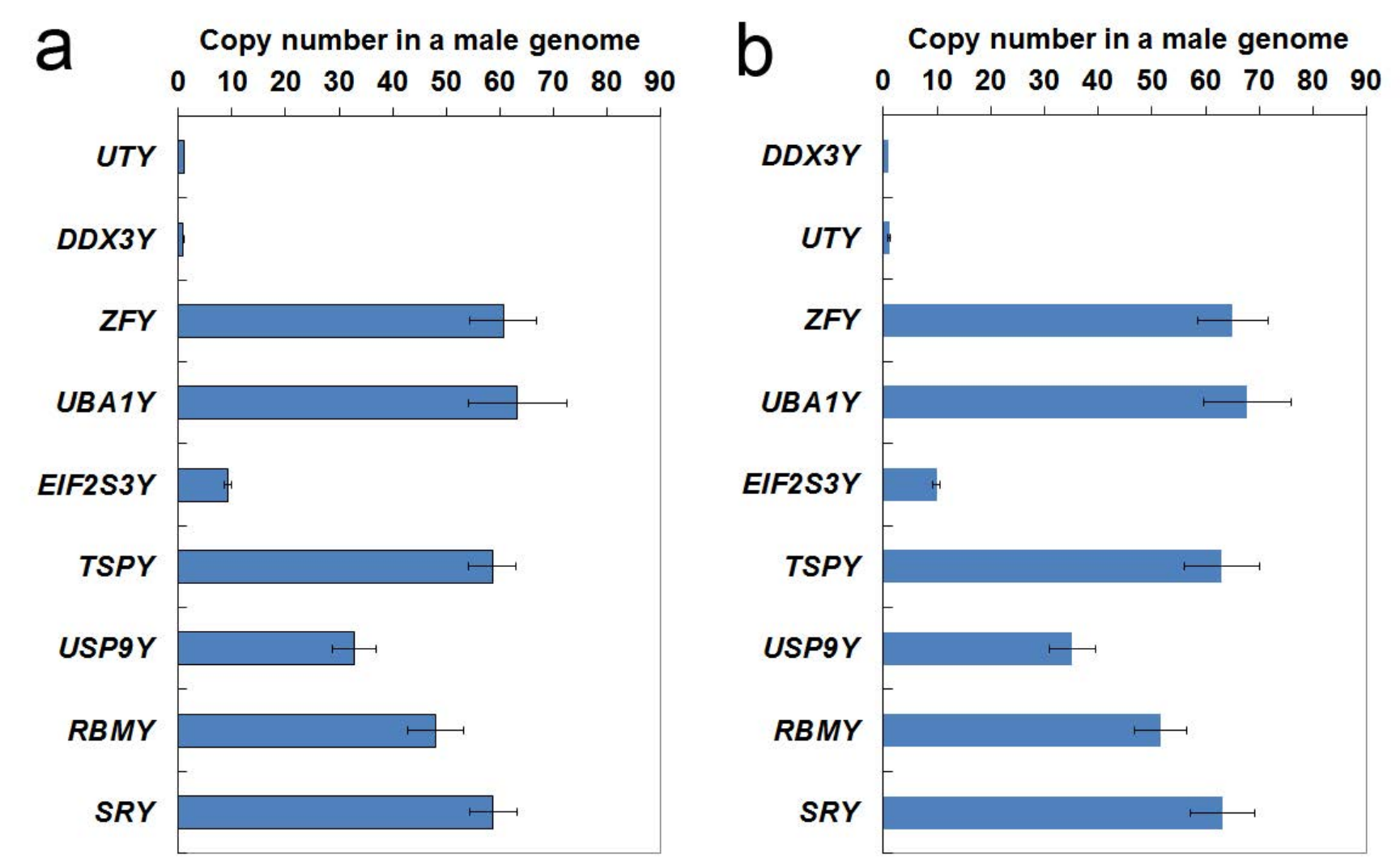
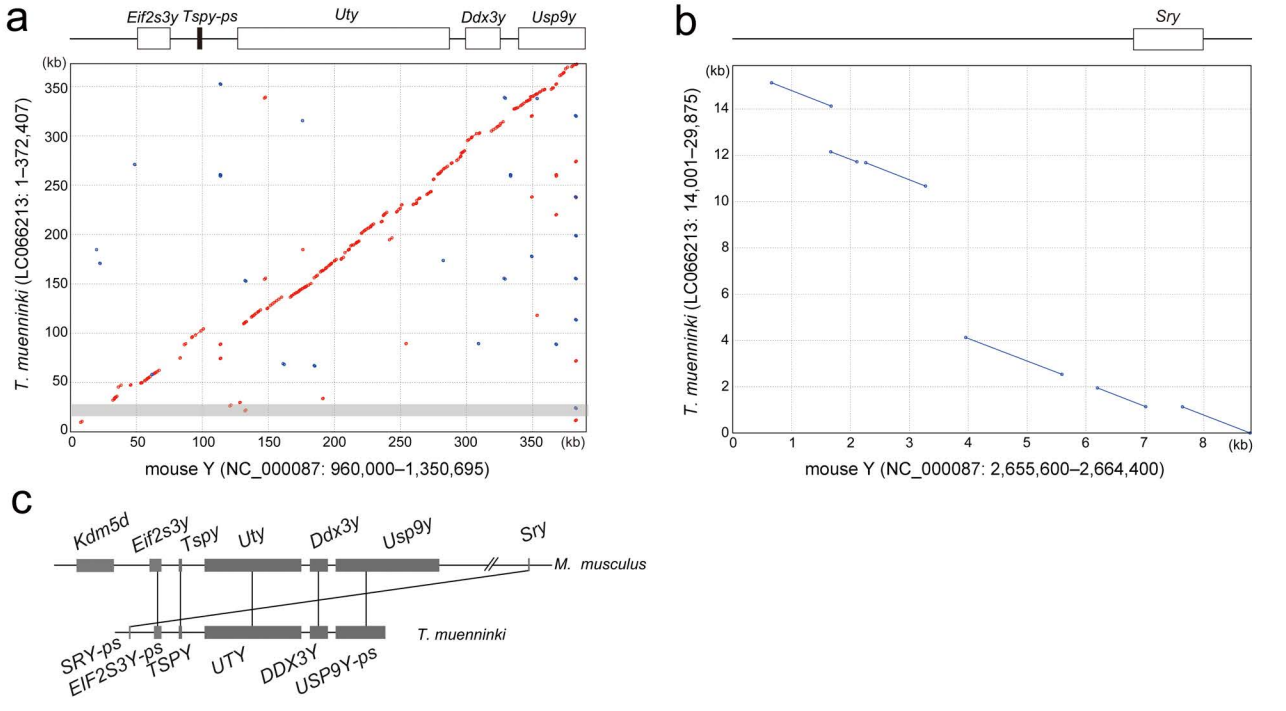
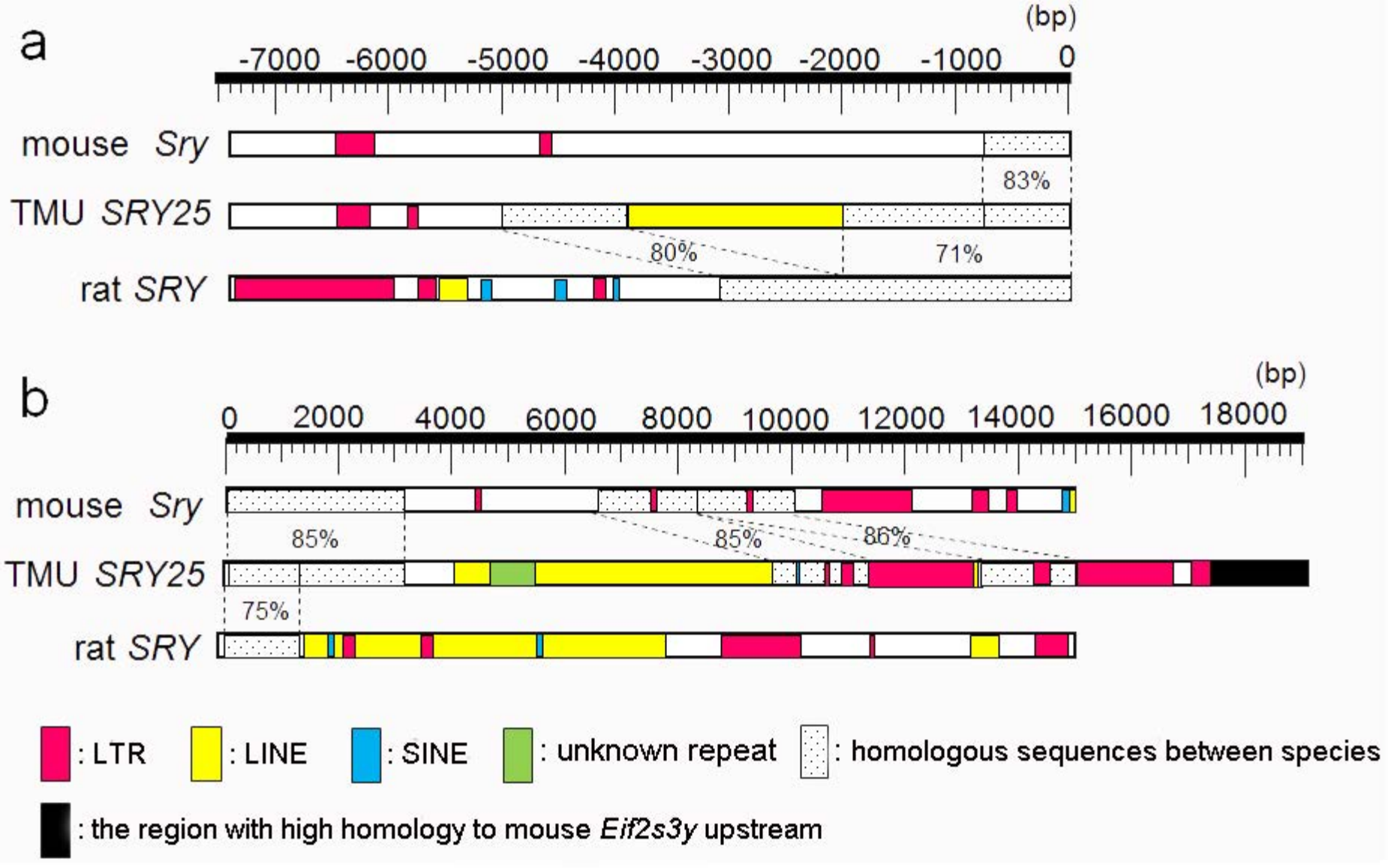
: *UBA1Y*

: *ZFY*


a
Copy number in a male genome
0 10 20 30 40 50 60 70 80 90
UTY
DDX3Y
ZFY
UBA1Y
EIF2S3Y
TSPY
USP9Y
RBMY
SRY
b
Copy number in a male genome
0 10 20 30 40 50 60 70 80 90
DDX3Y
UTY
ZFY
UBA1Y
EIF2S3Y
TSPY
USP9Y
RBMY
SRY


a
Eif2s3y
Tspy-ps
Uty
Ddx3y
Usp9y
(kb)
T. muenninki (LC066213: 1–372,407)
mouse Y (NC_000087: 960,000–1,350,695)
b
Sry
(kb)
T. muenninki (LC066213: 14,001–29,875)
mouse Y (NC_000087: 2,655,600–2,664,400)
c
Kdm5d
Eif2s3y
Tspy
Uty
Ddx3y
Usp9y
Sry
M. musculus
T. muenninki
SRY-ps
EIF2S3Y-ps
TSPY
UTY
DDX3Y
USP9Y-ps


a
(bp)
-7000 -6000 -5000 -4000 -3000 -2000 -1000 0
mouse Sry
TMU SRY25
rat SRY
83%
80%
71%
b
(bp)
0 2000 4000 6000 8000 10000 12000 14000 16000 18000
mouse Sry
TMU SRY25
rat SRY
85%
85%
86%
75%
: LTR
: LINE
: SINE
: unknown repeat
: homologous sequences between species
: the region with high homology to mouse Eif2s3y upstream

Y-to-X
euchromatic region
translocation

Y-to-autosome
gene
transposition

Amami spiny rat
T. osimensis
2*n*=25, XO/XO

Tokunoshima spiny rat
T. tokunoshimensis
2*n*=45, XO/XO

X Y

Y-loss
SRY-loss

Sex-autosome
fusion

Y-to-Y
Y-to-X
gene
translocations

Okinawa spiny rat
T. muenninki
2*n*=44, XX/XY

Transposed Y gene

Proto-Y
(Translocated Y region)

Autosome X

Neo-X

Neo-Y

Y-to-X

Ancestral-Y

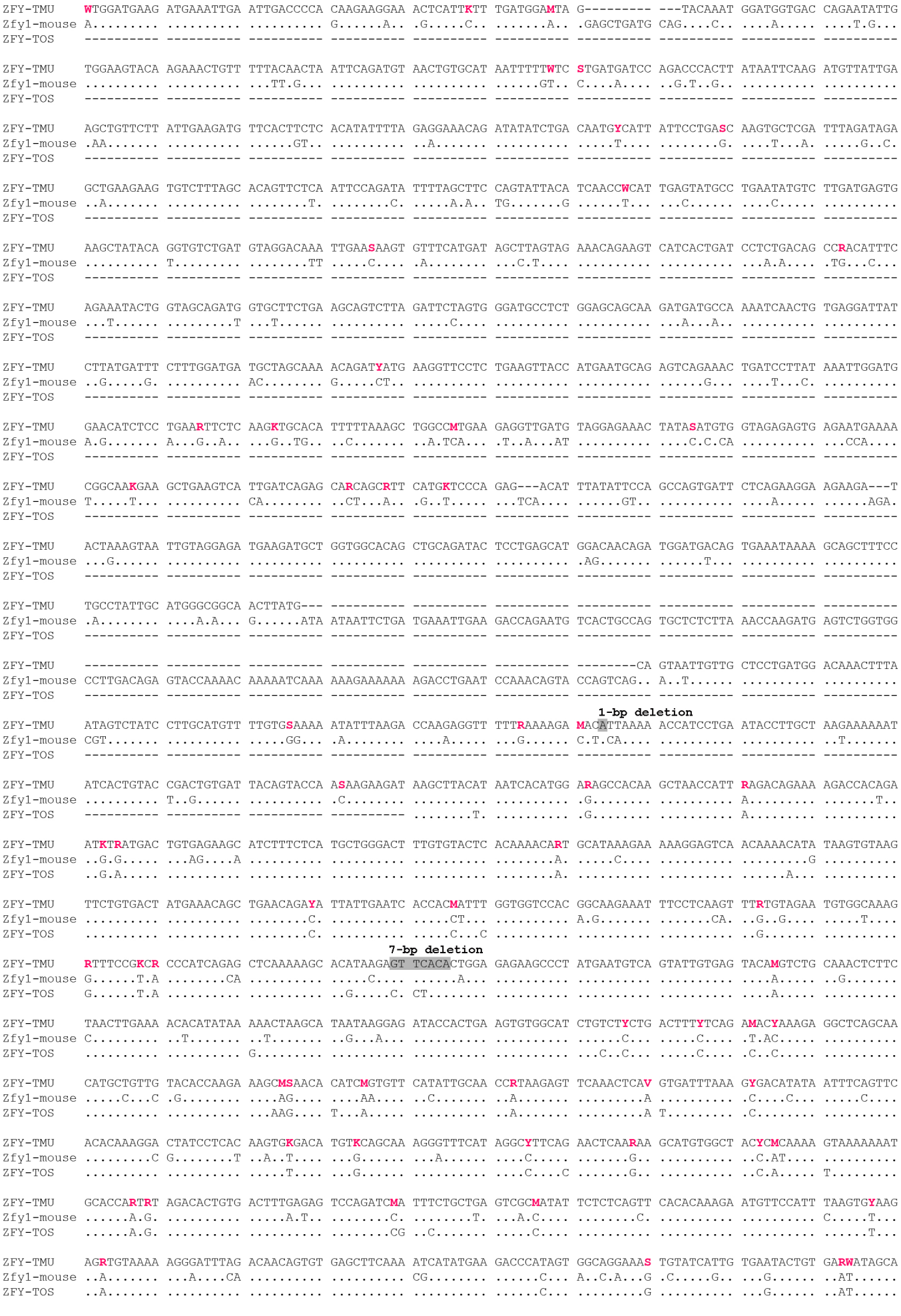
Ancestral-X

Heterochromatin

Y-to-Y

X Y

a



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ZFY-TMU    CCACAGATAC CTCAGGCTTT AAGCRGCASG TTATCTCCAT TCATAYGAAA GACTATCCTM ACCGCTGTGA GTAYTGCAGG AAAGGATTCT GGAGACCCTC
Zfy1-mouse ...A....G. ......T... ....GC..C. .......... .....C.... .........C ..A....... C.TC....A. .........C ..........
ZFY-TOS    .........T .......... ....G...C. .......... .....C.... .........C .......... ...C...... .........C ..........

ZFY-TMU    GGAAAAGART AAACACATAA TACGACATCA TAAAGAAGTT GSCGTGYCCT AA
Zfy1-mouse ........A. C......... .G........ ....---... .G.C..C... ..
ZFY-TOS    ........T. .......... .G........ .......... .G....C... ..
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b

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ZFY-TMU-mRNA1 MDEDEIELTP QEGNSFFDGL ----VQMDGD QNIVEVQETV FTTNSDVTVH NFYPDDPDPL IIQDVIEAVL IEDVHFSHIL EETDISDNAI IPDQVLDLDR
ZFY-TMU-mRNA2 .......... .....L...I ----...... .......... .......... ..FR...... .......... .......... ........V. ..E.......
ZFY-TMU-mRNA3 .......... .....L...I ----...... .......... .......... ..FR...... .......... .......... ........V. ..E.......
Zfy1-mouse    .......... E.EK.....I GADA.H..S. .IV....... .LA....... ..V..N.GSV .......N.. .....C.... ........V. ..E...N.GT

ZFY-TMU-mRNA1 AEEVSLAQFS IPDILASSIT STTLSMPEYV LMSEAIQVSD VGQIEQVFHD SLVETEVITD PLTANISEIL VADGASEAVL DSSGMPLEQQ DDAKINCEDY
ZFY-TMU-mRNA2 .......... .......... ..S....... .......... .......... .......... ....D..... .......... .......... ..........
ZFY-TMU-mRNA3 .......... .......... ..S....... .......... .......... .......... ....D..... .......... .......... ..........
Zfy1-mouse    .........L .....T.G.. ..S.T...H. ......H... ..HF...I.. .......... .I..DT.D.. ...CV..... .......... .ND.......

ZFY-TMU-mRNA1 LMISLDDASK TDYEGSSEVT MNAESETDPY KLDGTSPEVL KVHIFKAGPE EVDVGETIHV VESENENGKE AEVIDQSSSI HVPRD-IYIP ASDSQKEE-D
ZFY-TMU-mRNA2 ........N. ..H....... .......... .......... .......... ........D. .......... .......... .G...-.... ........-.
ZFY-TMU-mRNA3 ........N. ..H....... .......... .......... .......... ........D. ........N. .......... .G...-.... ........-.
Zfy1-mouse    ..M...EP.. A.L....... .....G..SS ...EA....I ..C.L..DS. VDEL.....A ....TK..N. ...T...T.. R...VN..MS ........E.

ZFY-TMU-mRNA1 TKVIVGDEDA GGTVADTPEH GQQMDDSEIK AAFLPIAWAA TY-------- ---------- ---------- ---------- ---------A VIVAPDGQTL
ZFY-TMU-mRNA2 .......... ...A...... .......... .......... ..-------- ---------- ---------- ---------- ---------. ..........
ZFY-TMU-mRNA3 .......... ...A...... .......... .......... ..-------- ---------- ---------- ---------- ---------. ..........
Zfy1-mouse    .E........ ...A...... E....V.... ........T. A.DNNSDEIE DQNVTASALL NQDESGGLDR VPKQKSKKKK RPESKQYQS. IF........

ZFY-TMU-mRNA1 IVYPCMFCGK IFKTKRFLKR HIKNHPEYLA KKKYHCTDCD YSTNKKISLH NHMESHKLTI KTEKTTEFDD CEKHLSHAGT LCTHKTMHKE KGVNKTYKCK
ZFY-TMU-mRNA2 ........A. .......... .......... .......... .......... .......... .......CN. .......... ......V... ..........
ZFY-TMU-mRNA3 ........A. .......... .......... .......... .......... .......... .......C.. .......... ......V... ..........
Zfy1-mouse    R......... K......... .T........ N......E.. .......... .......... .......C.. .R.N...... ........T. ......C...

ZFY-TMU-mRNA1 FCDYETAEQI LLNHHHLVVH GKKFPQVCVE CGKGFRHPSE LKKHIRVHTG EKPYECQYCE YKSANSSNLK THIKTKHNKE IPLKCGICLL TFSDTKEAQQ
ZFY-TMU-mRNA2 .........T .....N.... .......... ...D..R... .......... .......... .......... .......... .......... ..........
ZFY-TMU-mRNA3 .........T .....N.... .......... ...D..R... .......... .......... .......... .......... .......... ...E......
Zfy1-mouse    .........T .....L.... R....HI.G. .........A .......... .......... ....D..... ....S..S.. .......... ....N.....

ZFY-TMU-mRNA1 HAVVHQESRT HQCSYCNHKS SNSSDLKRHI ISVHTKDYPH KCDMCSKGFH RPSELKKHVA THKSKKMHQC RHCDFESPDP FLLSRHILSV HTKNVPFKCK
ZFY-TMU-mRNA2 ........T. .......... ...R...... .......... .......... .......... .......... .......... .......... ..........
ZFY-TMU-mRNA3 ........T. .P........ ...R...... .......... ....F..... .......... .P.......Y .........Q .......... ..........
Zfy1-mouse    ...L...... ....H..... .......... ......A... .......... .......... .......... .....N.... ....H....A ..........

ZFY-TMU-mRNA1 RCKKGFRQQC ELQNHMKTHS RRKVYHCEYC EYSTTDISGF KRHVISIHTK DYPHRCEYCR KGFRRPSEKN KHIMRHHKEA GVP*
ZFY-TMU-mRNA2 .......... .......... G......... ......T... .......... .......... ...W...... .........V ....
ZFY-TMU-mRNA3 .......... .......... G......... ......T... ..Q....... .......... ...W.....S ...I.....V A.S.
Zfy1-mouse    ....E.Q... ...T...... S....Q.... ....K.A... .......... ....S.DF.K .......... Q.......-V .L..
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C

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UBA1Y-TMU   NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN
Uba1y-mouse ATGTCCAGCT CAGTCCTGTC CAAGAAACGC AAAGTGTCTG GACCTGACTC TAGTCTGGAT TCTAGTTGGT CTCCTACCTA TTCTGTGATG TTTGGCGTGC
UBA1Y-rat   ATGTCCAGCT CGGTCCTGTC CAAGAAACGC AGAGTGTCTG GACCTGACTC TGAGGTGGAT TCTAGTTGGC CTTCTACCCA TTCTGTGATG TTTGGCGCAC

UBA1Y-TMU   NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN
Uba1y-mouse CCCCAGGGCC AACCAACGAA ATGTCAAAAA ATAAGGAAAT GGACATAGAT GAAAGCCTTT ACTCCCGCCA GCTGTATGTA CTCGGCCATG AGGCAATGAA
UBA1Y-rat   CCCCAGGGCC AAACAGCGGA ATGTCAAAAA ACAAGGAAAT GGATATAGAT GAAAGCCTTT ACTCCCGCCA GCTGTATGTA CTAGGTCATG AGGCAATGAA

UBA1Y-TMU   NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN
Uba1y-mouse GCATCTCCAG GCTTCCAGTG TACTGATATC AGGCCTGCAG GGTCTGGGTG TGGAAATTGC CAAGAATATC ATCCTTGGTG GGGTCAAGGC TGTCACTCTC
UBA1Y-rat   ACATCTCCAG ACGTCCAGTG TACTGATATC AGGCCTGCAG GGTCTGGGTG TGGAAATTGC CAAGAATATC ATCCTTGGTG GGGTCAAGGC TGTCACTCTC

UBA1Y-TMU   NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN
Uba1y-mouse CATGATCAGG GCATTGCCCA GTGGGCTGAT CTGTCTTCCC AGTTCTGCCT ACGTGAGGAA GATATTGGTA AAAACCGAGC TGAGATATCC CAACCACGCC
UBA1Y-rat   CATGACCAGG GCACTGCCCA GTGGGCTGAT CTGTCTTCCC AGTTCTACCT GCATGAGGAA GATATTGGCA AAAATCGAGC TGAGGTATCC CAACCACGCC

UBA1Y-TMU   NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN
Uba1y-mouse TTGCTGAACT CAACAGTTAC GTTCCTGTGT TTGCCTACAC TGGACCTCTA ATTGAGGAGT TCCTTAGTGG TTTTCAGGTA GTGGTTCTTA CCAACACTCC
UBA1Y-rat   TTGCTGAACT CAACAGTTAT GTTCCTGTGC ACACTTACAC TGGACCTCTA GTTGACGACT TCCTTAGTGG TTTTCAGGTG GTGGTTCTTA CCAACACTCC

UBA1Y-TMU   NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN
Uba1y-mouse TTTGGAATAT CAGCTGCAGG TGGGTGAATT CTGCCATAGC CACGGAATCA AGCTGGTAGT AGCAGACACT CGGGGCCTAG TTGGACAACT CTTCTGTGAC
UBA1Y-rat   TTTGGAATAT CAGCTGCAGG TGGGTGAATT CTGCCATAGC CATGGAATCA AGCTGGTAGT AGCAGACACT CGGGGCCTAG TTGGACAACT CTTCTGTGAC

UBA1Y-TMU   NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN
Uba1y-mouse TTTGGAGAGG AAATGATTCT CACTGATTCA AATGGAGAAC AGCCACTCAG TGCCATGGTT TCAATGATCA CTAAGGAGAA TCCAGGGATT GTCACCTGCC
UBA1Y-rat   TTTGGAGAGG AAATGATTCT CACTGATGCA AATGGAGAAC AGCCACTCAG TGCTATGGTT TCAATGATCA CTAAGGAGAA TCCAGGGATT GTCACCTGCT

UBA1Y-TMU   NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN
Uba1y-mouse TGGAGGACTC CCGGCATGGA TTTGAAAGTG GTGACTTTAT CTCTTTCACA GAAGTTCAGG GCATGAGTGA ACTCAATGGC ATTGGTCCCA TAGAGATCAA
UBA1Y-rat   TGGAGGAAAC CCGGCATGGA TTTGAAAGTG GTGACTTTGT CTCTTTCACA GAAGTTCAAG GCATGAGTGA ACTGAATGGC ATTGGTCCTA TGGAGATCAA

UBA1Y-TMU   NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN
Uba1y-mouse AGTTCTGGGT CCCTATACCT TTAGTATCTG TGATACCTCC AGCTTCTCTG AGTACATCCG TGGAGGCATT GTCAGTCAAG TGAAAGTACC TCGGAAGATC
UBA1Y-rat   AGTTCTGGGT CCCTATTCCT TTAGTATCTG TGATACCTCC AGCTTCTCTG AGTACACCCG TGGAGGCATT GTCAGTCAAG TGAAAGTATC TCAGAAGATC

UBA1Y-TMU   NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN
Uba1y-mouse AATTTTAAAC CCCTACTTGC CTCGCTGGCA GAGCCAGAGT TTGTGGTCAC AGATTTTGCT AAGTGCTGTC ATCCTGCTCA GCTCCACATT GGCTTCCAGG
UBA1Y-rat   AGTTTTAAAT CCCTAGTTGC CTCGCTGGCA GAGCCAGAGT TTGTGATAAC AGATTTTGCT AAGTGCTGTC GCCCTGCTCA GCTCCACATT GGCTTCCAGG

UBA1Y-TMU   NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN
Uba1y-mouse CCCTGCATCA GTTCTGTACT CAGCACAGCA GGCCTCCTCG GCCCCATAAT GAGGAGGATG CTGAGGAATT GGTGACCTTA GCCCAATCTG TGAATGCTCA
UBA1Y-rat   CCCTGCATCA GTTCTGTACT CAGCACAGCA GGCCTCCTCG GCCCCATAAT GAGGAGGATG CTGCAGAAAT GGTGACCTTA GCACAGGCTG TGAATGCTCA

UBA1Y-TMU   NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN
Uba1y-mouse AGCTTTGCCA GCAGTTCAGC AGGATTGCCT GGACATCGAC CTTATCCGGA AGTTGGCCTA TGTAGCAGCT GGGGACCTGG CACCCATGAA TGCCTTTTTT
UBA1Y-rat   ATCTTTGCCA GCAGTCCAGC AGGATTGCCT AGATATAGAC CTCATCCGGA AGTTGGCCTA TGTAGCAGCT GGGGATCTGG CACCCATGAG TGCTTTCATT

UBA1Y-TMU   NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN
Uba1y-mouse GGTGGTTTGG CTGCCCAGGA GGTCATGAAG GCTTGTTCTG GAAAGTTTAT GCCCATTAGG CAGTGGCTGT ACTTTGATGC CCTTGAATGT CTCCCAGAGC
UBA1Y-rat   GGTGGTTTGG CTGCCCAGGA GGTCATGAAG GCTTGTTCTG GAAAGTTTAT GCCTATTAGG CAGTGGCTGT ACTTTGATGC CCTCGAATGT CTCCCGGAGC

UBA1Y-TMU   NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN NNNNNNNNNN
Uba1y-mouse ACAGAGTGGC CTTCATGGAA GATAAGTGCC TGCCACACCA GAACCGTTAT GATGGGCAAG TGGCTGTATT TGGATCAGAC CTACAAGAGA AGCTTGGCAA
UBA1Y-rat   ACAGAGTGGC CTTCATGGAA GATAAGTGCC TGCCGCGCCA GAACCGTTAC GATGGGCAAG TGGCGGTATT TGGATCAGAC CTACAAGAGA AGCTTGGCAA

UBA1Y-TMU   NNNNNNNNNN NNNNNGGTAG GTGCAGGTGC TATTGGCTGT GAGCTGCTCA AGAACCTTGC CATGATTGGT CTTGGTTGCG GGGAGGTTGG AGAAATCACA
Uba1y-mouse GCAGAAATAC TTCCT..... .......... C......... .......... .....T.... .........C ..C..C..T. ......A... ..........
Uba1y-rat   GCAGAAGTAC TTCCT..... .......... C.....T... .......... .....T.... .........C .....C..T. .A....G... ..........

UBA1Y-TMU   GTTACAGACA TGGACACCAC TGAGAAGTCA AACCTGAACA GACAGTTTCT CTTCCGTCCC CGGGACGTCA CAAAGTTAAA GTCTGAGACT GATGCTGCTG
Uba1y-mouse .......... .........T .......... .......... .G........ .......... T.....A... ....A..... .......... .C......A.
Uba1y-rat   .......... .........T .......... .......... .......... ...T..C... T....T.... ....A..... .......... .C......A.

UBA1Y-TMU   CAGTGCATGA CATAAATCCA CACATCAGGG TATTCAGCCA CCAGAATCGA GTAGGCTCTG AGACAGAACA CATCTATGAT GATGACTTCT TCCAAAACCT
Uba1y-mouse ......G... .......... .........A .......... .......... ......C.A. .......... .G........ .......... .......G..
UBA1Y-rat   ....A.G... .......... .......... .G.G...... .......... ......C... .......... .G........ .......... ..........

UBA1Y-TMU   GGATGGTGTG GCCAATGCCC TAGACAATGT GGATGCTCGA TTGTACATGG ACCGCCGTTG TGTTTACTAT TGTAAGCCTC TCCTGGAATC AGGCACTCTG
Uba1y-mouse .......... .......... .......... .......... ..A...G... .......... .......... C......... .G........ ......G...
UBA1Y-rat   .......... .......T. .......... .......... ..A....... .......... .......... C......... .G........ C.....AT..

UBA1Y-TMU   GGCACCAAGG GCAACGTGCA GGTGGTTGTT CCCTTCTTGA CAGAATCTTA CAGCTCAAGC CAGGACCCAC CTGAGAAATC CATCTCCATC TGCACACTGA
Uba1y-mouse .......... .......... .......... .......... .......... .......... .......... .......... ....C..... ..........
UBA1Y-rat   .......... .......... A......... .......... .......... ......T... .......... .......... ....C..... ..........

UBA1Y-TMU   AGAACTTCCC TAATGCCATT GAGCACACCC TGCAGTGGGC TCGGGATGAG TTTGAAGGGC TCTTCAAGCC GTCAGCCGAA AACATTAACC AGTACCTCAC
Uba1y-mouse .......... .......... .........G .......... .......... .......... .........A ......T... ..TG...... .A........
UBA1Y-rat   .......... C......... .....T.... .......... .......... ........A. .G.......A .......... ..TG...... .A.......T

UBA1Y-TMU   GGACCCCAAG TTCATGGAGA GGACACTGCA GCTAGCTGGC ACCCAGCCTT TGGAAGTACT GGAGGCTATA CAGTACAGCC TGGTCCTGCA CAGGCCACAG
Uba1y-mouse .......... .........C .......... ......C... .......... .......... ......C... ..C.G..... .......... G.........
UBA1Y-rat   .......... .........C .......... .......... .......... .......... .......... ....G..... .......... G.........

UBA1Y-TMU   ACTTGGGCCG ACTGTGTGAA GTGGGCCTAC CAGCACTGGC ACACCCAGTA TTCTCACAAC ATCCAGCAGT TACTGCACAA CTTCCCTCCA GACCAGCTTA
Uba1y-mouse .......... .........C T......... .......... .......... .......... .......... .G........ .......... .CA.......
UBA1Y-rat   .......... .........C T......... .......... .......... .......... .......... .G........ .......... ..........
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UBA1Y-TMU   CAAGCTCTGG AGCACTTTTT TGGTCAGGAC CAAAACGCTG TCCACATCCC CTCACCTTTG ACACAAACAA TCCCCTACAT CTGGATTATG TGATGGCTGC
Uba1y-mouse .G..... ..... ..... ..... .....T ..... ...T..... .....G... ..... .....
UBA1Y-rat   ..... G.T..... ..... .T..... .....A ..... ..... ..... ..... .T.....

UBA1Y-TMU   TGCCAACCTG TTTGCTAAGA CATATGGACT AGGAGGGTCC CAGGACTGTG CTGCAGTGGC CAAAGTCCTG CAGTCTCTGC CGGTCCCCAA GTTTGCCCCC
Uba1y-mouse ..... .....C... ....C..... ..... ..... ...TG..... ....C..... ..... .A..... .....T...
UBA1Y-rat   ..... .....C... ....C..... ..A..... ..... ....T...A. ..C.C..... ..... .A.C..... .....T...

UBA1Y-TMU   AAGTCTGGCA TCAGGATCCG TGTTTCTGAG CAGGAGCTGC AGAGTACCAG TGCCACTGCC AATGTTG--- ---------- ---------- ----------
Uba1y-mouse ..... .....A ..... ..... ..... ......CA.. .T..A..ACA GCCACCTGGA GGAACTCAAG ACTGCACTTC
UBA1Y-rat   ..... .....A ..... ..... ..... ......A.T. G...ACA--- GCCACCTAGA GGAACTCAAG ACTTCACTAC
                                                                  Internal stop codon
UBA1Y-TMU   ---------- ---------- ---------- ---------- ---------- ---GATGACG ACAGCAACTT TGACATGGAT TTCATTGTGG CAGCATCCAA
Uba1y-mouse CTACTCCAGA CAAGCTGCTT GGATTCAAGA TGTACCCCAT TGACTTTGAG AAG..... ..... .C..... ..... .....
UBA1Y-rat   CTACTCCAGA CAAGATGCTT GGATTCAAGA TGCATCCCAT TGATTTCGAG AAG.....T. ..... .C..... ..... .....

UBA1Y-TMU   CCTCCGAGCT GAAAACTATG GCATTTCCCC TGCAGACCGG CACAAGAGCA AACTGATTGC AGGCAAGATC ATCCCAGCCA TTGCAACCAC CACATCAGCT
Uba1y-mouse .....A ..... ..... A..... ..T..... ..... ..... ..... ..... .....T...
UBA1Y-rat   .....G..A ..... A....C.... ..... ..T..... ..... ...G..... .....T. ..... T.....

UBA1Y-TMU   GTAGTTGGCC TTGTGTGTCT GGAGCTCTAC AAGGTGGTTC AGGGCCACCA ACAGCTAGAG TCTTATAAAA ACAGTTTTAT CAACTTGGCT CTGCCTTTCT
Uba1y-mouse A..... ..... .....G... ..... ..... ...A..G... ..... ..... ..... .....G.
UBA1Y-rat   ..... .C..... .....G..T ..... ....T..... ...A..G..T ....T..... ..... ..... .....

UBA1Y-TMU   TTAGCTTCTC TGTACCTCTG GCTCCAGCGG GTCACCAGTA CTATGATCAA GAGTGGACAT TGTGGGATCA CTTTGATGTG CAGGGACTGC AACCTAGCGG
Uba1y-mouse .....T.. ..C..... .....A.T ....T..... ..... .....C ..... .....G ..... ..A..... .....
UBA1Y-rat   .C..... ..C.....A .....G.T A..... ..... .....A.. A..... .....G T..... ..... .....T..

UBA1Y-TMU   TGAGGAGATG ACCCTCAAGC AGTTTATAGA CTACTTTTAA GACAGAGCAC AAACTGGAAG TCACCATGCT GTCCCAGGGT GTGTCCATGC TCTATTCCTT
Uba1y-mouse ..... ...T.G.... .....C.... .....-.. ..... ..G..... ...T..... ..... ..... .....TG.
UBA1Y-rat   ...A..... ..... .....C.... .....-.. ..... ..G.....A .....A.. A..... ..... .....

UBA1Y-TMU   CTTGATGCCA GCCACCAAGC TCAAGGAACG GTTGGATCAG CTGATGACAG AGATCGTGAG CCGTGTGTCA AAGCGAAAAC TGGGCCATCA TGTGAAGTCC
Uba1y-mouse ...T..... ....G..... ....A..... ..... .C..... ....T..... .T..... ....A..... ..... .....A
UBA1Y-rat   ...T..... .....A. ..C..... ..... .C..... ....T..... ..... .....G.... .....G.. C.....

UBA1Y-TMU   GTTGTGTTTG AGATGTGCTG CAACAATGAG AGTGGAGAAG ACATCGAGGT TCCTTATATA TGATACATCA TCCGCTGA
Uba1y-mouse C..... ..C..... .....G...C .....C. ....A..... .....G.. C..... .....
UBA1Y-rat   C..... ..C..... ..... .....C. ....T..A.. .....G.. C..... ..G.....
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d

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UBA1Y-TMU   ?????????? ?????????? ?????????? ?????????? ?????????? ?????????? ?????????? ?????????? ?????????? ??????????
Uba1y-mouse MSSSVLSKKR KVSGPDSSLD SSWSPTYSVM FGVPPGPTNE MSKNKEMDID ESLYSRQLYV LGHEAMKHLQ ASSVLISGLQ GLGVEIAKNI ILGGVKAVTL
UBA1Y-rat   MSSSVLSKKR RVSGPDSEVD SSWPSTHSVM FGAPPGPNSG MSKNKEMDID ESLYSRQLYV LGHEAMKHLQ TSSVLISGLQ GLGVEIAKNI ILGGVKAVTL

UBA1Y-TMU   ?????????? ?????????? ?????????? ?????????? ?????????? ?????????? ?????????? ?????????? ?????????? ??????????
Uba1y-mouse HDQGIAQWAD LSSQFCLREE DIGKNRAEIS QPRLAELNSY VPVFAYTGPL IEEFLSGFQV VVLTNTPLEY QLQVGEFCHS HGIKLVVADT RGLVGQLFCD
UBA1Y-rat   HDQGTAQWAD LSSQFYLHEE DIGKNRAEVS QPRLAELNSY VPVHTYTGPL VDDFLSGFQV VVLTNTPLEY QLQVGEFCHS HGIKLVVADT RGLVGQLFCD

UBA1Y-TMU   ?????????? ?????????? ?????????? ?????????? ?????????? ?????????? ?????????? ?????????? ?????????? ??????????
Uba1y-mouse FGEEMILTDS NGEQPLSAMV SMITKENPGI VTCLEDSRHG FESGDFISFT EVQGMSELNG IGPIEIKVLG PYTFSICDTS SFSEYIRGGI VSQVKVPRKI
UBA1Y-rat   FGEEMILTDA NGEQPLSAMV SMITKENPGI VTCLEETRHG FESGDFVSFT EVQGMSELNG IGPMEIKVLG PYSFSICDTS SFSEYTRGGI VSQVKVSQKI

UBA1Y-TMU   ?????????? ?????????? ?????????? ?????????? ?????????? ?????????? ?????????? ?????????? ?????????? ??????????
Uba1y-mouse NFKPLLASLA EPEFVVTDFA KCCHPAQLHI GFQALHQFCT QHSRPPRPHN EEDAEELVTL AQSVNAQALP AVQQDCLDID LIRKLAYVAA GDLAPMNAFF
UBA1Y-rat   SFKSLVASLA EPEFVITDFA KCCRPAQLHI GFQALHQFCT QHSRPPRPHN EEDAAEMVTL AQAVNAQSLP AVQQDCLDID LIRKLAYVAA GDLAPMSAFI

UBA1Y-TMU   ?????????? ?????????? ?????????? ?????????? ?????????? ?????????? ?????????? ?????????? ?????????? ??????????
Uba1y-mouse GGLAAQEVMK ACSGKFMPIR QWLYFDALEC LPEHRVAFME DKCLPHQNRY DGQVAVFGSD LQEKLGKQKY FLVGAGAIGC ELLKNFAMIG LGCGEDGEIT
UBA1Y-rat   GGLAAQEVMK ACSGKFMPIR QWLYFDALEC LPEHRVAFME DKCLPRQNRY DGQVAVFGSD LQEKLGKQKY FLVGAGAIGC ELLKNFAMIG LGCGEGGEIT

UBA1Y-TMU   ?????????S NLNRQFLFRP RDVTKLKSET DAAAVHDINP HIRVFSHQNR VGSETEHIYD DDFFQNLDGV ANALDNVDAR LYMDRRCVYY CKPLLESGTL
Uba1y-mouse VTDMDTIEK. ..... W.I..... A....R.... ...I..... ..P....V.. .....K.... ..... ..V..... R.....
UBA1Y-rat   VTDMDTIEK. ..... W..... A....R.... ....C..... ..P....V.. ..... ..... ..... R.....

UBA1Y-TMU   GTKGNVQVVV PFLTESYSSS QDPPEKSISI CTLKNFPNAI EHTLQWARDE FEGLFKPSAE NINQYLTDPK FMERTLQLAG TQPLEVLEAI QYSLVLHRPQ
Uba1y-mouse ..... ..... .....P. ..... ...V..... .....Q... .V..... ..... ..... HC....Q...
UBA1Y-rat   ..... ..... .....P. ..... ..... .....Q... .V....M... ..... ..... .C....Q...

UBA1Y-TMU   TWADCVKWAY QHWHTQYSHN IQQLLHNFPP DQLTSSGALF WSGPKRCPHP LTFDTNNPLH LDYVMAAANL FAKTYGLGGS QDCAAVAKVL QSLPVPKFAP
Uba1y-mouse ......T... ..... ..... A..... ..... ....I..... ..... ..Q..... ....V...L. .....
UBA1Y-rat   ......T... ..... ..... .....V.. ..... ..... ..... ..Q....E.. ......TTL. ....A.....

UBA1Y-TMU   KSGIRIRVSE QELQSTSATA NVG*
Uba1y-mouse ......H... .....T IDDSHLEELK TALPTPDKLL GFKMYPIDFE KDDDSNFHMD FIVAASNLRA ENYGISPADR HKSKLIAGKI IPAIATTTSA
UBA1Y-rat   ......H... .....V DD-SHLEELK TSLPTPDKML GFKMHPIDFE KDDDSNFHMD FIVAASNLRA ENYDIPPADR HKSKLIAGKI IPAIATTTSA

Uba1y-mouse IVGLVCLELY KVVQGHQQLE SYKNSFINLA LPLFSFSAPL APECHQYYDQ EWTLWDRFDV QGLQPSGEEM TLKQFLDYFK TEHKLEVIML SQGVSMLYSV
UBA1Y-rat   VVGLVCLELY KVVQGHQQLD SFKNSFINLA LPFFSFSAPL APGYHQYYDK KWTLWDRFDV QGLQPSGEEM TLKQFLDYFK TEHKLEITIL SQGVSMLYSF

Uba1y-mouse FMPASKLKER LDQP-MTEIV SCVSKQKLGH HVKSLVFELC CNSDSGDDIE VPYVRYIIR*
UBA1Y-rat   FMPATKLQER LDQP-MTEIV SRVSKRKLGQ HVKSLVFELC CNNESGDDIE VPYVRYIIG*
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e

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EIF2S3Y-TMU-mRNA  ATGGCGGGCG GCGAGGCTGG AGACTCTCTT GGGCAGCCGC ATCTTTCTCG TCAGGATCTT GCTAMTTTGG ATGTTACCAA GTTGACKCCG CTTTCACATG
EIF2S3Y-TMUgenome ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------
EIF2S3Y-TOS       .......... .......... ....A....C .......... .......... .......... ....C..... .......... ......T... ..........
Eif2s3y-mouse     .......... .A..A..C.. T.T.A....C .......... .......... .......... ..C.C..... .......... ......T... .......G..
EIF2S3Y-rat       .......... .A..A..C.. ..TGA....C .......... .C........ .......... ..C.C..... .......... ......T... ..A.......

EIF2S3Y-TMU-mRNA  AAATTATCAG CAGACAAGCC ACAATTAATA TAGGCACAAT TGGTCATGTT GCTCATGGGA AATCTACAGT TGTAAAAGCC ATTTCTGGTG TTCACACTGT
EIF2S3Y-TMUgenome ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------
EIF2S3Y-TOS       .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Eif2s3y-mouse     .......... .......... .......... .......... .......... ........A. .......... .......... .......... ..........
EIF2S3Y-rat       .......... .......... .......... .......... .......... .......... .......... .......... .......... ....T.....

EIF2S3Y-TMU-mRNA  CCGATTCAAA AATGAACTAG AAAGGAATAT TACCATAAAA CTTGGATATG CTAATGCCAA GATTTATAAG CTTGATGACT CAAGTTGTCC TCGACCAGAA
EIF2S3Y-TMUgenome ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------
EIF2S3Y-TOS       .......... .......... .......... .......... .......... .......... .......... .......... ......A... ..........
Eif2s3y-mouse     .......... .......... .......... .......... .......... .......... A......... .......... .......... ..........
EIF2S3Y-rat       .......... ........G. .......... .......... .......... .......... .......... .......... .......... ..........

EIF2S3Y-TMU-mRNA  TGCTACAKAT CTTGTGGAAG TAGTACACCT GATGAGTTTC CTTCAGATAT TCCAGGGACC AAAGGAAACT TCAGACTAGT CAGACATGTT TCCTTTGTTG
EIF2S3Y-TMUgenome ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------
EIF2S3Y-TOS       .......G.. .......... .......... .......... .......... .......... .......... .......... .......... ..........
Eif2s3y-mouse     ..T....G.. .......... .......... .......... .......... .......... .......... .......... .......... ..........
EIF2S3Y-rat       ..T....G.. .......G.. .......... .......... .......... C.......T. .......... .......... .......... ..........

EIF2S3Y-TMU-mRNA  ATTGTCCTGG TCATGATATT TTGATGGCAA CTATGCTGAA TGGGGCAGCA GTGATGGATG CAGCTCTTCT GTTGATAGCT GGTAATGRAT CTTGTCCTCA
EIF2S3Y-TMUgenome .......... .......... .......... .......... .......... .......... .......... .......... .......... .......A..
EIF2S3Y-TOS       .......... .......... .......... .......... .......... .......... .......... .......... .......... .......A..
Eif2s3y-mouse     .......... .......... .......... .......... .......... .......... .......... .......... .......... .......A..
EIF2S3Y-rat       .......... .......... .......... .......... ...T...... .......... .......... .......... .......... .......A..

EIF2S3Y-TMU-mRNA  ACCTCAGACT TCTGAACACC TAGCAGCCAT TGAAATTATG AAGCTAAAAC ATATTTTGAT TCTGCAAAAT AAAATTGATT TGGTGAAAGA AAGTCAGGCT
EIF2S3Y-TMUgenome .......... .......... .......... .......... .......... .A........ .......... .......... ..A....... ..........
EIF2S3Y-TOS       .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Eif2s3y-mouse     .......... .......... .G..T..... .......... .......... .......... .......... .......... .......... ...C......
EIF2S3Y-rat       .......... .......... .G..T..... .......... .......... .......... .......... .......... .......... ..........

EIF2S3Y-TMU-mRNA  AAAGAACAGT ATGAACAGAT ACTTGCATTT GTACAGGGTA CAGTAGCTGA AGGAGCTCCT ATTATTCCAA TTTCTGCTCA GTTAAAATAC AATATTGAAG
EIF2S3Y-TMUgenome .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
EIF2S3Y-TOS       .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Eif2s3y-mouse     .......... .......... .......... .......... .......C.. .......... .......... .......... .......... ..........
EIF2S3Y-rat       .......... ....G..... .......... .......... .......... .......... .......... .......... .......... ..........

EIF2S3Y-TMU-mRNA  TTGTATGTGA ATATATA--G TAAAGAAAAT TCCAGTACCT CTAAGAGACT TTACTTCAGA ACCCCGACTT ATTGTTATTC GGTCTTTTGA TGTTAACAAA
EIF2S3Y-TMUgenome .......... .......TA. .......... .......... .......... .......... .......... .......... .......... ..........
EIF2S3Y-TOS       .......... .......--. .......... .......... .......... .......... .......... .......... .......... ..........
Eif2s3y-mouse     .......... G......--. .......... .......... .......... .......... .......... .......... .......... ..........
EIF2S3Y-rat       ....T..... G......--. .......... A......... .......... .......... .......... .......... .A........ ..........

EIF2S3Y-TMU-mRNA  CCTGGCTGTG AAGTTGATGA CCTTAAAGGG GGTGTAGCTG GTGGTAGTAT TTTAAAAGGT GTATTAAAGG TGGGCCAAGA GATAGAAGTG AGACCTGGGA
EIF2S3Y-TMUgenome .......... .......... .......... A......... .......... .......... .......... .......... .......... ..........
EIF2S3Y-TOS       .......... .......... .......... .......... .......... .........C .......... .......... .......... ..........
Eif2s3y-mouse     .......... .......... .......... .......... .......... .........C .......... ....A..... .......... ........T.
EIF2S3Y-rat       .......... .......... T......... .......... .......... .........A .......... .......... .......... ........T.

EIF2S3Y-TMU-mRNA  TTGTTTCTAA AGATGGAGAA GGGAAGCTTA TGTGTAAACC AATCTTTTCC AAGATTGTAT CGCTTTTTGC AGAACACAAT GATCTTCAGT ATGCTGCTCC
EIF2S3Y-TMUgenome .......... .......... .......... .......... .......... ..A....... .A........ .......... .......... ..........
EIF2S3Y-TOS       .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Eif2s3y-mouse     .......... ...C...... .......... .......... .......... .......... .C........ .......... .......... ..........
EIF2S3Y-rat       .......... ...CA.T... .......... .......... .......... ........G. .A........ .......... .......... .......A..

EIF2S3Y-TMU-mRNA  AGGTGGTCTT ATTGGAGTTG GAACAAAAAT TGACCCAACA TTATGCCGAG CAGATAGAAT GGTTGGGCAG GTCCTTGGTG CTGTTGGAGC ATTACCTGAG
EIF2S3Y-TMUgenome .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
EIF2S3Y-TOS       .......... .......... .......... .........G .......... .......... .......... .......... .......... ..........
Eif2s3y-mouse     .......... .......... .......... .........G .......... .......... .......... .......... .......... ..........
EIF2S3Y-rat       .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........

EIF2S3Y-TMU-mRNA  ATATTCACAG AATTAGAAAT TTCCTACTTC CTACTAAGAC GGCTCCTAGG TGTACGTACA GAAGGAGACA AAAAAGCAGC AAAAGTTCAA AAACTATCCA
EIF2S3Y-TMUgenome .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
EIF2S3Y-TOS       .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Eif2s3y-mouse     .......... .G........ .......... .....G.... .......... .......... .......... .G........ .......... ..G.......
EIF2S3Y-rat       .......... .......... .......... T....G.... .A........ ...T...... .......... .......T.. ...G...... ..........

EIF2S3Y-TMU-mRNA  AGAATGAAGT ACTCATGGTA AACATAGGGT CCTTGTCGAC AGGAGGCAGA GTTAGTGCAG TCAAGGCAGA TTTGGGTAAA ATTGTTCTCA CCAATCCTGT
EIF2S3Y-TMUgenome .......... ....G.A... ..T....... .......A.. .......... .......... .......... .....A.... .......... ..........
EIF2S3Y-TOS       .......... .......... .......... .......... .......... .......... .......... .......... .......... ........A..
Eif2s3y-mouse     .......... .........G .......... .......T.. .......... .......... .......... .......... ........A. .......A..
EIF2S3Y-rat       .......... .......... ........A. ....A..T.. .......... .......... .......... .......... .......... ..........

EIF2S3Y-TMU-mRNA  GTGCACAGAA GTAGGAGAAA AAATTGCTCT TAGCCGACGA GTTGAGAAGC ACTGGCGTTT AATCGGTTGG GGCCAGATAA GAAGAGGAGT GACTATCAAG
EIF2S3Y-TMUgenome ....T..... .......... .......... .....A.... .......... .......A--- ---------- ---------- ---------- ----------
EIF2S3Y-TOS       A......... .......... .......... .......... .......... .......... .......... .......... .......... ...C......
Eif2s3y-mouse     A......... .......... .......... A......... ........A. .......... ...T...... .......... .......C.. ..........
EIF2S3Y-rat       .......... .......... .......... ...T...... .....A.... .......... ...T...... .......... .......... ..........

EIF2S3Y-TMU-mRNA  CCAACAATAG ATGATGAATG A
EIF2S3Y-TMUgenome ---------- ---------- -
EIF2S3Y-TOS       .......... .......... .
Eif2s3y-mouse     .......... .......... .
EIF2S3Y-rat       .......... .......... .
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f

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EIF2S3Y-TMU-mRNA1 MAGGEAGDSL GQPHLSRQDL ATLDVTKLTP LSHEIISRQA TINIGTIGHV AHGKSTVVKA ISGVHTVRFK NELERNITIK LGYANAKIYK LDDSSCPRPE
EIF2S3Y-TMU-mRNA2 .......... .......... .N........ .......... .......... .......... .......... .......... .......... ..........
EIF2S3Y-TOS       ........T. .......... .......... .......... .......... .......... .......... .......... .......... .....Y....
Eif2s3y-mouse     .......VT. .......... .......... ..R....... .......... .......... .......... .......... .......... ..........
EIF2S3Y-rat       .......VT. .......... .......... .......... .......... .......... .......... .......... .......... ..........

EIF2S3Y-TMU-mRNA1 CYRSCGSSTP DEFPSDIPGT KGNFRLVRHV SFVDCPGHDI LMATMLNGAA VMDAALLLIA GNESCPQPQT SEHLAAIEIM KLKHILILQN KIDLVKESQA
EIF2S3Y-TMU-mRNA2 ..I....... .......... .......... .......... .......... .......... .......... .......... .......... ..........
EIF2S3Y-TOS       .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Eif2s3y-mouse     .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
EIF2S3Y-rat       .......... .........I .......... .......... .......... .......... .......... .......... .......... ..........

EIF2S3Y-TMU-mRNA1 KEQYEQILAF VQGTVAEGAP IIPISAQLKY NIEVVCEYIV KKIPVPLRDF TSEPRLIVIR SFDVNKPGCE VDDLKGGVAG GSILKGVLKV GQEIEVRPGI
EIF2S3Y-TMU-mRNA2 .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
EIF2S3Y-TOS       .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Eif2s3y-mouse     .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
EIF2S3Y-rat       .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........

EIF2S3Y-TMU-mRNA1 VSKDGEGKLM CKPIFSKIVS LFAEHNDLQY AAPGGLIGVG TKIDPTLCRA DRMVGQVLGA VGALPEIFTE LEISYFLLRR LLGVRTEGDK KAAKVQKLSK
EIF2S3Y-TMU-mRNA2 .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
EIF2S3Y-TOS       .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Eif2s3y-mouse     .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
EIF2S3Y-rat       ....S..... .......... .......... .......... .......... .......... .......... .......... .......... ..........

EIF2S3Y-TMU-mRNA1 NEVLMVNIGS LSTGGRVSAV KADLGKIVLT NPVCTEVGEK IALSRRVEKH WRLIGWGQIR RGVTIKPTID DE*
EIF2S3Y-TMU-mRNA2 .......... .......... .......... .......... .......... .......... .......... ...
EIF2S3Y-TOS       .......... .......... .......... .......... .......... .......... .......... ...
Eif2s3y-mouse     .......... .......... .......... .......... .......... .......... .......... ...
EIF2S3Y-rat       .......... .......... .......... .......... .......... .......... .......... ...
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g

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TSPY-TMU-mRNA  ATGGAGAGTC TGGAGGAGGG GAGTGTGAGG GTGGCTGCCC AGGAATTCCA GGATCTGGTC CTCCTGGAAA GCAGCYTTCT GGAGGGASGG ACTCCAATAC
TSPY-TMUgenome .......... CT........ .......G.C ...C...... .......... ........G. A......... .....C.... .......G.. ..........
TSPY-ASY       .......... CC..A..... .......G.C ......C... .........G ...G...... .......... .....C.... .......GA. G......C..
TSPY1-rat      .......A.T CT.......A .......G.C ......C... .........G ...A.....A .......... .....C.... .......GA. C.CT..G...

TSPY-TMU-mRNA  AGGCACCAGA GCAGAGCCCA GGTGCTCCTG CTGAGGACAA TCGGATGGTG AAGCCTCTGG TAATAGGTGG TCCTGTGGAG GACTGGATAA CCCTTCCAGA
TSPY-TMUgenome .......... .......... .......--- --.G...... ..A....... ........A. .......... ...G...... ..........C ..........
TSPY-ASY       .......C.. .......T.. .......... ...G...... ..A....... .......... .......... .......... ......G..C ..........
TSPY1-rat      .......... .......... .......... ...G...... C.A....... .......... .....T.... .........A ..T...G..C .....G....

TSPY-TMU-mRNA  GGTGCTACTT CACCCTGACA CGTGTGAGGA GGGGCACGTG GCTGCTGTCG GGGAGTTGAA AGTCACAGAG GTGGCRGATG TCAAGAATGA SGGTGAAGAA
TSPY-TMUgenome .......... .......... .......... .......... ........G. .......... .......... .....G.... .......... G.........
TSPY-ASY       .......... .......... .......... ....G...A. .T....AAG. .......... .......... ....TG.... .......... GA..C....G
TSPY1-rat      ........A. .......... .......A.. ...TG...A. ...A..A.G. C......... .......... ....TA.... .......... G........G

TSPY-TMU-mRNA  ATAGAGCAGC ACAAGCAGGA TGGTGAAGAA GATCAGCAGC CTGAGCTGGA AAAGGACCCA GAGCAGGCCT GTGAGTCTAA AGACCAGCAT GGGATACAAA
TSPY-TMUgenome ...A...... ...G...... .........- --........ ...C...... .......... .......... ....T..... .......... ..........
TSPY-ASY       G..A..G... .......... A......... ........A. ....AT.... .......... .......... ....T..... ...T...... ..........
TSPY1-rat      G..A..G.T. .G..A..... A......C.. .......... .......... ....A..... .....A.... ....T..... ...T.....A ..........

TSPY-TMU-mRNA  GGCTGCCACA GCCAAGGGGA GGAACTGCAG AAAGCAGATC TAAAATGGAG GAGCTGGAAC TTCTTCAACT GGAGCTCAGC TTTGTGAATG CCCGCTATAG
TSPY-TMUgenome .......... .T.C.....T ..G....... .......... .......... .......... .......... .......... .......... ..T.......
TSPY-ASY       .......... .T..G.T..T ..G....... ..C....... .......... .......... .......... .......... .........T ......G...
TSPY1-rat      ....T....T .T..G.T..C ..GC...... ..C....... .......... .......... .......... .......... .......... ......G...

TSPY-TMU-mRNA  TGGAGATCTT GCACGGATTA AAACCCAGGT GGCCAAAATG CGCAAACCTC ACTTTGAGGG CAGAAAGACC ATAATCGAGG GCATCCCAGG CTTCTGGGCC
TSPY-TMUgenome .....C.... ....A..... ....AA.... .......... .......... ........C. .......... .....T.... .......... .........T
TSPY-ASY       .....C.T.. ..C....... ..G.AA.... .......... .......... ........C. .......... ......C... .......... .........T
TSPY1-rat      C....C.T.. ..C....... ..G.AA.A.. .......... ....G..... .......TC. .......... ......C... ....T..... .........T

TSPY-TMU-mRNA  AAAGCTATGA TGAACCATCC CCAGATGTCA TCCATCATCA GCAACCAAGA TGAAGATTTA TTAAGTTACA TGTTGAGCTC GGAGGTGGAG GAGTACAACC
TSPY-TMUgenome .......... .......... .........G .......... .......... ......C... ..G..C.... .........T .........CA ..........
TSPY-ASY       .......... .......... .......... ..T.....T. ....T..... ......C... ..G..C.... .........T .......... ..........
TSPY1-rat      .......... .......... .........C .......... .......... ......C... C.G..C.... .........T .......... ........T.

TSPY-TMU-mRNA  CTGGGCCAAG GATGTGCAGA ATGATGTTTT CCTTTAGTGA AAACCCATAT TTCCGGAATG ATGTTGTCAC TAAAGACTAT CAACTCAGTG TCATTGGATA
TSPY-TMUgenome .......... .......... .......... T....C.... .........C .......... .CA......T ...G...... .........CA ..........
TSPY-ASY       ......TG.. .......... .......... T......... .........C .....A.... .......... ...G...... .........CA ..........
TSPY1-rat      .....TTG.. .........G .......... T......... .........C .......... .CA....... ...G..T... .........CA ..........

TSPY-TMU-mRNA  CAAAGAGTCA GATTCCAGTA CAATAGAGTG GGTTGGACAG GCTGAGAATG GATATGCCAA CTGCATGCAA GAAACTACCA GACTCACTTT CTTCAACTGG
TSPY-TMUgenome .......... .....T.... .......... .A........ ....T.C... ...G...... .......... ..T....... ....T..... ..........
TSPY-ASY       ......A... .......... .......... .A........ ......C... .T........ .......... ..C....... .......... ..........
TSPY1-rat      .......... .....G.... .......... .A........ ......C... .T........ .......... ..C....... .......... ..........

TSPY-TMU-mRNA  TTGTGTGCAC ACAAATTCCC TGGATCCAAC AGGATTGCTG AGATAATCAT GGATGACCTG TGGCCAAATC CTCTTTACTA TTACCCTAAG GAAGACCACT
TSPY-TMUgenome .......... .......... .......... .A........ .......... .......... .......... ..G....... .......... .....T....
TSPY-ASY       .......... .......... ...C...... .......... .......... .......... .......... .......... .......... ..........
TSPY1-rat      C.......... ....N..... ...C...... .......... .......... .......T.. .......... .......... C......... ..........

TSPY-TMU-mRNA  CATGA
TSPY-TMUgenome .....
TSPY-ASY       .....
TSPY1-rat      .....
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h

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TSPY-TMU-mRNA1 MESLEEGSVR VAAQEFQDLV LLESSLLEGG TPIQAPEQSP GAPAEDNRMV KPLVIGGPVE DWITLPEVLL HPDTCEEGHV AAVGELKVTE VADVKNEGEE
TSPY-TMU-mRNA2 .......... .......... .....F...R .......... .......... .......... .......... .......... .......... ......D...
TSPY-ASY       ...P.....G ..P...RE.. .........E A.T......S ....G..Q.. .......... ..VP...... ........DE V.K....... .V.....SQ.
TSPY1-rat      ..NS..E..G ..P...RE.. .........E PSV....... ....G..Q.. .....C.... ..VP.A...H ........DE .TMA...... .V........

TSPY-TMU-mRNA1 IEQHKQDGEE DQQPELEKDP EQACESKDQH GIQRLPQPRG GTAESRSKME ELELLQLELS FVNARYSGDL ARIKTQVAKM RKPHFEGRKT IIEGIPGFWA
TSPY-TMU-mRNA2 .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
TSPY-ASY       VKE...E... .......... ....D..... .......SG. ....R..... .......... ...S.C..AF ....AK.... ......R... ..Q.......
TSPY1-rat      VKDQ..E..Q ........N. ....D....Q ......LSG. .P..R..... .......... .....C..AF ....AK.... .R...DR... ..Q.......

TSPY-TMU-mRNA1 KAMMNHPQMS SIISNQDEDL LSYMLSSEVE EYNPGPRMCR MMFSFSENPY FRNDVVTKDY QLSVIGYKES DSSTIEWVGQ AENGYANCMQ ETTRLTFFNW
TSPY-TMU-mRNA2 .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
TSPY-ASY       .......... .......... ......L... .....L.... ...F...... .......... ...I...... .......I.. ..H....... D.........
TSPY1-rat      .......... .......... ......L... .....L.... ...F...... ....I..... ...I...... .......I.. ..H....... D.........

TSPY-TMU-mRNA1 LCAHKFPGSN RIAEIIMDDL WPNPLYYYPK EDHS*
TSPY-TMU-mRNA2 .......... .......... .......... .....
TSPY-ASY       .......... .......... .......... .....
TSPY1-rat      ....?..... .......... .......... .....
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i

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UTY-TMU          ATGAAATCAT ACGGATTGCC TTTCACTACT GCTTTCCTAG GTAATGAGGA AGAGAAAATG GCGGCGAAAA AGGCTAGAGG CGAGGGCGAA GAGGGGTCCT
UTY-TMUgenome    .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Uty-mouse        ........T. .......AT. .C........ ...GC..... .......... .A........ ......G... .......... .......... ..........
UTY-rat          ...C.GATGC CA.TGAA.TT .GAT.T.CTA AGAA..TCG. .......... .A........ ......G... .......... .......... ..........

UTY-TMU          TCAGTCTCAC AGTCGAAGAG AAGAAGGCGC TTTGTGGATT AGATAGCAGC TTCTTCGGGT TCTTGAGCCG ATGCAAAGAG GGCGCCAAGA TGAAGACGCT
UTY-TMUgenome    .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Uty-mouse        .......... .......... .......... .......... .......... .......... ......C... .........T .......... ..........
UTY-rat          .......... .......... .......... .......... ...C...... .......... .........T .........T ....T..... ..........

UTY-TMU          GTTGAACAAG GCAATTCGCT GCTATGAATC TTTAATAGTA AAAGCTGAAG GAAAAGTGGA ATCAGAATTC TTTTGTCAAT TAGGTCACTT CAATCTCTTA
UTY-TMUgenome    .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Uty-mouse        .......... .......A.. T......G.. .......... .......... .......... ......T... .......... .......... ..........
UTY-rat          .......... .......AT. .......... .......... .......... .......... ......T... .......... .......... ..........

UTY-TMU          CTGGAAGATT ATTCAAAAGC ATTGTCATCT TACCAGAGAT ACTACAGTTT ACAGACTGAC TACTGGAAGA ATGCTGCCTT TTTATATGGT CTTGGTTTGG
UTY-TMUgenome    .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Uty-mouse        .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
UTY-rat          .......... .......... .......... .......... .......... .......... .......... .......A.. .......... ..........

UTY-TMU          TCTACTTCTA CTACAATGCA TTTCAGTGGG CAATTAGAGC ATTTCAAGAA GTCCTTTATG TTGACCCTAA CTTTTGCAGA GCCAAGGAAA TTCATTTACG
UTY-TMUgenome    .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Uty-mouse        ....T..... .......... .......... .......... T.....G... .......... .......... T......... .....A.... .C........
UTY-rat          .......... .......... .....A.... .......... .......... .......... .......... T......... .......... ..........

UTY-TMU          ACTTGGTTTT ATGTTCAAAG TGAATACAGA TTATGAGTCT AGTTTAAAGC ATTTTCAACT GGCACTGATC GACTGTAATG TATGCACCTT GTCCAGTGTC
UTY-TMUgenome    .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Uty-mouse        .......... .........A .......... ......A... .......... .......G.. ...C...... .......... .C..T..T.. ..........
UTY-rat          ......C... .......... .A........ .......... .......... .......G.. ...T..A.GG ....C..... .C..T..... .........T

UTY-TMU          GAAATTCAGT TTCATATTGC CCACTTATAT GAAACCCAGA GGAAATACCA TTCTGCAAAG GCAGCCTATG AACAACTTTT ACAAATTGAA AGCCTGCCAT
UTY-TMUgenome    .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Uty-mouse        .......... .......... T......... .......... .......... .......... .......... .......... ...G...... ...T......
UTY-rat          .......... .......... T......... ....TT.... .......... .........A .......... .......... .......... .....T....

UTY-TMU          CACAAGTCAA AGCAGCTGTA TTGCAACAGT TAGGTTGGAT GCACCATAAT ATGGATCTAG TAGGAGACAA GACCATCAAA GAAAGATATG CTATTCAGTA
UTY-TMUgenome    .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Uty-mouse        .......... ....A..... .......... .......... .......... .........A .T........ T....C.... ..G....... ..........
UTY-rat          .......... ....A..... .......... .......... .......... .......... .......... T....C.... .......... .C........

UTY-TMU          TCTCCAAAAG TCATTGGAGG AAGATCCTAA TTCTGGTCAA TCTTGGTATT TCCTTGGGAG GTGTTACTCA TGTGTTGGGA AGATTCAAGA TGCCTTTGTA
UTY-TMUgenome    .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Uty-mouse        .......... .......... .......... .......... .......... .......A.. .......... ...A...... ..G....G.. ..........
UTY-rat          ...T...... .......... .......... ......C... .......... .......A.. .........G ...A...... .AG....G.. ..........

UTY-TMU          TCTTATAGGC AATCCATTGA TAAATCAGAA GCAAGTGCAG ACACATGGTG TTCAATAGGT GTGCTCTATC AACAGCAAAA TCAACCTATG GATGCTTTGC
UTY-TMUgenome    .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Uty-mouse        .....C.... .......... .......... .......... .......... .......... .....G.... .......... .......... .....C....
UTY-rat          .......... .......... .......... .......... .......... .......... .....T.... .......... .......... .....C....

UTY-TMU          AAGCCTATAT ATGTGCTGTA CAGTTGGATC ATGGACATGC AGCAGCCTGG ATGGACTTAG GTATTCTTTA TGAATCTTGC AACCAACCCC AGGATGCCAT
UTY-TMUgenome    .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Uty-mouse        .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
UTY-rat          ....T..... .......... .....A.... .......... .......... .......... .......... ......C... ......A... ....C.....

UTY-TMU          TAAATGCTAC TTAAATGCAA CTCGGAGCAA AAGCTGTAGT AACACCTCTG CACTTATATC AAGAATTAAG TTTTTACAGG CTCAGTTATA TAACCTTCCA
UTY-TMUgenome    .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Uty-mouse        .......... .........G .......... .TCT....A. .......... ......C... .......... .......... .......G.G ..........
UTY-rat          .......... .......... .......... .......... .......... TG........ .......... .......... .......G.G ..........

UTY-TMU          CAAAATAGTC TACAGAATAA AACTAAATTA CTTCCTAGTA TTGAGGAGGC ATGGAGCCTA CCAATCCCCG CAGAGCTTAC CTCCAGGCAG GGTGCCATGA
UTY-TMUgenome    .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Uty-mouse        ....G..... .......... .......... .......... .......... .......... .....A.... .......... .......... ..........
UTY-rat          ....G..... .......... .......... .......... .......... .......... .......... .......... .......... ..........

UTY-TMU          ACACAGCACA GCAGAATGTT TCTGATAGCT GGAATAGTGT CCAGACAGCT TCTCATTATC CAGTACAGCA GAAACTTTAT GCACAGTGCT TCACAACACA
UTY-TMUgenome    .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Uty-mouse        .......... .....G.... .......C.. .......... .......... ..A...C..T .......... ....G..... A......... .....G....
UTY-rat          .......... .......... .......... .......... ...C...... ..A...C... .......... .C..G..... A......... ..........

UTY-TMU          GAAATTACAG AGTCTAGGTA AAGATCAGAA ACCTCCCTTT CAAACTGGGC CCACACGATA TCTCCAAGCA GCTAACACTA ATAATCAGAA TCAGAATGGA
UTY-TMUgenome    .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Uty-mouse        .......... ...T.T.... .......AC. ......A... .........T .......... .......... ....G..... ..G....... ..........
UTY-rat          .......... G....T.... .......AC. ..G...G... .........T .T.....G.. .A....T... ...TG.GT.G .......... ..........

UTY-TMU          ACTCATACTC TGCCTCAAAA TTTAAAGGGG GATGCTCAAA ACTACTACTC CTTACATATT CCTACCTCAG AAGAACAAAA GATCATTAAC TTTACCAAAG
UTY-TMUgenome    .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Uty-mouse        .A........ .......G.. ..C...A... .......... .TC..---.T .....G.... .......... .......... .......... ..........
UTY-rat          .A.......T .......... ..C....... ...A...... ..C...CT.. .......... .......... .......... ....T..... ..........

UTY-TMU          AGAGAAAGGA TTCAAGAAGT AAACCCTTGA GTTCTAAAAC AAGTAGGCAA GATATAGACA CT---GATAT CTGTGTTTCT GCAAAGGAAC ATTCTAATCA
UTY-TMUgenome    .......... .......... .......... .......... .......... .......... ..---..... .......... .......... ..........
Uty-mouse        ....C..... .......... ...T...... C......... .......A.. ....G..... .ATCTA.... .......AA. ......A.G. ..........
UTY-rat          ....C..... .......... ...T...... ..C....... .......... ....G..... .ATCTA.... ..A....AA. .......... .......AA.

UTY-TMU          TATTCATCAG ATCTCATCAG TTCCTATTTC CACCCTTAAT AATGAAGATT CTGTGTCACC AAATCTAACA ATTGTAGACA ATCCTCAGCT TTCTGTCTTT
UTY-TMUgenome    .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Uty-mouse        ....T..... .......... ....G..... ..G....... ...A....A. .......... .G..T...T. .......... .......... .........A
UTY-rat          .......G.. ...A...... .......... ..GT...... .......... .......... ....T...T. ........T. .......... .........G
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UTY-TMU          GTTGAAGAAG CTATTGATAA TGTGGACCAT GACACTGGAA CTTGTGACAA AGTCAATAAT GTTCATCCAG CCATTCACAA AAAGTCTGAT AATCTTACTG
UTY-TMUgenome    .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Uty-mouse        ....G....A ....C..C.. .......... ....T..... .......... .......... .......T.. .......... ....C..... ......T...
UTY-rat          ..A.G..... ....G..C.G .......... ...T..A... .......... .......... .........A ..C....... ....C..... ..........

UTY-TMU          CTTCTTCACC CTCTTCAGCC AGTTCCACAG AAACACTTTC TCCAAAGCTC ACTGAGCAG- -----ACACA CATTGTTACC AGCTTTACCA GTCCTCACAG
UTY-TMUgenome    .......... .......... .......... .......... .......... .........- -----..... .......... .......... ..........
Uty-mouse        .......... .......... .T........ .......... ..T....... .....A...- -----..... .......... .......T.. ..........
UTY-rat          .......... .......... .......... .......... ..T....G.. .........A TGCAC..... .......... .......T.. .C...A....

UTY-TMU          TGGATTACAC ACAACTAACG GAGAA---CA CAAGAACTCA GAGAGTTCTG TCAGTGTTAA TGTTGGCCTC AGACCTAGAC CTCAAATCAT ACCATCAATG
UTY-TMUgenome    .......... .......... .....---.. .......... .......... .......... .......... .......... .......... ..........
Uty-mouse        .......... ....T...T. .....GGG.. TG......T. .......... C......... .......... .........T .......... ..........
UTY-rat          .......G.. ....T...T. ...G.---.C TG........ ..A......A .........G .......... .......... .......... ......G...

UTY-TMU          TCTGTGTCCA TATACTCCAG TTCAACAGAA GTTCTGAAAG CATGCAGGAA TTTAGGTAAA AATGGCTTAT CTGATGGTCA TATTCTCTTG GATATATGCC
UTY-TMUgenome    .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Uty-mouse        .......... .......... .......... .......... .G.......G .C........ .....T..G. ..A....... .......... ..........
UTY-rat          .......... .......G.. .......... .......... .......... .C........ .....T..G. .AA....... .......... ..........

UTY-TMU          CACCTCCAAG ACCACCAACT TCACCATATC CTCCCTTACC AAAGGAGAAG TTGAATCCAC CAACTCCTAG TATTTATTTG GAAAATAAGC GTGATGCCTT
UTY-TMUgenome    .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Uty-mouse        .......... .......... ..G....... ....T..... .......... .......... .C........ .......... .......... .A........
UTY-rat          .......... .......... ........C. .......... ...A...... .......... .C........ .......... .......... ..........

UTY-TMU          TTTCCCTCCA TTACATCAAT TTTGTATCAA TCCAAAAAAC CCTGTTACAG TAATACGTGG TCTTGCTGGA GCTCTTAAGT TAGATCTTGG ACTTTTCTCT
UTY-TMUgenome    .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Uty-mouse        ...T...... .......... ....C..... .......... .......... .......... ....T..C.. .......... .......... ..........
UTY-rat          .......... .......... ....C..... .......... ........G. ....T..... C......... .......... .......... ..........

UTY-TMU          ACCAAAACTT TGGTAGAAGT TAACAATGAG CATATCGTAG AAGTAAGGAC CCAATTGTTG CAGCCAGCTG ATGAAAACTG GGATCCCTCT GGAACAAAGA
UTY-TMUgenome    .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Uty-mouse        .......... .......G.C .........A .......... ....G..... .........A ........A. .......... .......... ..........
UTY-rat          .......... .........C .......... .......... ....G..... ......T... ........A. .......... ......T... ..........

UTY-TMU          AAATCTGGCG ATATGAAAAC AAATGTTCTC ATACTACAAT TGCTAAATAT GCACAGTACC AGGCCTGCTC CTTCCAGGAA TCATTGAGAG AAGAAACTGC
UTY-TMUgenome    .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Uty-mouse        .......... .........T ...A...... .......... .......... .......... .......... .......... .......... ......A..A
UTY-rat          .......... G.......T ...A...... .......... .......... .......... .......... .......... .......... ......A..A

UTY-TMU          GAGAAGAACA CAAGTTAAAG ACTATTCAGA TAATGAATCA ACATGTTCAG ATACTTCTGG AAGGAGACAG AAAGCACCCT TTAAAACCAT AAAATGTGGG
UTY-TMUgenome    .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Uty-mouse        A......... .......... .......... .......... .......... ...A...... .......... .......... .......... ..........
UTY-rat          .....A.... .......... .......... .......... .......... ...T...CA. .......... ....T...T. .......... .........C

UTY-TMU          ATTAACATTG ATCTATCTGA TAACAAAAAG TGGAAGTTGC AGTTACATGA GCTGACTAAG CTTCCTGCTT TTGTACGTGT TGTATCAGCA GGGAATCTTC
UTY-TMUgenome    .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Uty-mouse        .......... ....G..... .......... .......... .......... ......A... .......... .......... .......... ..........
UTY-rat          .......... .C..G..... .......... .......... .......... .......... .......... .......... .......... ..........

UTY-TMU          TCAGTCATGT TGGCTATACT ATACTGGGGA TGAATTCAGT TCAACTATGT ATGAAAGTTC CAGGGAGTAG AATACCAGGT CACCAAGAAA ATAACAATTT
UTY-TMUgenome    .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Uty-mouse        .......... .......... ...T...... .......... .......... .......... .......... .......... .......... ..........
UTY-rat          .......... .........C ........A. .......... ......T... .......... .......... G......... .......... .......C..

UTY-TMU          CTGTTCTGTT AACATAAATA TTGGTCCAGG TGATTGTGAA TGGTTTGTTG TACCCGAAGA TTATTGGGGT GTTCTGAATG ACTTCTGTGA AAAAAATAAC
UTY-TMUgenome    .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Uty-mouse        .......... .......... .......... .......... .......... ....T..... .......... .......... .......... .........T
UTY-rat          .......... .......... .......... .......... .......... ....T..... .......... .......... .......... .........T

UTY-TMU          TTGAATTTTC TGATGAGCTC TTGGTGGCCG AACCTTGAAG ATCTTTATGA AGCAAATGTT CCTGTTTATA GGTTTATTCA AAGACCTGGA GATTTGGTCT
UTY-TMUgenome    .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Uty-mouse        ........C. .......T.. .......... .......... .......... .......... .......... .......... .C.......T ........T.
UTY-rat          ........CT .......T.. .......... .......... .......... .......... .......... .......... .......... ..........

UTY-TMU          GGATAAATGC AGGCACTGTT CACTGGGTTC AAGCTATTGG GTGGTGTAAC AATATCACCT GGAATGTTGG TCCACTCACA GCCTTCCAGT ATAAATTGGC
UTY-TMUgenome    .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Uty-mouse        .......... .......... .......... .G........ .......... ..C....... .......... .......... ........A. ..........
UTY-rat          .......... .......... .......... .G........ .......... ..C....... .......... .......... .......... ..........

UTY-TMU          AGTTGAACGA TATGAATGGA ACAAATTGCA AAGTATCAAG TCAGTAGTAC CCATGGTTCA TCTTTCATGG AATATGGCAC GAAATATCAA AGTTTCAGAT
UTY-TMUgenome    .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Uty-mouse        .......... .......... .T........ ....G..... .......... .......... .......... .......... .......... ...C......
UTY-rat          .......A.. .......... .......... .......... .......... .......... .......... .......... .......... ...C......

UTY-TMU          CCAAAGCTTT TTGAAATGAT TAAATATTGT CTCCTCAAGA TTCTCAAGCA GTGTCAGACT TTGAGAGAAG CTCTTGTTGC AGCAGGGAAA GAGGTTTTGT
UTY-TMUgenome    .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Uty-mouse        .......... .......... .......... .......... ....A..... T......... .......... .......... .......... ..........
UTY-rat          .......... .......... ...G..A--- ---------- ---------- ---------- ---------- ---------- ---------- ----------

UTY-TMU          GGCATGGGAG GATAAATGAT GAACCAGCTC CTTACTGTAG CATCTGTGAG GTGGAAGTTT TTAATTTGCT TTTTGTCACT AACGAAAGCA ATTCTGAAAA
UTY-TMUgenome    .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Uty-mouse        .......... .......... .......... .......... .......... .......... .......A.. .......... ..T....... .....C....
UTY-rat          ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------

UTY-TMU          GACATATATA GTACATTGTC AGAACTGTGC ACGAAAAACA AGTGGAAATT TGGAAAATTT TGTGGTGCTA GAACAGTACA AAATGGACGA CCTGATTCAA
UTY-TMUgenome    .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Uty-mouse        .......... .......... .......... .......... .......... .......... ...A...... .....A.... .......G.. ..........
UTY-rat          ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------
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UTY-TMU          GTCTATGACC AATTTACATT AGCTCCTTCA TTATCGTCTG CATCTTAA
UTY-TMUgenome    .......... .......... .......... .......... ........
Uty-mouse        .......... .......... .........G .....T.... ........
UTY-rat          ---------- ---------- ---------- ---------- --------
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UTY-TMU      MKSYGLPFTT AFLGNEEEKM AAKKARGEGE EGSFSLTVEE KKALCGLDSS FFGFLSRCKE GAKMKTLLNK AIRCYESLIV KAEGKVESEF FCQLGHFNLL
Uty-mouse    ......SL.. .A.....K.. ..E...... .......... .......... .....T...D .......... ..HF...... ........D. ..........
UTY-rat      .QMPVKFDIL RIS....K.. ..E...... .......... .......... ......L..D .V........ ..H....... ........D. ..........

UTY-TMU      LEDYSKALSS YQRYYSLQTD YWKNAAFLYG LGLVYFYYNA FQWAIRAFQE VLYVDPNFCR AKEIHLRLGF MFKVNTDYES SLKHFQLALI DCNVCTLSSV
Uty-mouse    .......... .......... .......... .......... .......... .......... .......... ...M...... .......... ..........
UTY-rat      .......... .......... .......... .......... .......... .......... .......... .......... .........R .S........

UTY-TMU      EIQFHIAHLY ETQRKYHSAK AAYEQLLQIE SLPSQVKAAV LQQLGWMHHN MDLVGDKTIK ERYAIQYLQK SLEEDPNSGQ SWYFLGRCYS CVGKIQDAFV
Uty-mouse    .......... .......... .......... ........T. .......... ...I..N.T. .......... .......... .......... .I..V.....
UTY-rat      .......... .I........ .......... ........T. .......... ......N.T. .......... .......... .......... .I..V.....

UTY-TMU      SYRQSIDKSE ASADTWCSIG VLYQQQNQPM DALQAYICAV QLDHGHAAAW MDLGILYESC NQPQDAIKCY LNATRSKSCS NTSALISRIK FLQAQLYNLP
Uty-mouse    .......... .......... .......... .......... .......... .......... .......... ...A.....N .....T.... ......C...
UTY-rat      .......... .......... .......... .......... .......... .......... ..T....... .......... ...V...... ......C...

UTY-TMU      QNSLQNKTKL LPSIEEAWSL PIPAELTSRQ GAMNTAQQNV SDSWNSVQTA SHYPVQQKLY AQCFTTQKLQ SLGKDQKPPF QTGPTRYLQA ANTNNQNQNG
Uty-mouse    .S........ .......... .......... ........S. ..T....... ..HS....V. T....A.... .F....Q... ...S...... .S..D.....
UTY-rat      .S........ .......... .......... .......... .......H.. ..H....QV. T......... G.....QR.. ...S...IH. .CVD......

UTY-TMU      THTLPQNLKG DAQNYYSLHI PTSEEQKIIN FTKERKDSRS KPLSSKTSRQ DIDT-DICVS AKEHSNHIHQ ISSVPISTLN NEDSVSPNLT IVDNPQLSVF
Uty-mouse    N......S.. ....H-F.R. .......... ....S..... .S.T.....K .R..SN...N ..K.....Y. .......S.. .KE....D.I .........L
UTY-rat      N......S.. .T..HS.... ........F. ....S..... .S..P..... .R..SN.Y.N .....KN..E .T.....S.. .........I .........L

UTY-TMU      VEEAIDNVDH DTGTCDKVNN VHPAIHKKSD NLTASSPSSA SSTETLSPKL TEQ--THIVT SFTSPHSGLH TTNGE-HKNS ESSVSVNVGL RPRPQIIPSM
Uty-mouse    .G.T...... .I........ ..L.....P. ..S..... I......L.. ...--..... ..I....... .I...G.E.L ...A...... ...S......
UTY-rat      .G..M.S... .SR....... ...TL...P. .......... .......L.V ...MH..... ..I..N...D .I..G-PE.. ...I..S... ..........

UTY-TMU      SVSIYSSSTE VLKACRNLGK NGLSDGHILL DICPPPRPPT SPYPPLPKEK LNPPTPSIYL ENKRDAFFPP LHQFCINPKN PVTVIRGLAG ALKLDLGLFS
Uty-mouse    .......... ......S... ....N..... .......... .......... .......... .......... .......... .......... ..........
UTY-rat      .......... .......... ....N..... .......... .......... .......... .......... .......... .......... ..........

UTY-TMU      TKTLVEVNNE HIVEVRTQLL QPADENWDPS GTKKIWRYEN KCSHTTIAKY AQYQACSFQE SLREETARRT QVKDYSDNES TCSDTSGRRQ KAPFKTIKCG
Uty-mouse    ......A... .......... .......... .......... .S........ .......... .....NE... .......... ....N..... ..........
UTY-rat      ......A... ........F. .......... .......... .S........ .......... .....NE.K. .......... ....I.R... .V........

UTY-TMU      INIDLSDNKK WKLQLHELTK LPAFVRVVSA GNLLSHVGYT ILGMNSVQLC MKVPGSRIPG HQENNNFCSV NINIGPGDCE WFVVPEDYWG VLNDFCEKNN
Uty-mouse    .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
UTY-rat      .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........

UTY-TMU      LNFLMSSWWP NLEDLYEANV PVYRFIQRPG DLVWINAGTV HWVQAIGWCN NITWNVGPLT AFQYKLAVER YEWNKLQSIK SVVPMVHLSW NMARNIKVSD
Uty-mouse    .......... .......... .......... .......... .......... .......... .......... .......... ........V. .......... ..........
UTY-rat      .......... .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........

UTY-TMU      PKLFEMIKYC LLKILKQCQT LREALVAAGK EVLWHGRIND EPAPYCSICE VEVFNLLFVT NESNSEKTYI VHCQNCARKT SGNLENFVVL EQYKMDDLIQ
Uty-mouse    .......... ......H... .......... .......... .......... .......... .....Q.... .......... .......... .....E....
UTY-rat      ........*- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------

UTY-TMU      VYDQFTLAPS LSSAS*
Uty-mouse    .......... ......
UTY-rat      ---------- ------
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DDX3Y-TMUmRNA      ATGAGTCAAG TGGAAGCTGA AAATACTGTC GGCCTAGACC AGCAATTCGC AGGTCTTGAC CTGAACTCAT CAGATAATCC AAATGGAGGA GGAAATACAG
DDX3Y-TMUgenome    .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Ddx3y_mouse        .......... ...C...G.. ..G...G.C. .....G.... .......T.T T......... .....A.... .........A G......... ..........
DDX3_rat           .C.......A ...CG..G.. ......G..T .....T.... .......T.. T......... A.........- --.......A G......... .TT.G.....

DDX3Y-TMUmRNA      AGAGCAAAGG GCGCTATATA CCTCCTCACT TAAGGAACAG AGAAACATCT AAAGGTGTCT GTGATAAGGA CAGTTCAGGA TGGAGTTGTA GTAAAGATAA
DDX3Y-TMUgenome    .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Ddx3y_mouse        .......... .......... .......... .......... .......... .....G.... .......... .......... .....C.... ..........
DDX3_rat           .A........ .......... .......... .......... .......... .......... .......... ..A......G ......---- --........

DDX3Y-TMUmRNA      AGATGCCTAT AGTAGTTTTG GATCTCGTGA TTCCAGAGGA AAGCCCAATT ATTTCAGTGA TCCTGGAAGT GGATCCAGGG GAAGATTTGA TGATCTTGGT
DDX3Y-TMUgenome    .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Ddx3y_mouse        .........C ..C....... .......... .........G .......... .......... ..G....... .......... ....G..C.. .....A....
DDX3_rat           .........C ..C....... .......G.. .......... ..A....... .......... ..G....... ........A. ....G..... .....C....

DDX3Y-TMUmRNA      CGAAGTGACT ATGATGGTAT TGGCGGTCGT GACCGAACTG GTTTTGGCAA ATTT------ ---------- --CGTTGGGG TGACAGATCA GATGAAGATG
DDX3Y-TMUgenome    .......... .......... .......... .......... .......... ....------ ---------- -- ......... .......... ..........
Ddx3y_mouse        ....A..... .......... ...T...... .......... .A........ ....GAACGG AGTGGTCATA GT......A. .......... ..........
DDX3_rat           .......... .....T.... ..C.A..... .......G.. .A........ ....GAACGA AGTGGTTATA GT........ .....A.... ..........

DDX3Y-TMUmRNA      ATTGGTCAAA ACCACTTCCA CCAAGTGAAC GCTTAGAGCA AGAACTTTTT TCTGGAGGAA ACACAGGAAT TAACTTTGAG AAATATGATG ATATACCAGT
DDX3Y-TMUgenome    .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Ddx3y_mouse        .C........ .......... .......... .......... .......... .......... .......G.. .......... .......... ..........
DDX3_rat           .......... .......... .......... .......... .........C ........T. .TG....G.. .......... .......... ..........

DDX3Y-TMUmRNA      AGAGGCAACT GGAAATAATT GTCCTCCTCA CATCGAAAAT TTCAGTGATG TTGAGATGGG AGAAATTATC ATGGGGAACA TTGAACTTAC TCGTTATACT
DDX3Y-TMUgenome    .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Ddx3y_mouse        ...A...... .......... .......... ...T...... .........A .......... .......... .......... .......... ......C...
DDX3_rat           .......... .......... .......... ...T...... .......... .......... .......... .....A.... .......... ......C...

DDX3Y-TMUmRNA      CGTCCTACTC CAGTGCAAAA ACATGCTATT CCAATTATTA AAGAAAAAAG GGACTTAATG GCTTGTGCTC AAACAGGGTC TGGAAAAACT GCTGCATTTC
DDX3Y-TMUgenome    .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Ddx3y_mouse        .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
DDX3_rat           .......... ....T..G.. .......... .......... .......... .......... .......... .......... ....TGT... ..........

DDX3Y-TMUmRNA      TACTACCCAT TCTCAGTCAG ATTTATACAG ATGGTCCAGG AGAGGCTTTG AAGGCTATGA AGGAAAATGG AAGATATGGT CGCCGTAAAC AGTACCCAAT
DDX3Y-TMUgenome    .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Ddx3y_mouse        .......... .......... .......... .......... .......... .......... .......... .......... .......... .A........
DDX3_rat           .......... .......... .....C.... .......... .......... .......... .......C.. .......... .......... .A........

DDX3Y-TMUmRNA      CTCCTTGGTG TTAGCCCCAA CAAGAGAATT GGCTGTGCAG ATCTATGAGG AAGCCAGAAA ATTTTCATAC AGGTCTAGAG TCCGTCCTTG TGTAGTGTAT
DDX3Y-TMUgenome    .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Ddx3y_mouse        .......... .......... .......... .......... .......... .......... .......... C.A....... .......... ..........
DDX3_rat           .......... .....T.... .......... .......... .......... .......... ......G... C......... ....A..... ..........

DDX3Y-TMUmRNA      GGCGGGGCTG ATACTGTTCA GCAGATTCGG GACTTAGAAC GCGGATGCCA CTTGTTAGTT GCCACACCCG GACGTCTAGT GGACATGATG GAAAGAGGAA
DDX3Y-TMUgenome    .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Ddx3y_mouse        ..T..T.... .......... .......... .......... .T........ .......... ........A. .......... .......... ..........
DDX3_rat           ..T..T.... .......... A......... .......... .A........ .......... ........A. .......... .......... ..........

DDX3Y-TMUmRNA      AGATTGGATT AGATTTCTGC AAATACTTAG TGTTGGATGA AGCCGATAGG ATGTTGGATA TGGGATTTGA ACCTCAGATA CGTCGTATAG TTGAGCAGGA
DDX3Y-TMUgenome    .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Ddx3y_mouse        .......... .......... .......... .A........ ...T...... ...C...... .......... ......A... .......... ..........
DDX3_rat           ........C. .......... .......... .......... ...T..C... ...C...... .......... .......... .......... ....A.....

DDX3Y-TMUmRNA      CACAATGCCA CCAAAGGGGG TTCGTCATAC TATGATGTTT AGTGCTACTT TCCCTAAGGA GATACAGATG CTTGCTCGTG ACTTTTTAGA TGAATATATC
DDX3Y-TMUgenome    .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Ddx3y_mouse        .......... .......... .......C.. .......... .......... .T........ .......... .......... .......... ..........
DDX3_rat           ...T...... ........A. .......C.. C......... .......... .T........ .......... .......... .......... ......C...

DDX3Y-TMUmRNA      TTTCTGGCTG TAGGCAGAGT AGGCTCTACT TCTGAGAACA TCACACAGAA AGTAGTATGG GTTGAAGAAC TAGATAAACG GTCATTTTTA CTTGATCTCT
DDX3Y-TMUgenome    .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Ddx3y_mouse        .......... .......... ...A...... .......... .......... .......... .......... .......... ....C..... ..........
DDX3_rat           .......... .......... .......... .......... .......... .......... .........GT ....C..... .......... ..........

DDX3Y-TMUmRNA      TAAATGCAAC AGGGAAGGAT TCATTGACTT TAGTATTTGT GGAGACTAAA AAGGGTGCAG ACTCCCTGGA GAACTTCTTA TTTCAAGAAA GATATTCGTG
DDX3Y-TMUgenome    .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Ddx3y_mouse        .......... .......... .......... .......... .......... .......... .T..G..... .......... .......... .....G....
DDX3_rat           .......... .....G.... .......... .......... .......... .......... .T..A..... .......... .......... .....G.A..

DDX3Y-TMUmRNA      TACTAGTATT CATGGAGACA GATCACAGAA AGATAGAGAA GAGGCCCTTC ATCAGTTCCG CTCAGGGAGA AAGCCAATTT TGGTGGCTAC AGCTGTGGCA
DDX3Y-TMUgenome    .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Ddx3y_mouse        .......... .......... .......... ....C....G .......... .C.....T.. .......C.. .......... .A........ ..........
DDX3_rat           .......... .......... .G........ ....C....G .......... .C..A..... .........G .......... .A........ ..........

DDX3Y-TMUmRNA      GCAAGAGGTC TAGACATTTC AAATGTGAAA CATGTTATCA ATTTTGATTT GCCAAGTGAT ATTGAAGAAT ACGTGCATCG TATTGGCCGC ACAGGACGTG
DDX3Y-TMUgenome    .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Ddx3y_mouse        .......... .......... .......... .......... .......... ......C... .......... .T........ A......... ..........
DDX3_rat           .......... .......... .......... .......... .......... .......... .......... .T........ .......... .........C.

DDX3Y-TMUmRNA      TAGGAAACCT TGGACTTGCC ACCTCTTTTT TCAATGAGAG GAATTTGAAC ATTACAAAAG ATTTGTTAGA TCTCCTTGTT GAGGCTAAAC AAGAAGTTCC
DDX3Y-TMUgenome    .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Ddx3y_mouse        .......... .......... .......... .......... .......... ..C....... .......... .......... .......... .......G..
DDX3_rat           .......... ...T...... .....C.... .......... .......... .....G.... .......... ......C... .......... .......A..

DDX3Y-TMUmRNA      ATCTTGGTTG GAAAGTATGG CCTACGAACA CCACTACAAG GGAAGCAGCC GGGGACGTTC AAAAAG---A TTCAGTGGAG GATTTGGAGC CAGAGACTAT
DDX3Y-TMUgenome    .......... .......... .......... .......... .......... .......... ......---. .......... .......... ..........
Ddx3y_mouse        T......... .......... .T..T..... .......... .....T.... .T........ T.....CAG. .......... .......... ..........
DDX3_rat           .......... .......... .T..T..... .......... ....A..... .......... T.....CAG. .......... .......T.. ..........
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DDX3Y-TMUmRNA      CGACAGAGCA GTGGTTCTGC AAATTCTGGG TTTAATACTA ATCGTGCAAA CAGCAGCCGA AGTAGTGGTA GTAGCCACAA CAGAGGATTT GGTGGAGGTG
DDX3Y-TMUgenome    .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Ddx3y_mouse        .......... .......... ....G....A .......G.. .C.....C.. T......... .......... .......... .......... ..........
DDX3_rat           .......... ......A... .......... .......A.. .......C.. .......... .C........ ...C...... .......... ..........

DDX3Y-TMUmRNA      GCTATGGAAG TTTCTACAAT AATGATGGAT ATGGAGGAAA TTATAACTCC CAGGCAGTTG ACTGGTGGGG CAATTGA
DDX3Y-TMUgenome    .......... .......... .......... .......... .......... .......... .......... .......
Ddx3y_mouse        ........G. .......... ........T. .......... .......... .......... .......... .......
DDX3_rat           ........G. ........GC .....A.... .......... .......... .......... .......... .......
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DDX3Y-TMUmRNA      MSQVEAENTV GLDQQFAGLD LNSSDNPNGG GNTESKGRYI PPHLRNRETS KGVCDKDSSG WSCSKDKDAY SSFGSRDSRG KPNYFSDPGS GSRGRFDDLG
Ddx3y_mouse        ....A..S.A ......V... .K....Q... .......... .......... .......... .......... .......... .......R.. .........H.
DDX3_rat              MA..... .......... M..-..Q... VS........ .......... .......N.. ..--...... .......... .......R.. .........P.

DDX3Y-TMUmRNA      RSDYDGIGGR DRTGFGKF-- ----RWGDRS DEDDWSKPLP PSERLEQELF SGGNTGINFE KYDDIPVEAT GNNCPPHIEN FSDVEMGEII MGNIELTRYT
Ddx3y_mouse        .N........ ........ER SGHS..S... .......... .......... .......... .......... .......... ...I...... ..........
DDX3_rat           .....C.AS. ..S.....ER SGYS....K. .......... .......... ....A..... .......... .......... .......... ..........

DDX3Y-TMUmRNA      RPTPVQKHAI PIIKEKRDLM ACAQTGSGKT AAFLLPILSQ IYTDGPGEAL KAMKENGRYG RRKQYPISLV LAPTRELAVQ IYEEARKFSY RSRVRPCVVY
Ddx3y_mouse        .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
DDX3_rat           .......... .......... ........C. .......... .......... .......... .......... .......... .......... ..........

DDX3Y-TMUmRNA      GGADTVQQIR DLERGCHLLV ATPGRLVDMM ERGKIGLDFC KYLVLDEADR MLDMGFEPQI RRIVEQDTMP PKGVRHTMMF SATFPKEIQM LARDFLDEYI
Ddx3y_mouse        .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
DDX3_rat           .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........

DDX3Y-TMUmRNA      FLAVGRVGST SENITQKVVW VEELDKRSFL LDLLNATGKD SLTLVFVETK KGADSLENFL FQERYSCTSI HGDRSQKDRE EALHQFRSGR KPILVATAVA
Ddx3y_mouse        .......... .......... .......... .......... .......... .......... .....A.... .......... .......... ..........
DDX3_rat           .......... .......... .......... ........R. .......... .......... .....A.... .......... .......... ..........

DDX3Y-TMUmRNA      ARGLDISNVK HVINFDLPSD IEEYVHRIGR TGRVGNLGLA TSFFNERNLN ITKDLLDLLV EAKQEVPSWL ESMAYEHHYK GSSRGRSK-- FSGGFGARDY
Ddx3y_mouse        .......... .......... .......... .......... .......... .......... .......... .......... ........SR ..........
DDX3_rat           .......... .......... .......... .......... .......... .......... .......... .......... .N......SR ..........

DDX3Y-TMUmRNA      RQSSGSANSG FNTNRANSSR SSGSSHNRGF GGGGYGSFYN NDGYGGNYNS QAVDWWGN*
Ddx3y_mouse        ........A. ..S....... .......... ......G... .......... .........
DDX3_rat           .....Y.... ..N....... T...T..... ......G..S .E........ .........
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USP9Y-TMU-mRNA  ATGACAATCA CAACTCGTGA CTCTCCAGTA GGAGAGAATG AGAGCCAGGG ACAGGTTCCA GATGGACAAC CTCAGCCTTC TTTCCAACAG AACCAG----
USP9Y-TMUgenome ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------
USP9Y_mouse     .......... .........G .......... .......... .......... T...AC.T.T .......... .......... .......... ......ATCT
USP9Y-rat       .....GG... .........G .........G .......... .......... .....C...T .....G...T .......... .......... ......ATCT

USP9Y-TMU-mRNA  ---------- ---------- ---AATTCCC CAGCAACTCC TCCAGATGAA CAAGGCCAAG GTGATGCTCC ACCGCAGCAC GAAGAAGAAG ATCCTTCATT
USP9Y-TMUgenome ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------
USP9Y_mouse     CATCATCTGA TTCTTCCAAT GAG.C...T. ..A....... ....T..... .......... .......... ...A.....T ........G. ..........
USP9Y-rat       CATCATCTGA TTCTTCCAAT GAG.....T. .......... ...T...... .......... .......... ...A.....T .......... .C...A....

USP9Y-TMU-mRNA  TCCTCATGCT GACCTGGCAA AGTTAGAAGA CATGATCAAC AGGCCCCAAT GGGTGGTTCC TGTTTTGCCA ATGGAGGAAT TAGAAGTACT TTTAGAAGCT
USP9Y-TMUgenome ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------
USP9Y_mouse     .......A.. .......... .......... .......... ...T...G.. .......... .......... .A..GA.... .......... ..........
USP9Y-rat       .......... .......... .......... .......... ..A....G.. .......... .......... .A..G..... .G........ ...G......

USP9Y-TMU-mRNA  TCTATGGATC TTACTAAAAA AGGCCTTGAT GCTAAAAGTG AAGCATGCCA GCGGTTTTTT CGAGATGTTT TAACTGTGTC TTTTACCAAA ATCCTCATGG
USP9Y-TMUgenome ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------
USP9Y_mouse     .....T.... .......... ...T..G... .T........ .......... ...T...... ..C....... .......... ....T..... .....T....
USP9Y-rat       .....T.... .......... ....A..... ..A....... .......... .........C .......... .......... .......... .....T.C..

USP9Y-TMU-mRNA  ATGAGGCAGT GACTAGATGG AAGTCTGAAA TTCATAGGTG TATTATTAAC AATATGCATC GCCTAGTGGA GCTTTGTGTG GCCAAATTGT CTCAGGACTG
USP9Y-TMUgenome ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------
USP9Y_mouse     .......T.. ..G.G..... ....T..... .......... .......... ....CC.... ....G..... .......... .......... ..........
USP9Y-rat       .......... ..G.G..... ....T..... .......... C.....A... ..C.CC.... .....A.... .......... .......... ..........

USP9Y-TMU-mRNA  GTTTCCATTT CTAGAACTTC TAGCAATGGC ATTAAATCCT CATTGCAAAT TTCATGTCTA TAATGGTGCA CGTCCATGTG AATCTGTGTC CTCAAGTGTT
USP9Y-TMUgenome ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------
USP9Y_mouse     .......... .......... .......T.. .......... .......... .......... .......... .......... ....A..... ..........
USP9Y-rat       .......... .......... .G.....A.. .......... .......... .......... ...C...... .......... ....A..... ..........

USP9Y-TMU-mRNA  CAGTTTCCTG AAGGTGATCT CTTTGCCTGT TCTCCAGACC TACATTCACC TAAGGGTTGG TTAGTGGATC TCATTAACAC ATTTGGCACA CTAAATGGTT
USP9Y-TMUgenome ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------
USP9Y_mouse     .......... ...A...A.. .......... .......... .......... A..A...... ........C. .......... .......... ..........
USP9Y-rat       .......... ...A...A.. .......... .......... ...G...... A..A...... C......... .......... ......T... ..........

USP9Y-TMU-mRNA  TCCAGATTTT GCATGATCGT TTCACTAGTG GATCAGCATT AAATGTTCAA ACAATTGCAG CTCTTATTAA GCCATTTGGA CAGTGCTATG AATTTCTTAC
USP9Y-TMUgenome ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------
USP9Y_mouse     .......... .......... .......... .......... .......... .......... .......... ...G...... .......... .G.....C..
USP9Y-rat       .......... .......... ....T..... .......... ......C... .......... .......... .......... .......G.. .G.A......

USP9Y-TMU-mRNA  TCAACATACA TTAAAAAAGT ACTTCGTTCC GGTTATAGAA GTGGTTCCAC AATTACTACA AAAATTAAGT AATGAAGAAC TGAAAAAAGA AGCAAAGAGT
USP9Y-TMUgenome ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------
USP9Y_mouse     .......... ....G..... .....A.... A......... ..A....... ..A..T.... ........C. .......... .......... .A......C.
USP9Y-rat       .......... .......... .....A.... A......... ..T....... .......... ........CC .......... .......... ........C.

USP9Y-TMU-mRNA  GAAGTGAAAA ATGATGCCAT TTCAGTAATT ATTAAATTTC TAAAAAATTT AGCTTCAAGA ATTCCAGGAC AAGAGGAAAC TGTTAAAGAT TTAGAAATTT
USP9Y-TMUgenome ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------
USP9Y_mouse     .......... .....A.... ....A.G... .......... .......... .......... .......... .......... .......A.. .......C..
USP9Y-rat       .......... .......T.. ....A.G... .......... .......... ...C...... .......... ....A..... .......A.. ..........

USP9Y-TMU-mRNA  TTAGGCTAAA AATGATACTC AGATGGCTAC AGATTTCTTC TTTTAGTGGA AAGATGAATG CACTGAATGA AATTAACAAA GTGCTGTCAA GTGTGTCATA
USP9Y-TMUgenome ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------
USP9Y_mouse     .....T.... .......... ....T..... .......... .....A.... .......... .......... .......... .....T.... ..........
USP9Y-rat       .....T.... .......... ....T..... .......... .....A...G .......... .G........ .........G .....A.... ..........

USP9Y-TMU-mRNA  T---ACACAT CAACATGGTA ATTTTGAAGA AGAGTGGCTA ACCGCTGAGC AAATGGCAGA GTGGATACAA CAGAACAATA TCTTATCCAT TGTTTTGCAA
USP9Y-TMUgenome ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------
USP9Y_mouse     .TAT..G... GG........ ...C...... .......... ....T..... G....A.... .........G .......... .......... ...C......
USP9Y-rat       .TAT...... .GG....... ...C...... .......... .......... G......... A......... ..A..T.... .......... A..C......

USP9Y-TMU-mRNA  GATAGCCTAC ATCAACCACA GTATGTGGAA AAGCTAGAGA AGATTCTTCG TTTTATAATT AAAGAAAAGG CTCTTACTTT ACAGGATCTT AATAATATCT
USP9Y-TMUgenome ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------
USP9Y_mouse     .......... .......... .......... ..AA...... .......... ....G..... .......... .......... .......... ..........
USP9Y-rat       ........T. .......... .......... .......... .......... ....G..... .......... .......... .......... ..........

USP9Y-TMU-mRNA  GGGCAGCACA GGCTGGAAAA CATGAAGCTA TTGTGAAGAA TGTACATGAT TTACTAGCAA AATTGGCTTG GAATTTTTCT TCTGAACAAC TTGATCATCT
USP9Y-TMUgenome ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------
USP9Y_mouse     .......... .......... .......... .......... ...T...... .......... .......... .......... C......... ..........
USP9Y-rat       .......... .......... .......... .......... .........C .......... .......... .G........ C......... ..........

USP9Y-TMU-mRNA  TTTTGATTGC TTTAAGGCAA GCTGGATGAA TGCAAGTAAA AAACAACGTG AAAAACTCCT TGAATTGATA CGTCGCCTTG CAGAAGATGA TAAAGATGGT
USP9Y-TMUgenome ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------
USP9Y_mouse     C......... .......... ......CA.. .......... .......... .......A.. .......... .....T.... .......... ..........
USP9Y-rat       .......... ........T. ......CA.. .......... .......... .......... .......... .....T.... .C........ ..........

USP9Y-TMU-mRNA  GTAATGGTAC ACAAAGTGTT GAATCTTCTT TGGAATCTAG CTCACAGTGA TGATGTACCT GTAGATATCA TCGATATGGC TCTCAATGCC CACTTAAAAA
USP9Y-TMUgenome ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------
USP9Y_mouse     .......C.. .......... .......... .......... .......... ...C..G... .......... .G...C.... .....G.... ...A......
USP9Y-rat       .......C.. .G........ .......... ........T. .......... ......G... .......... .G...C.... .....G.... ...A......

USP9Y-TMU-mRNA  TACTCAATTA TAGTTGTTYC CAGGATCGAG ATACACAAAA AATCCAGTGG ATAGACTGCT TTATTGAAGA ATTTCGAACA AATAATAAAT GGGTTATCCC
USP9Y-TMUgenome ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------
USP9Y_mouse     ....GG.... ........C. .......... .......... .......... .......... ....A..... .......... .......... ....C.....
USP9Y-rat       ....TG.... C.......C. .......... .......... G......... ........T. ....A..... .......... .......... ..........

USP9Y-TMU-mRNA  TGCATTAAAA CAAATTAAAG AAATTTGTAG CTTGTTTGGT GAAGCACCTC AAAATTTAAG TCAAACTCAT CAAAGTCCTC GTGTATTTTA TCGTCATGAT
USP9Y-TMUgenome ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------
USP9Y_mouse     .......... .......... .......... T......... .......... .......... .......... ........C. .......... .........C
USP9Y-rat       .......... .......... .G........ T......... .......... .......... ...G...... .G........ A......... ...C.....C
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USP9Y-TMU-mRNA  TTAATCAACC AACTTCAGCG TAATCATGCT TTAGTTACYT TGGTAGCAGA AAACCTTGCA ACTTACATGA ACAGCATAAG ATTGTATGCT AAAGACCATG
USP9Y-TMUgenome ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------
USP9Y_mouse     .......G.. .........A .......... ........T. .......... .......... G......... .......... .......... .G...T....
USP9Y-rat       .......G.. .........A .......... ........T. .......... .......... .......... ........C. .......... .G...T....

USP9Y-TMU-mRNA  AAGATTATGA CCCACAAACT ATAAGACTAG GAAGTAGATA CAGTCATGTT CAAGAAGTCC AAGAACGACT TAATTTCCTC AGATTCTTAT TGAAGGAAGG
USP9Y-TMUgenome ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------
USP9Y_mouse     .......... .......... G......... .......... .......... .......... .......G.. .......... .........C .......T..
USP9Y-rat       .......... .......... .......... .......... .......... .......... .......G.. .......... .......... .......T..

USP9Y-TMU-mRNA  TAAACTCCGG CTCTGTGCTT CCCAAGCAAA ACAAATATGG AACTGTTTAG CAGAGAACGC AGTTTACTTT ACTGATCGAG AAGCCTGTTT TATGTGGTAT
USP9Y-TMUgenome ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------
USP9Y_mouse     .C.....T.. .......T.. ....G..... .......... .......... .......T.. C......... .G........ .......... ..........
USP9Y-rat       .C.....T.. .......... ....G..... .......... C......... .......T.. .......... .G........ .......... ..........

USP9Y-TMU-mRNA  TCCAAATTAA TGGGCGATGA ACCTGAATTA GAGCCTGATC TTAATAAGGA GTTTTTTGAA AGTAATGTGC TTCAGCTTGA TCCTTCTCTA TTAACTGAAA
USP9Y-TMUgenome ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------
USP9Y_mouse     .......... ....A..... ......C... C.T......A .......... ......C... .......... .......... .......... ..........
USP9Y-rat       .......... ....A..... ......C... ..T......A .C........ .......... .......... .......... .......... ..........

USP9Y-TMU-mRNA  ATGGAATAAA ATGTTTTGAA AGATTTTTCA AAACTGTTAA TTGGCGAGAA GGGAAACTAA TGATAAAAAG AAAAGTCTAT ATGATGAATG ATTTGGACTT
USP9Y-TMUgenome ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------
USP9Y_mouse     ....G..G.. .......... .......... .......... ...T...... .......... .......... ....A..... ......G... ..........
USP9Y-rat       .......... .......... .......... .......... ...T...... .......... .......... .........C ......G... ..........

USP9Y-TMU-mRNA  AATAGGATTA GACTACCTTT GGAAGGTTGT GATTCAGAGT AATGACAATA TTTCAAGCAG AGCAATAGAT CTCCTTAAAG AGATACACAC AAGCCTTGGT
USP9Y-TMUgenome ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------
USP9Y_mouse     .......... .......... .......... .......... .....TG... .......... .......... ..T....... .....T.... ..........
USP9Y-rat       .......... .....T.... .......... .......... .....TG... ......A... ...T...... .......... .....T.T.. ..........

USP9Y-TMU-mRNA  CCCAGATTAC AAGCTAATCA GGTAGTAATT CATGAAGACT TTATTCAATC TTGCTTTGAT CGCTTGAATG CCTCATATGA TACACTGAAT GTTTTGGATA
USP9Y-TMUgenome ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------
USP9Y_mouse     ....A...G. .......... .......... .......... .......... .......... ........A. .......... ....T..TG. ..........
USP9Y-rat       ..T....... .......... ......T... .......... .......... .......... ..TA....AT .G........ ...GT..C.. ..........

USP9Y-TMU-mRNA  GTGATAAAGA CAATATTTTT AGCTGTGCAA GACAAGAAGC CATCCAGATG GTTCGCATAT TAACGGTTTT AAGGGAATAC ATAAATGAAT GTGACAGTGG
USP9Y-TMUgenome ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------
USP9Y_mouse     ....A..... .......... .......... .......... .....GA... .......... .......C.. .......... ....G..... A........A
USP9Y-rat       .......... .......... .......... .......... .....GA..A .......... ....T..... .......... ....G..... .........A

USP9Y-TMU-mRNA  TTATCATGTG GAAAGGATGA TTCTACCTAT GTCAAGAGCA TTTCGTGGGA ATCACTTGTC TTTTACAGTT CGATTTCCAA ACCAGGGCAA AGAAGTTGAT
USP9Y-TMUgenome ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------
USP9Y_mouse     ........A. .......... .......... .......... ........A. .A........ .........A .......... .......... ...G.....G
USP9Y-rat       ...C....A. .......... ....G..... .......... ..C....... .A........ .C........ .......... .....A.... ..........

USP9Y-TMU-mRNA  GATTTGGACA TAATGTCTCA TACAAATGCT ACCATTGGTT CTGTGCGACG CTGCATTCTC AATTGTATGA ATGCCAATGT AGCCCATACA GAAATTGAAC
USP9Y-TMUgenome ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------
USP9Y_mouse     .......... ..T....... ......C... .......... .......... .......... ...C...... ...T...... .......... A.........
USP9Y-rat       .......... ..C....... ...G...... .......... .......... .......... ...C....T. .......... .A........ A.........

USP9Y-TMU-mRNA  TGTTTATAGA TGGAGAGTTG ATCGATTCTG AAGATGACAG AAAGTTAGTT GAACAGTTAA ATTTGAAAGA TAAATCACTA ATTACTGCTA AATATATACA
USP9Y-TMUgenome ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------
USP9Y_mouse     .........G .........A G.A.CG.... .......... ....C..... .......... .......... .......... .......... ...T......
USP9Y-rat       .........G .........A ..A....... .......... ....C..... .......... .......... .......... .......... ...T.T....

USP9Y-TMU-mRNA  AATAAATTCC ATTATGCCTT CAAGTCCTGA TAGCTCATCT GATTCGTCAG CTGAATCTCC TGGAAACCAC AGTCATTA-- -CTACAGAGA TGTTCCCAAT
USP9Y-TMUgenome ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------
USP9Y_mouse     .......... .AC....... .......... .......... .......... ...G.C.... .........T ......A.TA A......... ....T.....
USP9Y-rat       .......... .AC....... .......... .......... .....A.... .C.G...... .......... T.....A.TA A...T..... ..........

USP9Y-TMU-mRNA  CCAGAAATGG AAAAATGTTT ACCTGGGGTG ATAATGTCAC TGCAGCCCAG ATACATATCT TTCCTTTGGC AAGTTGCAGA CTTAGGTAGC ATGTTGACTA
USP9Y-TMUgenome ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------
USP9Y_mouse     .......... ....G..... .......... .......... .......... .......... .......... .......... .......... .........G
USP9Y-rat       T......... ....T..... ...A..C... .......... .......... .......... .......... .......... ......A... ..........

USP9Y-TMU-mRNA  TGCCAAGTCT TAGAGATGGG GCAAGAATAC TTATGAAACT TATGCCACCA GATAGCACAA TCATAGAAAA ATTAAGAGCT GTATGTTCAG ACGATTTAAA
USP9Y-TMUgenome ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------
USP9Y_mouse     ......C... .........A .......... .......... .......... .......... CTC.....C. .......... C......... ..C..G....
USP9Y-rat       ......C... .........A .......... .......... A......... ....A..... CT........ C......... .......... ..C.......

USP9Y-TMU-mRNA  TCTTGGAGAA AGTCAACTTG GTCAGTCTCT TCACTCACTT TTCTTTGGTT CTTCTGCCTC TGAAGCACTA TATTTAACAG AGGTAGTTTA TATCTTGTTA
USP9Y-TMUgenome ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------
USP9Y_mouse     .......... ..A.G..... ....A..... .......... .......... .......... CC...TG... .......... .......... C.C.......
USP9Y-rat       .......... ..A.G..... ....A..... .......... .......... .......... CA...T.... .......... .......... C...C.....

USP9Y-TMU-mRNA  ATGCCTGCTG GTGGACCTCT GGCTGATATT TCCTATGACT TTCAGTATCA CTTCTTGAAA AGTGGTGGCT TACCCTTGGT ACTTAATATG CTGATTCAAA
USP9Y-TMUgenome ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------
USP9Y_mouse     .......... ...C...... .......... ....C..... .......... .......... .......... ....TC.T.. ...A.G.... .....A....
USP9Y-rat       ........C. ...C..A... A......... ....C..... .......... ......A... .A........ .G...C.T.. ...A.G.... .....A....

USP9Y-TMU-mRNA  ATAACTTCTT ACCAAATACA GATGTTGAAA CTCGTAGAGA TGCTTACGTC TGTGCCCTTA AAATAGCCAA ACTGTTGTTA ACTACCATTG GCTATGGCCA
USP9Y-TMUgenome ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------
USP9Y_mouse     .......... .......... .......... .......... .......T.. A......... .......... .......... ....T.G... ..........
USP9Y-rat       .......... .......... .......... .......... .......T.. A....T.... .......... .......C.. .......... ..........

USP9Y-TMU-mRNA  TGTCCAGGCC ATAGCAGAAG CTTGTCAGCC TGTTGCAGAT GATACAGATC CTAAAACACC GATTAACCAG GTTACTCATG ATCAAGCTGT GGTGCTACAA
USP9Y-TMUgenome ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------
USP9Y_mouse     ...T...... .......... .......... .......... .GC....... .......... .......... .......... .......... ..........
USP9Y-rat       .......... .......... .......... .......... .G........ .......... .........A .C........ .......... ...AT.....
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USP9Y-TMU-mRNA  AATGCCCTTC AGAGCATTCC TAATCCTTCT TCTGAGTGTA TACTTAGAAA CGTATCAATG CGCCTTGCTC AACAGATATC AGGGTTGGCC TCAAGATATA
USP9Y-TMUgenome ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------
USP9Y_mouse     .......... .A........ .......... ........C. .G........ T......G.T .AT....... ....A..... .......... ..........
USP9Y-rat       .......... .......... .......... ........C. .G.....G.. T.....G..T ..T....... .C..A..... .......... ........C.

USP9Y-TMU-mRNA  TACCTGATAT TTGTGTTATT ARAGCTATAC AAAAAATTAT CTGGGCAACA GGATGTGGAT CATTAGAACT AGTATTTAGT CCAAATGAAG ATATCACAGA
USP9Y-TMUgenome ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------
USP9Y_mouse     .......... .......... .G........ .......C.. .......G.. .......... .......G.. ...T...... .......... ..........
USP9Y-rat       .......... .......... .G........ .......C.. T...A..T.. .......... .......G.. .......... ...C.C.... ........A.

USP9Y-TMU-mRNA  AACTTACAAG ATGACCACCA GTACAAGGGG TAATCCAGAA GTGAAAGATG AGCAGGTTTG CTGTGAAGCC CTGGAAGTGA TGACATTATG TTTTGCTTTA
USP9Y-TMUgenome ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------
USP9Y_mouse     .......... .......... ........A. .....T.... .......... .A..A..... .......... .......... .......... ..........
USP9Y-rat       .......... .......... ....T...A. .......... .......... ....A..... .......... ..A.....A. .......... ..........

USP9Y-TMU-mRNA  ATTCCAACTG CGTTGGATTC ACTCAGTAAA GAAAAGGCCT GGCAGTCATT TGTCATTGAC TTACTATTGT ACTGTCCTAG CAAAACTATA CGGCAGTTGG
USP9Y-TMUgenome ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------
USP9Y_mouse     .......... .TA....... .....A.... .......... .......... .......... .......... .......... .......G.. ..........
USP9Y-rat       .......... .T........ .......... .......... .......... .........T .......... .......... .......... ..........

USP9Y-TMU-mRNA  CACAGGAGCA GTTCTTTTTA ATATGTACCA GATGTTGCAT GGGACATAGA CCTTTGCTGT TCTTCATTAC TCTACTCTTT ACCATTTTGG GGAGTGCAAC
USP9Y-TMUgenome .......... .......... .......... .......... .......... .......... .......... .......... .........A T.........
USP9Y_mouse     .......... .......... .......... .......... .......... .......... .......... .......... .......... ..G.....G.
USP9Y-rat       .......... .......... .......... .......... .......... .......... .......... .......... .......... .......G.

USP9Y-TMU-mRNA  AGCAGAGAAG GGTAAACATT CCGATGTCTA CTTCCCTCTT TTAAGGCGTC TTCTCACTTA TGCTTACAAC AGGAACATTC ATATACCCAA TGTTGACGTT
USP9Y-TMUgenome .A......-- ---------- ----...... ......A... .......... .......... ......A... .......... .......... ..........
USP9Y_mouse     TAAC...... .......... .A........ ....A.A... .......... .......... .......... ..C....... .AG.T..... ......T...
USP9Y-rat       .A.G...... .......... .A........ ...T..A... .......... .......... .......... ..C....... .C........ ......T...

USP9Y-TMU-mRNA  CTTCTCAATG AAGAAATTGA TTGGCTTAAA AGGGTTAGGG ATTACATTAA AAATACAGGA GAAACAAATA TAGAAGACCC AGTCTTAGAA GGCCATCTTG
USP9Y-TMUgenome .......... .......... .......... ...T...... .......... .......... .......... .......... .A........ ..........
USP9Y_mouse     .......... .C........ .......... .......... .......... .......... .........G .......... .A........ ........G.
USP9Y-rat       .......... .C......A. .......... .......... .......... .........C .....C.G.G .......... .A....G... ........G.

USP9Y-TMU-mRNA  GAGTAACAAA AGAGTTACTG TCTTTTCAGA CTCCTGAGGA AAAATATCAT ATTGGTTGTA AAACAGGAGG TGCTAATCTT GTTAAAGAAT TAATTGATGA
USP9Y-TMUgenome ...C...... .......... .......... ........A. .......... .......... .......... ..G....... .......... ..........
USP9Y_mouse     .......... .......... .........T .......AA. ...G...... .......... .......... .......... .......... ........T.
USP9Y-rat       .......... .......... ..C.....A. ....C..TA. ...G...... .......... ..G....... .........A .......... ..........

USP9Y-TMU-mRNA  TTTCATCTTT CCTGTATCCA AAGCTTACCT ACAATATATG AGAAGTGGGG AACTGCCAAT CAAGCAAGCC GTTCCAGTTT GTGGTTCACC AGCTACAGTT
USP9Y-TMUgenome .......... ...AC..... .......... .......... .......... .......... .......... .......... .......... ..........
USP9Y_mouse     .......... ....C..... .......... G......... .......... ..T....... .........T A.......C. .......... .......A..
USP9Y-rat       .......... ....C..... .......... G......TCA .......... ..A......A ...A...A.T A.......C. .......... ...C......

USP9Y-TMU-mRNA  AATGCTGGTT TTGAACTCCT TGTAGCTTTG GCTTTTGGTT GTGTAAGGAA TCTTGAACAG ATAGTAAACT GTTTGACTGA ACTGTTTTAC ATAGGCACAT
USP9Y-TMUgenome .......... .......... .......... .......... .......... ....A..... .......... .......... .......... .........C
USP9Y_mouse     .......... .......A.. .......... .......... .......... ....A..... .......... .......... .......... .........C
USP9Y-rat       .......... ....G..A.. .......... .......... .....C.... ....A..... .......... .......... .A........ .........C

USP9Y-TMU-mRNA  CAGTAACTAC TTGTGAAGCC CTTGCTGAAT GGGAATATTT ACCACCTGTT GAATCCCGCC CACCAAAAGG ATTTGTGGGA CTGAAAAATG CTGGTGCTAC
USP9Y-TMUgenome .......A.. .G........ .......... .......... .......... .G........ .......... .......... .......... ..........
USP9Y_mouse     .......... .......... G...G..... .......... ...C...... .GGC...... .......... .......... .......... ..........
USP9Y-rat       .T....A... A--------- .......... .......... .......... .G.C...... .......... .......... ..C....... ..........

USP9Y-TMU-mRNA  TTGTTATATG AACTCTGTGA TCCAACAGCT ATACATGATT CCTTCAATCA GGAACAGTAT TCTTGCAATT GAAAGCATAT GCAATGATAC AGATGGTGAT
USP9Y-TMUgenome .........C. .......... .......... .......... .......... .......... .......... .......... .G........ ..........
USP9Y_mouse     .......... .......... .......... .......... .......... .......... .......... .......... ..T....... .G.G...... .....A....
USP9Y-rat       .......... ..T....... .T........ G......... .......... .......... .......... ....A..... .G........ .....A....

USP9Y-TMU-mRNA  ATATTCAAAG ATGGAAAGCA CGACAGTGAG AATAATGCTG ATCCCAGAGA TGATGTATTC GGATATCCTC ATCAATTTGA AGATAAACCA ACATTAAGTA
USP9Y-TMUgenome .......... ...A...... G......... .......T.. .......... .......... A......... .......... .......... ..........
USP9Y_mouse     .......... G..A...... G......... .......T.. .......... .......... A......... .......... .......... ..........
USP9Y-rat       .C........ ...A..G... G........A ......AT.. .......... .........T .......... .......... .......... .......A..

USP9Y-TMU-mRNA  AAGTAGAAGA TCGAAAAGAG TACAACATTG GTGTTTTAAA GCACCTTCAG ATAACCTTTG GACATTTAGC TGCTTCACAA CTACAATACT ATGTGCCAAG
USP9Y-TMUgenome .......... .......... .......... .......... .......A.. .......... .......... ......T... ........T. ..........
USP9Y_mouse     .......... ...G...... .......... .......C.. A......... .......... .......... ......T... .......... .........A
USP9Y-rat       ...C...... .........C .......... .......... A......... .......... .......G.. ......T... .....G.... ..........

USP9Y-TMU-mRNA  AGGATTTTGG CAGCAATTCA GGCTTTGGGG TGAACCTGTT AATCTTCGTG AGCAACATGA TGCATTAGAG TTTTTTAATT CTTTGGTGGA TAGTTTAGAT
USP9Y-TMUgenome .......... .......... .......... .......... .......A.. .A........ .A.....A.. .......... .......... ..........
USP9Y_mouse     .......... ..A....... .......... .......... .....C.... .......... .......... .......... .......... ..........
USP9Y-rat       .......... ..A....... .......... ...G...... .....C.... .......... .......... .......... .......... ..........

USP9Y-TMU-mRNA  GAAGCTTTTA AAGCATTAGG ACATCCAACT GTGCTAAGTA AAGTTTTTGG AGGATCATTT GCTGATCAGA AGATTTGCCA AGGCTGCCCA CATCGGTATG
USP9Y-TMUgenome .......... .......... .......... .......... .......... .......... .......... .......... .......... ...A......
USP9Y_mouse     .......... ....T..... .T........ .....T.... ....CC.... ......C... .......... .......... .......... ...A......
USP9Y-rat       .......... ....T..... .......... C......... .....C.... ......C... .........A .......... .......... ...A......

USP9Y-TMU-mRNA  AATGTGAAGA ATCTTTTACA ACATTAAATA TAGACATTAG AAATCACCAA AATCTTCTTG ATTCTTTGGA ACAGTATATT AAAGGAGATT TATTAGAGGG
USP9Y-TMUgenome .......A.. .......... .......... .......... .......... .......... .......... .........C .......... .......T..
USP9Y_mouse     .......... .......... .........G .......... .......... .......... .......... .......... G......G.. .......... ..........
USP9Y-rat       .......... .......... ..T.....CG .......... .......... .......... .......... ....C..... .......... .......... ..........

USP9Y-TMU-mRNA  TGCAAATGCA TATCACTGTG AAAAATGTGA TAAAAAGGTT GATACAGTAA AACGCTTACT GATTAAAAAA CTACCTTCTG TTCTTGCTAT CCAACTGAAA
USP9Y-TMUgenome .......... .T........ ......A... .......... ....T..... .......... .......... .......... C......... ..........
USP9Y_mouse     .......... .......... .......... .......... .......... ........T. .......... .......... .....A.... ..........
USP9Y-rat       .......... .......... .......... .......... .......... .G...C.... .......... .......... .......... ..........
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Internal stop codon

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USP9Y-TMU-mRNA  CGATTTGACT ATGATTGGGA AAGAGAATGT GCAATTAAAT TCAATGATTA CTTTGAATTT CCAAGAGAAT TGGATATGGA GCCTTATACA RTAACAGGTG
USP9Y-TMUgenome .......... .......... .......... .......... .......... .......... .........A .......... .......... G.........
USP9Y_mouse     .......... .......... .......... .....C.... .......... .......... .......... .......... A.....C... G..G......
USP9Y-rat       ..T....... .......... .......... .....C.... .......... .......... .......... .A........ A......... G.GG......

USP9Y-TMU-mRNA  CCACAAAGCT GGAAGGTGAC AGTATAAGTC TACAGACTCA ATTGATTAAG CAGAATGAGC AGTCTGAAAG TGTGATACCA GGAAGCACAA AATATAGACT
USP9Y-TMUgenome .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
USP9Y_mouse     .......... .......... ...G...A.. C......... .........A .......... .......... ...C...... .......... .....C....
USP9Y-rat       .......... .......... G..G..CA.. C......... .......... ..A....... .........A ....G..... .......... ....C.....

USP9Y-TMU-mRNA  TGTTGGTGTA CTTGTTCACA GTGGTCAAGC AAATGGTGGA CATTATTATT CTTATATCAT TCAAAGAAAT GGTAAAGACA GTAAGAGAGG TCGCTGGTTC
USP9Y-TMUgenome .......... .......... .......... .......... .........C .......... .......... .......... .......... ..........
USP9Y_mouse     .........G .....A.... .......... .......... .......... ....C..... .......... .......... ........A. ..A.......
USP9Y-rat       .........G .....A.... ....C..... .......... .......... .......... ......G... .......... A...T..... ..........

USP9Y-TMU-mRNA  AAATTTGATG ATGGAGATGT AACGGAATGT AAAATGGATG ATGATGAAGA AATGAAAAAT CAATGCTTCG GTGGAGAGTA TATGGGAAAA GTTTTTGATC
USP9Y-TMUgenome .......... .......... ........A. .....A.... G......... .......... .......... .......... .........T ..........
USP9Y_mouse     .......... .......... ...A...... .......... .......... .......... ..G.....T. .......... .......G.. ..........
USP9Y-rat       .......... .......C.. ...A...... .......... .......... .......... ..G.....T. ....G..... .......G.. ..........

USP9Y-TMU-mRNA  ATATGATGAA GCGGATGTCA TACAGGCGTC AGAAAAGATG GTGGAATGCT TATATTCTTT TTTATGAACG GATTGATATA ACTGATGAAA ATGATGAGAT
USP9Y-TMUgenome .......... .......... ......T... .......... .......... .......... .......... .......... .......... ..........
USP9Y_mouse     .......... ...T...... .......... .......... .......... ..C....... .......... ...G...... .........G ..........
USP9Y-rat       .......... ...T...... .......... .......... .......... .......... .......... ...G...... .........G ..A.......

USP9Y-TMU-mRNA  GATAACATAT ATATCAGAGC TAACAATCAC CAGACCTCAT CAGATTATGC CACCTGCCAT TGAGAGAATT GTGTGGAAAC AGAATGTGCA GTTCCTGCAT
USP9Y-TMUgenome .......... .......... .......... ...G...... .......... .........A..G. .......... .......... A.........
USP9Y_mouse     A......... .......... .....T.T.. .......... .........T ....A..... ........G. ..A....... .......... A.........
USP9Y-rat       .......... T.G....... .......T.. .......... .........T ....A..T.. ........G. ..A....... .......... A.........

USP9Y-TMU-mRNA  AACCAAATGC AGTTTAGCTT AGAATATTTT CAGTTTATTA AAAAACTGCT TACATGTAAT GCTGTGTACT TAAACCCTGC TCCAGGACAA GATCACTTGC
USP9Y-TMUgenome .......... .......... .......... .......... .......... A......... .......... .......... .......... ......A...
USP9Y_mouse     .......... .......... .......... .......... .......... .......... .......... ...G...... .......... ..........
USP9Y-rat       ..T....... .A........ .......... .......... .......... .......... .....A.... .......... .......... ........A.

USP9Y-TMU-mRNA  TTCCAGAAGC AGAAGATATT ACAATGATCA GCATTCAGCT TGCTGCTAGA TTCCTCTTTA CTACTGGATT TCATACTAAG GAAATAGTCC GTGGACCTGC
USP9Y-TMUgenome .......... .......... .......... .......... .......... .......... .......... .......... A......... A....T...T
USP9Y_mouse     .A........ .........C .......... .......... ....T..... .......... .......... .......... A.....A... ..........
USP9Y-rat       CA........ .........A .......... .......... .......... .......... .A........ .......... A....T.... .C........

USP9Y-TMU-mRNA  CAGTGACTGG TATGATGTGC TGTGTATTCT CCTCCGTCAC AGCAAGAATG TACGTTTTTG GTTTGCTCAT AATGTCCTTT TTAATGTATC AAATCGCTTC
USP9Y-TMUgenome .......... .......... .....G.... .......... .......... .......... .......... .......... .......... ..........
USP9Y_mouse     ..A....... .....C.CA. .T........ .......... .......... .......... .....T.... ........C. .......... ......T...
USP9Y-rat       ..A....... .....C.C.. ....C..... .......... .......... .G........ .......... .......... .......... ...C..T...

USP9Y-TMU-mRNA  TCTGAGTACC TTCTGGAATG TCCAATTGCA GAAGTCAGAG ATACATTTGC AAAGCTTATA GTATTTATTG CACATTTTTC TTTGCAAGAT GGATCTTCCC
USP9Y-TMUgenome .......... .....A.... .......... .......... .......... .......... .......... .......... .......... ..........
USP9Y_mouse     .......... .......... .....G.... ...A...... G......... .......... .......... .......... .......... ........T.
USP9Y-rat       .......... .......... .....G.... .......... G......... .......... ..T....... .......... .......... ........--

USP9Y-TMU-mRNA  CTTCACCTTT TACATCACCT TTTGCAACTC CAGGACCTTA TAGTCAGATC TATAACAATT TAAGCTTGAG CGATCATTTA CTAAAAACAG TATTAAGTCT
USP9Y-TMUgenome .......... .......... .......... .......... .......... C..G...... .......... .......... .......... ..........
USP9Y_mouse     .......... .......... .......A.. .......... .......... ...G...... .......... T......... ......G... ..........
USP9Y-rat       -....T.... .G........ .......... .......... .......... ...G...... .G..T..... .........G ......G... ..........

USP9Y-TMU-mRNA  CTTGAGAAGG GAAGTTTCTG AACATGGGCG ACATTTACAA CAATATTTCA ATTTATTTAT AATGTATGCC AGTTTAGGTT TGGCAGAAAA GACCCAGCTT
USP9Y-TMUgenome .........A .......... ........T. .......... .......... .......... .......... .......... .......... ..........
USP9Y_mouse     .......... .......... .......... .......... .......... .......... .......... ..C....... .......... ..........
USP9Y-rat       .......... .......... .......... .......... ..G....... .......... .......... .......... .......... ..........

USP9Y-TMU-mRNA  CTAAAACTGA ATGTACCTGC AACCTTTATG CTTGTGTCTT TGAATGAAGG ACCAGGCCCC CCAATTCAAT ATCAATATGC TGAATTAAGC AAGTTACATT
USP9Y-TMUgenome .......... .......... .......... .......... .......... .......... .....T.... .......... .........A. ..........
USP9Y_mouse     .......... ....T..... .......... .......... .......... ..G....... .........T ...G..A... .......... ...G..G... ..........
USP9Y-rat       .......... ........A. .......... .......... .......... ..G....... C.....T..T ......A... .......... ...G...G.. ...C......

USP9Y-TMU-mRNA  CAATAGTGTC TCAGCTGATC CGATGTTGCA GTGTCTCATC AAGAATGCAG TCTTCTATTA ATGGTAACCC CCCTCTTCCA AATCCATTTG GTGACCCAAA
USP9Y-TMUgenome .......... .......... .A........ .......... ..C....... ........A. .........A .......... .......... ..........
USP9Y_mouse     ..G....... .......... A......... .......... .......... ........C. .......... .......... .......... ..........
USP9Y-rat       ..G....A.. .......... .......... .......... .......... ..C.....A. .......T.. .......... .......... ..........

USP9Y-TMU-mRNA  TTTGTCACAA CCTATAATGC CAATTCAGCA GCATGTGGCT GACATTTTAT TTATGAGAAC CACTTATATG AAGAAAGTTA TTGAAGACTG CAGTAACTCA
USP9Y-TMUgenome .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
USP9Y_mouse     ...A...... ..G....... .......... AA........ .......... .......... .......... ......C... .......... .......... ..........
USP9Y-rat       ...A.....G .......... .......... .A........ .......... .......... .......... .......G.. ..A...A... .......... ..........

USP9Y-TMU-mRNA  GAAGAACCAG TCAAATTACT TCGATTCTGC TGCTGGGAGA ATCCCCAGTT TTCATGTTCT GTTCTCAGTG AACTGCTGTG GCAGGTTGCA TATTCATATG
USP9Y-TMUgenome ..G....... .......... .......... .......... .......... .......... .......... .......... .......... ..........
USP9Y_mouse     ..G..TA... .......... ..TC...... .......... .......... .......... .....T..C. .......A.. .......... C.....C...
USP9Y-rat       ..G...A... .......... ...C...... .......... .......... .......... ........C. .......A.. .......... ..........

USP9Y-TMU-mRNA  AATATGAACT TCAACCATAT TTGGATCTAC TTTTGCAAAT TATTCTGCTT GAAGACTCCT GGCAAGCTCA CAGAATTCAT AATGCACTTA AA-GGAATTC
USP9Y-TMUgenome .......... .......... .......... .......... .......... .......A.. .......... .......... ..C....... ..A......
USP9Y_mouse     C......... .......... .......... .......... .......T.. ..G....... .......... .......... .......... ..-.......
USP9Y-rat       C......... ...G.....C .......... .......... .......... .......... .......... .......... .......... ..-.....A.

USP9Y-TMU-mRNA  GAAATGATCG AGACGGACTG TTTGATACAA TCCAGCGCTC CAAAAATCAC CATCAAAAAA AAGCCTATCA ATGTATAAAA TGGATGGTAA CTCTCTTTAA
USP9Y-TMUgenome ........T. .......... .......... .......... .......... .......... .......... .......... ...T...... ..........
USP9Y_mouse     C........A ...T...... .......... ......A... T......... .......... G......... .......... .......... ..........
USP9Y-rat       C..G...... ...T...... .......... ......A..T T........T .....G.... G......... .......... .......... ..........
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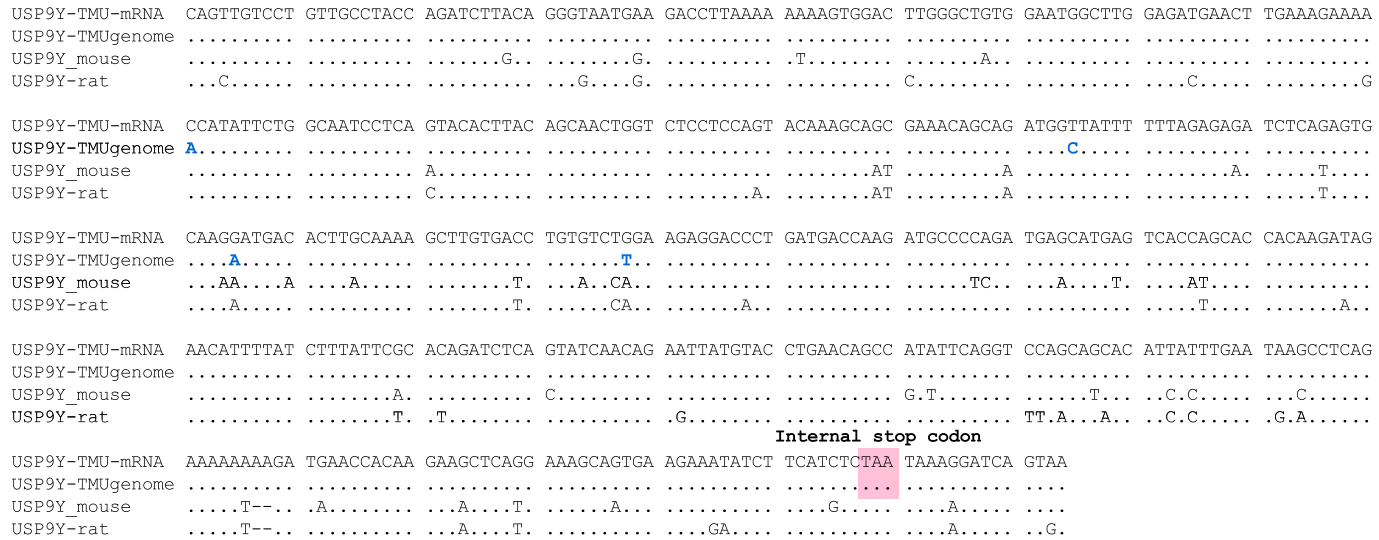

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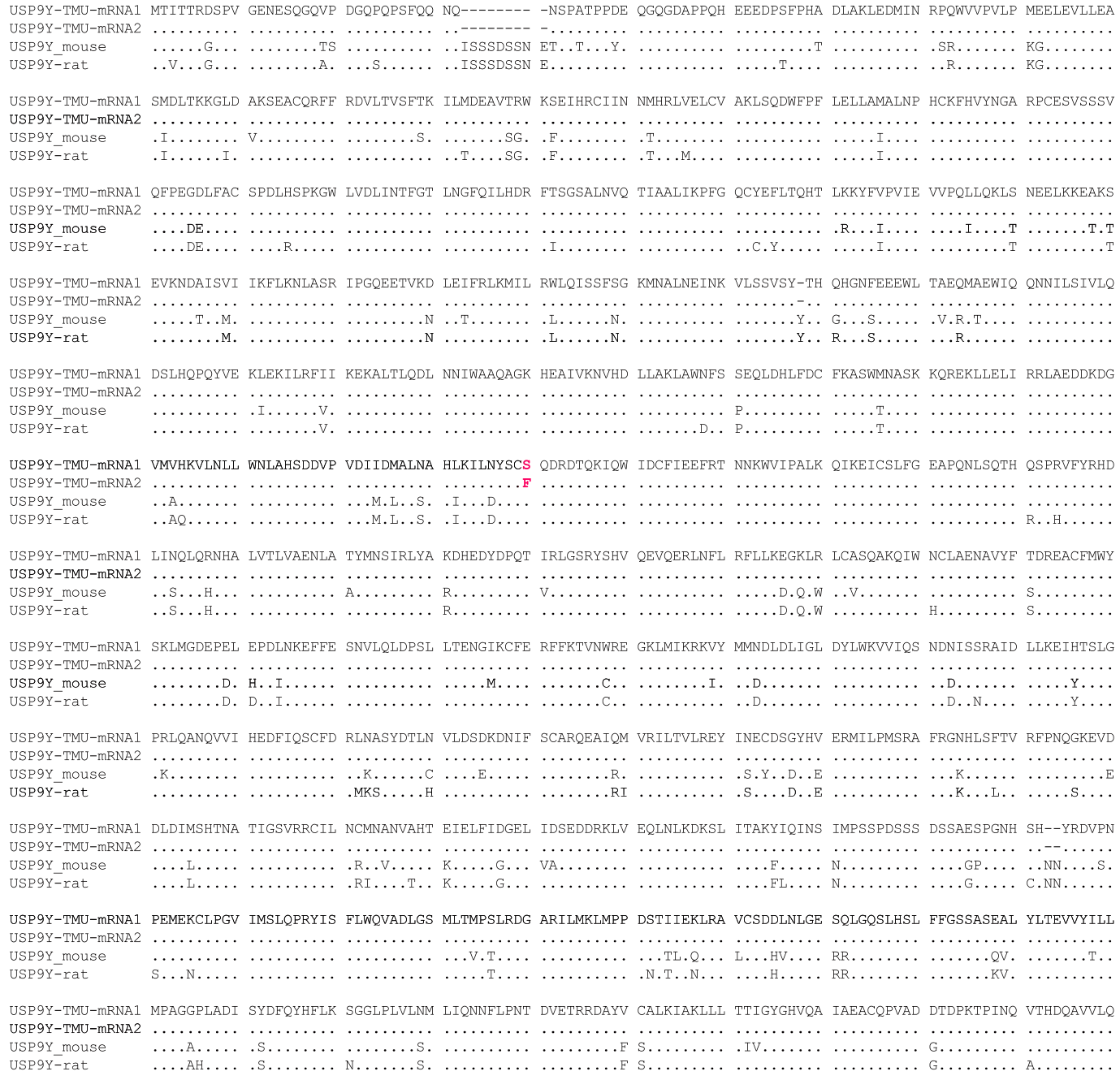

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USP9Y-TMU-mRNA1 NALQSIPNPS SECILRNVSM RLAQQISGLA SRYIPDICVI RAIQKIIWAT GCGSLELVFS PNEDITETYK MTTSTRGNPE VKDEQVCCEA LEVMTLCFAL
USP9Y-TMU-mRNA2 .......... .......... .......... .......... K......... .......... .......... .......... .......... ..........
USP9Y_mouse     .......... ...M.....V H......... .......... .........A .......... .......... ......S.L. .......... ..........
USP9Y-rat       .......... ...M.....I ...H...... .......... ........TS .......... .H....K... ......S... .......... ..........

USP9Y-TMU-mRNA1 IPTALDSLSK EKAWQSFVID LLLYCPSKTI RQLAQEQFFL ICTRCCMGHR PLLFFITLLF TILGSATAEK GKHSDVYFPL LRRLLTYAYN RNIHIPNVDV
USP9Y-TMU-mRNA2 .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
USP9Y_mouse     ....M...N. .......... .........V .......... .......... .......... ....G.AN.. ........T. .......... S..QV.....
USP9Y-rat       .......... .......... .......... .......... .......... .......... ......AT.. .......... .......... S.........

USP9Y-TMU-mRNA1 LLNEEIDWLK RVRDYIKNTG ETNIEDPVLE GHLGVTKELL SFQTPEEKYH IGCKTGGANL VKELIDDFIF PVSKAYLQYM RSGELPIKQA VPVCGSPATV
USP9Y-TMU-mRNA2 .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
USP9Y_mouse     ...D...... .......... ...V...I.. .......... ...S..K... .......... ......Y... .A........ .......... I........I
USP9Y-rat       ...D..N... .......... ..SV...I.. .......... .....DK... ....A..... .......... .A.......S ....M.N..T I.........

USP9Y-TMU-mRNA1 NAGFELLVAL AFGCVRNLEQ IVNCLTELFY IGTSVTTCEA LAEWEYLPPV ESRPPKGFVG LKNAGATCYM NSVIQQLYMI PSIRNSILAI ESICNDTDGD
USP9Y-TMU-mRNA2 .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
USP9Y_mouse     .......... ........K. .......... ...P...... VG........ GP........ .......... .......... .......... D..WS...D.
USP9Y-rat       .......... ........K. .......M.. ...P.N.--- .......... GP........ .......... .......... .......... .N.W....D.

USP9Y-TMU-mRNA1 IFKDGKHDSE NNADPRDDVF GYPHQFEDKP TLSKVEDRKE YNIGVLKHLQ ITFGHLAASQ LQYYVPRGFW QQFRLWGEPV NLREQHDALE FFNSLVDSLD
USP9Y-TMU-mRNA2 .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
USP9Y_mouse     ...GE.Q... ..V....... R......... .......... ...A...... .......... ......K... .......... .......... ..........
USP9Y-rat       T...ERQ... ..I....... .......... ..N.A....D .......... .......... .......... .......... .......... ..........

USP9Y-TMU-mRNA1 EAFKALGHPT VLSKVFGGSF ADQKICQGCP HRYECEESFT TLNIDIRNHQ NLLDSLEQYI KGDLLEGANA YHCEKCDKKV DTVKRLLIKK LPSVLAIQLK
USP9Y-TMU-mRNA2 .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
USP9Y_mouse     .......Y.. .....L.... .......... .......... ...V...... .........V .......... .......... .......... .....T....
USP9Y-rat       .......... L....L.... .......... .......... ...V...... .......... .......... .......... .......... ..........

USP9Y-TMU-mRNA1 RFDYDWEREC AIKFNDYFEF PRELDMEPYT VTGATKLEGD SISLQTQLIK QNEQSESVIP GSTKYRLVGV LVHSGQANGG HYYSYIIQRN GKDSKRGRWF
USP9Y-TMU-mRNA2 .......... .......... .......... .......... I......... .......... .......... .......... .......... ..........
USP9Y_mouse     .......... .......... .......... .A........ .VNP...... .......... .......... .......... .......... ......SH..
USP9Y-rat       .......... .......... .......... .A........ GVHP...... ......N.V. .......... .......... .......... ...NN.....

USP9Y-TMU-mRNA1 KFDDGDVTEC KMDDDEEMKN QCFGGEYMGK VFDHMMKRMS YRRQKRWWNA YILFYERIDI TDENDEMITY ISELTITRPH QIMPPAIERI VWKQNVQFLH
USP9Y-TMU-mRNA2 .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
USP9Y_mouse     .......... .......... .........E .......... .......... .......M.. ...D..I... .....F.... ...S.....S ..........
USP9Y-rat       .......... .......... .........E .......... .......... .......M.. ...DN..... L......... ...S.....S ..........

USP9Y-TMU-mRNA1 NQMQFSLEYF QFIKKLLTCN AVYLNPAPGQ DHLLPEAEDI TMISIQLAAR FLFTTGFHTK EIVRGPASDW YDVLCILLRH SKNVRFWFAH NVLFNVSNRF
USP9Y-TMU-mRNA2 .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
USP9Y_mouse     .......... .......... ....S..... .......... ........S. .......... K.I....N.. ..A....... ........V. ..........
USP9Y-rat       .......... .......... .......... ...P...... .......... .......... K......N.. ..A....... .......... ..........

USP9Y-TMU-mRNA1 SEYLLECPIA EVRDTFAKLI VFIAHFSLQD GSSPSPFTSP FATPGPYSQI YNNLSLSDHL LKTVLSLLRR EVSEHGRHLQ QYFNLFIMYA SLGLAEKTQL
USP9Y-TMU-mRNA2 .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
USP9Y_mouse     ........S. .I.G...... .......... .......... ..N....... .D........ ..A....... .......... .......... ..........
USP9Y-rat       ........S. ...G...... .......... ...-.S.A.. .......... .D........ ..A....... .......... .......... ..........

USP9Y-TMU-mRNA1 LKLNVPATFM LVSLNEGPGP PIQYQYAELS KLHSIVSQLI RCCSVSSRMQ SSINGNPPLP NPFGDPNLSQ PIMPIQQHVA DILFMRTTYM KKVIEDCSNS
USP9Y-TMU-mRNA2 .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
USP9Y_mouse     .......... ....D..... .VK....... ....V..... .......... .......... .......... .......... .......N.. .......... ..........
USP9Y-rat       ......T... ....D..... ..K......G ....V..... .......... .......... .......... .......N.. .........V ..I.......

USP9Y-TMU-mRNA1 EEPVKLLRFC CWENPQFSCS VLSELLWQVA YSYEYELQPY LDLLLQIILL EDSWQAHRIH NALKGIRNDR DGLFDTIQRS KNHHQKKAYQ CIKWMVTLFN
USP9Y-TMU-mRNA2 .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
USP9Y_mouse     .DT....L.. .......... .......... H.HA...... .........F .......... ......P..Q ........H. ......R... ..........
USP9Y-rat       ..T....... .......... .......... ...A...... .......... .......... ......PS.. ........HF ......R... ..........

USP9Y-TMU-mRNA1 SCPVAYQILQ GNEDLKKKWT WAVEWLGDEL ERKPYSGNPQ YTYSNWSPPV QSSETADGYF LERSQSARMT LAKACDLCLE EDPDDQDAPD EHESPAPQDR
USP9Y-TMU-mRNA2 .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
USP9Y_mouse     .......... ..G...N... ..M....... .......... .......... ..N...N... ..K.H..K.K .T.....YP. ........L. ..V.H.....
USP9Y-rat       .......... ..G....... .......... .........H .........I ..N...N... ....H..... ........P. ..T....... ........E.

USP9Y-TMU-mRNA1 TFYLYSHRSQ YQQNYVPEQP YSGPAAHYLN KPQKKKMNHK KLRKAVKKYL HL*
USP9Y-TMU-mRNA2 .......... .......... .......... .......... .......... ...
USP9Y_mouse     .........H .......... F....S.H.. N...NDKPQE THESNEEISS C.IKDQ*
USP9Y-rat       .......... ...S...... ...LTE.H.. E...NDEPQE THESSEEESS S.IKDQ*
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RBMY-TMU    ATGGCAGAAA CTCACCTGCC TGGGAAAATT TTTGTAGGAG GCCTCAACAT AGAGACCAGA CAAAAGAGAC TTCAAGTAAT ATTTGAGAGA TTTGGACCAG
Rbmy-mouse .......... ..A.T.A... .......... ...A...... .......... .A........ .......CT. ......A... .....G.... ..........
RBMY-rat   .......... G.G.G.A... .......... ...A...... ........T. .......... .......CT. .......... .....G.... .........A

RBMY-TMU    TATTAAATGT TATTTTGATG AGAGATCGTG AAACTAAGAA GTCAAGAGGC TTTGCTTTSA TTACTTTTAA ACATGCTGCA GATGCTAAGA ATGCTGTCAA
Rbmy-mouse ..GC.CG... .......... .......... .......... ...T...... ........CC .......CCG ..GCCT.... .......... ..........
RBMY-rat   ..AC.C.... .......... .......... .......... ...T...... ........CG ........C. .TG.C...G. .......... ..........

RBMY-TMU    AAAAATGAAT GGAGTGTCTT TGGATGGAAA AAGAATTAAA GTAGAACAAG CCARGAGACC ATCATCACTT GAACGTGGTA GAAAGCAGAG ACCACAATCC
Rbmy-mouse .G........ ......AT.. .......... .......... ...A...... ...G...... .......... ...A...... .C...A.... .....C....
RBMY-rat   .G.......C .......... .......... .......... .......... ...GA..... .......... ...A.GA... .C...AG... ....TT....

RBMY-TMU    TTTTTAAGA- ---------- ---------- ---------- ---------- ---------- GCAAGAGGTT GTCCTTCACG TGGAGGGAAT TTGGATGGTG
Rbmy-mouse ....C....A CCAGAGGTGC TTCAAGAATT CTGAAATGTG GAAGAGGAGG AAGAAGCAGA ......A..G ........T. ..A......C ....G.....
RBMY-rat   ....C....A CCAGAGGTGC TTCAAGAATT CTGAAATGTG GAAGAGGAGG AAGTAGCAGA .G....A..C .......... ..A.A.A... .......A..

RBMY-TMU    ATAGATATAC T--------- GACATGAGTT CTTCTGGGAG ACACTTTACA GTTAAAAGAA GTTCATCTTC AAGAAGGAAT GGTGGTCCTT CTAAAAGACC
Rbmy-mouse .......... .CCTAATTTC A.TG...... .......... .......G.. .......... A.C....... ..A....G.. .A.CCC.... ........T.
RBMY-rat   G......... .CCTAATTTC A....A.... .......... .......G.. .......... ..A....... ..A....G.A ...CC..... ........T.

RBMY-TMU    TGCAACTTCT GCTCTGACAA G---CAATTC TGGACTGAGA GGACAAGAAC CACGTGGAAG ACAGATTTCT AGAAATATGC CAAGAAGAGA ACCAGTGGCT
Rbmy-mouse .......... ....A..... .AAG....A. ......A... ....G..... ...A.C.... .G........ .......... .....G.... G..G.C.T..
RBMY-rat   .......... .......... .AGC....C. .......... ....G..... ...A...... .G..G.G... .......... .......... G.....AT..

RBMY-TMU    TCCAGAACAG ATGAGTATCC ATTACCAAGG GATGATGACC AGTCTTCTAA TGATAGTTAT TCAAGGAGAT ACGCATCAAC ATCCAGAGGA TACAGTGACT
Rbmy-mouse .......G.. .......... .......... ...T...G.. .......... ......G--- ------.A.. .T.A...... .......... ...T......
RBMY-rat   .......G.. ....A..... .......... .......GTT .T........ .......... .......... .T..G..... ...T...... ...C......

RBMY-TMU    ATGGCAATTA CAGCTCTCAG GAGGAACATA CATCTGAAGT CTTCAGTGAT CATGCTGGCT ATCGTGGGGG ACATAATAGA GATTTCTCTG ACTATTTAAG
Rbmy-mouse .......... .CA.....G. ..A...AG.G .....A.... .......... .......... ...T...A.. ..G.G..... .......... .A........
RBMY-rat   .......... ........G. ..T......G .....A.... .......... .......... .C.A...... ..G.G..... .......... .A..C.....

RBMY-TMU    TGGAAGTTCA TACAGAGATA CAGATGGAAG TTACGGAAAC TTCCGTGAGG CACCATCTGC AAGATGGACT TACACTGGAA ACAACCACTA TGATGATTAT
Rbmy-mouse .....A.... .......... ..T.CA.... ...T....GA ....A..... .......... ....G.T--- ------.... ......G... ......C...
RBMY-rat   .......... .......... ..T...A... ...T....G. ....A..... A......... ....G.C... .......... ......G... ......C...

RBMY-TMU    AGTAGCTCAC GAGATGGATA TGGTGGAAGA CGAGAAATAT ATTCAAGCGG CCAAAGTGAC ATATATTCAA GTGGCTATGA ACATAGTGGT AGA---GAAG
Rbmy-mouse ....A..... A......... .......... G.....CC.. .CAT....AA ..G....A.. .....C.... ...A...... ..G....... ...CAA....
RBMY-rat   .......... ...G...... .......G.. .......CT. .C...G..A. ..G....... .....----- -......... ..G....... ...C...---

RBMY-TMU    TGCTTCCACC T---GTTGAT AGGGAATACT TTGAGCATGG AGGCAGACAA GGAAGAGGGC ATTCACCTAT GGATAGGCTG TATATTGCAT CTCGTGAAAC
Rbmy-mouse .......... ACCTA..... .......... ....C.G..A G........G .A........ .........A ....G..... ....G..... ........T.
RBMY-rat   .......... ---.A..... .......... C.....G... G......G.. .A........ .C........ .......... ....GG.... ...A....T.

RBMY-TMU    ATATAATAGT AGT-TCAAGA TTTGGGGGAT CCCATGGAGA TCATGCAGAA AGCAGATATG AAAGTGGAGG CAGGTACTAA
Rbmy-mouse ...C.G.... .A.AC..... .......... .........G ..G..G.... .........- .......... ..........
RBMY-rat   ...C.C..T. ...-.....G .......... A........G .....A.... .C........ .......... ....CGT...
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RBMY-TMU-mRNA1 MAETHLPGKI FVGGLNIETR QKRLQVIFER FGPVLNVILM RDRETKKSRG FAFITFKHAA DAKNAVKKMN GVSLDGKRIK VEQARRPSSL ERGRKQRPQS
RBMY-TMU-mRNA2 .......... .......... .......... .......... .......... ..L....... .......... .......... ....K..... ..........
Rbmy-mouse     ....NQ.... .I.....K.. ..T..E..G. ....AR.... .......... ...L..RRL. .......E.. ..I....... .K........ .S.S.K..P.
RBMY-rat       ...SEQ.... .I....L... ..T.....G. ...ITH.... .......... ..FV..QCPG .......E.. .......... ....R..... ..SS.R..L.

RBMY-TMU-mRNA1 FLR------- ---------- ARGCPSRGGN LDGDRYT--- DMSSSGRHFT VKRSSSSRRN GGPSKRPATS ALT--NSGLR GQEPRGRQIS RNMPRREPVA
RBMY-TMU-mRNA2 ...------- ---------- .......... .......--- .......... .......... .......... ...--..... .......... ..........
Rbmy-mouse     .S.TRGASRI LKCGRGGRSR ..SG..CE.. .G.....PNF NV.......A ...NP..K.D DP....S... .Q.RS.T... .R..HR.E.. .....G..AS
RBMY-rat       .S.TRGASRI LKCGRGGSSR G.SR...ER. ..DG...PNF NI.......A ....T..K.E .P....S... ...RA.P... .R..H..EV. .........S

RBMY-TMU-mRNA1 SRTDEYPLPR DDDQSSNDSY SRRYASTSRG YSDYGNYSSQ EEHTSEVFSD HAGYRGGHNR DFSDYLSGSS YRDTDGSYGN FREAPSARWT YTGNNHYDDY
RBMY-TMU-mRNA2 .......... .......... .......... .......... .......... .......... .......... .......... .......... ..........
Rbmy-mouse     ..R....... .YG.....R- --K.E..... .C.....H.R ..SA.K.... ....L..RD. ...E....N. ....YR...R .H......G- --...R....
RBMY-rat       ..R....... ..GY...... .......... .R.......R D..A.K.... ....H..RD. ...E...... ....YE...S .H.E....G. .....R....

RBMY-TMU-mRNA1 SSSRDGYGGR REIYSSGQSD IYSSGYEHSG R-EVLPP-VD REYFEHGGRQ GRGHSPMDRL YIASRETYNS SSRFGGSHGD HAESRYESGG RY*
RBMY-TMU-mRNA2 .......... .......... .......... .-.....-.. .......... .......... .......... .......... .......... ...
Rbmy-mouse     .N.Q...... G.P.I.NR.N ....D..R.. .Q.....PI. ....DRE... E.....K.G. .S....S.S. NTKIW.IPWR SWRKQI*
RBMY-rat       ....G....G ..T..GSR.. .---...R.. .Q--..--I. ...S.R...E E......... .R..H.S.TI ......Y..G .E.T...... .R.
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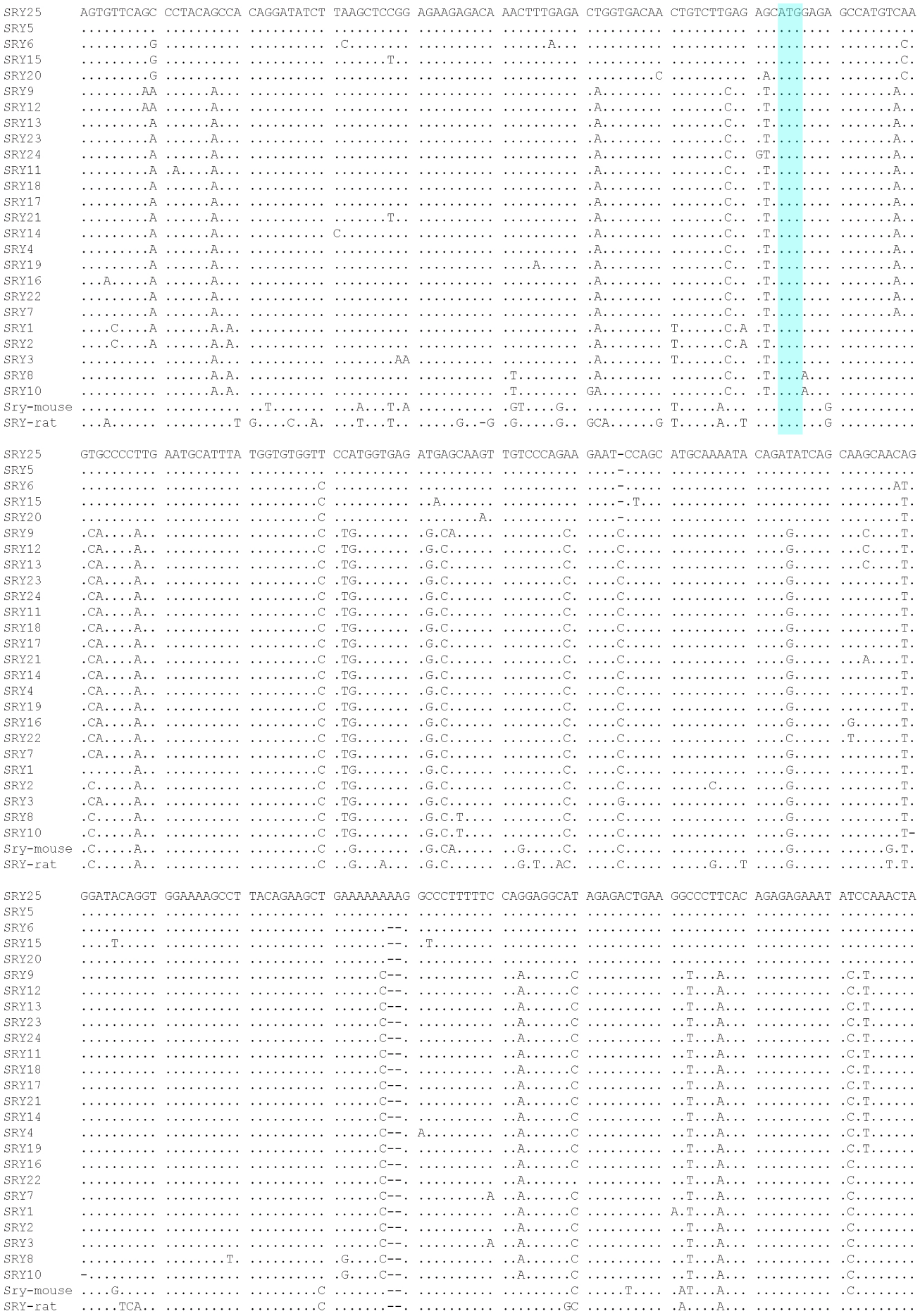
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SRY5 ....
SRY6 .....G .... .C..... ....A.. ....C.
SRY15 .....G ....T.. ....C.
SRY20 .....G ....C ..... .A..... .....C.
SRY9 .....AA .....A... .... .A..... .....C.. .T..... .....A..
SRY12 .....AA .....A... .... .A..... .....C.. .T..... .....A..
SRY13 .....A .....A... .... .A..... .....C.. .T..... .....A..
SRY23 .....A .....A... .... .A..... .....C.. .T..... .....A..
SRY24 .....A .....A... .... .A..... .....C.. GT..... .....A..
SRY11 .....A .A....A... .... .A..... .....C.. .T..... .....A..
SRY18 .....A .....A... .... .A..... .....C.. .T..... .....A..
SRY17 .....A .....A... .... .A..... .....C.. .T..... .....A..
SRY21 .....A .....A... ....T.. .... .A..... .....C.. .T..... .....A..
SRY14 .....A .....A... ..... C..... .... .A..... .....C.. .T..... .....A..
SRY4 .....A .....A... .... .A..... .....C.. .T..... .....A..
SRY19 .....A .....A... ....A..... .A..... .....C.. .T..... .....A..
SRY16 ...A.....A .....A... .... .A..... .....C.. .T..... .....A..
SRY22 .....A .....A... .... .A..... .....C.. .T..... .....A..
SRY7 .....A .....A... .... .A..... .....C.. .T..... .....A..
SRY1 ....C....A .....A.A. .... .A..... T.....C.A .T..... .....
SRY2 ....C....A .....A.A. .... .A..... T.....C.A .T..... .....
SRY3 ....A... ....AA .... .A..... T.....C.. .T..... .....
SRY8 ....A.A. .... .T..... .A..... .....C.. .T....A... .....
SRY10 ....A.A. .... .T..... GA..... .....C.. .T....A... .....
Sry-mouse ....T..... ...A...T.A ..... .GT....G.. ..... T.....A... .....G .....
SRY-rat ...A..... .....T G....C..A. ...T...T.. .....G..-G .G.....G.. GCA.....G T.....A..T .....G .....
SRY25 GTGCCCCTTG AATGCATTTA TGGTGTGGTT CCATGGTGAG ATGAGCAAGT TGTCCCAGAA GAAT-CCAGC ATGCAAAATA CAGATATCAG CAAGCAACAG
SRY5 ....-..... ....
SRY6 ....C ....-..... ....AT.
SRY15 ....C ....A..... ....-.T... ....T.
SRY20 ....C ....A. ....-..... ....T.
SRY9 .CA....A.. ....C .TG..... .G.CA..... .....C. ....C..... ....G..... ...C....T.
SRY12 .CA....A.. ....C .TG..... .G.C..... .....C. ....C..... ....G..... ...C....T.
SRY13 .CA....A.. ....C .TG..... .G.C..... .....C. ....C..... ....G..... ...C....T.
SRY23 .CA....A.. ....C .TG..... .G.C..... .....C. ....C..... ....G..... .....T.
SRY24 .CA....A.. ....C .TG..... .G.C..... .....C. ....C..... ....G..... .....T.
SRY11 .CA....A.. ....C .TG..... .G.C..... .....C. ....C..... ....G..... .....T.
SRY18 .CA....A.. ....C .TG..... .G.C..... .....C. ....C..... ....G..... .....T.
SRY17 .CA....A.. ....C .TG..... .G.C..... .....C. ....C..... ....G..... .....T.
SRY21 .CA....A.. ....C .TG..... .G.C..... .....C. ....C..... ....G..... ...A....T.
SRY14 .CA....A.. ....C .TG..... .G.C..... .....C. ....C..... ....G..... .....T.
SRY4 .CA....A.. ....C .TG..... .G.C..... .....C. ....C..... ....G..... .....T.
SRY19 .CA....A.. ....C .TG..... .G.C..... .....C. ....C..... ....G..... .....T.
SRY16 .CA....A.. ....C .TG..... .G.C..... .....C. ....C..... ....G..... .G.....T.
SRY22 .CA....A.. ....C .TG..... .G.C..... .....C. ....C..... ....C..... .T.....T.
SRY7 .CA....A.. ....C .TG..... .G.C..... .....C. ....C..... ....G..... .....T.
SRY1 .....A.. ....C .TG..... .G.C..... .....C. ....C..... ....G..... .....T.
SRY2 .C.....A.. ....C .TG..... .G.C..... .....C. ....C..... .....C.... ....G..... .....T.
SRY3 .CA....A.. ....C .TG..... .G.C..... .....C. ....G..... ....G..... .....T.
SRY8 .C.....A.. ....C .TG..... .G.C.T.... .....C. ....C..... ....G..... .....T.
SRY10 .C.....A.. ....C .TG..... .G.C.T.... .....C. ....C..... ....G..... .....T-
Sry-mouse .C.....A.. ....C ..G..... .G.CA..... ..G.....C. ....C..... ....G..... .....G.T.
SRY-rat .C.....A.. ....C ..G...A... .G.C..... ..G.T..AC. ....C..... .....G...T ....G..... .....T.T.
SRY25 GGATACAGGT GGAAAAGCCT TACAGAAGCT GAAAAAAAAG GCCCTTTTTC CAGGAGGCAT AGAGACTGAA GGCCCTTCAC AGAGAGAAAT ATCCAAACTA
SRY5 ....
SRY6 ....--. ....
SRY15 ....T..... ....--. .T..... ....
SRY20 ....--. ....
SRY9 ....C--. ....A.....C ....T...A... ..... .C.T.....
SRY12 ....C--. ....A.....C ....T...A... ..... .C.T.....
SRY13 ....C--. ....A.....C ....T...A... ..... .C.T.....
SRY23 ....C--. ....A.....C ....T...A... ..... .C.T.....
SRY24 ....C--. ....A.....C ....T...A... ..... .C.T.....
SRY11 ....C--. ....A.....C ....T...A... ..... .C.T.....
SRY18 ....C--. ....A.....C ....T...A... ..... .C.T.....
SRY17 ....C--. ....A.....C ....T...A... ..... .C.T.....
SRY21 ....C--. ....A.....C ....T...A... ..... .C.T.....
SRY14 ....C--. ....A.....C ....T...A... ..... .C.T.....
SRY4 ....C--. A..... ..A.....C ....T...A... ..... .C.T.....
SRY19 ....C--. ....A.....C ....T...A... ..... .C.T.....
SRY16 ....C--. ....A.....C ....T...A... ..... .C.....
SRY22 ....C--. ....A..... ....T...A... ..... .C.....
SRY7 ....C--. .....A ..A.....C ....T...A... ..... .C.....
SRY1 ....C--. ....A.....C ..... A.T...A... ..... .C.....
SRY2 ....C--. ....A.....C ....T...A... ..... .C.....
SRY3 ....C--. .....A ..A.....C ....T...A... ..... .C.....
SRY8 ....T. ..... .G....C--. ....A.....C ....T...A... ..... .C.....
SRY10 -..... .... .G....C--. ....A.....C ....T...A... ..... .C.....
Sry-mouse ....G..... ....C .....--. ....C .....T.... .AT...A... ..... .C.....
SRY-rat .....TCA.. ....C .....--. ....GC ..... .A....A... ....

Internal stop codon

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SRY25      TAAATATCAG CCTCGTCAAA GGGCTAAAGT GCCACAGAGG AGTGGCACTT TGTAGCCTGC AGTTGCCTCA ACAAAAGTGT ACAACATGCT GCAGTGGGAC
SRY5       ...
SRY6       
SRY15      
SRY20      
SRY9       
SRY12      
SRY13      
SRY23      
SRY24      
SRY11      
SRY18      
SRY17      
SRY21      
SRY14      
SRY4       
SRY19      
SRY16      
SRY22      
SRY7       
SRY1       
SRY2       
SRY3       
SRY8       
SRY10      
Sry-mouse  
SRY-rat    

SRY25      AGGAACCCAC ATGCCATCAC ATACAGACAA GACTGGGCTA GAACTGTACA CCTGTCTTCC AAGAACCAGC AAAGCTTTTA TTTGCAGCCT GTGGACATCA
SRY5       
SRY6       
SRY15      
SRY20      
SRY9       
SRY12      
SRY13      
SRY23      
SRY24      
SRY11      
SRY18      
SRY17      
SRY21      
SRY14      
SRY4       
SRY19      
SRY16      
SRY22      
SRY7       
SRY1       
SRY2       
SRY3       
SRY8       
SRY10      
Sry-mouse  
SRY-rat    

SRY25      CCACTGG--- ---------G CAGAAGCAGC AGCAGCAGCA GCA------- ---------- -------ACA CCAGCAGCAG CACAAGTTCC ACCAGCAGCA
SRY5       
SRY6       
SRY15      
SRY20      
SRY9       
SRY12      
SRY13      
SRY23      
SRY24      
SRY11      
SRY18      
SRY17      
SRY21      
SRY14      
SRY4       
SRY19      
SRY16      
SRY22      
SRY7       
SRY1       
SRY2       
SRY3       
SRY8       
SRY10      
Sry-mouse  
SRY-rat    
```

```
SRY25     CCTCACCTAC TTACTGACAG CTGACATCAC TGGTGAGCA- ---TACACCA TAGCAGGAGC ACCTCAGCAA AGCCCTGTGG TAGGCAGTCT CATAACACTG
SRY5      .......... .......... .......... .........- ---....... .......... .......... .......... .......... ..........
SRY6      .......... .......... .......... .........- ---....... .......... .......... .......... .......... ...G......
SRY15     .......... .......... ....T..... .........- ---....... .........T .......... .......... .......... ...G......
SRY20     .......... .......... ....T..... ..C......- ---....G.. .......... .......... .T...A.... .......... ...G......
SRY9      .......... .......... .......... .........- ---....... .......... .......... .T........ .......... ...G......
SRY12     .......... .......... .......... .........- ---....... .......... .......... .T....A... .......... ...G......
SRY13     .......... .......... .......... .........- ---....... .......... .......... .T........ .......... ...G......
SRY23     .......... .......... .......... .........- ---....... .......... .......... .T........ .......... ...G......
SRY24     .......... .......... .......... .........- ---....... .......... .......... .T........ .......... ...G...T..
SRY11     .........A .......... .......... .........- ---....... .......... .......... .T........ .......... ...G......
SRY18     .........A .......... .......... .........- ---....... .......... .......... .T........ .......... ...G......
SRY17     .........A .......... .......... .........- ---..T.... .......... .......... .T........ .......... ...G......
SRY21     .........A .......... .......... .........- ---....... .......... .......... .T........ .......... ...G......
SRY14     .......... .......... .......... .........- ---....... .......... .......... .T........ .......... ...G......
SRY4      .......... .......... .......... .........- ---....... .......... .......... .T........ .......... ...G......
SRY19     .......... .......... .......... .........- ---....... .......... .......... .T........ .......... ...G......
SRY16     .......... .......... .......... .........- ---....... .......... .......... .T........ .......... ...G......
SRY22     .......... .......... .......... ..T......- ---....... .......... .......... .T........ .......... ...G......
SRY7      .......... .......... .......... .........- ---....... .......T.. .......... .T........ .......... ..GG......
SRY1      .......... .......... .......... .........- ---....... ...........  .......... .T........ .......... ...G......
SRY2      .......... ........T. .......... .........- ---....... ...........  .......... .T........ .......... ...G......
SRY3      .......... .......... .......... .........- ---....... ...........  ......A... .T........ .......... ...G......
SRY8      .......... .......... .......... .........- ---....... .......... .......... .......... .......... ...G......
SRY10     .......... .......... .......... .........- ---....... .......... .......... .......... .......... ...G......
Sry-mouse G..T...... .....A.... .......... .........- ---....... ..C....... .........C .......... .T........ ..G.....
SRY-rat   G..A...... ......T.CA ....T..... ......C.GA CCC.G..... .......... .......... ..A......A ....T.A... ...G.T....

SRY25     GCCTCTG---
SRY5      .......---
SRY6      .......---
SRY15     .......---
SRY20     .......---
SRY9      .......---
SRY12     .......---
SRY13     .......---
SRY23     .......---
SRY24     .......---
SRY11     .......---
SRY18     .......---
SRY17     .......---
SRY21     .......---
SRY14     .......---
SRY4      .......---
SRY19     .......---
SRY16     .......---
SRY22     .......---
SRY7      .......---
SRY1      .......---
SRY2      ....G..---
SRY3      .......---
SRY8      .......---
SRY10     .......---
Sry-mouse ....T.T---
SRY-rat   ..TCTGCTCC
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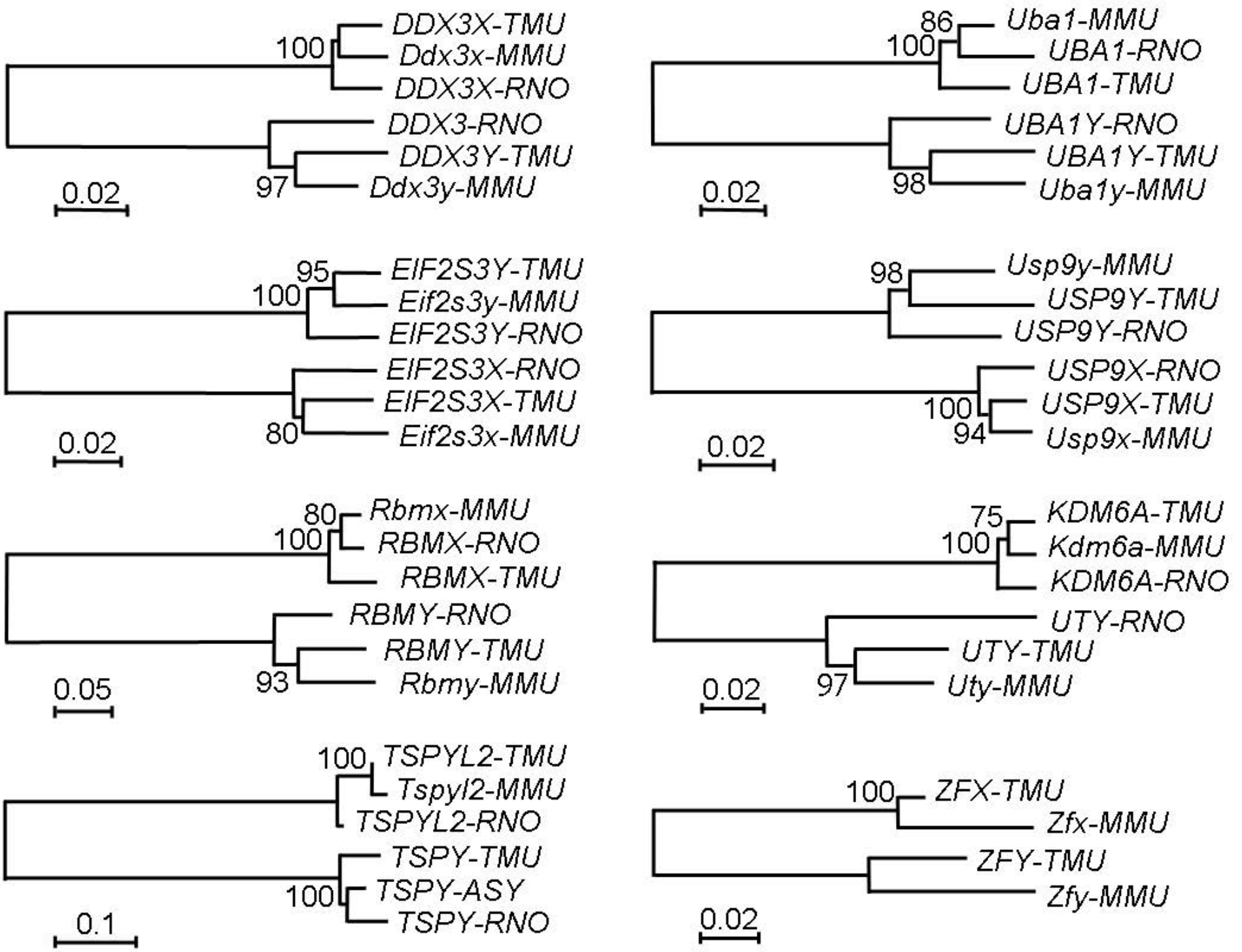
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86
Uba1-MMU
100
UBA1-RNO
UBA1-TMU
UBA1Y-RNO
UBA1Y-TMU
98
Uba1y-MMU
0.02
95
EIF2S3Y-TMU
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EIF2S3Y-RNO
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80
Eif2s3x-MMU
0.02
98
Usp9y-MMU
USP9Y-TMU
USP9Y-RNO
USP9X-RNO
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USP9X-TMU
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Usp9x-MMU
0.02
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Rbmx-MMU
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RBMX-TMU
RBMY-RNO
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93
Rbmy-MMU
0.05
75
KDM6A-TMU
100
Kdm6a-MMU
KDM6A-RNO
UTY-RNO
UTY-TMU
97
Uty-MMU
0.02
100
TSPYL2-TMU
Tspyl2-MMU
TSPYL2-RNO
TSPY-TMU
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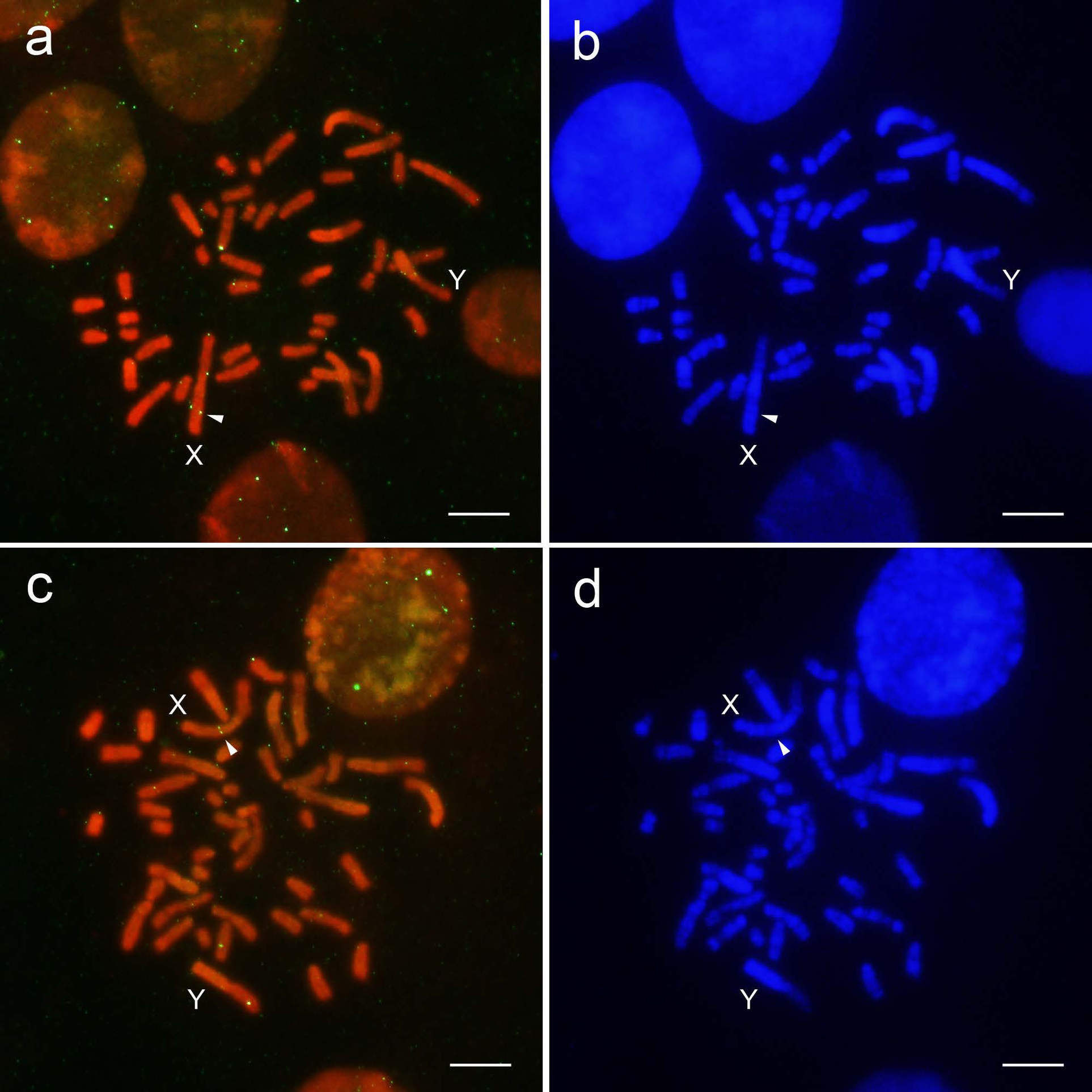
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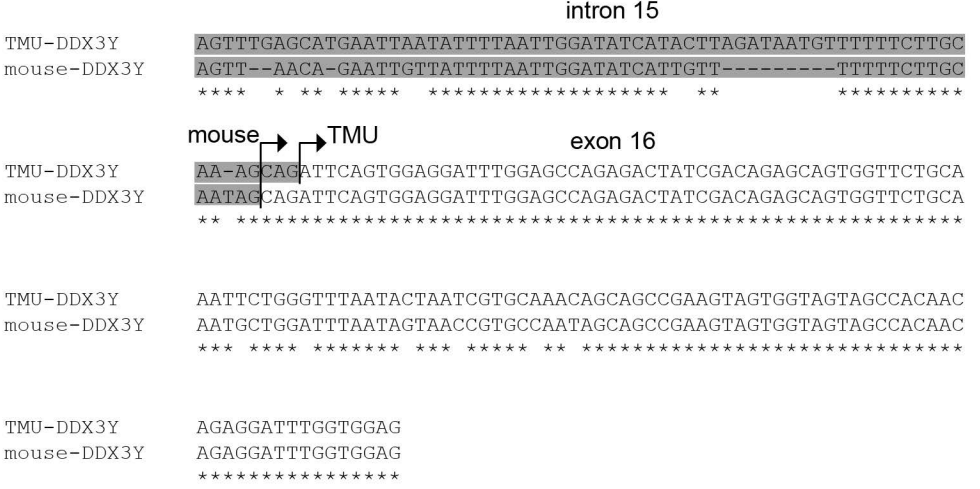
intron 15
TMU-DDX3Y AGTTTGAGCATGAATTAATATTTTAATTGGATATCATACTTAGATAATGTTTTTTCTTGC
mouse-DDX3Y AGTT--AACA-GAATTGTTATTTTAATTGGATATCATTGTT---------TTTTTCTTGC
mouse
TMU
exon 16
TMU-DDX3Y AA-AGCAGATTCAGTGGAGGATTTGGAGCCAGAGACTATCGACAGAGCAGTGGTTCTGCA
mouse-DDX3Y AATAGCAGATTCAGTGGAGGATTTGGAGCCAGAGACTATCGACAGAGCAGTGGTTCTGCA
TMU-DDX3Y AATTCTGGGTTTAATACTAATCGTGCAAACAGCAGCCGAAGTAGTGGTAGTAGCCACAAC
mouse-DDX3Y AATGCTGGATTTAATAGTAACCGTGCCAATAGCAGCCGAAGTAGTGGTAGTAGCCACAAC
TMU-DDX3Y AGAGGATTTGGTGGAG
mouse-DDX3Y AGAGGATTTGGTGGAG

a

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upstream-TMU   AAAGCTCCCA AAGACTGGGT TCTCTTAAGT CATTATCACA TTCTTTTGGG ATGGTCCTGT GAGGAGAATT ATAAAGGCAG AGGCTATCCA TGGAGTAGAC
upstream-rat   ....TA.T.. .G...CT... .....-...C ........T. ..G....... G.A....C.. .......... .......... .......... ....A..A.T
upstream-mouse ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------

upstream-TMU   AACAGGCAAA ATAGAAATCA TGGCCGCAGT GCACCCCATA CACGTTACCA TAGGTCTCTA TTAGGAAAGT TAGAAAGATG TCTCAGGTAG GGAGTCTTCT
upstream-rat   .G........ .-....TC.. .....A-... ...G..TG.. ...A.....T .........G C......... .....G.CA. .......... ......C...
upstream-mouse ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------

upstream-TMU   GATACCCTCA GTGGGAGAAG GAGAACTCTG GGTGCTATGA TCTAGATGAC TGGCATTGTG TGGATGGAGT AAAGAGGCAG GACTGGGACT GACATGCCAT
upstream-rat   ...G....TG A........A AG.G...... .A..T..... ...G....G. .....GC... ..CT.....A ..T...A... .......... ..........
upstream-mouse ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------

upstream-TMU   TCAGTGACTC TACTCCCTCA GTAACTAGTA GTCATTAGGT GTTATTCAGG TGAGGGAAGT AGGGGTGCAG CAAAACTGTA CTCGAGGGCA CAAACTGGCA
upstream-rat   ........A. .......... .C....G... .....G.... .....C..C. .....A..-. ......AG.. A.GGG..... ..T....AT. ....G.....
upstream-mouse ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------

upstream-TMU   AGACGCCAAA ACATGCCCTT ACCAAAGGAT AGAAAGCCAA AAGCCAAGGG TGTTGGCTCA ACAACA---- ---GTGCCCT TTCTTGAGCT GAGTGAGGAA
upstream-rat   ...T...... .......T.. G...GT.... G..G...... .-........ CAG.....TG G..GA.TGGA CGG.G.T... ....G..... ..........
upstream-mouse ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------

upstream-TMU   GTCAGAGAGA GAAAGGCATT CCCTCCCCCT AATAGGCAAC AAGCACTCTC TTAGGCAGAC ACCCCAGAGA GCATCAGTGC CTCCATCAGG AGGGATCAGG
upstream-rat   .........- -.....T... T.....T... ......TT.. .T........ .....A.... .G.A..C... T.....A... ........A. ..CA.C....
upstream-mouse ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------

upstream-TMU   TAATAACAGG CTACCAGGGA AGGCACTTGG GGCACTTTGA CTCCTGCTTC AGAGTCCTGG AGGAC----- -------CCT GCCTCAAATG GGAAAGCCCA
upstream-rat   C......... .......AC. .....T.... ........A. .......... .......... .....ACACT AATTGCT... ......TG.. .....T.A..
upstream-mouse ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------

upstream-TMU   GAATATTGGC CTTGACAGAA TTCCCTGTGA AGCCCTCACA ACCAAGGCCC CAGTGTAGTT CTCAGTGTTG GAACATGTCC TCACTAGCCA CCCTGACTCA
upstream-rat   ....G..... .......... .........C CA.....--- ---------- .....C.... ....A..... .G.TG..A.. .......A.. .A........
upstream-mouse ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------

upstream-TMU   GGCACAGGTT TCAAGTGTCA AGTTTGGGGT TCTGAGAGTG ACCCAGATAT CCCAGCCCCA CAGTGTGTTG AAAGGTGCAG GTTCACTGTC AGCAC-----
upstream-rat   .A......A. .....A..A. ...C....T. .........A .......A.G ....T....G ......TAG. .......... C....T.... .....GGTTA
upstream-mouse ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------

upstream-TMU   ------TCCC CAACTTCCAT TCCACCAGTA CCTTCAAAGC CCACCCTTTC CCCACAAGAA TCACCTTTTC CTTTTATTGT GTCCAAAACA CAGGAAAACT
upstream-rat   TGGAAC.... .......... C...T....G ..C.....-- -.TGA..... T.......T. ...G...... ..G....... .....C.... ..........
upstream-mouse ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------

upstream-TMU   G--//  LINE insertion //--TGCCCC TCC-TCTAAT CTTGTATTGA CATGGCAGCT TTCAGAGCAG AGACTGATGA GCCCTGTCTG AGTATCACGG
upstream-rat   .CTAGGCCTC ACCTGGGTGT CTGG...... ...AC..TGA ...A...... ..CA...... G-........ G..T...... .-----.... ..G....T..
upstream-mouse ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------

upstream-TMU   GGGCTGCTCC AGACCTTCCC TTGCTGATTA TCAACAAGGT GTGCTAGCTT CCTACT---- ---CTCCTTC TTCAAGGATG GACAAAGACC AAGCAGGCAA
upstream-rat   .....T.... ...T...... .........C ....G...A. ...GCCC... ......TCAA GGT..A.... .......TCT .G.C...... T.T.C....T
upstream-mouse ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------

upstream-TMU   GAATTAACAC ACAATACAGA ATGCTGACAA ACAGCTGAGG CAGGCTGACT GGAGATTACT CACAATG--- --------CT TACTTGACTT GAGTTGACAC
upstream-rat   ....AT.... .G....TGA. ....CAG..C ...AT..... A......G.. ..GC...... ....G..TTT CCTTTACT.. .......A.. ...C..G..T
upstream-mouse ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------

upstream-TMU   TCAGATCTCT CTGCCTGAAG TGACCTTGAC TCAGCATCTG CTGAACTCC- ---------- -----TGATA GTCTTC---- ---------- ----------
upstream-rat   .....-.... .........A G...T...C. .......T.C .C....A..C TTCCTGCCCA AGAGC...C. AA.C.TTCTG TGCCTTCTTG GGCCAAGCAG
upstream-mouse ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------

upstream-TMU   ------GTGC TCCTCCATTA TTGAGCCAAG ---------- ---------- ------AGCC AAGAAACCTT AGCATG---- -CCTTCCTAG GCAAAGCAGA
upstream-rat   ACAAAT...G .AGGT.C..C ...G...... CAGACATATG TGGTAGGTCC ATCTTGG... ...C.GA.A. .TGTG.TAGG T.....T.G. ..C.......
upstream-mouse ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------

upstream-TMU   CATATACCTC AGGGAACTT- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----ACCAGA
upstream-rat   .....GTGGT ...TC.T.CT TGGGCCAAGC ACACATATGT GGTAGGTCAT TCTTGGGCCA AGCACACATA TGTGGTAGGT CCTTCTTGGG CAAAG.AGAC
upstream-mouse ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------

upstream-TMU   GGATGTGGTA GGTCCTAGAA GCAAACTGGG GGAGGCATAG GCTTGGCTGT GGACTTGGAC TTTGAGGAAT ACCTGTTTTC GTATCTCACA GGAGGACCCA
upstream-rat   AT........ ......T... .......... A...A..... .......... ...T------ G.....A... ......CC.A T.GC..... .....GA..T
upstream-mouse ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------

upstream-TMU   TTG--AAATT GGGTTTCCCT GACCTGGAGG TCTGCATCTT CTGCTACGCA CATTACTCTA TTTACCTACA TTGGCTCCTC TGGTAGGCCA TCTACCATCC
upstream-rat   C.AGC....G .CT....... .......... .......... .....C.C.. .......... C....TG... .......... .......... ......G...
upstream-mouse ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------

upstream-TMU   ATTGGACTAA GAGATTGTTT ACCCACTGGC ACTCTGTGAA GTTTCTCTGC CTGTGCTAAG ATTCCCCCCT ATGATCTCCT GCATACACTG CACATATTCT
upstream-rat   .C..A..... .....G.... GT.---.... .......... ..G....... .......... .G......-. .CC...C.T. .T........ .TT..C....
upstream-mouse ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------

upstream-TMU   GGACGGTGTG -TCAAGTCCA GAGCTGTCAG CCACTTTTCC CACATTCATA TACCAGGCAG CTCAGCCAGT GTTCCATGTG TTCTCCCTCC AGCCCTCT-T
upstream-rat   ....AA.... G....A.... ..TAG..... .AG..C.GTG A..TC..... ........T. .........G A..G...... .......... ....T...C.
upstream-mouse ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------

upstream-TMU   CTGTCTAAGA GAAATACTTC AGGAGATCCA GCCAGAGGTC TTCTGCAAAG ACCTGGAAAG AGGTGAGGCA AGTGTCTCTC TAGCTTGTGG GATCAGTGTC
upstream-rat   G.CGGAG... ...G...... T...T..... .A....T... .......GT. .....----- -----..... .A......-- ......C... -..GG.G...
upstream-mouse ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------

upstream-TMU   CTTGTGCAGC CCAGCAGGAC TCATGACACT TTTGGTTTTT AGAGTATAGA GTTCAAAGGT ATGTGTTTGT TGTATTTGTC TGAGTGTCTA TGTGTTACTT
upstream-rat   .C.C...... ...C..A.G. ...G.....A ..---....C ......G... A.A..GTA.. G...T.C... .......... .........G ....A.TA.C
upstream-mouse ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------

upstream-TMU   CTTCCATGGG GTGTGTATAT GTGTGTGTTT GTGCATATCT GTAAATAATT GAGTCTCAGA CCCACATTAG AAAAGTGCCA GCAAAAACTG TAGGTTGTGT
upstream-rat   TGAATG...A ......---- --......C. ...TG..... .......... ........A. TG...T.... C..CA..... ....C..... .T..G...A.
upstream-mouse ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------
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upstream-TMU   AACAACAATC CTAATAAAAA ATAATTTTAA TATTACTATG TAAGAAATTT GTACAAATGA CTTTTATGAT GTATTGATAT GACAAAGAAA GGAAGAAAAG
upstream-rat   TGA..T.... .......C.. -..T.....G ........CA ..G....... .......C.. GC........ .C..GT.... ........G. .A.......C
upstream-mouse ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------

upstream-TMU   TTC-TTGAAA TCTTTGTTGA TATTTATTTC TTGTAGCTTG AAACTCCAAA GCTCAGCTTA TGCATTAGTA GTGATTTTGT TTTGTTTTGT TTTGTTGTTT
upstream-rat   ...C....G. .A.A..... GT....GG.T .........T C..T..T... AT....---- --.....C.. C..T..... ...TGG..-- ----------
upstream-mouse ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------

upstream-TMU   TGTTTTTCTC TTTCATGGTG TAGATATTTA TACTGGGCTA TTGAGCCAGG ACAGACTATT TCTTGGTGTT TTCCATGCCG CCTGCCTTAG TGGACTAGGA
upstream-rat   --...C.T.G .CC...T... .....C.... ..G....... ...C-..T.. .......GA. ....---T.. C.T......A T..CG....C ..........
upstream-mouse -------T.. ...T...... .......... .......... .A.......A .......... ....A..T.. ........T. ......G... .A.....T..

upstream-TMU   TACAAATGAT AGGTTTCATG CTTATCA--- TTAACT---- --ACTACTTT TGCATGCTTT TTTATTTCTC ATGTTGGTCC ATCCTAAATA TCATGTACAC
upstream-rat   .....T.... T.T....C.A ......T--- ..G..CCTCT CT..C....G .A.......G .......... .-...A.... ...A...... .......TG.
upstream-mouse ...T...... .....C.... .......TTT .....CCTTT CC........ ....A...CG ..A....... .C...A.... ........C. .T.G....GT

upstream-TMU   TACAGGGCAA AA-GAACTCA TTAGTATCCT TGTTGTTGTA TCTTAATCCT TGTTGTGACA GAATGATCAA ATAGAAAGAG ATATTTTTTT GAGGAGTGTA
upstream-rat   .......... ..A....GGC .......G.. CA........ .......G.. .....C..AG A.....C... .G......TT G...G..... A.........
upstream-mouse .......... ..-....... .C.....GT. CT....---. .......A.. .......... .......... .G.A....C. ....A..... A........G

upstream-TMU   AAAAAAAAAG TCTCTAACAT TCACAAACCA GAGGGGTTTT GGTTGACAAT GGGTAGGTGT GTTGTTGTTG TCTTAAAGTA GATTAGTTGG GTTGGACTAT
upstream-rat   ...GTGC.-. ......-... .T....T... C......... .......... AT......T. ..-..G..G. CT.C...... .......... .C.....GTG
upstream-mouse ...GTGC.-. .......... ........TT .......... .C......G. ATC.....TC A.-.AGA.G. CT.C...... .......... .C.......G

upstream-TMU   GAAGGTCCTG AAAGATGAGG GACAAATATT TTCTTGCACA CTTTAAAAAC TAAAATCTTT ATTACAAACA CATTTTGGAT AGTAGCTTTT AGCTCTTGCA
upstream-rat   .G......C. .GG..G.... ..TG...... .....A.... .......T.. .......... G......... ..G..C.... .C........ .C.C...A..
upstream-mouse .G........ ..G..G.... ..T....... .....A.... .G.....T.T .......... G......G.. ........TC ...G...... .......A..

upstream-TMU   CTTTAAGTTT TGACTTCTGT CTCTGTCTCT CTGTCTCTGT CTCTCTCTGT CT--CAATCT CTGTCTCTCT ---------- -----CATCT CATCTCTGTC
upstream-rat   .....TA.C. ...T...... ........G. ..C.G...C. ..G.T..... ..--.T.... .......... ---------- -----....C ........A.
upstream-mouse .......... .......... A.......G. ..C.G..... ....G..... ..GT.TG... T.....G... GTCTGTTTCT CTTTG....A ..........

upstream-TMU   TTCTTGGTTG CAATCATAAT TCTTCTACTA TGTTCAGCAT TTCAGATCTT GCTTTTTAGT GTTCAGCCCT ACAGCCACAG GATATCTTAA GCTCCGGAGA
upstream-rat   ...C...... .....C.... .....C.... CA........ .......... CAC...C... A......... ......TG.. ..C..A.... T...T.....
upstream-mouse ..T....... .......... .....C.T.. .......... .......... .A........ .......... .........T .......... A...T.A...

upstream-TMU   AGAGACAAAC TTTGAGACTG GTGACAACTG TCTTGAGAGC
upstream-rat   ..G..-G.G. ....G..GCA ......GT.. ...A..T...
upstream-mouse ........GT ....G..... .......T.. ...A......
```

b

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downstream-TMU   ---------- ---------- ---------- ---------- ---------- --------CA GGAGCACCTC AGCAAAGCCC TGTGGTAGGC AGTCTCATAA
downstream-mouse ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------
downstream-rat   CAGCAGCTAA CCTACTTACT GTCCACTGAT ATCACTGGTG ACCGACCCTG CACCATAG.. .......... .......A.. ....A....T .A......G.

downstream-TMU   CACTGGCCTC TGCTCCTACC TATGTCAACA CTCCCCTTTC CTATAGTTTT ACAGCCAGAG ATTCCTAGTT GATTTGGATA CTGATTTCCC TCCTGTTGTA
downstream-mouse .........T .T........ ....C..... ......C..G ....GA.... TA..T.T... .......... .G...A.... ....C..TT. ......CA.G
downstream-rat   T......-.. .......... ....C..... ......-..G ..G..A...- .....TT... .......C.. .G........ ....G..... ..TG......

downstream-TMU   ATAAATGTCC CGTTCCTGTT CCTTCCTT-- TAGGGTT--- ---------- CTGAAATGTG TGTAAAGATG CATAGTGTGT AAAGAAGGAG AATGCTTAGA
downstream-mouse .A...----- -.G.....C. ........-- .T.....--- ---------- ..A....... .......... TG........ ..G....... ..A.......
downstream-rat   .....G.... T.C...CC.. TT..TT..GG .G...G.GTG GGGGAGGAGA .CA....... .....G.... TG........ ..GA...... ..........

downstream-TMU   AATGCTA-CT ACATATTTTT TGTTTGACAC AGTGCTAGTT GCATTGTTGT GGCATTTGAT TCCACAGAAG AAACCAGAGG CTTAGACTGA GATCCAATTT
downstream-mouse ......T-.. .....G.... .....A...T ...A...... .......... .......A.. -.......... .........A T...C...C. ..........
downstream-rat   .GG...TA.. .....C.... .....A...G C..A...C.C .....C..A. .......A.. -.G.TG.... ....T.--.. T.......C. T.ATG.....

downstream-TMU   AGATGAAATG CATTTATTCT TACCCTAGCA CAAAGAGAGC AAACAGCTTT AGAGCCAATA CAGCATGCCA TGCTAAAGTG GGGGGTAGGG GTGGGTAAGG
downstream-mouse .......... .......... ..GG...... ......C... .......... GTG.T..... .......... .......... ....C..... ...A.....C
downstream-rat   .TC..T.C.. .---------- -----C...T .....GC... .......... .......... ....T..A.. .......... ..A...G..A .A...G....

downstream-TMU   TGGAGTTTTA AAGGAAACAT CAGAAGGG-- -------ATG -CAGATATCC AGCTGGGCAA GAAAATTCTT TTCATTCCTG TTATGTAGTA ATAAAAGACC
downstream-mouse .......... .......... .....T.ACA CATTGTA... A..A...... .......... .......... .......... .......... ..........
downstream-rat   .......... .......... ........TA CAC----.CT A..A...... .......... .G........ .......... ..TGA...G. .....G...T

downstream-TMU   ATTTCATCCC CTTTCCCAGA GTTTAGCAGG TTTTATAGGA ACAAAGGCAG CTGTTAATTC CTCACAGCCT ATTATCCTAT TCTTATCTCA CTCTATGGCC
downstream-mouse .......... ....T...C. .A.A.T.... .....C.... ......A... ....G..... .......... ....G..... .........T. ....T.....
downstream-rat   ......AT.. ........C. .A.A..A... .G...C.... ...G...... .......... T..C.....C ...G.T.... ....T...T. AA..CCA.A.

downstream-TMU   AATAGGATCT CCAAGGCATT CCAAGGCAAA GGTCAAGGGC CCTTACTGGA CGCCTGGCTG CATATTCAAT TTCTAGTGCC ACCACAGGGA ATATATATAA
downstream-mouse .......... ..C......C .......... ........A. T......A.. T......... ...G..A... ....TTA... .....C..AG .C.......T
downstream-rat   .G........ TGC....... ....T..... .........A .T........ T......A.. ......A..G ....TTA... ....AG.... .C....G...

downstream-TMU   ATT------- ---------- ---------- ---------- ------ATCA CTCAGGTTGA GCCTTCTAAT GACTGGGATG CAGCAATTCA TGTCTGAGCA
downstream-mouse ..ATATATAT GTGTATATAT ATATATAATA TGTAGGTATG TATATA.... T......... ..A....... .......... ...T.G.... .T........
downstream-rat   ...------- ---------- ---------- ---------- ------.... ..T....... .TA....... ....A....A ...T.G.... .T........

downstream-TMU   GACCAAAACA CAGTGTCATG AAATCAGGGC TGAT----GT GTCCTTATAG CATCACCATG GAAACCATTC TTTTTCTGTT AGTCTTCAAT ACTCATCTCT
downstream-mouse .G.A...... .....C...C GGG....... ..GCTGTCA. T......C.. .......... C.......-- -----....A .......... ........T.
downstream-rat   .G.A...... .....C.C.C .GG..T.... ..GCTGTCC. ......TC.. .......... .........G .C.A.....A ...GG...G. ..........

downstream-TMU   GAAAAACATA ACACAACTTA ATGCTGTGTA ACTACAGGCT GTGCCTCTTT AGCATTCAAA ACACTCCTTT GTAGATCAAA GGCAGGTCTC TGTTTAT-GG
downstream-mouse .......... C..-...... ..A.....A. .AC....... ...T...... ..T..G.... .......... ..T....... T...TC.T.. .......CA.
downstream-rat   .......... ------.A.. .......AA. .AC..T.... .....C.... .....G.... GT........ ........G. CA..TC.A.. .......CA.

downstream-TMU   AGTCTCTCTC T--AGTACAC AAAACTACAT CATGAGAATG TAAAGTCT-G T-AGTGTGTG TGGGGGGGGG GATATTT--- ---------- ----------
downstream-mouse G..T...... .--..C.... .......... ......T... ........T. .G........ ..T.T.T.T. TG.G.G.GTG TGTGTGTGTG TGTGTATGAC
downstream-rat   G......... .CC..C.... .......... ...ATATGAC .TCT...A-C .GT.....CT ..C.T.T.CA CG.GCG.GCC AGTGTGCATG GGTTTGTCTT

downstream-TMU   --TTCATTTC TCCTCCACAC TGTGCAAAGT CCAGTTGTTC CATGAAATAT GCTATGAGCT CTCTATAGTA TCTTTGGGTA AAATAAACCT TTATCTTGTG
downstream-mouse AT..T.G... .........A .........C .......... T.....C... .......... .......--- .....T.... ......G... ....------
downstream-rat   -T....C... ....T..... .A..AT..AC .......... TG.A..C.C. -------T.. ....G..AC. ..G..T.A.. ......G.A. ....ACAA-.

downstream-TMU   TTTATGCAGA AGAATTCACA CT--GCTTCT CTTTTATTAT CTATCTCTAA AGAATAAATA GTTTTATTAA CATAAAAATT TTTATGTAAA AGCACCACAG
downstream-mouse -----...T. ..G.G..... T.TT...... .......... .......... ......G.CT ........G. ....C.GC-- ---------- ----------
downstream-rat   ..C.....C. G...G..... T.CT...... ......---C ......T... ........C. ........C. ..---..... ...G.A.... ....GT....

downstream-TMU   AAAAAGCTAT GTTTTTGA-- -CTAGGTAAG AATCAGAAAC AAAATGAAGA GAGCAAGAAG CTACATAGGC AAGCTAAGTG GATGTACCAA CACTGAAGAT
downstream-mouse --......G. .....CA.GG G....C.... .......... ......G.C. A.-------- -----...T. .......... .......... ....T....C
downstream-rat   .C........ .....CC.GG A.CG...... .......... ......T.C. A....CC... T......... .......... ..-------- ----------

downstream-TMU   GCCACATATA ---------- GGAATA---C AAAAGGTGAA GGGGAGGGTA CAAAGTGTAA GTGGGGTGAA TCAATGGCCA TCTGCCCATG AGCTCAACTG
downstream-mouse A......... CAAAATGATT ......AAA. .......... ........C. ---...A..T .....A.... .G........ .....A...T .A........
downstream-rat   ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------

downstream-TMU   CCAGATTGTT CAGCTAAGGA ATGGTTCAAA GTTAACCACT CTGGTTCTTA CAAACAAGCT TAAAAGAGAA ATACACAGCA CACACAGCAA CGTCTGCAAC
downstream-mouse .......... A......... .......... ....C..... ..T..... ........T. ....-....C ...A.G..TG T.......... .CC.......
downstream-rat   ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------

downstream-TMU   AGCTTCTTTA AGAAAGATCA AGGAACTACA CATAGG-TTA TTGACAACAA ATTACA---- --AACCTTCA GTTAACCTGT TACAGTTTCA TCACATCACT
downstream-mouse ..T....... .......C.. .......... ......C..G ..A...C..T G.....TACC CAG....... ....G..... .C..A..... ......T.A.
downstream-rat   ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------

downstream-TMU   CTCCCACAAT GTTTCCCTCA CAGTAACTCC ATTAGCCATA AACTTTTGAA TCGACTATGT AACCCTGGCA CAACTCTAAA TTGTAGGTTG GAAGAAAATA
downstream-mouse .....T.G.. .C........ T.A......T G...C....G .......... .T..T..... .G.....T.. --........ ACA....... ..........
downstream-rat   ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------

downstream-TMU   GCAGTCATGG AGCCTCAACT GTGACATCTT ATACACTAAG TACTATACAA CAATATGGTG TATTATCTGA AGTATTGAAC TCTTTGAAAC AATTAAACTA
downstream-mouse ....C..A.. .......... .......... .........A .......... ...C...... .......... T......... ....G..... ..........
downstream-rat   ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------

downstream-TMU   CTGTTCTACA TGGTCTTAAG ACTTTTGAGT TTCAGTTGAT TTTCAAAACC ATCTATTGTG AACACTAAAT TACATGTATG ACTTTCTGTA GATGAGTTAA
downstream-mouse .......... .......... ..C....... ..T....... ........TT ..G....... ....G...C. .........T GT........ A..C......
downstream-rat   ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------

downstream-TMU   AAGACTGTTG TCTAGAATTC AATGGAACAG TTAATTGACA CT-------- TCTAAGTGGT TGATTTTT-- ---------- ---TTCAGCA CCATTTCAAT
downstream-mouse .........C .......... .......... ...GC..G.. ..ACTGGACT .......... .AG.....CT TTTAAGGGTA TGT....... ..........
downstream-rat   ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------

downstream-TMU   GCCAATAATC AATAAG---- ---------- ------GGTT TCTAACACAA ATGAATACAA ATATATCCAT CCCAGTGGGG GCTTATGTAG --TTCTCTGG
downstream-mouse .....C.... ......ACTT TATAAGCCTT TGGATA.A.. .......... .......... .......... ....A....T ........CT CT........
downstream-rat   ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------
```

Non-homologous regions between mouse and TMU.

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downstream-TMU   AATCTGAAAG GCATAACAAC TAAGTGAAAG GAGAAATGAA AGCAAGAGTA TGCAGTCTTG GTGTGGTAAT AAGGTAAAGA AGCACCGAGG GGAATAAAAG
downstream-mouse ......... ........GG ...C...... .......A.. .......... .TA....... .......... .......... ..T...A... ..G.A.....
downstream-rat   ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------

downstream-TMU   GACTATCCTA CTCCATGTAG GAAGAGCAGG CCTTTGAGAC ACAGTAGAAA CAGCAGACAT TAGGACTCAG TGTGGAATTC ATCTGCAGTT TCCCTCCCAG
downstream-mouse .......... T.T..G.... .......... .-........ ....A..... ......G... ....C..... .......... .......A.. ..........
downstream-rat   ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------

downstream-TMU   CTCTGTTAAG ATCAGGCTCT GGGCCTGCTT ATAATCTTAC AGCTTGGAGC TGAGTGGCCA TGTTGAAAAG GCTTAAAATG AACTG----- GGAGGAGAGG
downstream-mouse .......... C.TG..T... ......A... .......... .A........ .......... G.....G... .......... .....GGGTT ..G..G.G..
downstream-rat   ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------

downstream-TMU   TGTATGA--A AAGGGAGGGG AGTGATTAGT GGTTAGG--- -GAGGGAATG AGTGCCTCAG GTAGATGGAT CAGGGAGTGG GCCCAGGCCC TTTGCCATTT
downstream-mouse .AGG..GAT. ..A.ATA... ..G.G..... ......TGGG T......... .T..T..... ..G.G..... ....A..... .......... ....T.....
downstream-rat   ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------

downstream-TMU   CCTTTGAGAA AGTTATCACA GGTTGTCAGG AGCCATAA-- //       LTR insertion     //--AA CTGAAAGTTT AAAACTGTAG CCAGAGGTGG
downstream-mouse .....C.... .C......T. ....T.G.A. .-..T...-- ---------- ---------- --------.. T......... ....A...-- --------..
downstream-rat   ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------

downstream-TMU   CCATTATCCA AGAATATGGC CACATTTGAG TGGAAAACTG AGTTTGGTGG GCTGTAACTA ACTCTAAAGA CAGAGCTTTA GAGTCTACTT TTAAATTTGA
downstream-mouse ........TG .....G.... T.....C... ....T..... T......... A..------- ------.... .....T.... .....C.TC. ....G.C..G
downstream-rat   ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------

downstream-TMU   CTTAAATTTG ACATGTCAGG CAGGATTGGT AGGGTGTTTT ATCCCACTGC CACCCAGGTT TT-GGCAAAG ATGAAAATGA GCCCAGTATG GAATCCCCAA
downstream-mouse .......... ....A..... .....C.... .......... .......... .T..T.A... ..T..... .......... ....GA.G.. .C.C...A.T
downstream-rat   ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------

downstream-TMU   ACTCATAAGA CTGTGGTGAG A-TTTGTCAT GTCAGTCAA- ACCTGGAAGG TGCCTAGAAC CTCCAGGACG TTGTGATCCA GAGAAGGAAT GAGAAGCT--
downstream-mouse .......... .GA.....G. .C........ .........G .A....G... .......... .........A .......... .......... ........CC
downstream-rat   ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------

downstream-TMU   -------TCT GGGGTCAGGG CAGGGCACCT CTCAGTCAGA CCAGGATGGA TGGTTACCTT GCCATGCAGA ATTTGTTTGT GTGTGATGAT AAGAATCC--
downstream-mouse CACAGCT... ...A..T..T .......... .......... .......... CA........ .T.C....C. .....G--.. .......... .GC.....AA
downstream-rat   ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------

downstream-TMU   -----AAAGC TCTCTTATCC TGAGGATCAG AGACTCTAGG AAGAAGAGGT CAGAGAAAGA GTCTTAGGAA GATGGATGGA GAGGACTCAA AGGATAGCTG
downstream-mouse TGGAA..... .......... .......... .......T.- .......... .C...G.... .......... .........G .......... ....C.....
downstream-rat   ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------

downstream-TMU   AAAGGACTGT GATGACACTG GGCTGACAGA GAGTACAAAT ---GTCCCAG GAACTGAAAT TGCCCTCACA GGAAGGCAGA GGCCACAAGA GGACAGAGGT
downstream-mouse .GCAA..... .....AG... .......... ......T... GAA....... ..G....... ....T...T. A...A..... ...T...... ..C.....A.
downstream-rat   ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------

downstream-TMU   CTCAGGTTAC TCAGCTAACC ATTAGCCT-- -----GTGTT CAAAGAGTGT GTGTGCTTCC ATTTGAATGT CTGCAGAGGA GCATAAGAAA AATGAGGCAG
downstream-mouse ..-....C.. .T.AG..... ........AT GTAAT...C. ..G....... .......... .........A T......... A......... .........
downstream-rat   ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------

downstream-TMU   AGAGGCAGAC AGACTCAATT TTATCAGGCT GACAATGGGT TATTGGAACA TCAGTGAGAG GCCTCAATAT TTTGG----A CAAAGGTTAA GTGCACTTAA
downstream-mouse .......... ........A. .C........ ....G...T. .......... G......... .TG....... .....CAGC. TG...C.... ...T......
downstream-rat   ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------

downstream-TMU   ATGTCAAGAG ACAGGCTGAC TCCATGACAA GCTTCAAATA GGCAGTCCAG GGGATTATGC CTAAAAACCC AGGGCTAAAG CACCTCTTCA GAAACTGCCC
downstream-mouse ....G.T... .......... .........G .......... ......T... .......... .......... ...C..T... ..A..AA... C...A..TT.
downstream-rat   ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------

downstream-TMU   CAACACCTGG CCTGAAGTAT GGACAATCGA TCAGCAACAG TTTCCAGCCC TCATACCCAG GTAACAGTTC AGAATGTCCC TAAGAGAGAT AGCCACCTAC
downstream-mouse .TC...T... ...A.....A .....G.G.G .......... .......... ...C...... ....------ ---------- ---------- ----------
downstream-rat   ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------

downstream-TMU   CCCTGAAGTA TCCAACAACA GATTCCAGGT CGCATGTCCT GATAATGGTT TCGAATGTCC CTAAGAGATA GAGCAGGATA AGGTCACCTA CCCAGGAATT
downstream-mouse ---------- ---------- ---------- ----...T.. .--------- --........ ..G....... ...T...... .......... G........C
downstream-rat   ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------

downstream-TMU   CCCCTAATGT GCTTTAAATC AGGCCTGCTA GCTCACTCTG GCATCCATTT AA--AAATGG GAGACCCCAG CATGCAGGAC TTCTGCAGAA TGAAACAACC
downstream-mouse ...TG.G..C A.A....... ......---. A......... .TG..TC.C. .TCTT..... ....T..... T......... ..TG.G.... ..........
downstream-rat   ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------

downstream-TMU   TTTGCATTTA CTTGCTATTT ---------- -ATCATCCTT CAGTGAACCA TGGCTCCTTA CATTTAGGTG CTCATCCTGT GATTCACTGG AGAACCCCTG
downstream-mouse .....G.... .A.A..G... GAGTCTGGGG T.....T... .......T.. ...AC..... .T...G.... .....T.... ........CT ..T.....CC
downstream-rat   ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------

downstream-TMU   ACACTTATAG TGGAGGGTT- --TTTATAGT GGATCAGTAA ATCCAGGTGC CTGGCTGTCT TTTATCT--- ---TATCTAT GTTTCTATTT CTTGGTTAAT
downstream-mouse C.C.CA.CCC CC...A..GG AGGC.T.GTC T...TG.... G......... ....T.T.G. ..C....TTT CCA....... .......... ..CC......
downstream-rat   ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------

downstream-TMU   TCTGTTTCAG TTTTAATCTG AAGGCCAAGA AGGGCAAGGC CGGCTCATAT TTTGTTACCT GGCCAGGGCT AATGGATGAC TCCCCACCCC CACCTCCACA
downstream-mouse ........C. ........A. .......... G.AT...... ..A......- ---.......  ........G. ......A... ..T..----- ----------
downstream-rat   ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------

downstream-TMU   TCCCCCACAC CCCCATTCCC TACCAGAGGA AAGAGAACTT GCTAGTTCTC ATCTGGATCC TGAACTGGCT GTGAGTCTAT GGGTCTCCCA TGAGCAGGGC
downstream-mouse ---------- -.AG...... .......A.C .G........ .......... .....A.... .......... ...G...... ...C...... .........A
downstream-rat   ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------

downstream-TMU   AGAAGACTGT CTGAGACAGC CTTTTGGTTT TGGTTT---- --TCTGGAAA TGTCCTTCTG TGCCTGTCTG TGTGTATTCT GTTTGTGCTT GT-GTCCTCT
downstream-mouse G......CA. G.....T... .......... ......TGGT TT.....G.. .......T.. ..----.T.. ........T. .......... T.T.......
downstream-rat   ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ---------- ----------

downstream-TMU   TGTACAAATT TTTCTTTT-- --ATACCTAC AATGGGTCAG GCATAAAACA CCCCACTGGG CCTG
downstream-mouse .......T.. ...TC...TT TA..T....G G....AA-.. .......G.. .TG......A ....
downstream-rat   ---------- ---------- ---------- ---------- ---------- ---------- ----
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Table. S1 Accession number list of the nucleotide sequences used in this study.

Genes	Accession No.				
	T. muenninki	*M. musculus*	*R. norvegicus*	*T. osimensis*	*A. sylvaticus*
ZFY	LC066211	NM_009570	-	E14275	-
UBA1Y	LC066205	NM_011667	XM_008773685	-	-
EIF2S3Y	LC066199	NM_012011	NM_001167666	AB441819	-
TSPY	LC066203	NR_027507	NM_022923	-	AF205112
UTY	LC066209	NM_009484	XM_008773696	-	-
DDX3Y	LC066197	NM_012008	NM_001167665	-	-
USP9Y	LC066207	NM 148943	XM 008773701	-	-
RBMY	LC066201	NM_001166384	GATN01000006	-	-
ZFX	LC066212	NM_001044386	-	-	-
UBA1	LC066206	NM_001136085	NM_001014080	-	-
EIF2S3X	LC066200	NM_012010	NM_001100542	-	-
TSPYL2	LC066204	NM_029836	NM_001191618	-	-
KDM6A/UTX	LC066210	NM_009483	XM_008773047	-	-
DDX3X	LC066198	NM_010028	NM_001108246	-	-
USP9X	LC066208	NM_009481	NM_001135893	-	-
RBMX	LC066202	NM_011252	NM_001025663	-	-

Table. S2 Primer pairs for RT-PCR and qPCR.

Genes		No.	Primers for RT-PCR	Primers for qPCR
ZFY	*, **	1	GTCAGGAGCTGACTTAGTAAGAA	AAGTCATCACTGATCCTCTGACA
		2	GCTTCAAAGTCAACATCCCTA	CCAGAGGCATCCCACTAGAA
UBA1Y	*, **	1	GGAGGTTGGAGAAATCACAG	TGGAGAGGACACTGCAGCTA
		2	AGGTCTTAGGAACAAGGAGGT	ACAGTCGGCCCAAGTCTGT
EIF2S3Y	*, **	1	CTTTCGTGAGCGAGATGGC	TCTGAACACCTAGCAGCCATT
		2	CCTGGTTCCAGCCAAATGT	TGTTCATACTGTTCTTTAGCCTGA
TSPY	*, **	1	GGAAAAGGACCCAGAGCAG	GGAGGAACTGCAGAAAGCAG
		2	GCTCTTTATCCTGGCAAGGTC	GGGTTTTAATCCGTGCAAGA
UTY	*	1	ACAGCTGCTGCCGTAAAGTT	ATGTGGACCATGACACTGGA
		2	CAGAACTTCTGTTGAACTGGAG	AACTGGCTGAAGAGGGTGAA
		3	TACCAGTCCTCACAGTGGAT	—
		4	CAGGCTGAACAAGCAGGAAT	—
DDX3Y	*	1	CAAAGCTGTGTCCGTAAGCA	GTCGTGACCGAACTGGTTTT
		2	AACCAAGCAGGAAGGGAGTT	CTAAGCGTTCACTTGGTGGA
USP9Y		1	GGTGGTCACTTTGTGGTGTC	AGAGAGGTCGCTGGTTCAAA
		2	AGCATAAGTGAGAAGACGCCT	ACTCTCCACCGAAGCATTGA
	*, **	3	CCAGATGTTGCATGGGACA	—
		4	TTCTTCACTGCTTTCCTGAGC	—
RBMY	*, **	1	GACAAGAAAAATGGCAGAAAC	ATGGAGGCAGACAAGGAAGA
		2	CCTTTAGTACCTGCCTCCAC	GGATCCCCCAAATCTTGAAC
SRY			—	AACCCACATGCCATCACATA
			—	TGTCCACAGGCTGCAAATAA
ZFX	**	1	GCGTATAACTGTGCCCAGC	—
		2	GAGTACCATCAAGGTAAGCAAGG	—
UBA1X	**	1	GAGGACAAAGAGGCTCTGACA	—
		2	ACTCCTTCCTTCCCTGGAGA	—
ACTB	*	1	GGCATCCTGACCCTGAAGTA	—
		2	TCTCAGCTGTGGTGGTGAAG	—

*The primer pairs used for RT-PCR using multiple tissues to reveal the expression profiles.
**The primer pairs used for FISH analysis.

A

Table. S3 Sequence homology of *T. muenninki* X-linked genes with the orthologues of mouse

Gene name	Homology of nucleotide with mouse (%)	Homology of amino acid with mouse (%)
ZFX	94.6	95.4
UBA1	96.2	97.6
EIF2S3X	96.2	99.4
TSPLY2	94.9	93.4
KDM6A/UTX	97.6	96.2
DDX3X	97.2	99.4
USP9X	97.6	99.4
RBMX	91.8	96.9