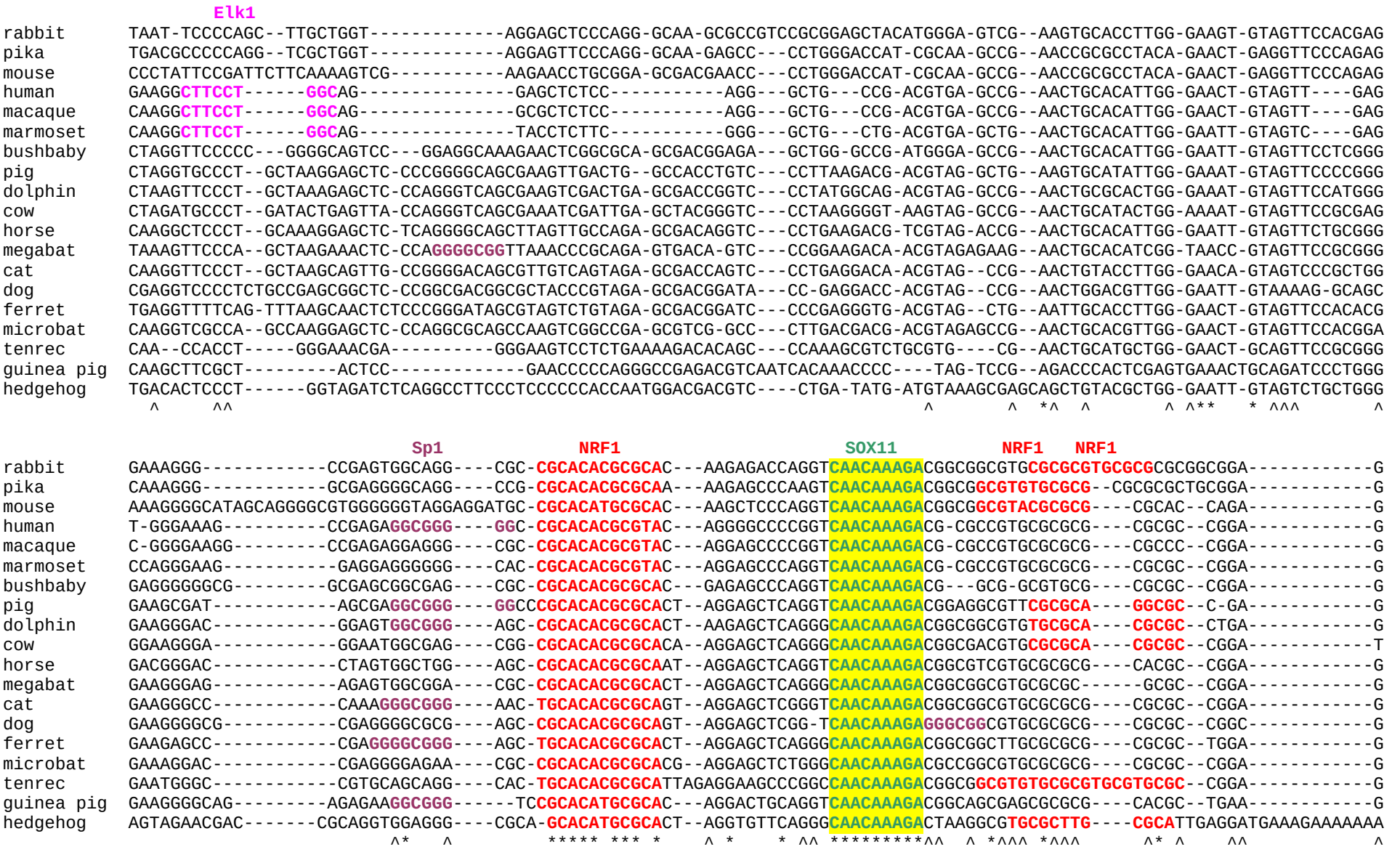
Figure S1. Evolutionary conservation of the *SPAST* promoter in mammals identifies putative transcription factor (TF) binding sites. A) Multisequence alignment for 19 eutherian mammalian species of the full-length *SPAST* promoter region into exon 1 and including the first translational start codon (a segment of 474-nt in human). In addition, *Spast* promoter-exon 1 sequence alignments are shown for **B)** tenrec and elephant, and **C)** western European hedgehog and a marsupial, *Monodelphis domestica* (the tenrec and hedgehog sequences are also shown in **A**). Despite a high degree of nucleotide conservation, elephant is the only sequenced mammalian genome in which the SOX11 motif has been lost and replaced by duplications (shown by gray shading) of putative NRF1 binding site elements. Sequences were aligned using ClustalW 2.1 and manually adjusted as needed for maximum parsimony. The putative TF binding sites identified in this study are indicated in bold type (red, NRF1; green, SOX11; purple, Sp1); a poorly conserved putative site for Elk1 (Canbaz et al. 2011) is also shown (pink); yellow shading, highly conserved SOX11 motif; gray shade (human sequence), upstream transcription start site (TSS); INIT, translation start site for the 68 kDa spastin isoform; *, nucleotide positions conserved in all aligned sequences; ^, nucleotide positions conserved in 90% (17 of 19) of aligned sequences.

A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Sp1
NRF1
SOX11
NRF1
NRF1
rabbit
pika
mouse
human
macaque
marmoset
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pig
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cow
horse
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cat
dog
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tenrec
guinea pig
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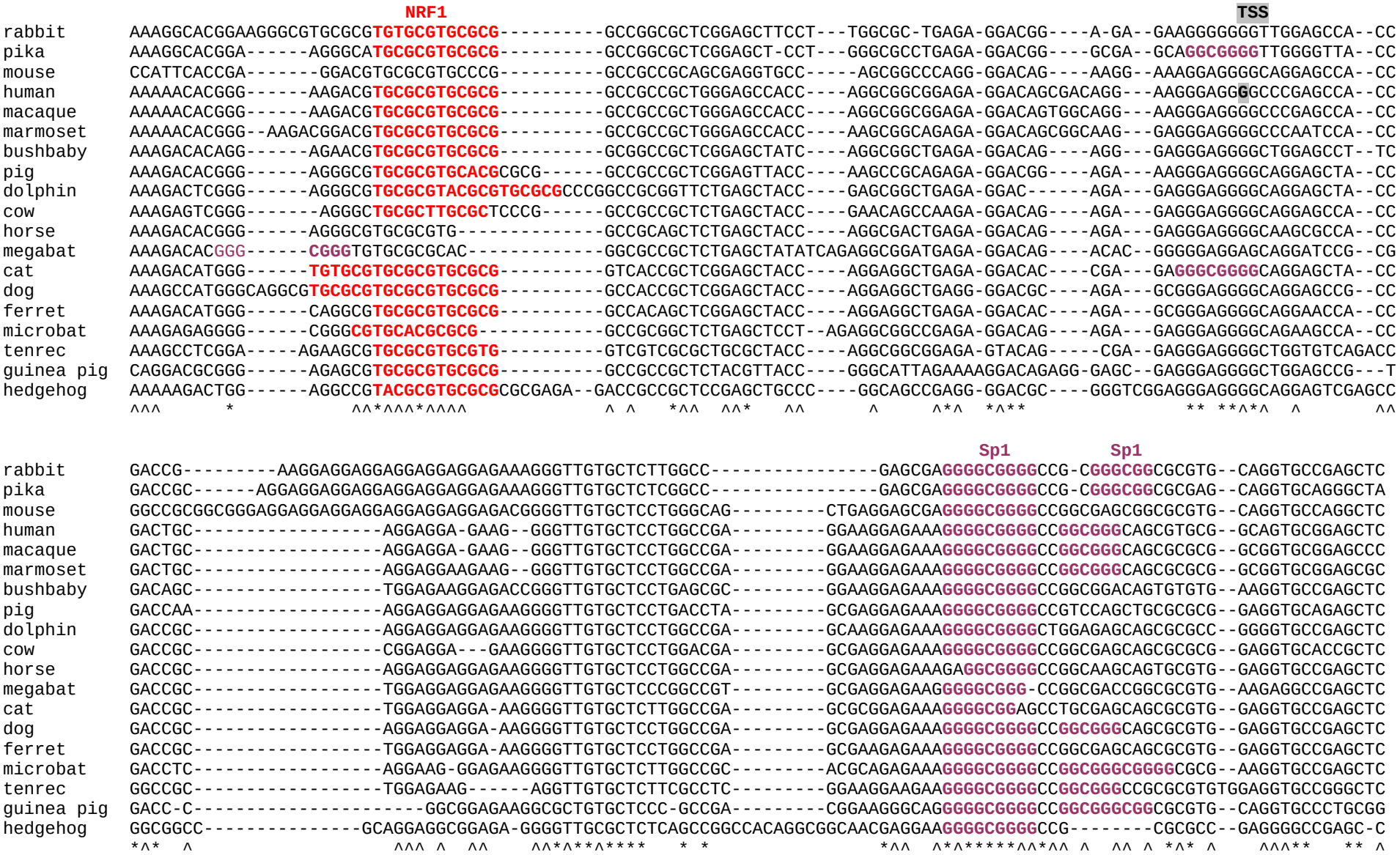
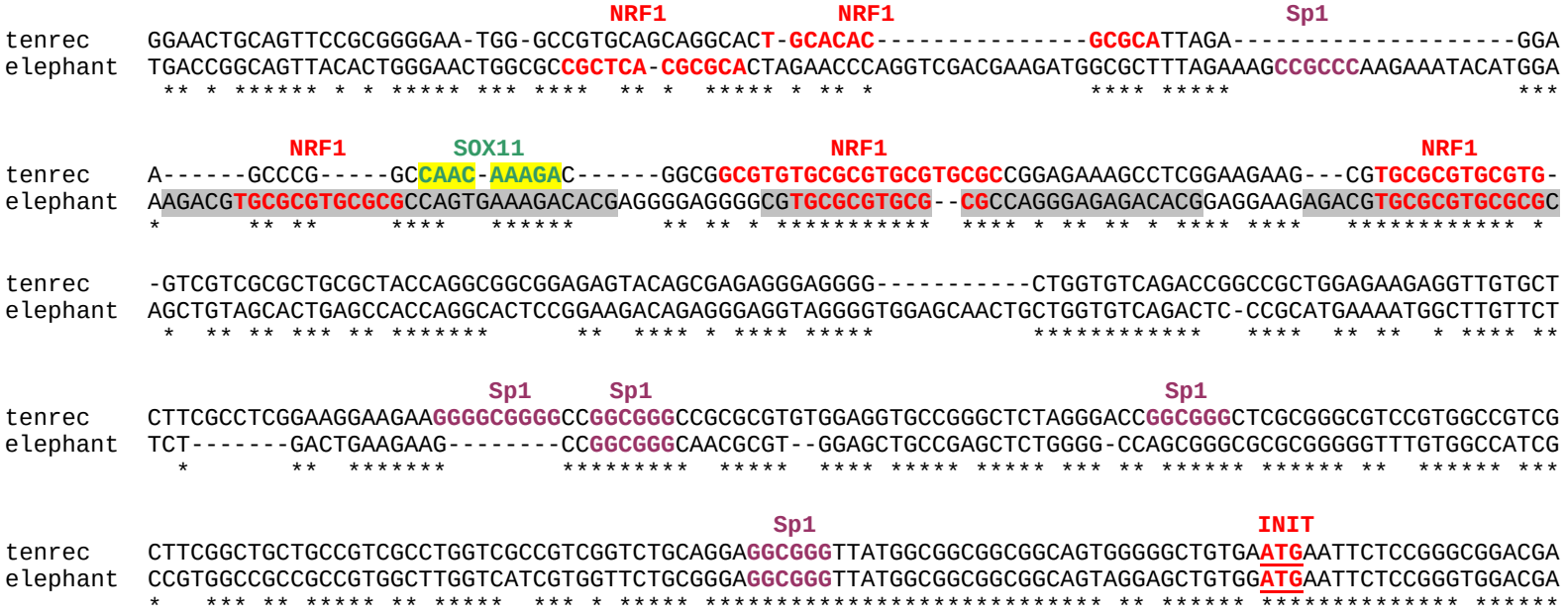

NRF1
TSS
Sp1
Sp1
rabbit
pika
mouse
human
macaque
marmoset
bushbaby
pig
dolphin
cow
horse
megabat
cat
dog
ferret
microbat
tenrec
guinea pig
hedgehog

```
                  Sp1                                 Sp1
rabbit      CTGGGACCCG-C-CGGCGTGCGGG-GGGTCT-GTGG-CCCTCGCCGTCGAAGCA---GTCGCCGCCGTCGCTG------GGTCG
pika        CCGGGACCCG-C-CGGCGCGCGAG-GG-TCC-TTGG-CCCTCGCCGTCGCAGCAA--GTGGCCGCCGTCGCTG------GGTCG
mouse       CCGGGCTCCGGC-TGGCGCGCGAGCGGCTCC-GTGGGCCCCCGCCGCCGCAGTGGCAGTGGCCGCCGCCGCCGCCGCTTGGTCG
human       CTGAGA-CCGGC-GGGCACACGGG--GGTCT-GTGG-CCCCCGCCGTAGCAG------TGGCTGCCGCCGTCG---CTTGGTT-
macaque     CTGAGA-CCGGC-GGGCACACGGG--GGTCT-GTGG-CCCTCGCCGTCGCAG------TGGCCGCCGCCGTCG---CTTGGTT-
marmoset    CGGAGG-CCGGC-GGACAGGCGGG--GGTCT-GTGG-CCCTAGCCGTCGCAG------TGGCCGCTGCTGTCG---CTTGGTTG
bushbaby    TCGCGG-CCGGC-CGGCGCGCGGG--CGTGT-GTGG-CCCTAGCCGTCGCAG------TGGCCGTCGCCGTCC---TTTGGTC-
pig         CTGGGA-CCGGC-GGGCGCGCGGG--GGTCTCGTGG-CCCTTGCCCTCCCAG------TAGCCGCCGCCGTCG---CTTGACCG
dolphin     CTGGGA-CCGGC-GGGCGCGTGGG--GGTCT-GTGG-CCCTTGCCGTCCCAG------TGGCCGCCGCCGTCG---CTTGGCTG
cow         CTGGGA-CCGG--GGCCGCCCGGG--GGTCT-GTGG-CCCTTGCCGTCCCAG------TGGCTGCCGCCGTCG---CTTGGCCG
horse       CTGGGA-CCGGC-GGGCGCGCGGG--GGTCT-GTGG-CCCTCGCCGTCCCAG------TGGCCGCCGCCGTCG---CTTGGTCG
megabat     CCGGGG-CCGGC-CGGCGCGCGGT--AGTCT-GTGG-CCCTCGCTGTCCCAG------TATCCGCTGCCGTCG---CTTGGTCG
cat         CTGGGA-CCGGC-GGGCGCGCGGG--CGTCT-GTGG-CCCTCGCCGTCCCAG------TGGCCGCCGCCGTCG---CTTGGTCG
dog         CTGGGC-CCGGC-GGGCGCGCGGG--CGTCT-GTGG-CCCTCGCCGTCCCAG------TGGCCGCCGCCGTC----CTTGGTCG
ferret      TCGGAA-C-GGC-GGACGCGCGGG--CGTCT-GCGG-CCCTCGCTGTCCCAG------TGGCCGCCGCCGTGG---CTTGGTCG
microbat    CCGGGA-CCGGC-CGGCGCGCGGG--GCTCT-GCGG-CCCTCGCCGTCCCAG------TGGCCGCTGCCGTCA---TTTGGTCG
tenrec      TAGGGA-CCGGC-GGGCTCGCGGG--CGTCC-GTGG-CCGTCGCT-TC-----------GGCTGCTGCCGTCG---CCTGGTCG
guinea pig  CTGAGC----------CCCGCGGG--GGTCC-GTGG-CCCTCGC-GTGGCAG------TGGCCGCCGCTGAAG---CTTGGTCG
hedgehog    CCGGGA-CCGGCGGGGGGCGCGGG-------------CCCTCGTCGTGCCGG-------GGCCACCACCGCCG---CTCGGTGG
              * ^  ^^^ ^  ^ ^ ^ ^*^^    ^^  ^ ^^ **^^ *^ ^^  ^^^      ^ ^* ^^ ^^^*         *^

                                  Sp1                                         INIT
rabbit      ----CCGTCGGTCTGCGGA-AGGCGGGTTATGGCGGCGGCGGCGGC---AGTGGGAGCTGTGAATGAATTCTCCGGGTGGACGA
pika        ----CCTTGGGTCTGCGGG-AGGCGGGTTATGGCGGCGGCGGCGGCGGCAGTGGAAGCCGTGAATGAATTCTCCGGGTGGACGA
mouse       ----CCGTCGGTCTGCGGG-AAGCGGGTTATGGCGGCGGCGGC------AGTGGGAGCTGTGAATGAGTTCTCCGGCCGGACGA
human       ---CCCGTCGGTCTGCGGG-AGGCGGGTTATGGCGGCGGCGGC------AGTGAGAGCTGTGAATGAATTCTCCGGGTGGACGA
macaque     ---CCCATCGGTCTGCGGG-AGGCGGGTTATGGCGGCGGCGGC------AGTGAGAGCTGTGAATGAATTCTCCGGGTGGACGA
marmoset    GTTCCCGTTGGTCTGCGGG-AGGCGGGTTATGGCGGCGGCGGC------AGTGAGAGCTGTGAATGAATTCTCCGGGTGGACGA
bushbaby    ---CCCGTCGGTCTGCGGG-AGGCGGGTTATGGCGGCGGCGGC------AGTGGGAGTTGTGAATGAATTCTCCGGGTGGACGA
pig         ----CCGTCGGTCTGCGGA-AGGCGGGTTATGGCGGCGGCGGC------AGTGGGAGCTGTGAATGAATTCTCCGGGTGGACGA
dolphin     ----CCGTCGGTCTGCGGG-AGGCGGGTTATGGCGGCGGCGGC------AGTGGGAGCTGTGAATGAGTTCTCCGGGTGGACGA
cow         ----CCGTCGGTCTGCGGG-AGGCGGGTTATGGCGGCGGCGGC------AGTGGGAGCTGTGAATGAATTCTCCGGGTGGACGA
horse       ----CCGTCGGTCTGCGGGGAGGCGGGTTATGGCGGCGGCGGC------AGTGGGAGCTGTGAATGAATTCTCCGGGAGGACGA
megabat     ----CCGTCGGTCTGCGGA-AGGCGGGTTATGGCGGCGGCGGC------AGTGGGAGCTGTGAATGAATTCTCCGGGTGGACGA
cat         ----CCGTCG-TCTGCGGG-AGGCGGGTTATGGCGGCGGCGGC------AGTGGGAGCTGTGAATGAATTCTCCGGGTGGACGA
dog         ----CCGTCGGTCTGCGGG-AGGCGGGTTATGGCGGCGGCGGC------AGTGGGAGCTGTGAATGAGTTCTCCGGGTGGACGA
ferret      ----CCGTCGGTCTGCGGG-AGGCGGGTTATGGCGGCGGCGGC------AGTGGGAGCTGTGAATGAATTCTCCGGGTGGACGA
microbat    ----CCGTCGGTGTGCGGG-AGGCGGGTTATGGCGGCGGCGGC------AGTGGGAGCTGTGGATGAATTCTCCGGGTGGACGA
tenrec      ----CCGTCGGTCTGCAGG-AGGCGGGTTATGGCGGCGGCGGC------AGTGGGGGCTGTGAATGAATTCTCCGGGCGGACGA
guinea pig  ----CCGTCGGTCTGAGGG-AGGCAGGTTATGGCGACGGCGGC------AGTGGGAGCTGTGAATGAGTTCTCCGGGTGGACGA
hedgehog    ----CCCTGGCTCT---------CGGGTTATGGCGGCGGCGGC------AGCGGGGGCTGTGAATGAATTCTCCCGGCCGACGA
                ** * *^*^*^^^^  ^^^*^**********^*******      **^* ^^*^^^**^**** ******^*^ ^*****
```

B


NRF1
NRF1
Sp1
tenrec GGAACTGCAGTTCCGCGGGGAA-TGG-GCCGTGCAGCAGGCACT-GCACAC----------GCGCATTAGA----------GGA
elephant TGACCGGCAGTTACACTGGGAACTGGCGCCGCTCA-CGCGCACTAGAACCCAGGTCGACGAAGATGGCGCTTTAGAAAGCCGCCCAAGAAATACATGGA
NRF1
SOX11
NRF1
NRF1
tenrec A------GCCCG-----GCCAAC-AAAGAC------GGCGGCGTGTGCGCGTGCGTGCGCCGGAGAAAGCCTCGGAAGAAG---CGTGCGCGTGCGTG-
elephant AAGACGTGCGCGTGCGCGCCAGTGAAAGACACGAGGGGAGGGGCGTGCGCGTGCG--CGCCAGGGAGAGACACGGAGGAAGAGACGTGCGCGTGCGCGC
tenrec -GTCGTCGCGCTGCGCTACCAGGCGGCGGAGAGTACAGCGAGAGGGAGGGG----------CTGGTGTCAGACCGGCCGCTGGAGAAGAGGTTGTGCT
elephant AGCTGTAGCACTGAGCCACCAGGCACTCCGGAAGACAGAGGGAGGTAGGGGTGGAGCAACTGCTGGTGTCAGACTC-CCGCATGAAAATGGCTTGTTCT
Sp1
Sp1
Sp1
tenrec CTTCGCCTCGGAAGGAAGAAGGGGCGGGGCCGGCGGGCCGCGCGTGTGGAGGTGCCGGGCTCTAGGGACCGGCGGGCTCGCGGGCGTCCGTGGCCGTCG
elephant TCT-------GACTGAAGAAG--------CCGGCGGGCAACGCGT--GGAGCTGCCGAGCTCTGGGG-CCAGCGGGCGCGCGGGGGTTTGTGGCCATCG
Sp1
INIT
tenrec CTTCGGCTGCTGCCGTCGCCTGGTCGCCGTCGGTCTGCAGGAGGCGGGTTATGGCGGCGGCGGCAGTGGGGGCTGTGAATGAATTCTCCGGGCGGACGA
elephant CCGTGGCCGCCGCCGTGGCTTGGTCATCGTGGTTCTGCGGGAGGCGGGTTATGGCGGCGGCGGCAGTAGGAGCTGTGGATGAATTCTCCGGGTGGACGA

C


NRF1
SOX11
NRF1
hedgehog GCAGCACATGCGCACTA--GGTGTTCAGGGCAACAAAGACTAAGGCGTGC----------GCT----------TGCGCAT-------
Monodelphis GCTGCACAAGCGCACCGCGGGAGCTCCTGTCAACAAAGACCGAGGCACGCTCGTGAGCAACGGAGGCTAAGATCCTATGGAACGTGCGCGTGTCCGCC
NRF1
hedgehog ----TGAGG------ATGAAAGAAAAAAAAAAAAGACTGGAGGCCGTACGCGTGCGCGCGCGAGAGACCGCCGCTCCGAGC---TGCCCGGCAGCCGA
Monodelphis TCGCTGAGGCAGCCTATGAAAGGAGAAAGAAAG-GACAGGA---------CGTGCGCGCGACTGTC-CCATTGC-CTGAGCCTATGGCCGTGAGA-GA
Sp1
hedgehog GGGGACGCGGGTCGGAGGGAGGGGCAGGAGTCGAGC--CGGCGGCCGC--AGGAGG----CGGAGAGGGGTTGCGCTCTCAGCCGGCCACAGGCGGCAA
Monodelphis GGAGG-GCGGGGCGGGACCAGGA-CAGATCCCGGACATCAAAACCTGCTCGGGAGGGTTTCGGAGAGGGG--GAGGTGAC-----GCTTCGGACCG---
Sp1
hedgehog CGAGGAAGGGGCGGGGCCGCGCGCCGAGG-GGCCG-AGCCCCGGGA------CCGGCGG-----------GGGGCGCGGGCC--------CTCGTCGT
Monodelphis -AGGGAAGGGGCGGGGCCGGGTGTGGAGGTAGCTGGAGCTCTGAGGACTCGCCCGACTGCTACCCAGCTTTGGTGCTTTGCCCTTCTCCTCCTCCTCTT
INIT
hedgehog GCCGG-GGC-----CACCACCGCCGCTCGGTGGCCCTGGCTCTCGGGTTATGGCGGCGGCGGCAGCGGGGGCTGTGAATGAATTCTCCCGGCCGACGA
Monodelphis GCCTGCAGCACCAGCACCACCGCCGCTTGGTCG--CTGACAG-CGGGTTATGGCGGCGGCGGCAGTGGGAGCCGTGAATGAATTCTCCGGGCGGCCGA