

1   **Supplemental figure legends**

2   **Supplemental Fig. S1.** Nucleotide sequence and deduced amino acid sequence of  
3   FOXL2 in *E. sinensis* (Es-FOXL2). The underlined sequences indicate the forkhead  
4   domain of the FOX family. The initiation and stop codons are in bold.

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6   **Supplemental Fig. S2.** Multiple alignment of FOXL2 amino acid sequences from *E.*  
7   *sinensis* and other representative invertebrates and vertebrates. The sequence  
8   alignment was performed with software ClustalX software. The forkhead domain and  
9   the alanine polymer region are marked in red boxes. The green boxes indicate the  
10   nuclear localization signal. The sequences were downloaded from NCBI with  
11   following accession numbers: *Homo sapiens*: P58012, *Mus musculus*: Q2TVT7,  
12   *Gallus gallus*: Q5J7N5, *Rattus norvegicus*: D4A0S1, *Bos Taurus*: Q6VFT7,  
13   *Oryctolagus cuniculus*: Q6VFT5, *Xenopus tropicalis*: F7E8K4, *Danio rerio*:  
14   NP\_001038717.1, *Strongylocentrotus purpuratus*: ABB89483.1.

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16   **Supplemental Fig. S3.** Phylogenetic analysis of Es-FOXL2 and FOXL2 homologs  
17   from other invertebrates and vertebrates. The phylogenetic tree was constructed using  
18   the neighbor-joining method in Mega 4.0 software. Numbers in the phylogram nodes  
19   indicate percent bootstrap, and the bar at the bottom indicates 5% amino acid  
20   divergence in sequence.

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22   **Supplemental Fig. S4.** Nucleotide and deduced amino acid sequence of FTZ-F1 in *E.*  
23   *sinensis* (Es-FTZ-F1). The DNA binding domain and the ligand binding domain are  
24   shown in bold and italicized, respectively. The two zinc fingers (amino acid residues  
25   33–53 for ZF1 and 69–88 for ZF2) are boxed. The underlined region is the FTZ-F1  
26   box. The AF2 core is shown in a bold box.

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28   **Supplemental Fig. S5.** Phylogenetic analysis of Es-FTZ-F1 and other members of the  
29   NR5As subfamily. The phylogenetic tree was constructed using the neighbor-joining  
30   method in Mega 4.0 software. Numbers in the phylogram nodes indicate percent  
31   bootstrap, and the bar at the bottom indicates 10% amino acid divergence in sequence.

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33   **Supplemental Fig. S6.** Particle Nucleotide and deduced amino acid sequence of  
34   DDX20 in *E. sinensis*.

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36   **Supplemental Fig. S7.** Analysis of recombinant forkhead domain protein. (A)  
37   Recombinant plasmid amplified by PCR; M, marker; RP, recombinant pET-32a-EsFH  
38   plasmid; D, plasmid digested by *Eco*RI and *Xho*I restriction enzymes. The band at ~5  
39   kb is the empty plasmid, and the lower band is the forkhead domain sequence (not  
40   observed due to low concentration). (B) Recombinant forkhead domain protein  
41   analysed by Coomassie Blue staining. M, marker; Un, lysates of BL21 cells with  
42   recombinant pET-32a-EsFH plasmid; In, BL21 cells with recombinant pET-32a-EsFH  
43   plasmid induced with IPTG. (C) Forkhead domain protein analysed by western blot.  
44   FH, forkhead domain protein; EP, empty plasmid.

45 **Supplemental figures**

## 46 Supplemental Fig. S1.

1	10	20	30	40	50	60
1	GAGTACATGAGCCCTTCGCCTACGAGAGTCATGACATAAAATCCTACGACAAAATGTAC					
	M S P S A Y E S H D I K S Y D K M Y					
	70 80 90 100 110 120					
61	TCGTCGAAGGCCACTGGAGTCTCGATTGATGACTCTCGTATGACAAGTGCTGCGAC					
19	S S K S H L E S P I D D S S Y D K C C D					
	130 140 150 160 170 180					
121	AAGACCTACAAGACCCTCGAGACGCTGGACCCATAACAAGAACGCCCTATTCTACGTG					
39	K T Y K T L E T L D P N K K P P Y S Y V					
	190 200 210 220 230 240					
181	GCGCTTATCACCATGGCGATCAAGGAGAGGCCGGAGCGGAGACTGCAGCTGAGCAGATC					
59	<u>A L I T M A I K E S P E R R L Q L S E I</u>					
	250 260 270 280 290 300					
241	TACCACTGGATCGCCAACAAGTCCCTTCTACGCCAAGGAGAGCGCCAAGGAGAACAG					
79	<u>Y Q W I A N K F P F Y A K E S A K E K Q</u>					
	310 320 330 340 350 360					
301	GGCTGGAAGAACTCGATCCGGCACAAACCTGAGTCTGAACGAATGTTCCAGAACAGCCC					
99	<u>G W K N S I R H N L S L N E C F Q K Q P</u>					
	370 380 390 400 410 420					
361	CGGGATGGCGGGCGGGAGGAAAGGGCAACTACTGGACGCTAGACCCCCAGCACGAG					
119	<u>R D G G G G G K G N Y W T L D P Q H E</u>					
	430 440 450 460 470 480					
421	AACATGTTGAGAATGGCAACTTCACCGCGGCCGACGGATGAGACGCGCGCTAACCTC					
139	<u>N M F E N G N F T R R R R M R R A A N L</u>					
	490 500 510 520 530 540					
481	CTGCGACAGCCCTACCCCTCCCTATCCCATCTTCAGATCTCCCCGAGCAGCAGCTGGGG					
159	<u>L R Q P Y P P Y P I F Q I S P S S S W G</u>					
	550 560 570 580 590 600					
541	CTCGGCCAAATCCAAGCGGGAACTCGCAGCTATACACAGGGCACGCGCATGCACACC					
179	<u>L G Q I Q G G N F A S Y T Q G T R M H T</u>					
	610 620 630 640 650 660					
601	CCGCACTCTACACCTACCCGAGATGAACAGCAGCTTCAGGGCAGATGCGAGCTCGCGGC					
199	<u>P H S Y T Y P Q M N Q L Q G Q M Q L G G</u>					
	670 680 690 700 710 720					
661	GGCTACCACTGGCGCTCCCTGGGACGGCGCGCTCACTTCAGGCGCTTGGGG					
219	<u>G Y Q Q I G G S L G T A P L T S G A L G</u>					
	730 740 750 760 770 780					
721	TCAAGCTCTCCAGTCACCTGGGGGGACTCTGGGAGCTCCCTCGCCGCCATTTCG					
239	<u>S S F S S H L G G L L G S S S P A I S</u>					
	790 800 810 820 830 840					
781	TCCCCGAGCAGCTCCCACACCTCCCTCGGCACCGCGCCTGAACACTCTGGACATCGGGTCG					
259	<u>S P S S S H T S L G T A P E L W T S G S</u>					
	850 860 870 880 890 900					
841	AGCACAAGCCTTCACTCTAGCATCGGAGCCAGTAGTTTTAAAGCCCCAGCAGTACTTTA					
279	<u>S T S L H S S I G A S S F L S P S S T L</u>					
	910 920 930 940 950 960					
901	GGATCGCCTCTACTACACCCCTCCAGCCTCTTCGGTAGGGCAGCGGCAGTACGGC					
299	<u>G S P S T T P F Q P S F G S G S G S Y G</u>					
	970 980 990 1000 1010 1020					
961	CGGGGGCTGAACCGCGTGCCTGGCGACAGGGAGGGCACACACCTCCAGCTTGGCC					
319	<u>A G L N A C R R Q G E A T T S S Q L A P</u>					
	1030 1040 1050					
1021	CTCTCCTACTACGGCTGGACGGATTCCAAGCCCTAA					
339	<u>L S Y Y G W T D S K P *</u>					
	CAGATTTGGACCGCGACGCCCTGGCTTGACCGGGCGGGCGAGGGGGTGAACCCACAGCACCTGATGA					
	CTGACGGGACTGACCACTCAGCGCTGGCGGAGACTGACCAAGCCACACACCTTAACTACTGTT					
	ATGAAAAGCAGATGTAGGACCTCCGACTACCAAAGGAGCGACGCCAATCTGGACAAGTAACCGAAAGGAT					
	CGTAATCTGGGAAGTCAAAGTAAATCATGTGCTGCGAAAATCAAATAACGAGCTGGACGACGGCGAA					

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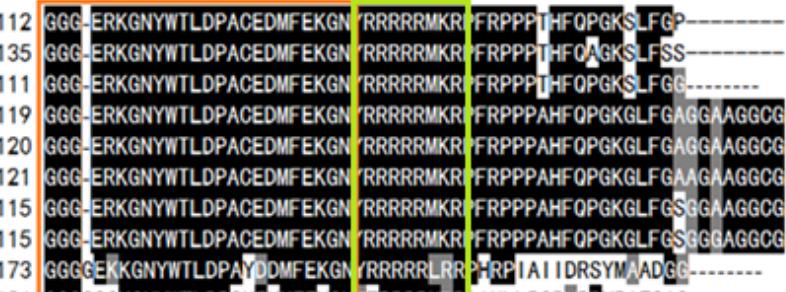
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54 Supplemental Fig. S2.

Gallus	6	ADGEEDAVAILAHGCGGSKEPERGKEELSAEKG-----PEKPDFSQKPPYSYVAL
Xenopus	29	QSPEOGTVALI THNSNGNKEAERSKEDLLPEKG-----QEKPDFSQKPPYSYVAL
Danio	6	PGHEDNGMILLDTTSS-SAEKDRTKDEAPPEKG-----PKSDPTOKPPYSYVAL
Homo	6	PEPEDAAGALLAPEIGRTVKEPEEGPPP-SPGKGGGGGG-----TAPEKPDFAQKPPYSYVAL
Bos	6	PEPENASGALLAPEIGRAAKEPEAPPSPGKGGGGGTG-----TAPEKPDFAQKPPYSYVAL
Oryctolagus	6	PEPEEAAGALLAPEGRAAKEPEAPP-SPGKGGGGGGGGSAE-----EKPDPAQKPPYSYVAL
Mus	6	PEPEDTAGTLLAPESGRAVKEAEASPP-SPGKGGG-----TPEKPDFAQKPPYSYVAL
Rattus	6	PEPEDTAGTLLSPESGRAVKEAEASPP-SPGKGGG-----TAPEKPDFAQKPPYSYVAL
Strongylocentrotus	61	AARAEQVSSPRLSSRSVOTDDVADDET[KDDREKKPIN-----DKPDFSQKPPFSYVAL
Eriocheir	5	AYESHDTIKSYDKMYSS[SHLESPIDDDSSYDKCCDKTYKT-----LETLDENKKPPYSYVA
		
Gallus	56	IAMAIRESAEKRLTLSGIYQYIISKFPFYEKN-----KKGWQNSIRNLSQLECFIKVPRE
Xenopus	79	IAMAIRESAEKRLTLSAIYQYIISKFPFYEKN-----KKGWQNSIRNLSQLECFIKVPRE
Danio	55	IAMAIRESEKRLTLSGIYQYIISKFPFYEKN-----KKGWQNSIRNLSQLECFIKVPRE
Homo	63	IAMAIRESAEKRLTLSGIYQYIIAKFPFYEKN-----KKGWQNSIRNLSQLECFIKVPRE
Bos	64	IAMAIRESAEKRLTLSGIYQYIIAKFPFYEKN-----KKGWQNSIRNLSQLECFIKVPRE
Oryctolagus	65	IAMAIRESAEKRLTLSGIYQYIIAKFPFYEKN-----KKGWQNSIRNLSQLECFIKVPRE
Mus	59	IAMAIRESAEKRLTLSGIYQYIIAKFPFYEKN-----KKGWQNSIRNLSQLECFIKVPRE
Rattus	59	IAMAIRESAEKRLTLSGIYQYIIAKFPFYEKN-----KKGWQNSIRNLSQLECFIKVPRE
Strongylocentrotus	117	IAMAIKDSPERKLTLSQIYQYIINKFSTYEKN-----KKGWQNSIRNLSQLECFLKJARE
Eriocheir	61	IAMAIKESPERRLQSEIYQIANKFPFYAKESAKEKGWQNSIRNLSQLNECFDQKOPRD
 		
Gallus	112	GGC-ERKGNYWTLDPACEDMFKEKGNVRRRRRMKRFRPPPTHFQPGKSLFGP-----
Xenopus	135	GGC-ERKGNYWTLDPACEDMFKEKGNVRRRRRMKRFRPPPTHFQAGKSLFSS-----
Danio	111	GGC-ERKGNYWTLDPACEDMFKEKGNVRRRRRMKRFRPPPTHFQPGKSLFGG-----
Homo	119	GGC-ERKGNYWTLDPACEDMFKEKGNVRRRRRMKRFRPPPAHFQPGKGLFGAGGAAGGCG
Bos	120	GGC-ERKGNYWTLDPACEDMFKEKGNVRRRRRMKRFRPPPAHFQPGKGLFGAGGAAGGCG
Oryctolagus	121	GGC-ERKGNYWTLDPACEDMFKEKGNVRRRRRMKRFRPPPAHFQPGKGLFGAAGAAGGCG
Mus	115	GGC-ERKGNYWTLDPACEDMFKEKGNVRRRRRMKRFRPPPAHFQPGKGLFGSGGAAGGCG
Rattus	115	GGC-ERKGNYWTLDPACEDMFKEKGNVRRRRRMKRFRPPPAHFQPGKGLFGSGGAAGGCG
Strongylocentrotus	173	GGGCEKKGNVWTLDPAYDDMFKEKGNVRRRRRLRPRPIAIIDRSYMDGG-----
Eriocheir	121	GGGGGGKGNVWTLDPOHENMFEKGNVRRRRRMRRANLLRQPYPYPPIFQIS-----
Gallus	163	DGYGYLSPPKYLQSTIFNNSWPLPOPPAPVYASCOMGGSVSPVN-----
Xenopus	186	DTGYGYLSPPKYLQSTIFNNSWPLPOPPAPMSYTISCOMAGGNVSPVN-----
Danio	162	EGYGYLSPPKYLQSGFINNSW-----SPAPMSYTISCOVSSESVSPVN-----
Homo	178	VAGAGADGYGYLAPPKYLQSGFLNNSWPLPOPPSPMPYASCOMAAAAAAAGGPG
Bos	179	VAGAGADGYGYLAPPKYLQSGFLNNSWPLPOPPSPMPYASCOMAAAAAAAGGPG
Oryctolagus	180	VAGAGADGYGYLAPPKYLQSGFLNNSWPLPOPPSPMPYASCOMAAAAAAAGGPG
Mus	174	VPGAGADGYGYLAPPKYLQSGFLNNSWPLPOPPSPMPYASCOMAAAAAAAGGPG
Rattus	174	VPGAGADGYGYLAPPKYLQSGFLNNSWPLPOPPSPMPYASCOMAAAAAAAGGPG
Strongylocentrotus	225	-----YPSYPRC10YGSSWICCPPPPPAVPSIHOUP-----
Eriocheir	173	-----PSSSWGLGQI QGENFASYTQGTRMHTPHSVTPYOMNQLMHTPHSYTYP-----

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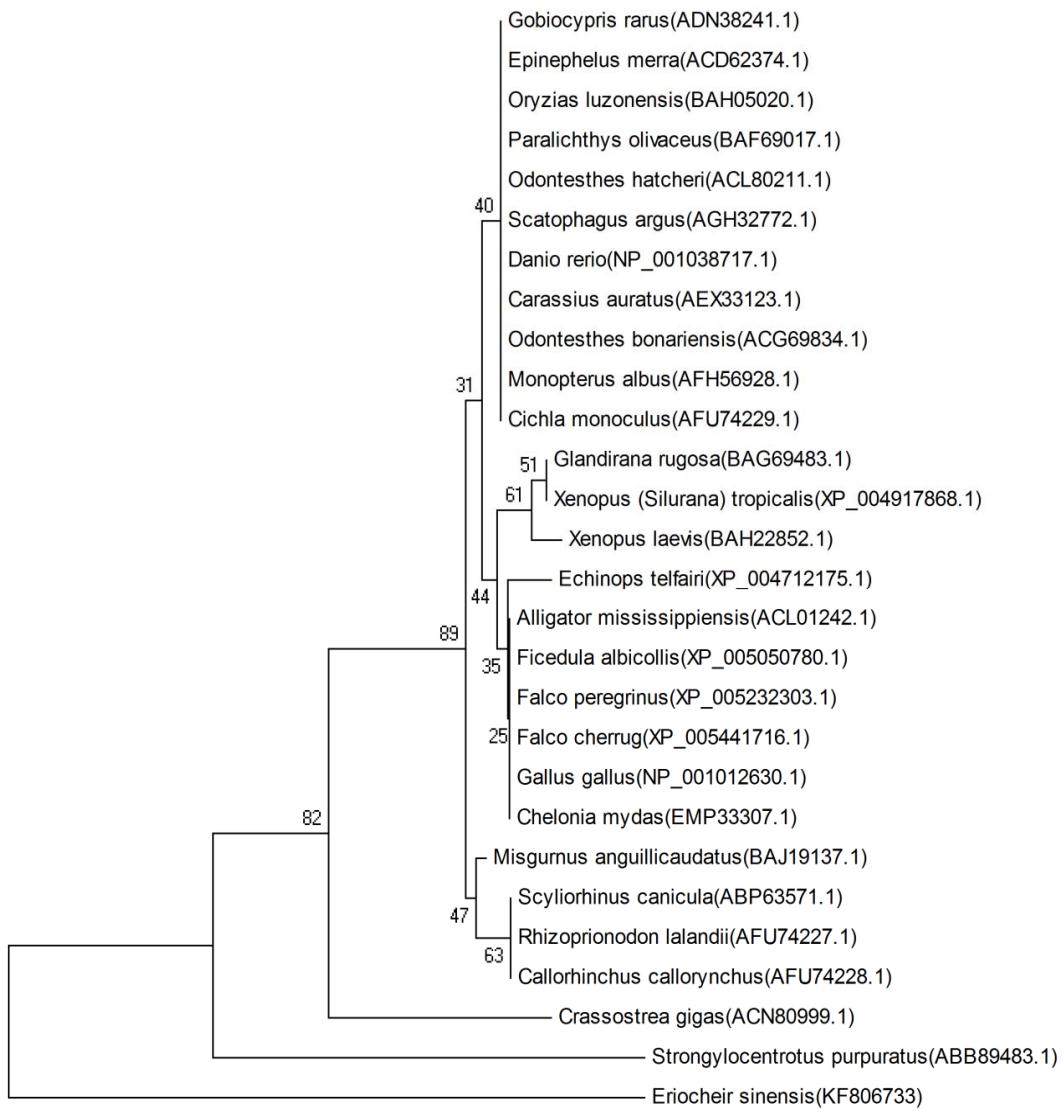
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66 Supplemental Fig. S3.



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## 82 Supplemental Fig. S4.

1 ATGGACTCTGGTCTCTCCGGCGTAGCTACCACCACTTGGACTACACCTCCTCACCG  
 1 M D S G L F P G V A T T T L D Y T S L T  
 61 GACATGCCGACAGAAAGAGGGCATCGAGGAACATGCCCTGTGTGGGACAAGGTG  
 21 D M P D T K E G I E E L C P V C G D K V  
 121 TCGGGCTACCACTACGGCTACTCACTTGTGAATCCTGCAAGGGCTTCTCAAGAGGACG  
 41 S G Y H Y G L L T C E S C K G F F K R T  
 181 GTTCAGAACAGAAGGTTTACGTGTGCGCCGACCGCTCTGTCAAGATTGATAAGACC  
 61 V Q N K K V Y T C V A D R S C Q I D K T  
 241 CAGAGGAAGAGGTGTCCGTACTGCCGCTTCCAGAAAATGCTAGAAGTGGGAATGAAACTA  
 81 Q R K R C P Y C R F Q K C L E V G M K L  
 301 GAAAGCCGTGCGGGCGGACCGCATGAGGGCGGCGCAACAAAGTCCGGGCCATGTACAAG  
 101 E A V R A D R M R G G R N K F G P M Y K  
 361 CGTGACCGCGCCAGGAAACTGCAGATGCTGAGACAACGTCAGCTGACCCACCCGGCAGC  
 121 R D R A R K L Q M L R Q R Q L T H P G S  
 421 CTGCTGTCCGGCGCCACCCCACCTCGGGCGTCGCGCTCACGTACTCCACCCGGG  
 141 L L S G G R H P T T S G V A S R T P P G  
 481 GCTACTCCTCGGCGCCATCCACACACGTCCACATCAAGGAAGAGATCCAGAGTCCCTTCC  
 161 A T P R R H P H T S T S R K R S R V P S  
 541 TCTCCTCGTCCACATCTCGCCAGACTCCTCGCCGTCCCCATGGCAATTGGCGGCCCTC  
 181 S P R P H L R Q T P R R P P W Q L G G L  
 601 GTCGCAGGCTCGGCGGGCTGGCGGCCATGTGCTCCGGCCCTGTGGCACCCATCCTC  
 201 V A G S G G R G R P S A S G P V A P I L  
 661 GCTGGGCCGATCCGCCCTGTGGTCACCAACGCCAATCCACGGCGGGTGGCGTAACA  
 221 A G P D P A L W V T N A Q S T A G G V T  
 721 ACGGGCACACCAACCTACGGGAGGGCTCGCGGTGGAGGAGGAGGAGGAGGCGGGGG  
 241 T G T P P T G G R R R W R R R R R R R G  
 781 AGGAGGACGCACAGTACGCCCTGTATCCCTCACCAATTATCCGAGAATTGGTGGAGACC  
 261 R R T H S T P L Y P F T I I R E L V E T  
 841 GTTGACGACAGGAGTGGCAGGCGTTGTCTCGCTGCTGAGAACAGACACTAACAC  
 281 V D D Q E W Q A L L F S L L Q N Q T Y N  
 901 CAGTGCAGGTGGACCTCTCGAGCTGATGTGCAAAGTCCTCGACCAGAACCTCTCGCG  
 301 Q C E V D L F E L M C K V L D Q N L F A  
 961 CAGGTCGACTGGGCCGAACTCCTGTTCTCAAGATCTCAAGGTTGATGACCAGATG  
 321 Q V D W A R N S C F F K D L K V D D Q M  
 1021 AAGCTCCTACAGCACTCTGGTCTGACTTGCTGATTCTGACCCACTACATCAGCGCATC  
 341 K L L Q H S W S D L L I L D H L H Q R I  
 1081 CACAACAGACTGCAGGACGGAGACCACCGCTGCCGAATGGTCAGAAGTTCGACCTGCTCC  
 361 H N R L Q D E T T L P N G Q K F D L L S  
 1141 CTGGCACTGCTCGGGACCAACAGTTGCCGACCGTTCCACACATCCTCAGCAAGCTT  
 381 L A L L G T T Q F A D R F H N I L S K L  
 1201 ATTGACCTCAAATTGACGTCCCCGAGTATATCTGCTCAAGTTCGTCATCCTCTCAAT  
 401 I D L K F D V P E Y I C L K F V I L L N  
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 421 P A E V R L L S D R R S V I T A H E Q V  
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 441 K Q A L L D Y I A N V Y P E D T E K Y Q  
 1381 AAGATGATGGACCTGCTCCCCGAACCTCACTTCATTGCCGACAACGGTAAAAATACCTT  
 461 K M M D L L P E L H F I A D N G E K Y L  
 1441 TACTTCAAGCACATAAACGGCGCTGCCACGCAAGACCTTGTAAATGGAGATGCTCCAC  
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 1501 ACTAAACGGAAATAG  
 501 T K R K \*

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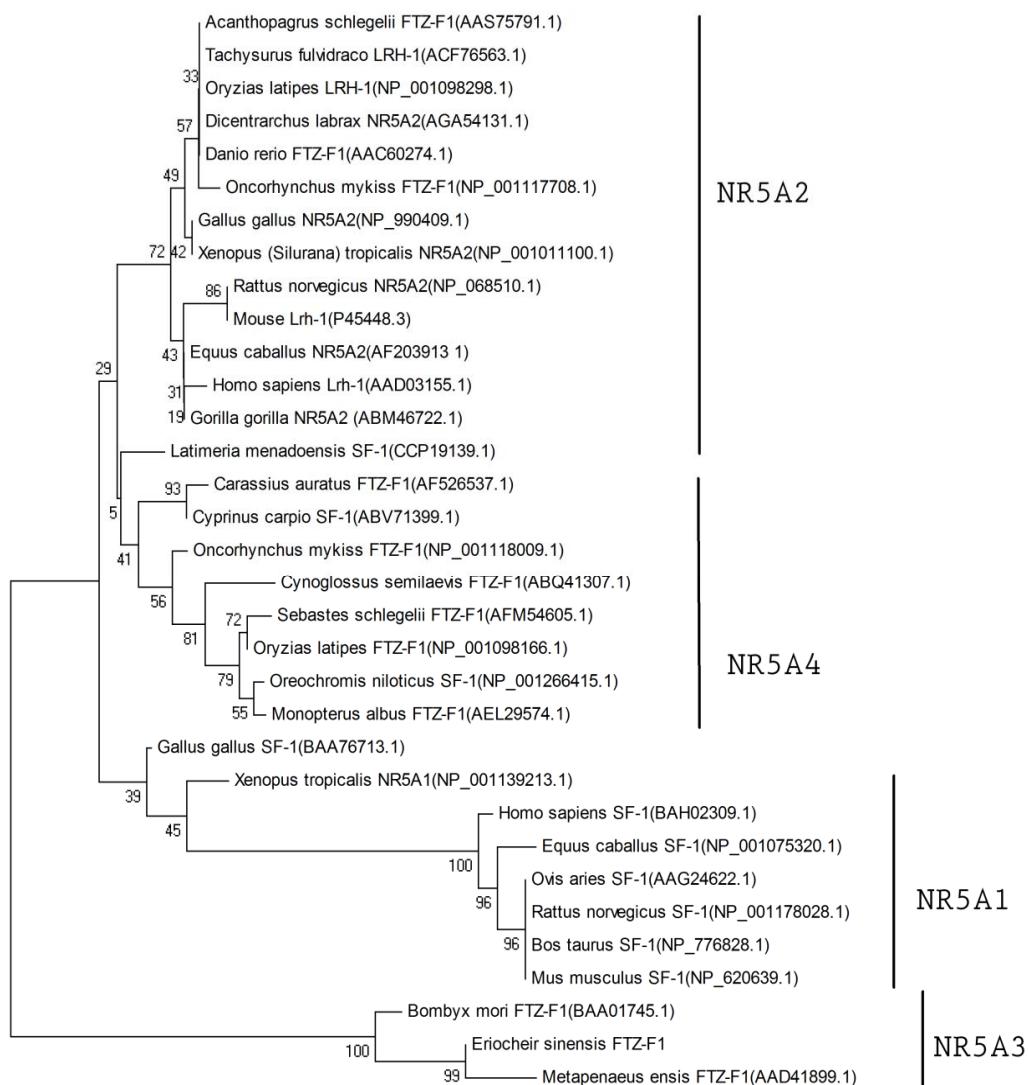
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89 Supplemental Fig. S5.

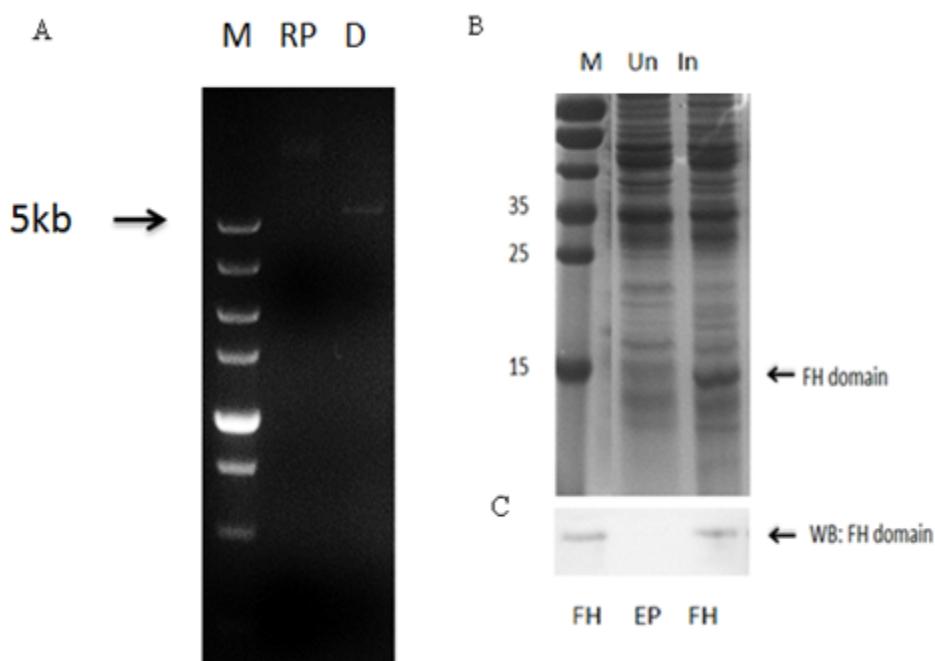


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## 106 Supplemental Fig. S6.

1 CTGGCAGTGCCTCTGAAACGGACAACACTCGAGAGCGGGGGCAACACCAGCATCGTGGC  
 1 L A V L L R T G Q L E S G R Q H Q H R G  
 61 GACCTGTGGGGGGTCATCGTGTGCTTCATCAATGTGCTGGACGCCAGGTGGTTCTCAA  
 21 D L W G V I G V L H Q C A G R R G G S Q  
 121 GTACAACCGCAGTACAAAAACCTTGCCATCCACCGCCAGCTGGCAGTGCCTCTGAAAC  
 41 V Q P Q Y K N L A I H R Q L A V L L R T  
 181 GGACAACCTCGAGAGCGGGCGGAAACACCCAGCATCGTGGCGACCTGTGGGGGTCACTGAC  
 61 G Q L E S G R Q H Q H R G D L W G V I D  
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 101 T A G E N L Y A Q V W S T L P L D D L G  
 361 CTCAAGACCATGQAACGTCCTCGCTGCAAGCTGGTGCACGATCCACAGTGCCTCTCACTCAC  
 121 L K T M N V L A A A G A R S T V L L I H  
 421 CCAGACGGGGGGATATGTTACCAAGATGTTCCGCACGGTCCCGATAAGGCTGGCGCTGTG  
 141 P D A G I C Y Q M F R T V P N K A A A V  
 481 ATATGTGGCTGCTCTCCACCCCTAACAGAAGGGGACATGGTTGTTGTGGTCACGGAGAC  
 161 I C A L L F H P K K A T W L F C G H E D  
 541 GGGCAGATCCAGCTGGGACATTGGCACTCCACCTTACCTGAGTACGAAGGTTAGCCAG  
 181 G Q I Q L W D I G T P T L P E Y E V S Q  
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 201 V H L L T I P P V A R D V Y N L A F S C  
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 221 T H D L L I G G C D G G L Y A W K V D L  
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 241 K K I E A G E S L E R L E F V L P E V D  
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 281 C A L H G Q I Y V F S V S K A L S V A K  
 901 CTTAATAAGAGTOGGTCAGATCTATGTTCTCCGTGCTAACGGCTTCCGTGGCCAAG  
 301 L N K S R S Q L Q A E V K T E M M V R L  
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 361 G R A T S P L T G G A P I P M V E P V K  
 1141 ATCCTGGAGTGGCTGAGCTGGAGGATGGGAGGTGGAAAAGGCCGTAAGCTTCACTG  
 381 I L E W P E L E D A E V E K A R K L R L  
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 421 A V T S N N M V C I W R H G K E E E E E  
 1321 GAGGAGGCAGGGTGAAGCAGGAGGAAGCGGAGAAGGATAAAAAAAAAAAAAAAA  
 441 E E A A V K Q E E A E K D K K K K K K K  
 1381 AAAAAAAAAATTCOOGGTGTGTTCCCCCCCAGGGCCCCAAAAAGGGAAAAAAA  
 461 K K K I S R C F P P G A P K K G K K K K

108      Supplemental Fig. S7.



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