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AUTÓNOMA DE
MÉXICO



Facultad de Medicina



Universidad Nacional Autónoma de México

Analysis of TATA binding Protein 1 (TBP1) of *Taenia solium* and core promoter elements In taeniidae family genes.

Oscar Rodríguez Lima

Taenia solium

Human
(Definitive Host)

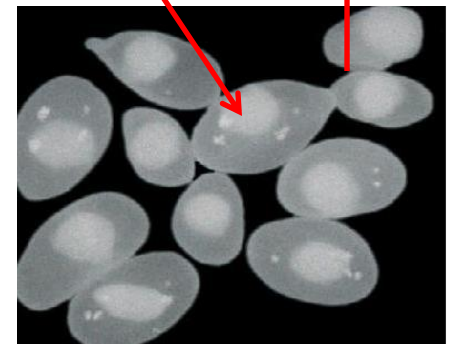
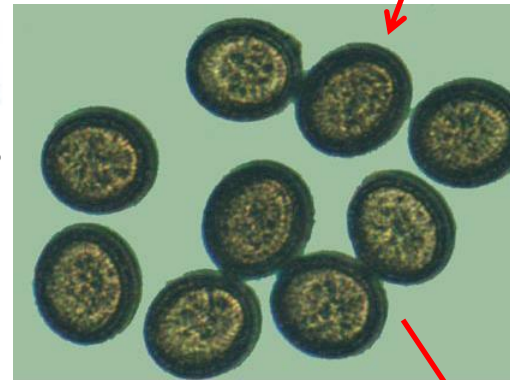
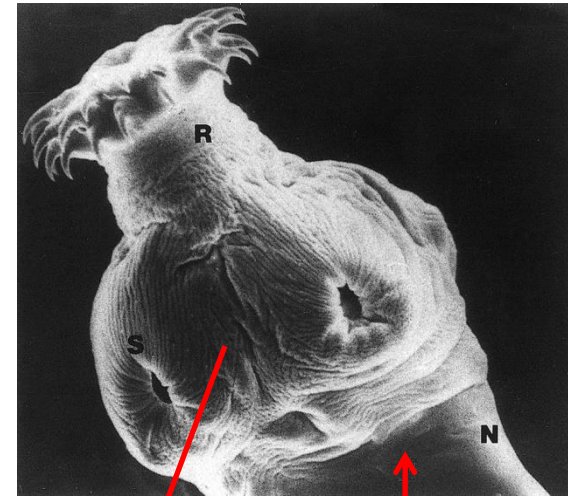
Infected swine
meat consumption

T. solium eggs
consumption
Human cysticercosis

T. solium eggs
consumption
Swine cysticercosis

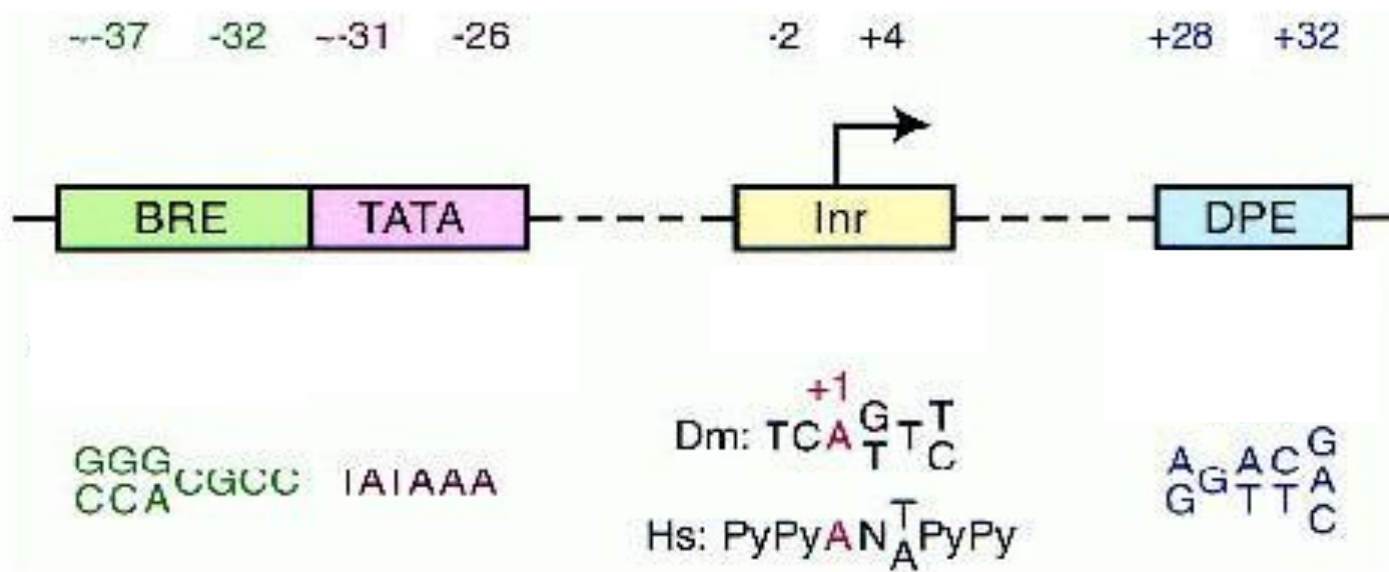
Swine
(Intermediate host)

Taeniosis



Transcription by RNA Pol II

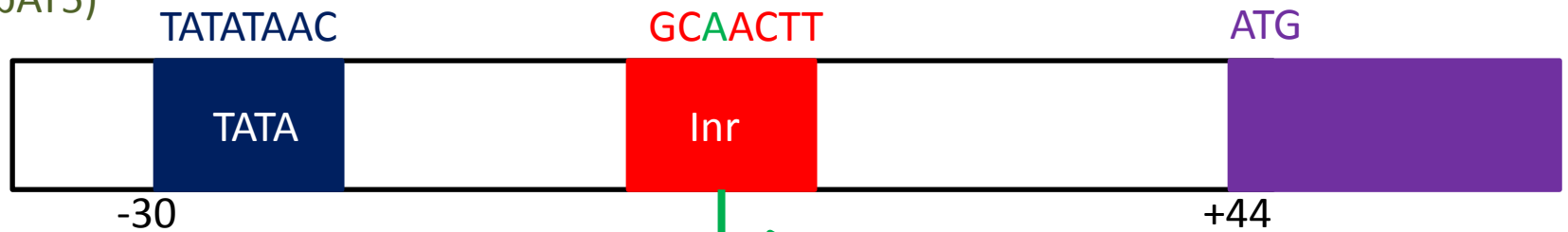
- The protein coding-genes are transcribed by RNA Pol II.
- RNA Pol II required the formation of pre-Initiation Complex in the core promoter.



Transcription in *Taenia* genus.

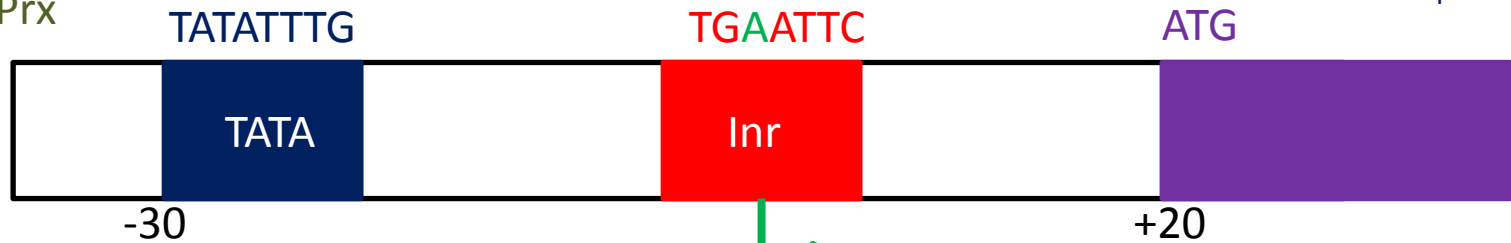
- Little is known about transcription in this genus.
- Only a few core promoter sequences have been published.

Actin (pAT5)



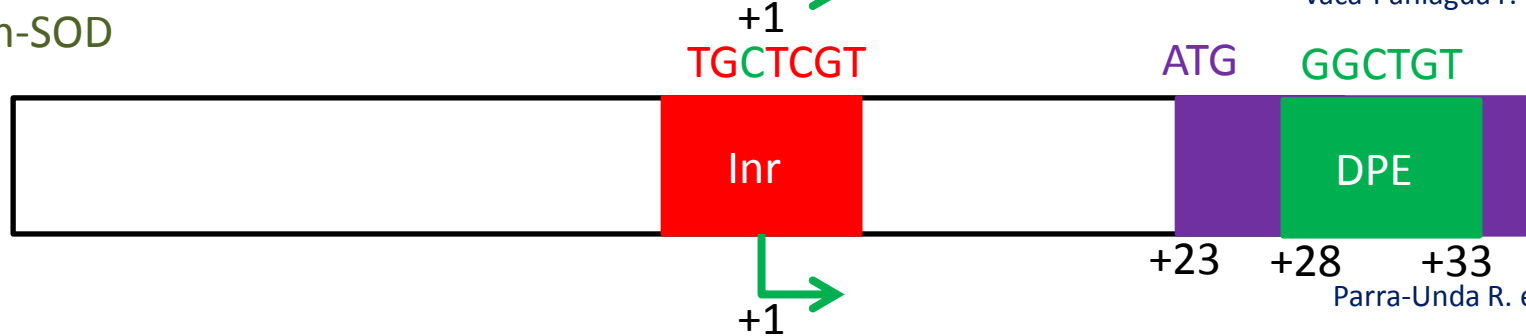
Campos A. et al, 1990.

Ts2-CysPrx



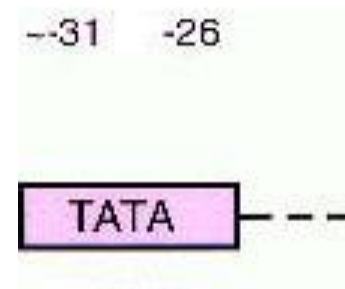
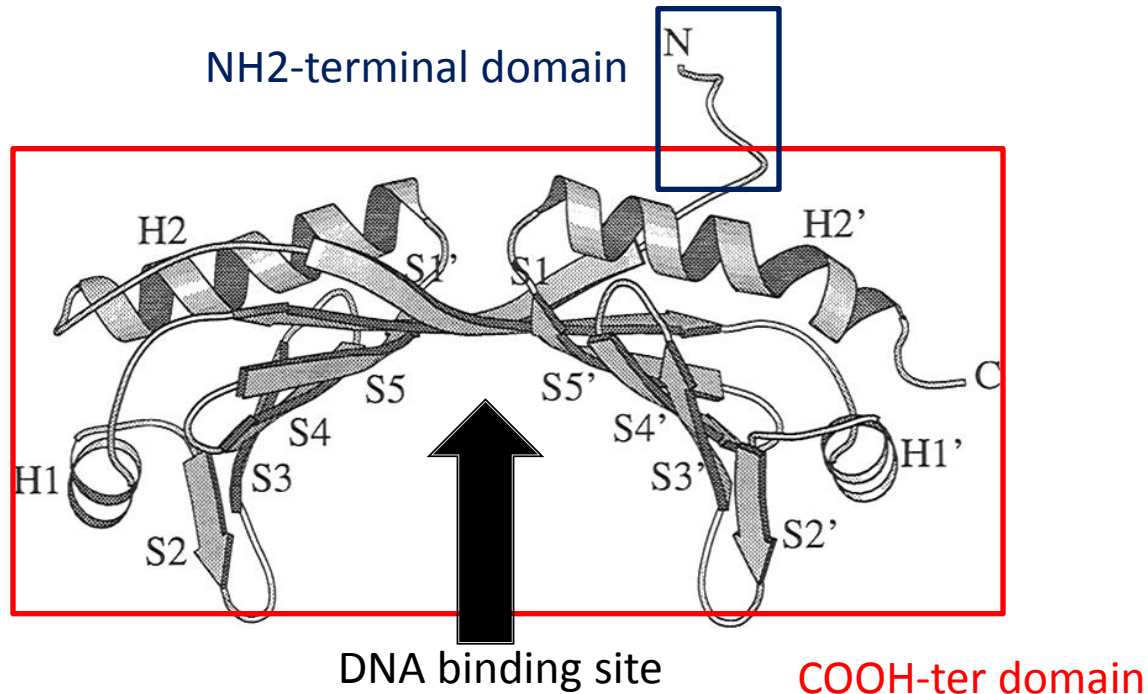
Vaca-Paniagua F. et al, 2009.

TsCu,Zn-SOD



Parra-Unda R. et al, 2011.

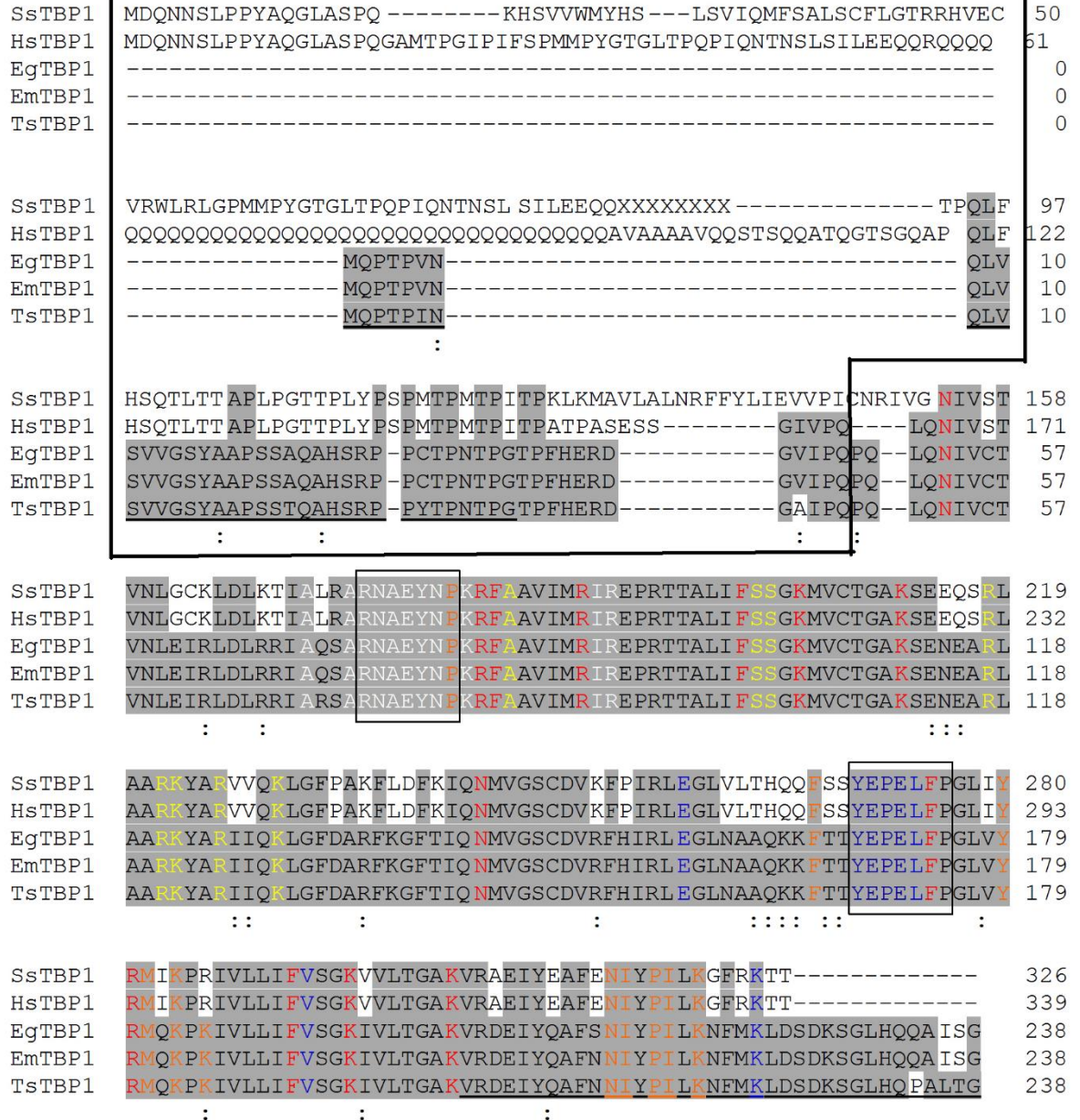
TATA-box Binding Protein (TBP)



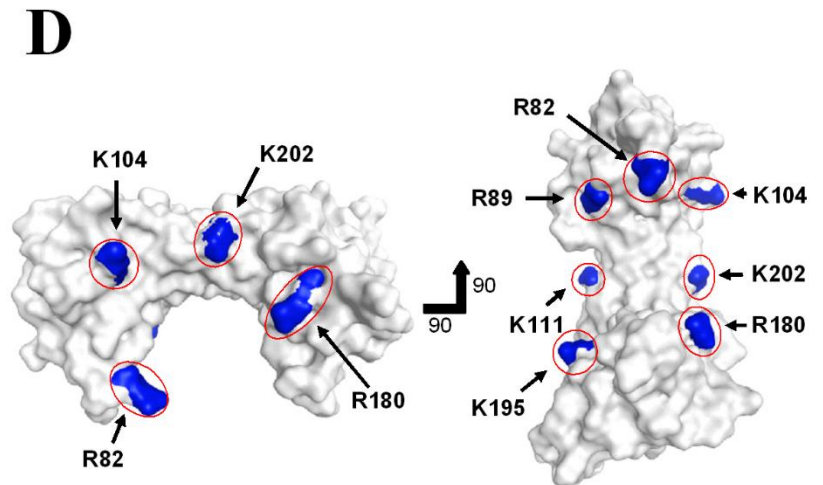
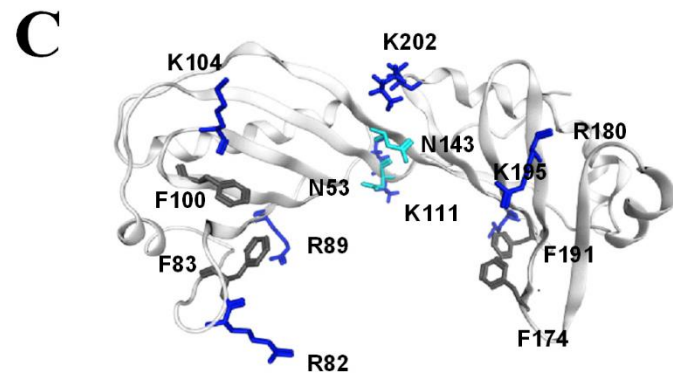
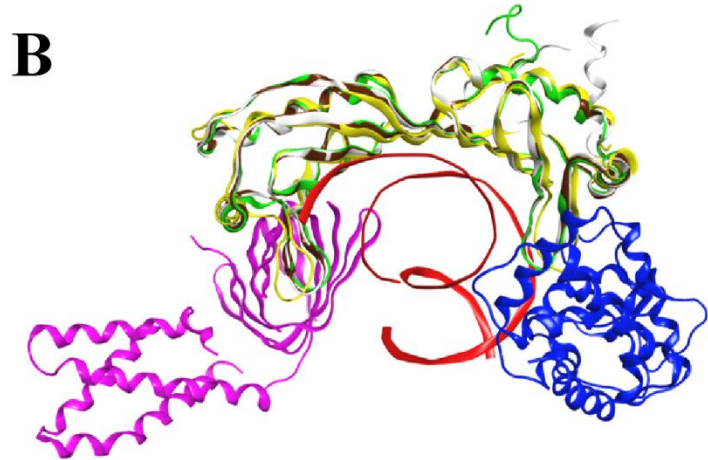
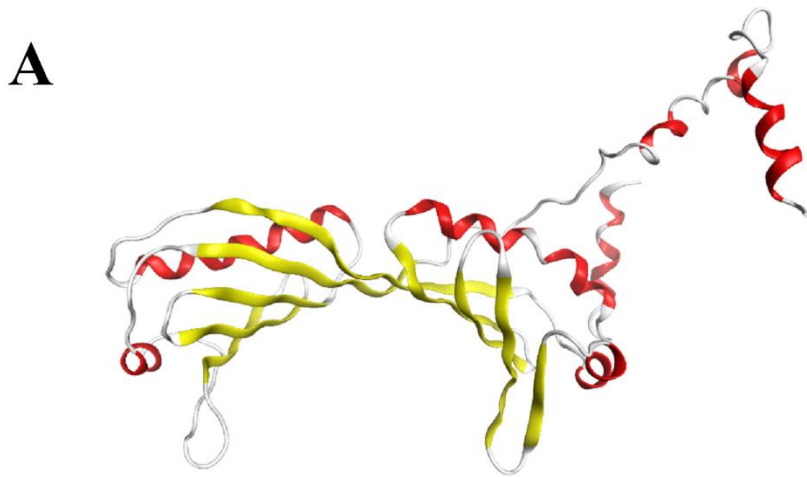
TATA**WAAR**

W = A or T
R = A or G

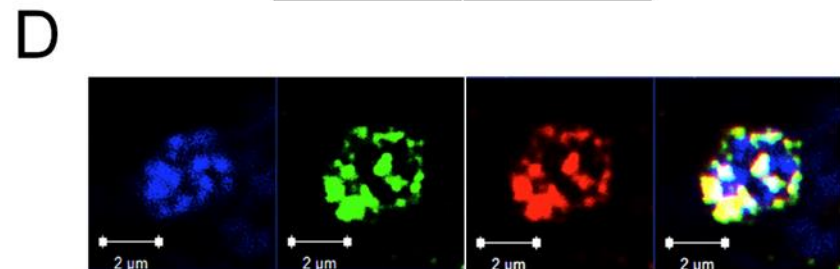
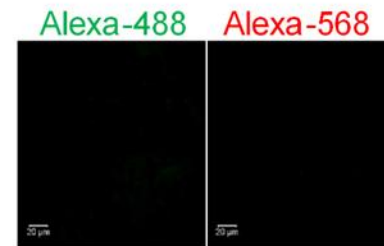
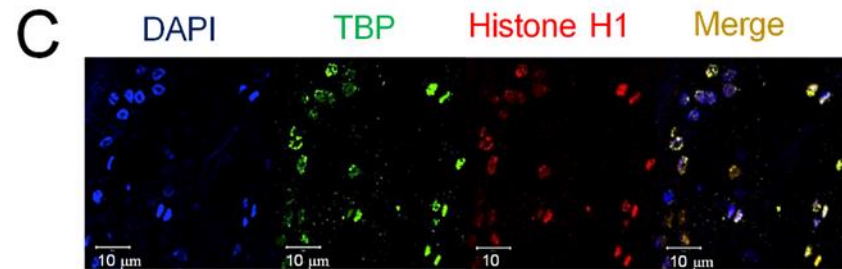
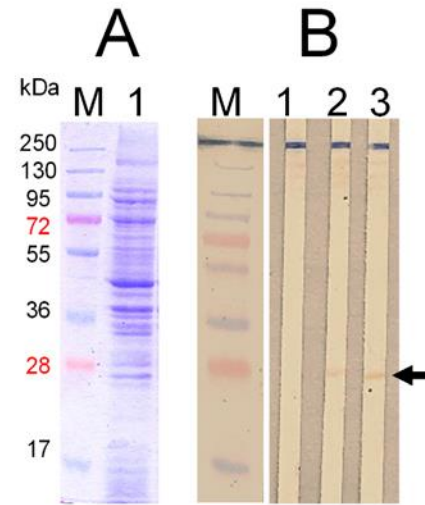
Multiple alignment of different TBP1s



TsTBP1 homology model.



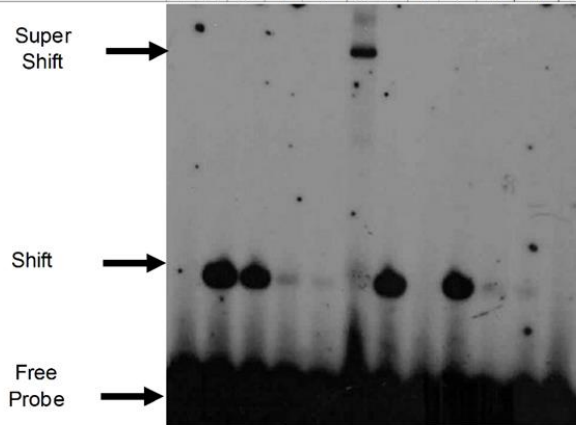
Identification of TsTBP1 by western blot and confocal microscopy.



TsTBP1 interaction with two putative TATA-box

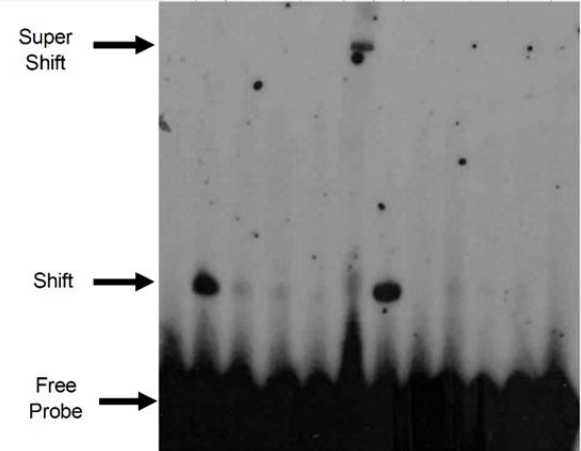
A pAT5

Nuclear extract (10µg)	-	+	+	+	+	+	+	+	+	+	-	
pAT5 TATA Box probe	+	+	+	+	+	-	-	+	+	+	+	
pAT5 TATA Box Cold probe	-	-	25X	50X	100X	-	-	-	-	-	-	
Consensus TATA Box probe	-	-	-	-	-	+	-	-	-	-	-	
Consensus mut TATA Box probe	-	-	-	-	-	-	+	-	-	-	-	
Ts2-CysPrx TATA Box Cold probe	-	-	-	-	-	-	-	25X	50X	100X	-	
Anti-TsTBP1 antibody	-	-	-	-	-	+	-	-	-	-	+	
	1	2	3	4	5	6	7	8	9	10	11	12

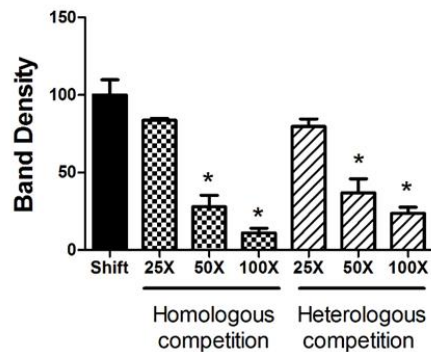


A Ts2-CysPrx

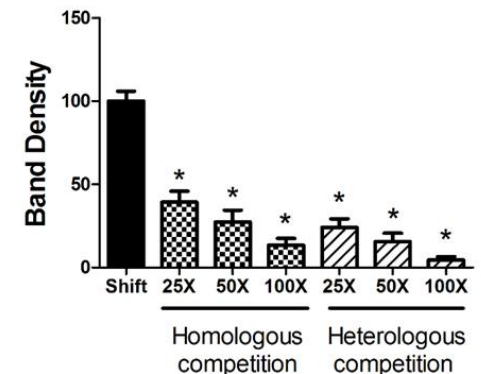
Nuclear extract (10µg)	-	+	+	+	+	+	+	+	+	+	-	
Ts2-CysPrx TATA Box probe	+	+	+	+	+	-	-	+	+	+	+	
Ts2-CysPrx TATA Box Cold probe	-	-	25X	50X	100X	-	-	-	-	-	-	
Consensus TATA Box probe	-	-	-	-	-	-	+	-	-	-	-	
Consensus mut TATA Box probe	-	-	-	-	-	-	+	-	-	-	-	
pAT5 TATA Box Cold probe	-	-	-	-	-	-	-	25X	50X	100X	-	
Anti-TsTBP1 antibody	-	-	-	-	-	+	-	-	-	-	+	
	1	2	3	4	5	6	7	8	9	10	11	12



B



B

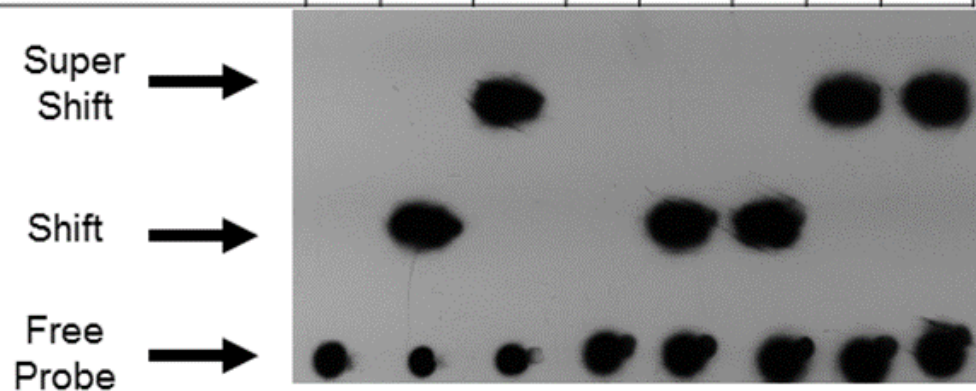


Probes used in EMSA

pAT5 TATA-box	5' – CCCAAATCT TATATAAA CCGTGGGT – 3' 3' – GGGTTTAGAATATATTTGGCACCCA – 5'
Ts2-CysPRX TATA-box	5' – GCGCTTCGCT TATATTTG GC GGTAAG – 3' 3' – CGCGAAGCGATATAAACCGCCATTC – 5'
Consenso TATA-box	5' – AAGGGGGGGCT TATAAAAG GGGGGTGGG – 3' 3' – TTCCCCCGATATTTTCCCCCACCC – 5'
Consenso TATA-box mutada	5' – AAGGGGGGGCT <u>TGTGAAAG</u> GGGGGTGGG – 3' 3' – TTCCCCCGATATTTTCCCCCACCC – 5'

TsTBP1 binding inhibition to the TATA-box of pAT5 by anti-pTsTBP1-C antibody.

Nuclear extract (10µg)	-	+	+	+	+	+	+	+
pAT5 TATA-box probe (biotin labeled)	+	+	+	+	+	+	+	+
Anti-TsTBP1-C antibodies	-	-	+	+	-	-	-	-
Anti-TsTBP1-N antibodies	-	-	-	-	-	-	+	+
Normal rabbit IgG	-	-	-	-	+	+	-	-
	1	2	3	4	5	6	7	8

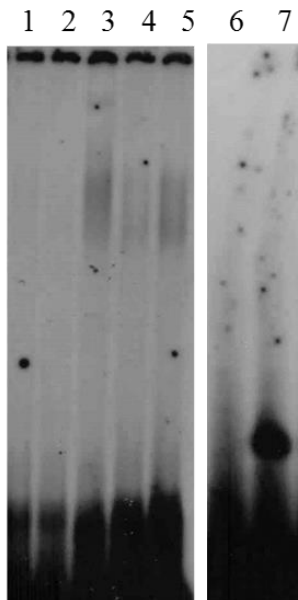


Analysis of core promoter of TsTBP1 and pAT5 coding genes

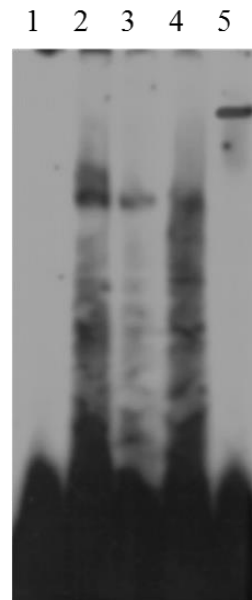
A



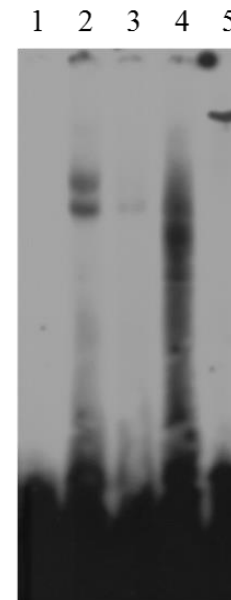
B



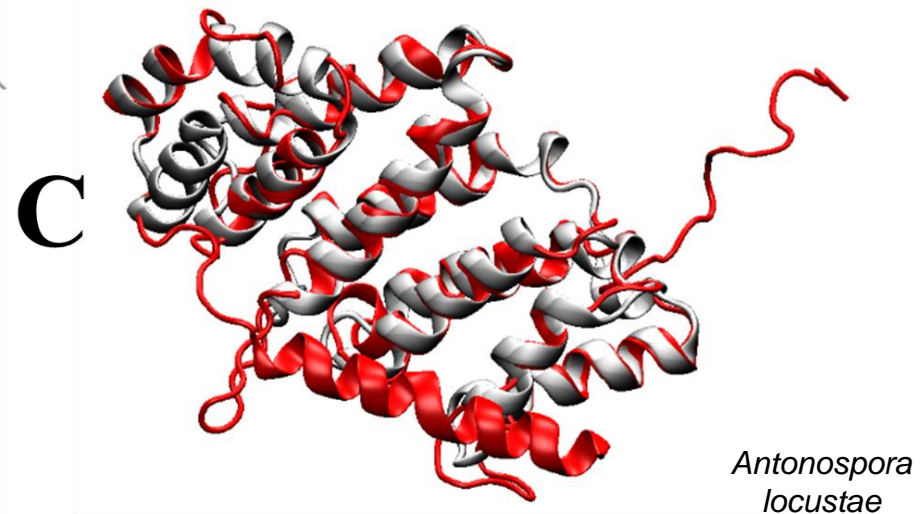
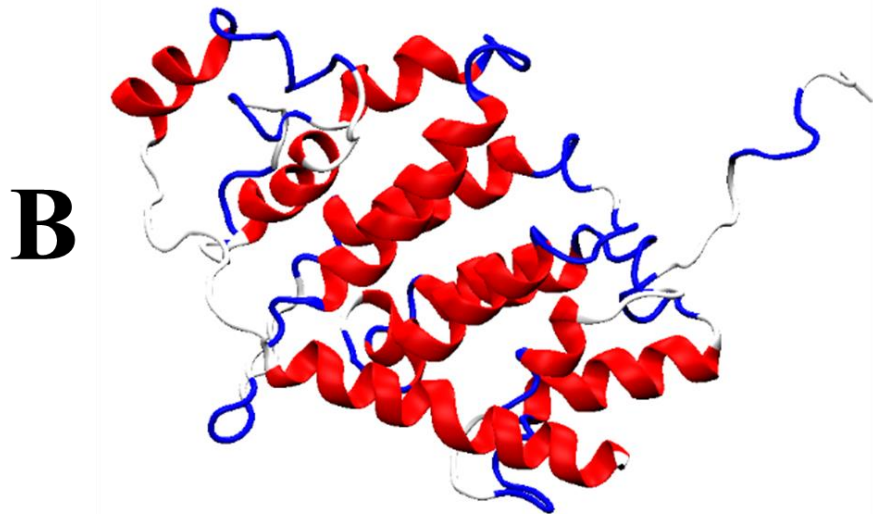
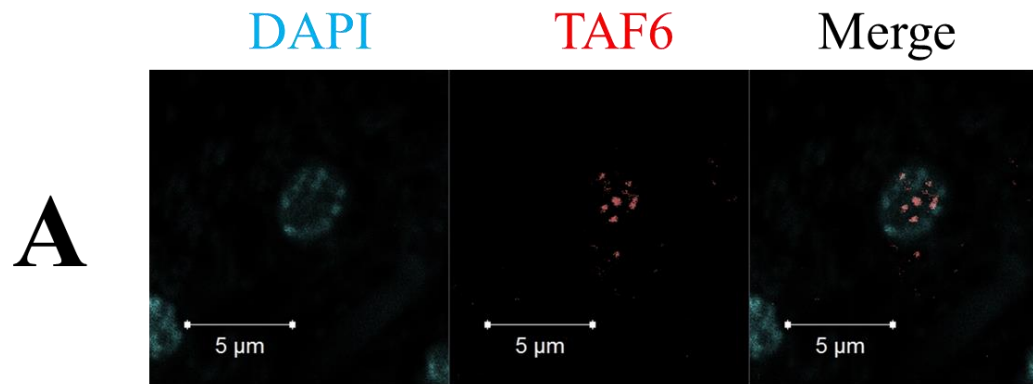
C



D



Identification of TsTAF6 and homology model.



Core promoter analysis in taeniidae family genes.

A

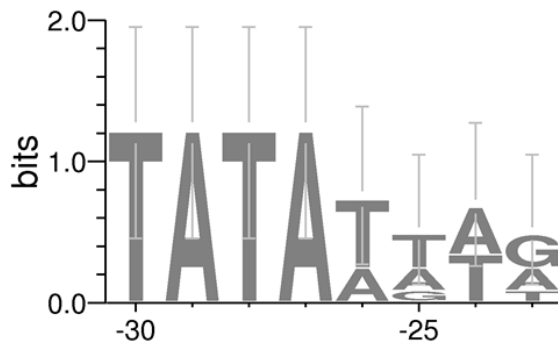
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pAT5	▼	▼	▼ ▼
pAT6	▼	▼	▼ ▼
TsPrx	▼	▼	▼ ▼
TcPrx	▼	▼	▼ ▼
Egact I	▼	▼	▼ ▼
Egact II	▼	▼	▼ ▼
TsTrx			
EgHbx1			
TsCu/ZnSOD			
TsTBP			

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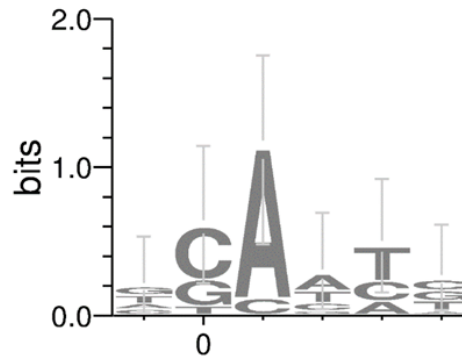
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TATATTTG GCGGTAAGAGCTGTGCGTGG TGAATT CCATTGTTTTCGTGTAATGGCTGCT TGCTGT CATCGGGAGGCCT
TATATTTG GCGGTAAGAGACGCTGTGGCTGT TGAATCC ATTGTCTGCTCGCGTTCAGTGAT GGTTGC CGTTATCGGGAGG
TATAAAAAG CCTAGAAATCACTAGAAGGAT TCACTAG AAGGATCACTTTTGGTTGAGTGCAGTAGAGAAGACAAATCCTTTGGTGAGCCATG
TATATTTTACGTCGAAACGGTGAACGTGG CCATTT TGATTTTACTCTTTGCTAGCCTCTCGATGGCGGACGAGGACACCGCA

TGAACGTCGGTGTAAATAAATAAATAATTA ACAATGC TTTAATGACCTTGAGGCTGTTTAAACAGAGCTCCTTGTAGCACGTTTCAAAGA//ATG
CGCACGAGGAGATAAGATAGTGAAACTC AGAGAGA CGAATCTGGTGACACTCACAAAACACTACGCGCGGCCACACAACCTTTTAGTTGGCTGGATG
CTAGCAGTGGCATCAAGGATCTGCAGGG CTCTCGT TTTTCTTTGGAGTCGCTATGAAG GGCTGT TTGTGTTATGCGAGGGGGAGGA
CCGCAACCAAGCAATGTAAGCATCGGCAG GCATCCT CAACAGCAACCCCTCCGGGTTCT TGTCGA GCAGATGCAGCCAACCCCATCAA
    
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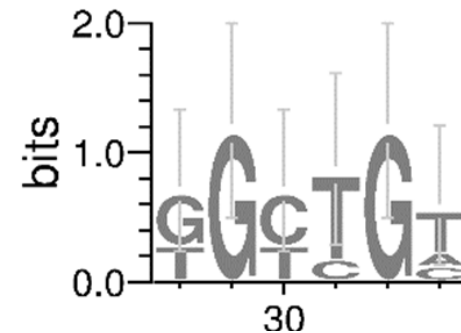
B



C



D



Thank you!

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