

Article

Nicotinic Acetylcholine Receptors Are Novel Targets of APETx-like Toxins from the Sea Anemone *Heteractis magnifica*

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108 MDYQRLFLFAVAMVITTTVALPQDTALMDGQLQKRGAPCKCGGYTGVYWLM
110 MDYQRLFLFAVAMVITTTVALPQDTALMDGQLQKRGAPCKCGGYTGVYWF
106 MDYQRLFLFAVAMVITTTVALPKDAALMDGQLQKRGAPCKCGGYTGVYWF
109 MDYQRLFLFAVAMVITTTVALPKDAALMDGQLQKRGAPYKCGGYTGVYWF
1078 MDYQRLFLFAVAMVITTTVALPKDAALMDGQLQKRGAPCKCGGYTGVYWF
111 MDYQRLFLFAVAMVITTTVALPKDAALMDGQLQKRGAPCGCHGYTGVYWF
107 MDYQRLFLFAVAMVITTTVALPKDAALMDGQLQKRGAPCGCHGYTGVYWF
1081 MDYQRLFLFAVAMVITTTVALPKDAALMDGQLQKRGAPCGCRGYTGVYWF
1097 MDYQRLFLFAVAMVITTTVALPQDTALMDGQLQKRGTPCKCLGYTGVYWF
102 MDYQRLFLFAVAMVITTTVALSKDTALMDGQLQKRGTPCKCHGYIGVYWF
104 MDYQRLFLFAVAMVITTTVALPKDTALMDGQLQKRGTPCKCHGYIGVYWF
1082 MDYQRLPFLFAVAMVITTTVALPKDTALMDGQLQKRGTPCKCHGYIGVYWF
1077 MDYQRLFLFAVAMVITTTVALPKDTALMDGQLQKRGTPCKCHGYIGVYWF
996 -----GVYWFMLAGCPNGYGYNLSCPYPFLGICCVKK
993/Hmg 1b-4 -----GVYWFMLSRCPNGYGYNLSCYFPMGICCVKR
1203 -----GVYWFMLSRCPNGHGYNLSCPYPFLGVCC---
992 -----GVYWFMITRCPNGHGYNLSCPYPFLGVCCVKK
994 -----GVYWFMVSRCPGGYGYNLSCPYPFLGVCCVKK
1217 -----GVYWFMVSRCPGGYGHNLSCPYPFLGVCCVKK
916 -----GVYWLMVSRCPGGYGYNLSCPYPFLGVCCVKK

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5' RACE

3' RACE

(a)

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Hmg 1b-2
104 5' RACE MDYQRLFLFAVAMVITTTVALPDTALMDGQLQKRGTPCKCLGYTGVYWF
996 3' RACE -----GVYWFMLAGCPNGYGYNLSCPYPFLGICCVKK
1593 MDYQRLFLFAVAMVITTTVALPDTALMDGQLQKRGTPCKCLGYTGVYWF
-----GVYWFMLAGCPNGYGYNLSCPYPFLGICCVKK

Hmg 1b-5
1097 5' RACE MDYQRLFLFAVAMVITTTVALPDTALMDGQLQKRGTPCKCLGYTGVYWF
992 3' RACE -----GVYWFMITRCPNGHGYNLSCPYPFLGVCCVKK
1547 MDYQRLFLFAVAMVITTTVALPDTALMDGQLQKRGTPCKCLGYTGVYWF
-----GVYWFMITRCPNGHGYNLSCPYPFLGVCCVKK

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(b)

Figure S1. (a) 3'- and 5' RACE translated sequences. (b) Full-length cDNAs of Hmg 1b-2 and Hmg 1b-5. Amino acid substitutions are shown by green (Hmg 1b-2) and yellow (Hmg 1b-5) colors.

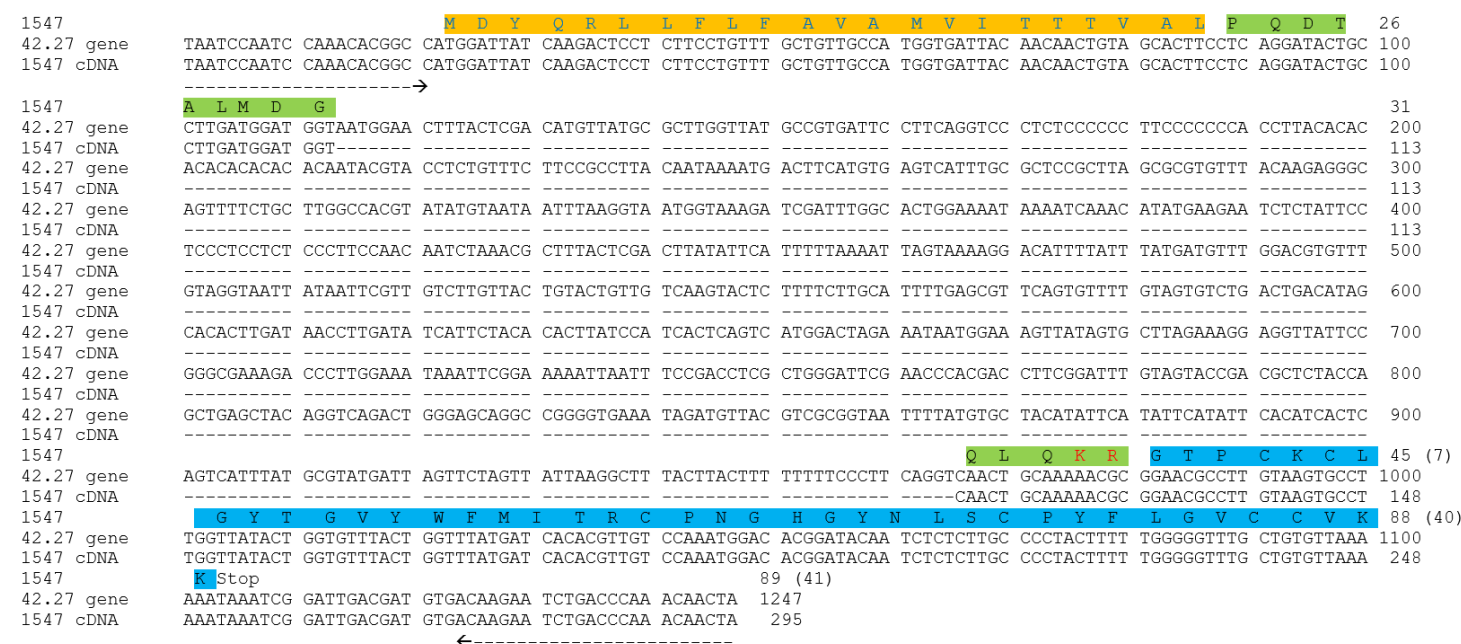


Figure S2. Structure of *HmgTx 1b-5* gene inferred by similarity of 42.27 gene sequence with HmgTx 1547 cDNA. The signal peptide region is highlighted by orange, the propeptide region is highlighted by green, and the mature chain is highlighted by blue. The furine proteinase site is colored by red. The mature peptide is highlighted by blue.

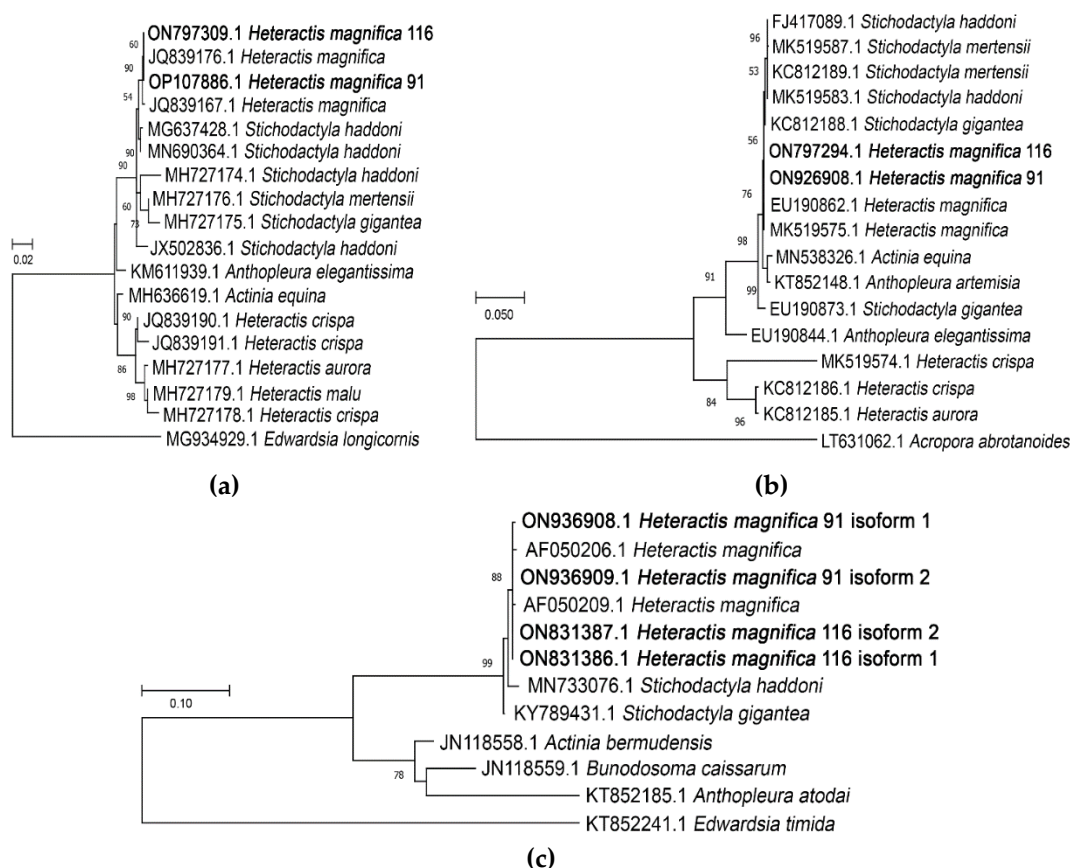


Figure S3. ML trees showing phylogenetic relationships of the sea anemone *H. magnifica* 91 (present study) and *H. magnifica* 116 (as *H. crispa* 116 in previous study [16]) in the Stichodactylidae family based on sea anemone COI gene sequences (a), 18S gene sequences (b), and ITS gene sequences (c). Sea anemones *Anthopleura elegantissima*, *Actinia equina*, *Anthopleura artemisia*, *Actinia bermudensis*, *Bunodosoma caissarum*, and *Anthopleura atodai* belong to the Actiniidae family. Bootstrap values (%) of 1000 replications. Nodes with confidence values greater than 50% are indicated.