

Supplementary information

Enlisting the *Ixodes scapularis* Embryonic ISE6 Cell Line to Investigate the Neuronal Basis of Tick-Pathogen Interactions

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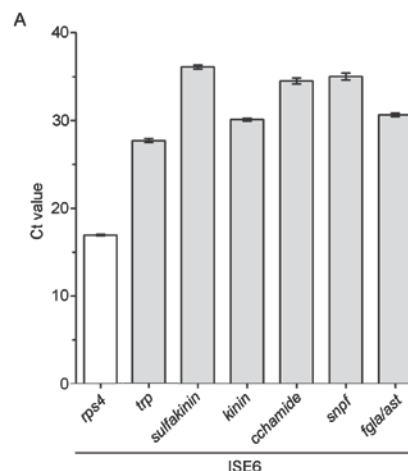
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Supplementary figure S1. (A) Relative expression of ribosomal protein S4 (*rps4*) transcript and six different neuropeptide transcripts in uninfected ISE6 cells. The Ct values (i.e., the mean-standard error) for the two biological replicates are shown.

A

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ISCI008383 -----MSIRARQGGKPKSKSEVGN---EFVGEVSLVEKLWDDGLDGG 41
EL516783 MDHFEMKVVALMSLVLLLTLSSVRTASFQSEVGN---FEVGEVSLVEKLWDDGLDGG 57
MW082607 MDHFEMKVVALMSLVLLLTLSSVRTASFQSEVGNLQSEVGEVSLVEKLWDDGLDGG 60
      :          :          :          :          :          :          :
      *****
ISCI008383 DDLEIAAAAADDEKRAFHAMRGGKDDPSLDWEADKRAFHAMRGRLLAPASALQAKN-IR 100
EL516783 DDLEIAAAAADDEKRAFHAMRGGKDDPSLDWEADKRAFHAMRGRLLAPASVDSFIAQLR 117
MW082607 DDLEIAAAAADDEKRAFHAMRGGKDDPSLDWEADKRAFHAMRGRLLAPASVDSFIAQLR 120
      :          :          :          :          :          :          :
      *****
ISCI008383 GPFLRLLGGEGFPCKLSRGRT--TRRFPKCAASK-----EGCSRHW- 141
EL516783 RAVLQGRGSGFFGMRGKRMSRTPGKEHPRSTFTVATRGRRSVLSAERSPYY 169
MW082607 RAVLQGRGSGFFGMRGKRMSRTPNKEHPRSTFTVATRGRRSVLSAERSPYY 172
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B

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ISCI008383 ----- 0
EL516783 CTTACGCGCGGGGGCCGCAAGTGAAGCAACCAGGTCGCGAACCTTTGCTCGCCCA 60
MW082607 -----AGTGTAAGCAAAACCCGGTGCAGAACCTTTGCTCGCCCA 41
      :          :
ISCI008383 ----- 0
EL516783 GTCGATCTCTCGTGCACACGAAGCTCGCGTCAGCCAGCCAGCACCGGGTCTTACGG 120
MW082607 GTCGGTCTCCTCGTGCACAAAGCTCGCGTTCGAGCCAGCCAGCACCGGGTCTTACGG 101
      :          :
ISCI008383 ----- 0
EL516783 TGGAAGCAGCAGCGCCCATCGGACAGCCGAAATGACACTCCGGAGATGAAGTTGTG 180
MW082607 TGGAAGCAGCAGCTCCCATCGGACAGCCGAAATGACACTCCGGAGATGAAGTTCTG 161
      :          :
ISCI008383 -----ATGTCGATACGCGGAGGCAAGGTCAAAAACCGTCG 36
EL516783 GCTCTGTGGTCTCTGGTCTGCTGCTCACGCTAAGCTC---AGTCAGAACTGCTCGTGT 237
MW082607 GCACTGTGGTCTCTGGTCTGCTGCTCACGCTAAGCTC---AGTCAGAACTGCTCGTGT 218
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ISCI008383 -----GGAGGTGCGCAACGAGCCGAGTGGGAGAATTCGCTGTT 87
EL516783 CAGAGTTCCGAGGTCGGCAACGAGCTCGAGTCCCGCAGGTGGGAGAATTCGCTGTT 288
MW082607 CAGAGTTCCGAGGTCGGCAACGAGCTCGAGTCCCGCAGGTGGGAGAATTCGCTGTT 278
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ISCI008383 GAAAAGTTGGGTTGGGAGCAGCGGCTTGGACGCGGTGACGACCTGGAGATCGCGCGGCA 147
EL516783 GAAAAGTTGGGATGGGAGCAGCGGCTTGGACGCGGTGACGACCTGGAGATCGCGCGGCA 348
MW082607 GAAAAGTTGGGCTGGGAGCAGCGGCTTGGACGCGGTGACGACCTGGAGATCGCGCGGCA 338
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ISCI008383 GCGGACGACGAGAAGCGGCTTCCAGCCCATCGGAGCAGAAAGACGCTTCCCTG 207
EL516783 GCGGACGACGAGAAGCGGCTTCCAGCCCATCGGAGCAGAAAGACGATCTTCCCTG 408
MW082607 GCGGACGACGAGAAGCGGCTTCCAGCCCATCGGAGCAGAAAGACGCTTCCCTG 398
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ISCI008383 GACTGGACGAGCGCCGACAAGAGGGCCCTTCCAGCCATGCGCCGGAAGCGGCTTTGGCC 267
EL516783 GACTGGACGAGCGCCGACAAGAGGGCCCTTCCAGCCATGCGCCGGAAGCGGCTTTGGCC 468
MW082607 GACTGGACGAGCGCCGACAAGAGGGCCCTTCCAGCCATGCGCCGGAAGCGGCTTTGGCC 458
      *  *  *  *  *  *  *  *  *  *  *  *  *  *  *  *  *  *  *  *
      *  *  *  *  *  *  *  *  *  *  *  *  *  *  *  *  *  *  *  *
ISCI008383 CCGGCCAGC-----GCACTCAGGCAAAAGACAT 296
EL516783 GGCTCGGGCTTCTTGGCATCGGGGCAAGAGGATGTCTGCACTCCAGGCAAAAGACAT 588
MW082607 GGCTCGGGCTTCTTGGCATCGGGGCAAGAGGATGTCTGCACTCCAAAGAAAGACAT 578
      *  *  *  *  *  *  *  *  *  *  *  *  *  *  *  *  *  *  *  *
      *  *  *  *  *  *  *  *  *  *  *  *  *  *  *  *  *  *  *  *
ISCI008383 CCGAGTCCACCTTGTGGGACTAGGGGGGGAAGGTCCCTTCTGCGAAGCTGATGTC 356
EL516783 CCGAGTCCACCTTGTGGGACTAGGGGGGGAAGGTCCCTTCTGCGAAGCTGATGTC 648
MW082607 CCGAGTCCACCTTGTGGGACTAGGGGGGGAAGGTCCCTTCTGCGAAGCTGATGTC 638
      *  *  *  *  *  *  *  *  *  *  *  *  *  *  *  *  *  *  *  *
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ISCI008383 AGGCCGTACTA---TAGAAGATTCCCAAGCCGTGTCAGCTTCAAGAAAGATGCAAGCG 416
EL516783 AGGCCGTACTA---TAGAAGATTCCCAAGCCGTGTCAGCTTCAAGAAAGATGCAAGCG 708
MW082607 AGGCCGTACTA---TAGAAGATTCCCAAGCCGTG----- 671
      *  *  *  *  *  *  *  *  *  *  *  *  *  *  *  *  *  *  *  *
      *  *  *  *  *  *  *  *  *  *  *  *  *  *  *  *  *  *  *  *
ISCI008383 CAACTG----- 423
EL516783 CAACTAGAAGCTCGAGCGCCCTCACACCAAGGAC 740
MW082607 ----- 671

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Supplementary figure S2. Protein (A) and nucleotide (B) alignment of *Ixodes scapularis* and *Ixodes ricinus* TRP. ISCI008383 – *I. scapularis* genome-predicted transcript; EL516783 - *I. scapularis* EST sequence; MW082607 – *I. ricinus* transcript (identified in this study). Please note the incorrect N-terminal sequence of ISCI008383 (A) caused by the incomplete 5'-end (B). In addition the shift in the 3'-end (B) reading frame caused an improper conceptual TRP translation (A) of ISCI008383.

Name	Forward primer (5'-3')	Reverse primer (5'-3')
<i>fgla/ast</i>	CACCATGCTGCTGAGCAA	CTACTTTCTCCGGAAGCAGG
<i>cchamide</i>	GTGTCTCTGACGTGATGC	GTCTATCAAGCCTCGCCTG
<i>short neuropeptide f</i>	GCCTGCATCCTTCTCGATAG	CTTCCGTTCCATGGTGTGG
<i>sulfakinin</i>	CTTTGTCTTCTTCGACACTC	GACGGGATCGATCATGTCC
<i>trp</i>	CTTGCGGATGGCATTGGTTC	CACCTATTGGCGTAACCCG
<i>kinin</i>	GAGGCAGGACAAAGAGAGC	CGAGAAGGTGCTGTCTTAG
<i>natalisin (1st set)</i>	AAACTCGGATCACAGACC	CATCGTGACGTCGAAAGAGT
<i>natalisin (2nd set)</i>	AGGTAGTATGTCGACCAAC	ACAGGTGACTACTGCGACAA
<i>natalisin (3rd set)</i>	CTTGCGGATGGCATTGGTTC	CACCTATTGGCGTAACCCG

Supplementary table S1. List of primers used in our qRT-PCR experiments.