

1       **tataaa**aaggaaacttagatctgagccagagcACATACTACTTTACCCTTTCACGAAC     60  
 61       TTTATAACTCAGAAAATAACACTCGTAACCTTCTTCCAGAGTATTCAATAATAATCGATT   120  
 121       m l p s r i i g v l l f c c i g g i  
 TAGAAAAATGTTACCATCCAGAATTATAGGCAGTCTTTGCTGTATTGGCGGCAT   180  
 181       i s C K A P N G A D G R S N G E A G G A  
 TATTAGCTGTAAGCTCTAACGGAGCTGATGGCAGAAGTAACGGCGAAGCCGGCGGGC   240  
 241       G T A G A N G C D G G D G G N G F P G T  
 GGGTACTGCTGGGCAAATGGTTGTGATGGTGGAGATGGAGGAATGGGTTCCCTGGAAC   300  
 301       G S A A G G A G G I G G V G G A G A K G  
 AGGGTCTGCAGCTGGTGGTGTGGCGGAATCGGCGGTGCGAGGAGCGGGTGCAGGAAAGG 360  
 361       T  
 G N G G K G G I G A S S E S A A G G A G  
 TGGAAATGGAGGAAAGGTGGTATTGGGCCAGTCGAATCCGCTGCTGGCGGGCAGG   420  
 421       G A G G A A G A G S S G P G G Q G G E G  
 TGGTGCAGGAGGAGCCGCTGGAGCTGGATCATCAGGACCTGGAGGAACAGGGGGTGAGGG 480  
 481       G K G G K A N G V T G T G G A G G I G G  
 TGGCAAAGGGGCAAAGCGAATGGAGTTACCGAACAGGTGGTGCAGGTGGAATAGGTGG 540  
 541       V G G A G S G G Q P G G N G G N A G L G  
 TGTTGGGGAGCGGGTTCGGGTGGACAACCCGGTGGTAATGGTGGAAACCGCGGGACTAGG 600  
 601       →  
 G D S V T A G G  
TGGTGTAGTGTAACTGCCGGGGtaattatcatattactgtgatattaatatgt   660  
 661       A G G T  
 aaataatataatatgtcacaaaataagtaaaatattattaacagCGCGGGTGGAAC 720  
 721       ←  
 G G A G G A G T P G G R G G N G A N G G  
 AGGAGGAGCTGGCGGAGCTGGTACACCCGGTGGCGTGGAGGCAATGGCGCAAATGGAGG 780  
 781       S G H G N P G G R G L P G A A G L P V G  
 AAGTGGTCATGGGAATCCAGGAGGCCGGGTTACCCGGGCTGCTGGTTGCCTGTTGG 840  
 841       G G G A  
 TGGAGGTGGTGCAGgtacatacacatattaaaggctaatacatcgtagatgtactcataca 900  
 901       G G I  
 aatcaaactacagaaatacttactactaatcacaattattgtataaaagGTGGAAT 960  
 961       ←  
 G G G G A \*  
TGGTGGCGCGGGTGGAGCGTAAGTAAGATGCAATGAGATAAGAAGTTCCATGTTAA 1020  
 1021      GTATTTCTGTACTGTATGCATAGTGAATTCTTCTCAACCATTGCTAGTTGTAAT 1080  
 1081      GTTACTGTTACTATTGTCTAAATTATTCTCTAACACAATT**AATAAA**TCGGAAAATAATGC 1140  
 1141      ATAAAAAAAAAAAAAAAAAAAA 1162

**Supporting Figure 1.** DNA sequence encoding the large snow flea AFP isoform. Sequences

appearing in mRNA are in uppercase font whereas introns and promoter sequences are in lowercase font. The deduced protein sequence is shown with the signal peptide in lowercase font. The presumed TATA box and polyadenylation signals are shown in bold font. The transcription start site at base pair 33 was predicted using the BDGP neural network prediction program ([http://www.fruitfly.org/seq\\_tools/promoter](http://www.fruitfly.org/seq_tools/promoter)). The positions of partially degenerate primers used to amplify the bulk of the coding sequence from cDNA are underlined with arrows showing orientation, whereas the primer used to obtain the 3' portion of the cDNA by anchor PCR is double underlined. The majority of the sequence was obtained from these two overlapping cDNA sequences. Genomic DNA was the template used to amplify 230 bases at the 5' end of the gene by inverse PCR (5' primer shown highlighted in grey) as well as a fragment of the gene containing two introns by using the anchor PCR primer (double underlined) with a 3' primer (grey highlighting).

**Supporting Table 1:** Comparison of observed and predicted tryptic peptide masses for the large snow flea AFP.

Observed Mass (Da) <sup>1</sup>	Predicted Mass (Da) <sup>2</sup>	Residue Range	Missed Cleavages
1046±1	1045.5	1-10	1
4474±4	4472.9	3-57	1
3738±3	3734.6	11-57	0
3487±3	3485.6	58-103	2
3245±3	3243.5	58-100	1
3016±3	3015.4	64-103	1
2774±3	2773.2	64-100	0
4735±5	4735.2	101-162	1
4492±4	4493.1	104-162	0
1424±1	1422.6	163-179	0
n.d.	1733.9	180-203	0

<sup>1</sup> Uncertainty was estimated as half of the width of the ion peak at 50% of maximum intensity, which averaged 0.1%.

<sup>2</sup> The predicted monoisotopic masses were calculated using PeptideMass (Wilkins, M.R., Lindskog, I., Gasteiger, E., Bairoch, A., Sanchez, J.-C., Hochstrasser, D.F., and Appel, R.D. *Detailed peptide characterisation using PEPTIDEMASS - a World-Wide Web accessible tool*; Electrophoresis 18(3-4), 403-408 (1997)).