

d

Angam/1-1020	1 MV <mark>R</mark> LLLLV <mark>S</mark> LGLCCLLA SAD <mark>R PPYK</mark> SSGIYEEEPQLPGASDLSGGVA <mark>S</mark> DEIGVA <mark>PAQ</mark> AVDER <mark>YRLP</mark> TT <mark>SIPIHYDLHLRTEIHR</mark>	85
Analb/1-775	1 MVR PLVIVSIGLWCLLGAGELLAERPPYKTDTVHAEEEP VVSAEQAFVPAQDVDESYRLPKASVPIHYDVHLTTNIHR	79
Angam/1-1020	86 E rt FTGT VGI QLQVVQAT DKLVMH NRGLVMS SAKVS SLPNGVT GAPT LIGD VQYSTDT TFEHITFTSPTILQPGTYLLEVAFQG <mark>R</mark> LAT	174
Analb/1-775	80 D RT FTGT VAI KLQVIEPT DKLVLH QRGLQLSAAR ISSLLAGI DGPAT PIGD VTFATET QRAHLTLTAAMVLQPGYYQVEID YSGRLST) 168
Angam/1-1020	175 DDGFYVSSYVADNGERRYLATTOFESTSARMAFPCYDEPGLKATFTVSITHSLSYKAISNMPOKTTTDIETDMRTTFFEKTPAMSTYLL	263
Analb/1-775	169 DDGFYVSSYVADNGERRYLATTOFESTSARRAFPCYDEPALKATFTITITHGALYGAISNTEOVS-RVVSGESATTKFKTTPLMSSYLI	256
Angam/1-1020	254 A F V V <mark>SD F QLR LS G A QR V V V R PNA F NEAT F A LEAG</mark> VKILK V LDDH LG I PYDT YMP <mark>KLDQ</mark> IA I PD F A A GAME NWGL V T Y REQALL F N PA V S	352
Analb/1-775	257 A F V V SD F LN R GSD KHR V F A R PNA F QELP F A LEAS KLILS V LD EHLY V PYMS YMEKIDQ VA LPD F A A GAME NWGL V T Y REQLL LF N PE V N	345
Angam/1-1020	353 TYRGKTNVATTIAHEYAHQWFGNLVSPEWWEYIWLNEGFATLYEFYALDMAYPGQEYWELFNQQVIQYAMGQDGQASTRPMNWNAATPO	441
Analb/1-775	346 TYRTKTNIATTIAHEYAHQWFGDLVTPQWWEYIWLNEGFATLYEYLATDMAYPGMEYWELFNTQVIQAAMVPDGLASTRPMNYNAATPO	434
Angam/1-1020	442 EISALFDRVAYDKSGSVLNMMRHVLGDDNWKAGLKAYLTDRALQGAVDEQLYAGLQSAIEGKGVLPNGVTVAQIMRTWTNEAGYPVLN	530
Analb/1-775	435 EISALFDRVAYPKSGSVLNMMRHVLGDDNWREGLRIYLLDRALKDATDEDLYNALQQAIEGKGVLPVGVTVGQIMRTWTNEAGYPVLN	523
Angam/1-1020	531 <mark>R R SY DT G D VII S Q E R F Y ND R K</mark> V PN T N I WMI PY NY VH QA K A D F N E F DD F QWL A T KA A R I E T T V PANEWI V F N K Q Q V G Y Y R V NY D E H NWE L	619
Analb/1-775	524 R R SY DT G E I I I S Q E R F Y S D R K I PNAN I WMI PY NY VH QA V A D F N E F DD F KWL A T KA DR FMT T V PAHEWI I F N K Q E V G Y Y R V NY D D H NWE L	612
Angam/1-1020	620 I TNA LHENWA <mark>SIHRINRAQLIDDAYWLARSGRIDIR</mark> VAIR FMTYIRNEREYAPWTAANVA LTYFNNRIRGTAEYHNFLIFVDALIEDIN	708
Analb/1-775	613 I TNALHASYGSVHRINRAQLIDDAYWLARSGRIDIRIAIRIMAYIQHEREYSPWASADVAISYFNSRIRGTEAYEHFSTFVQSIVVPIN	701
Angam/1-1020	709 <mark>S</mark> ILTIDAYS <mark>PDDTILHKYIVQTISTWAC</mark> SMGYT <mark>DC</mark> IM <mark>KT</mark> AALIKAEASGTGPAVHPDIASVTYCYGM <mark>R</mark> SALESEFQYIYRKMMNSKNIA	797
Analb/1-775	702 SILNIEAVDPQDSILHKYIVQTISSWACRIDHPDCIARTKALIATEASG-GAKVHPDIATVTYCYGMRTAGEQE - Y	775
Angam/1-1020 Analb/1-775	798 ERTMLIDSLGCSNNKEFLKAFLTTALGSGTGVEINYRADERRRVVQAIYSGGRTGVDALIEFLMDPALVNEFVSTLSTSTLNSALSAIA	886
Angam/1-1020 Analb/1-775	887 SRTNNVEEMNKLNALITALGSRVNSQTAANLRTTAQANLDWVNGFEGLMLSNFLAEFAAETLLTTTAAPETTTVTAGPTTTTTPSSAV	975
Angam/1-1020 Analb/1-775	975 TTTTGPTSVTTTVVQTTTEEDDGAATIGLSIAALLVSITVHLLMG	10 20

Supplemental Figure S2. Analysis of the Brush Border Microvilli (BBMV)Vesicle **Preparations.** (a) SDS-PAGE analysis of Brush Border Microvilli Vesicles (BBMV) revealed similarities in banding patterns between An. gambiae (AG) and An. albimanus (AA). (b) Immunoblot of An. gambiae (AG) and An. albimanus (AA) BBMV lysates. The upper arrowhead indicates the expected size (kDa) of the predicted full-length AnAPN1 product while the lower arrowhead indicates smaller isoforms. (c) BBMV enzymatic activity was assessed as a quality control check for microvillar aminopeptidase enrichment. The midgut microvilli surface antigen, AnAPN1 was detected in both species and aminopeptidase activity was observed to be enriched in the BBMVs as compared to the original lysates and non-BBMV fractions. Representative data are shown. Only samples meeting these quality control assessments were analyzed by LC-MS/MS. (d) Pairwise amino acid alignment of full-length An. gambiae APN1 and its putative ortholog in An. albimanus. Residues are color coded according to the "Clustalx" option in Jalview (http://www.jalview.org/help/html/colourSchemes/clustal.html). Briefly, identical amino acids are colored by biochemical property as follows: green = polar (NTSQ), cyan = large aromatic polar (HY), blue = hydrophobic (LVIMFWA), red = basic (KR), magenta = acidic (DE), pink = C, orange = G, yellow = P. Non-identical but similar amino acids follow the same color scheme but in a different shade. The signal peptide is underlined in orange, the 135-residue recombinant transmission-blocking vaccine antigen is underlined in red, the M1 peptidase domain is underlined in blue (overlap between antigen and peptidase domain double underlined in red/blue), the threonine-rich mucin domain is underlined in light blue, and the GPI anchor is underlined in black. The alignment was generated using the T-coffee web-based server (http://tcoffee.crg.cat/) and edited with Jalview 2.7 and Adobe Illustrator 14.0.