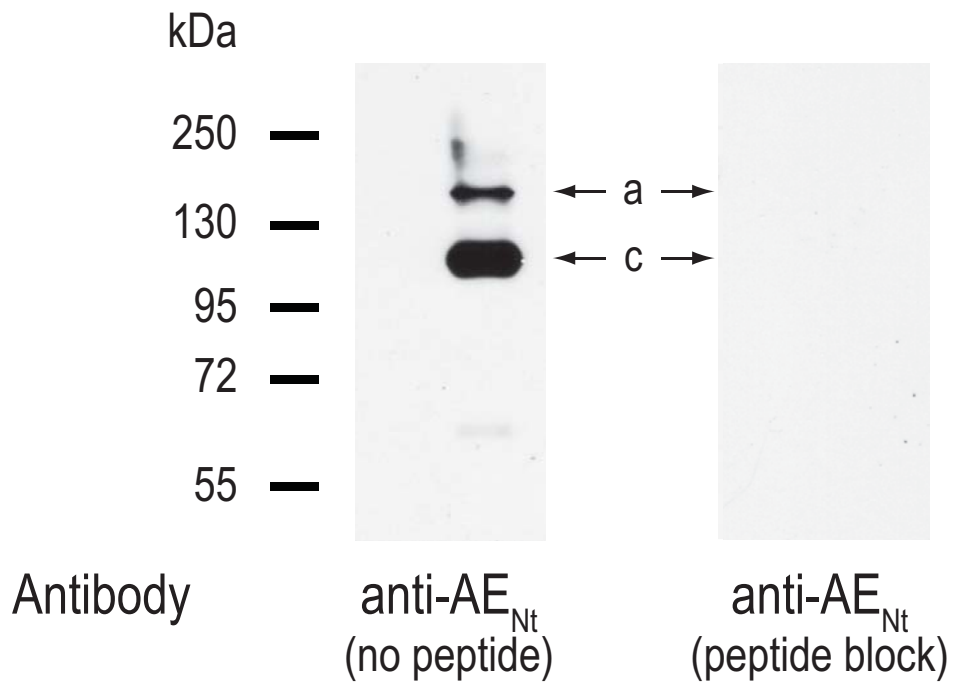
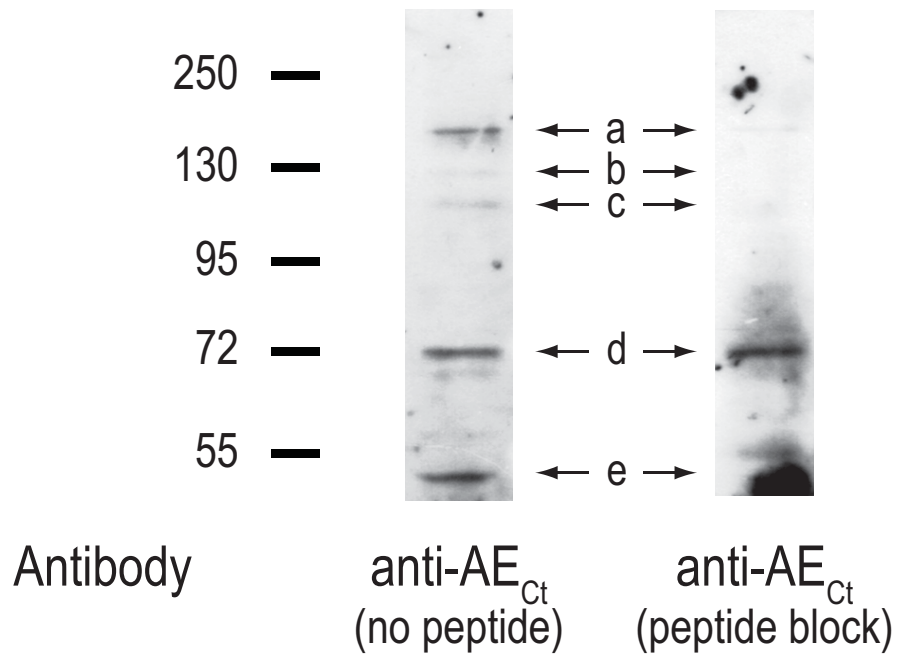


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AeAE	genome	AACGATGATCCCAACCACGTGCAGTTGGACGATGAGATGGAAAAGGTGTTTCGGATCGGTTGGGACCGATAAGGAGCGGTTTCAACTGAAG	360
AeAE	cDNA	CGCCTCAACGATGAGGTGCTGGTGGACAACTCGCCGTTAAAGTACGACGAATCACACCGATCCCGTGGATAATCGTCCGTTACTGTCTCTCG	450
AeAE	genome	CGCCTCAACGATGAGGTGCTGGTGGACAACTCGCCGTTAAAGTACGACGAATCACACCGATCCCGTGGATAATCGTCCGTTACTGTCTCTCG	450
AeAE	cDNA	GCGGCCCTTCGACCGATGGATAGCCCCGCTGCAGGCGGCGGAGGAGCAGGAGGTGGAGGAGGTGGTGGGCCAAGCAGTGGACAGCAGAAC	540
AeAE	genome	GCGGCCCTTCGACCGATGGATAGCCCCGCTGCAGGCGGCGGAGGAGCAGGAGGTGGAGGAGGTGGTGGGCCAAGCAGTGGACAGCAGAAC	540
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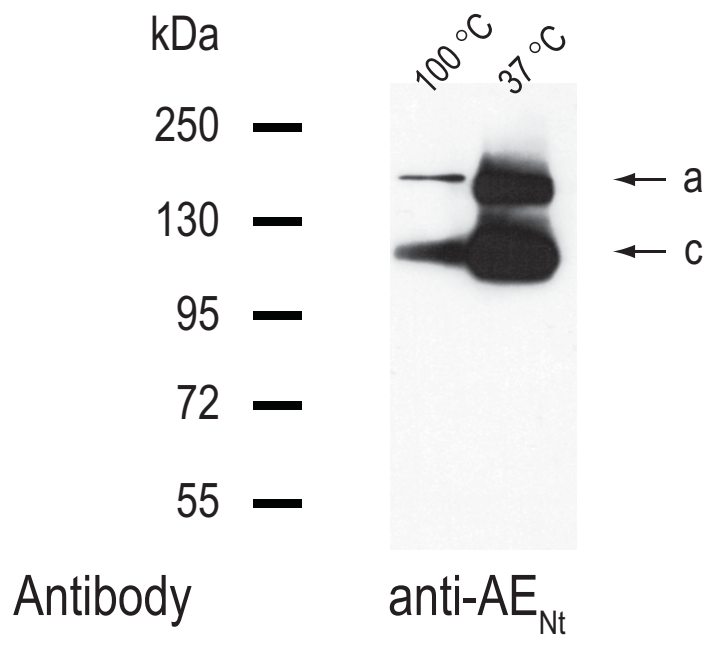
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AeAE	cDNA	AATAGAAACAATAAATTTCAAATAAAATTTGATACTTCTCTACGCTAAATTTCCCTT [*] TGAATGAAATTCATAC	4394
AeAE	genome	AATAGAAACAATAAATTTCAAATAAAATTTGATACTTCTCTACGCTAAATTTCCCTTCGAATGAAATTCATAC	4394



A)



B)



Supplemental Figure Legends

Supplemental Figure 1. Nucleotide sequence of *AeAE* cDNA. The nucleotides of the *AeAE* cDNA cloned in the present study from Malpighian tubules (accession # EU700988) are aligned with the corresponding nucleotides from the genomic DNA of *Aedes* larvae (44), using the ClustalW algorithm (38) within the BioEdit Sequence Alignment Editor software, Version 7 (24). The bold letters underneath asterisks indicate nucleotide differences with the genomic data of *Aedes* that do not affect the predicted amino-acid sequence. The red letters underneath the red bar indicate non-synonymous nucleotide differences with the genomic data. The green and blue bars overlay the first and last 10 nucleotides of the predicted open-reading frame, respectively. The orange and purple arrows overlay the locations of primers 1R and 1F, respectively (Table 1).

Supplemental Figure 2. Effect of antibody pre-adsorption on *AeAE*

immunoreactivity in *Aedes* Malpighian tubules. A) Western blots of crude lysate protein (30 μg per lane) from Malpighian tubules of adult *Aedes* females. The anti-AE_{Nt} antibody (1 $\mu\text{g ml}^{-1}$) was used in the absence (no peptide) or presence (peptide block) of the immunogenic peptide. For the 'peptide block', the anti-AE_{Nt} antibody was pre-adsorbed with its immunogenic peptide for 45 min at 37°C in a 7:1 molar ratio (peptide:antibody) before incubating it with the PVDF membrane. Migrations of the molecular mass markers (in kDa) are indicated to the left. Labeled arrows indicate the locations of the immunoreactive bands in the 'no peptide' blot and their expected locations in the 'peptide block' blot. Note the diminished immunoreactivity of bands 'a'

and 'c' in the 'peptide block' blot. B) Western blots of membrane protein (30 μg per lane) isolated from Malpighian tubules of adult *Aedes* females. The anti-AE_{Ct} antibody (2 $\mu\text{g ml}^{-1}$) was used in the absence (no peptide) or presence (peptide block) of the immunogenic peptide. For the 'peptide block', the anti-AE_{Ct} antibody was pre-adsorbed with its immunogenic peptide for 60 min at 23°C in a 2.5:1 molar ratio (peptide:antibody) before incubating it with the PVDF membrane. Note the reduced immunoreactivity of all bands in the 'peptide block' blot, with the exception of band 'd'. The immunoreactivity at the bottom of the 'peptide block' blot is background staining not associated with band 'e'.

Supplemental Figure 3. Effect of denaturation temperature on *AeAE*

immunoreactivity in *Aedes* Malpighian tubules. Western blots of crude lysates (30 μg protein per lane) from Malpighian tubules of adult *Aedes* females. The lysates were denatured in standard Laemmli sample buffer (36) at either 100°C or 37°C for 5 min. The anti-AE_{Nt} antibody (1 $\mu\text{g ml}^{-1}$) was used to detect *AeAE* immunoreactivity. Migrations of the molecular mass markers (in kDa) are indicated to the left. Labeled arrows indicate the locations of the immunoreactive bands. Note that denaturing at 37°C results in the anomalous migration of both bands 'a' and 'c' (relative to 100°C) as indicated by their broad, diffuse immunoreactivity. Band 'a' appears to be fully denatured at 100°C, whereas band 'c' requires the addition of urea to be fully denatured (see **Figure 7B**).