Identification of the *abdominal-A* homologue from *Aedes aegypti* and structural comparisons among related genes

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A homologue of *abdominal-A* has been isolated from the mosquito *Aedes aegypti* by exploiting sequence homology to homeobox containing genes from *Drosophila melanogaster*. A genomic DNA library was prepared from fourth instar larvae and screened at reduced stringency with a probe derived from the *Drosophila* homeobox gene, *Antennapedia*. Analysis of a plaque purified positive clone revealed a 1.76 kb *EcoRI* fragment which hybridised strongly to both the *Antp* probe and to a synthetic oligonucleotide whose sequence corresponded to the consensus for the most conserved region of the *Antp*, *Ubx* and *fz* homeoboxes. This fragment was digested in separate reactions with *HaeIII* and *AluI* and the products ligated into M13. Recombinant clones carrying overlapping fragments were sequenced on both strands using the dideoxy chain termination technique. The mosquito sequence carries a complete homeobox which is 81% identical to that from *abdominal-A* of *Drosophila*. Conceptual translation of this sequence yields a protein which is identical to ABD-A of *Drosophila* (1) throughout the homeodomain (Figure 1). 5' to the homeodomain, the fruit-fly and predicted mosquito ABD-A proteins are identical for a further five amino acids before similarity ends abruptly at a point where the *Drosophila* sequence has an 11–13 kb intron (1). At this point, the mosquito sequence closely resembles a consensus splice acceptor sequence [Py₄TTGCAG/G] reported by Breathnach and Chambon (2). 3' to the homeodomain, the fruit-fly and predicted mosquito ABD-A proteins are identical for a further nine amino acids and then similarity ends abruptly at a point where the *Drosophila* sequence has a 70 bp intron (1). At this point, the mosquito sequence matches exactly the splice donor consensus [PyAG/GTPuAGT] (2). This 3' intron in the mosquito gene, however, must be in excess of 530 bp since there is no further similarity between the *Drosophila abd-A* cDNA and the mosquito sequence downstream of this site.

Recently, *abd-A* homologues have also been isolated from *Schistocerca* (3), *Manduca* (4), *Bombyx* (5), *Tribolium* (6) and *Apis* (7). The predicted ABD-A proteins for all but one of these insects are identical throughout the homeodomain (Figure 1). The one variant appears to be the *Apis* sequence in which tyrosine replaces phenylalanine at position 22. All seven sequences are identical for five amino acids 5' of the homeodomain, ending at known or predicted intron/exon boundaries. Downstream of the homeodomain, the first nine amino acids (seven for *Apis*) are identical in all of the insects and, at this point, the *Drosophila*

nucleotide sequence has an intron of 70 bp whilst that of *Aedes* has a predicted intron in excess of 530 bp. This 3' intron is absent from all of the other insects and presumably has arisen relatively recently in insect evolution, after the divergence of the lepidoptera and the diptera. Whether this intron is specific to the diptera would require a knowledge of abd-A structure in other insect orders, particularly the aphaniptera (fleas) which post-date the lepidoptera and predate the diptera (8).

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Figure 1. Predicted sequence of the *Aedes aegypti* ABD-A protein and its alignment with homologous genes in *Drosophila*, *Schistocerca*, *Manduca*, *Bombyx*, *Tribolium* and *Apis*. Amino acid identity is denoted by asterisks. The homeodomain is boxed and the position of the 3' intron in the *Drosophila* and *Aedes* sequences marked by a vertical bar.