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Identification and characterization of cuticular collagens in the human filarial parasite *Onchocerca volvulus*

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Abstract

Cuticular collagen is a major structural protein of nematode cuticles encoded by a large multigene family. The cuticle of nematode parasites provides a barrier between the worm and its hosts. This study is focused on the collagen genes expressed in the third larval (L3) and the molting third larval (3M) developmental stages of a human filarial parasite, *Onchocerca volvulus*, the major causative agent of onchocerciasis, or African River Blindness, one of the four leading causes of blindness worldwide. ^ Based on the River Blindness Genome Project launched in Steven Williams' laboratory, EST analysis has identified 45 distinct cuticular collagen clusters derived from L3 and 3M stages, showing differential expression patterns through the life cycle stages. The collagens appear to be the most abundant gene family in the *O. volvulus* genome, reflecting their critical role in parasite development. ^ The genomic structures of the three most highly expressed cuticular collagen genes (*Ov-col-1*, *Ov-col-9*, and *Ov-col-10*) have been characterized. These three genes are similar in the size of the genomic copy of the gene, transcripts and open reading frame, but belong to three distinct subfamilies based upon the feature of conserved cysteine residues. ^ Due to its extremely high expression level and 3M-stage upregulation, *Ov-col-9* is considered a putative candidate for vaccine design. The gene is 2.6 kb long, has 4 introns, a 1017 by transcript, and encodes a protein 289 amino acids in length. Western blot analyses identified the native COL-9 protein at a mass between 64 kDa and 80 kDa, about 3-fold the predicted size, indicating a nonreducible covalent crosslink between the COL-9 and other polypeptides under reducing conditions. Immunogold EM localized the COL-9 to the L3 body channel, to the 3M hypodermis, and to uterine microfilariae in the adult female. Analysis of the human IgG and IgG subclass responses to COL-9

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demonstrated statistically similar levels between putatively immune (PI) and infected (INF) group except slightly elevated in IgG1 by the INF group.

^

Subject Area

Biology, Molecular|Health Sciences, Pathology

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