Association of axonal processes with contractile cells in salivary glands of the tick Ixodes ricinus

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When attached to a vertebrate, host tick salivary glands (SG) produce a variety of pharmacologically active compounds which significantly compromise the host immune system and thus represent a great route for tick-borne pathogens transmission. Tick SG is a highly innervated tissue, and over the last decade various axonal projections expressing signaling molecules or their receptors were identified within the SG acini (alveoli) [1, 2]. In presented study, we employed combination of immunogold labelling (Tokuyasu technique) and electron tomography to visualize the neuropeptide SIFamide along with its receptor, the neuropeptide pigment dispersing factor (PDF) and the invertebrate specific D1-like dopamine receptor (InvD1-L) in SG acini type II and III. We found, that three types of axonal processes expressing, SIFamide, PDF and InvD1-L were in close proximity to the acinar duct enclosed by sheets of highly convoluted myoepithelial (MC) and adluminal cells (AC). The SIFamide receptor was detected in MC close to the SIFamide-specific axon terminals. MC, containing numerous bundles of microtubules, is thought to possess contractile properties mediating the fluid expulsion from the acinus to the associated ducts. Stellate shaped AC, with unknown function, were located under the basement membrane of the type II and III acini, forming basket-like network around the secretory cells. The use of hafnium chloride *en-block* staining during freeze substitution instead of convectional uranyl acetate greatly enhanced the TEM contrast of the examined structures (Fig.1) allowing the spatial reconstruction of both MC and AC with their associated axonal processes in larger volumes.

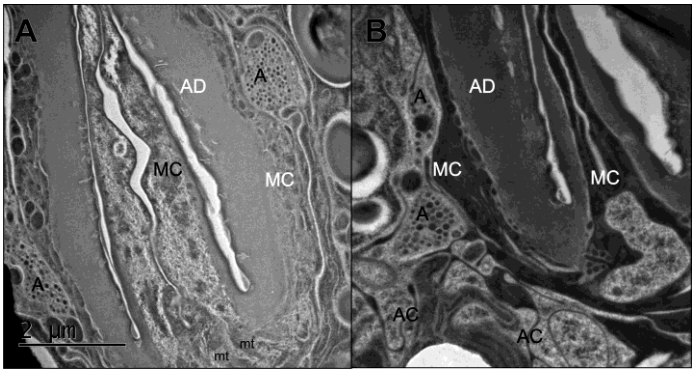

Figure 1. Salivary glands of acini type II. Unfed *Ixodes ricinus* female. Samples were prepared by HPF/FS technique using OTO protocol followed by *en-bloc* staining with either uranyl acetate (A) or hafnium chloride (B). Axonal projections (A) were enclosed by myoepithelial cell (MC) and were also associated with adlumenal cells (AC). Acinar duct (AD), microtubules (mt)

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[2] L. Šimo et al., J. Insect. Physiol. 58 (2012) 459-456.

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