



The natural insect peptide *Neb*-colloostatin induces ovarian atresia and apoptosis in the mealworm *Tenebrio molitor*.

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Background: The injection of Neb-colloostatin into *T. molitor* females causes gonadoinhibitory effects on ovarian development. This peptide inhibits intercellular space formation (patency) in follicular epithelium and results in slowed vitellogenesis, delayed ovulation, reduced number of eggs laid and presumably cell death in the terminal follicles. However, as does the form of cell death in the terminal follicle, the mode of action of Neb-colloostatin remains unknown.

Results: We tested Neb-colloostatin for a sterilizing effect on females of *Tenebrio molitor*. We report that injection of nanomolar doses of Neb-colloostatin induce ovarian follicle atresia in 4-day old females during their first gonadotropic cycle. Light microscope observations revealed morphological changes in the ovary: after Neb-colloostatin injection the terminal oocytes are significantly smaller and elicit massive follicle resorption, but the control terminal follicles possess translucent ooplasm in oocytes at different stages of vitellogenesis. A patency is visible in follicular epithelium of the control vitellogenic oocytes, whereas peptide injection inhibits intercellular space formation and, in consequence, inhibits vitellogenesis. Confocal and electron microscope examination showed that peptide injection causes changes in the morphology indicating death of follicular cells. We observed F-actin cytoskeleton disorganization, induction of caspase activity, changes in chromatin organization and autophagic vacuole formation. Moreover, the apical cytoplasm of follicular cells is filled with numerous free ribosomes, probably indicating a higher demand for protein biosynthesis, especially in preparation for autophagic vacuole formation. On the other hand, the process of polyribosomes formation is inhibited, indicating the contributing effect of this hormone.

Conclusion: Neb-colloostatin induces atresia in the mealworm ovary. Degeneration of *T. molitor* follicles includes changes in morphology and viability of follicular cells, and oosorption as a consequence of these changes.

Słowa kluczowe

Insect ovary, Neb-colloostatin, Gonadoinhibitory peptide, Follicle atresia, Apoptosis, Autophagy

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