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DISSERTATION SUMMARY

Expression and a novel function of filamin-240 in lamellocyte development in *Drosophila*

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Drosophila has a very effective innate immune system with striking similarities to innate immunity in vertebrates. Cellular immune defense in *Drosophila* is mediated by three different classes of hemocytes: the plasmatocytes, the lamellocytes and the crystal cells. Lamellocytes represent a unique population of hemocytes, both in terms of morphology and function. The lamellocytes are large, flat, adherent cells, involved in encapsulation and neutralization of intruders or of abnormally developed tissues, too large to be engulfed by plasmatocytes. These cells are nonphagocytic and their cytoplasmic constituents are relatively sparse. Very few lamellocytes are observed in wild-type *Oregon-R* larvae. Parasitization by the Hymenopteran wasp, *Leptopilinia boulardi,* initiates the rapid differentiation of lamellocytes, which subsequently adhere to and surround the egg capsule**.** This capsule subsequently melanizes, locally generating cytotoxic compounds and creating a barrier. Bacterial challenge does not induce lamellocyte differentiation, but this can be induced artificially by objects placed in the larval haemocel that are too large to be phagocytated (Evans et al. 2003). They develop from small (8-10 μm), round stem cells and become large (40-50 μm) flattened cells capable of binding and encapsulating large foreign particles. The development of lamellocytes from stem cells to effector cells involves two major steps: first, the differentiation of small stem cells to the immediate precursors of the fully differentiated large cells. This step requires cell division and is terminated by the expression of the L1 antigen. Second, the terminal differentiation of the immediate precursors to the large flattened cells. This process does not require cell division but is characterized by the expression of L4 and L6 antigens and the most characteristic morphological changes. In this work we studied the role of a lamellocyte antigen in lamellocyte development, which turned out to be a unique, actin-binding component of the cytoskeleton in lamellocytes.

By screening *Drosophila* cDNA expression libraries with an antibody reacting preferentially with lamellocytes, positive clones were obtained and 5 DNA fragments were isolated. The analysis of the generated sequences showed that these correspond to 8 exons of *cheerio* gene, which encodes filamin, a protein that organizes filamentous actin in networks and stress fibers and also anchors various transmembrane proteins to the active cytoskeleton. All of our isolated cDNA sequences contain a so-called filamin-folding domain, a consensus motif profile, generated from the 25 existing filamin domain sequences. Filamin exists in two isoforms in *Drosophila*, as a result of alternative splicing. The longest form, filamin-240, contains an N-terminal actin-binding domain and so far it has been known to be specific for the ring canals in the ovaries. Our studies revealed that this isoform is expressed in lamellocytes too, and is possibly involved in the organisation in the actin-network during the terminal stage of lamellocyte development. To get further insight into the role of filamin-240 in lamellocyte development, we studied the possible effect of the lack of this protein in the *cher1/cher1* loss of- function type mutant stock. The results show that the lack of the isoform results in abnormal number and proportion of developing lamellocytes, suggesting that filamin-240 is involved in the regulation of lamellocyte differentiation, perhaps as an organizer of the rearrangement of the cytoskeleton.

References

Andó I, Laurinyecz B, Nagy I, Márkus R, Rus F, Váczi B, Zsámboki J, Fehér L, Gateff E, Hultmark D, Kurucz É (2003) Our archaic heritage: the innate immunity. The cellular immunity of *Drosophila*. Magy Immunol 2:39-45.

Evans CJ, Hartenstein V, Banerjee U (2003) Thicker than blood. Conserved mechanisms in *Drosophila* and vertebrate hematopoiesis. Dev Cell 5:673-690.

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