Dissertation abstract

NILSSON HELÉN Pigment Organelle Localization in Fish Melanophores. Zoophysiology Zoology Göteborg University, Sweden.

Fish chromatophores are able to rapidly aggregate pigment organelles to the cell center or to disperse them uniformly throughout the cell. This process is under neuro-humoral regulation and is the reason behind the physiological color change reaction. The present study aimed to increase our knowledge of roles of microtubules and microfilaments in aggregation, maintenance of the aggregated state, dispersion and in maintenance of the uniformly dispersed state.

Primary cultures of melanophores from Atlantic cod, *Gadhus morhua*, were used for immunocytochemistry, microinjections and video-enhanced motion analysis of pigment organelle dynamics. Pigment organelle movement was analysed in melanophores and erythrophores present on scales from Spotted triplefin, *Grahamina capito*. An *in vitro* assay for pigment organelle aggregation was developed using microtubule asters from oocytes from the surf clam, *Spisula solidissima,* and liberated pigment organelles from killifish, *Fundulus heteroclitus*, melanophores. Aggregation was stimulated by noradrenaline. Dispersion was stimulated by depletion of noradrenaline or by increasing intracellular cAMP levels using forskolin.

The results allowed me to propose a principal model for the relative contribution of the microtubule-dependent dynein, the antagonistic putative kinesin 2, the putative actin-dependent myosin V, timing of their regulation and signal pathways involved in melanophore pigment organelle positioning. In this model, microtubules and cytoplasmic dynein are responsible for the noradrenaline-mediated aggregation and maintenance of the aggregated state. During aggregation, kinesin and myosin are inactivated; kinesin by decreased cAMP and myosin by a different noradrenaline-mediated signal pathway. Dispersion is initiated by increased intracellular cAMP that inactivates dynein and activates the putative kinesin 2, the motor protein responsible for dispersion. To disperse pigment uniformly throughout the cell, dynein and myosin are reactivated by a cAMP-independent feedback signal system located in cell periphery that becomes activated upon arrival of pigment organelles. The thesis presents the hypothesis that myosin can be regulated to be inactive during both aggregation and dispersion in order to optimize the rates of the bidirectional microtubule-dependent transport. Such scenario would be of great advantage for the fish, since the color change reaction would be faster that way.

In addition, aggregating pigment organelles presenting dynein motors on their surfaces, were found to organize short microtubules into asters in cod melanophores. Upon redispersion, these ordered microtubules became randomly distributed as the pigment. This shows the power of organelles to self-organize the track upon which they move along and may be an important mechanism for dynein-dependent organisation of the cytoplasm in other cells as well.

Key words: melanophore, erythrophore, organelle transport, dynein, kinesin, myosin, actin, cAMP, noradrenaline, norepinephrine, self assembly, signal transduction, crosstalk

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