1. Abstract

Refractive index gradient in the cubozoan eye: specific gene expression analysis

Lenses are spread through animal kingdom as an improvement of different eye types. Despite conservation of some key regulators and shared use of photopigment opsin, eyes and their lenses develop by variable mechanisms impeaching their monophyletic origin. *Tripedalia cystophora* (*T. c*), a cubozoan jellyfish, is an emerging new model for studying eye evolution. The presence of advanced lens-containing eyes (firstly incident within metazoans in this phylum of *Cnidaria*), the two types of lesser eyes, the use of *pax* gene and vertebrate type of phototransduction cascade for eye establishment make this jellyfish an useful tool for comparing eye development and different evolutionary strategies.

We focused on the lenses of *T.c.* and studied formation of their refractive index gradient to reveal its mechanism. Using new antibodies raised against J1 and J2 crystallins (proteins of the *T. c.* lens), TEM and histology we found that graded refractive index is of protein origin and formed by unequal accumulation of proteins (particularly J1 and J2 crystallins) in different layers of the lens. We have shown that J1 crystallin occurs also in the lesser eyes (*ocelli*) suggesting how the lens mass can evolved. The synthesis of J2 crystallin in lens development has been examined. Furthermore we have prepared J2 crystallin fused to GFP and described the cellular localization of this novel crystallin. The experiment confirmed its cytoplasmatic localization and thus a biologically sensible expression regarding its role as a crystallin. Finally we have determined the expression of several interesting genes using RNA *in situ* hybridization. For example, we determined the expression of selenoprotein O, one of many genes lost in insects or nematodes but conserved among *T. c.* and vertebrates. RNA *in situ* hybridization for MITF, transcription factor crucial for eye development, revealed its presence in the eyes of *Tripedalia cystophora.*.

In summary, it seems that *T. c.* and vertebrate share much more than a fine lens within an eye. Recent studies, this one included, shed light on the conservation of cnidarian and vertebrate genomes and support the hypothesis of the maintenance of early evolving gene set in these two lineages and massive loss in the others, such as of insect or nematodes.

Key words: eye, lens, J1 crystallin, J2 crystallin, evolution, refractive index gradient, gene set conservation

Klíčová slova: oko, čočka, J1 krystalin, J2 krystalin, evoluce, gradient refrakčního indexu, konzervace genových sad