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Ocean acidification promotes cellular burst on photosynthetic (kleptoplastic) sea slugs

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Ocean acidification is known to trigger deleterious effects on several marine photosynthetic invertebrates [1]. Photosymbiosis, whereby photosynthetic microorganisms or organelles live inside an animal (host) is widespread in the marine biota, underlying a wide range of ecologically and biogeochemically significant processes that remain largely unclear [1]. One of the most remarkable symbiosis is between sacoglossan molluscs and algal chloroplasts [2]. These organisms are able to “steal” functional chloroplasts (termed kleptoplasts) from their algal prey and keep them functional inside digestive diverticula [2]. The aim of this study was to investigate the impact of environmental hypercapnia on cellular structures of kleptoplastic animals. *Elysia viridis* (Sacoglossa) were exposed to different pH conditions (pH 8, 7.5, 6.8 and 6.1) for 24 h. Six animals were anaesthetized per treatment [3] and the integrity of kleptoplasts was determined (*in vivo*) under optical and stereomicroscope (Figs. 1, 2). Morphological modifications to the normal condition of digestive diverticula, chloroplasts and mortality, were checked every 8 h Under normal conditions (pH 8.0) the symbiotic chloroplasts are packed tightly in the tubule cells, particularly close to cell walls (Fig. 2A, arrow). Tubule cells ramify throughout the body, giving its green appearance. The same condition was verified for pH 7.5 (Fig. 2B). Sea slugs subjected to pH 6.8 presented fragmented clusters of chloroplasts and overall color variation (Figs. 1, 2C). Mortality was only verified in animals exposed to pH 6.1 (100% mortality), presenting severe impacts at cellular level after 24 h of exposure, including cellular burst (Fig. 1). Chloroplasts and their plastoglobuli spread across host cytoplasm (Fig. 2D, arrow) and cellular layers surrounding the plastid were absent. These preliminary results suggest that *E. viridis* may resist to environmental hypercapnia down to pH 6.8, where the first sings of cellular modifications where detected. At pH 7.5 (future scenario, 2100 [4]) kleptoplastic sea slugs retain their endosymbionts in opposition to other well-established photosymbiotic groups.

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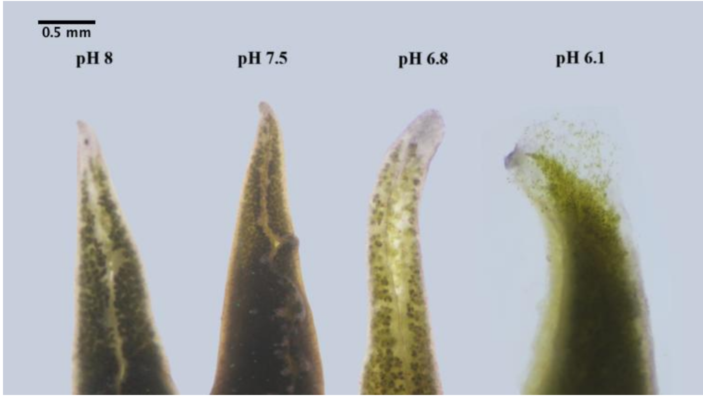

Fig. 1 – Posterior sections of *E. viridis* exposed to different pH conditions for 24h (8, 7.5, 6.8 and 6.1) observed under optical stereomicroscope; green coloration is given by chloroplasts and evident burst is shown under pH 6.1. Scale bar: 0.5 mm.

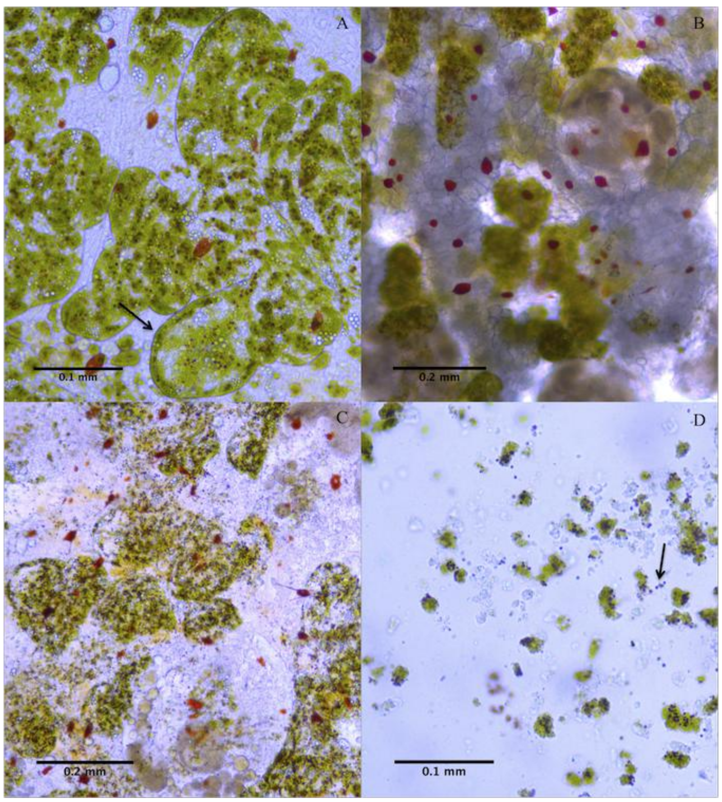

Fig. 2 – Digestive diverticula from posterior sections of *E. viridis* exposed to different pH conditions for 24 h (A – pH 8; B – pH 7.5; C – pH 6.8; D – pH 6.1), observed *in vivo* under optical microscopy. Normal pH conditions (A) show numerous algal plastids within a cell (black arrow, A), contrasting with the absence of cellular layers surrounding the plastids, and plastoglubuli (black arrow) spread across host cytoplasm at the lowest pH (D). Scale bars: 0.1 mm (A, C) and 0.2 mm (B, D).

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