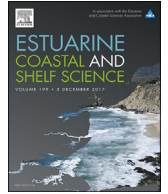




Contents lists available at ScienceDirect

## Estuarine, Coastal and Shelf Science

journal homepage: [www.elsevier.com/locate/ecss](http://www.elsevier.com/locate/ecss)

## Target gene expression studies on *Platynereis dumerilii* and *Platynereis* cfr *massiliensis* at the shallow CO<sub>2</sub> vents off Ischia, Italy

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## ARTICLE INFO

## Article history:

Received 28 October 2016

Received in revised form

3 November 2017

Accepted 12 November 2017

Available online xxx

## Keywords:

Ocean acidification

Gene expression

CO<sub>2</sub> vents

Polychaeta

*P. dumerilii*

Sibling species

## ABSTRACT

Many studies predict negative effects of ocean acidification on marine organisms, potentially leading to loss of biodiversity and ecosystem function. Research on species inhabiting naturally high pCO<sub>2</sub> environments, such as volcanic CO<sub>2</sub> vents, offers an opportunity to understand the molecular mechanisms involved in high pCO<sub>2</sub> regulation. Here we investigate the relative expression of *NADH dehydrogenase*, *sodium-hydrogen antiporter (NHE)*, *carbonic anhydrase (CA)* and *paramyosin* genes from two non-calcifying sibling Nereididae polychaetes species, *Platynereis* cfr *massiliensis*, collected in the shallow CO<sub>2</sub> vents off Ischia (Italy; 40°43'52.0"N 13°57'46.2"E and 40°43'55.5"N 13°57'48.4"E), and *P. dumerilii* collected in an area nearby (40°43'34.51"N; 13°57'35.7"E). The origin of the worms was confirmed using restriction enzyme digest. *NHE* and *paramyosin* expressions were both significantly increased in *P. dumerilii* relative to the *P. cfr massiliensis* vent populations. Furthermore, a seven day laboratory transfer experiment to lower/higher pCO<sub>2</sub> conditions was conducted to investigate the effects on the short term gene expression. The transfer experiment of the non-vent worms to high pCO<sub>2</sub> conditions showed no significant effect on any of the genes analysed, however, two genes (*NADH dehydrogenase* and *NHE*) from worms of the vent population were significantly down-regulated under low pCO<sub>2</sub>. These findings will help to gain further insights into the cellular mechanisms affected by pCO<sub>2</sub> changes in two polychaete species.

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### 1. Introduction

Since the Industrial revolution, atmospheric carbon dioxide (CO<sub>2</sub>) levels have increased from 280 ppm in pre-industrial times to over 380 ppm, contributing to ocean acidification (OA), also termed the 'other CO<sub>2</sub> problem' (IPCC, 2013). Over the last decade, OA has become the focus of numerous studies examining the effects of changing seawater conditions on marine organisms (Gattuso and Hansson, 2011). From pre-industrial times to the 1990s, the ocean surface mean pH has decreased from 8.2 to 8.07 and models predict a further pH drop by 0.4 pH units by the year 2100 (Caldeira and Wickett, 2003; Gattuso and Lavigne, 2009). To predict the impact of OA on organisms, it is essential to understand its effects on molecular processes that form the basis of physiological functions.

Several transcriptomic and proteomic studies have been

conducted in which marine organisms have been exposed to experimental OA conditions. The majority of these studies focus on differential expression in calcifying organisms such as sea urchins, corals, barnacles, oysters and coccolithophores (Todgham and Hofmann, 2009; O'Donnell et al., 2010; Martin et al., 2011; Parker et al., 2011; Stumpp et al., 2011; Tomanek et al., 2011; Wong et al., 2011; Dineshram et al., 2012, 2013; Moya et al., 2012; Jones et al., 2013; Pespeni et al., 2013; Vidal-Dupiol et al., 2013). These studies reveal that genes involved in biomineralisation, lipid/energy metabolism, acid-base regulation and ion transport processes exhibit the most significant changes in gene expression patterns in response to OA (O'Donnell et al., 2010; Dineshram et al., 2012; Moya et al., 2012; Pespeni et al., 2013; Vidal-Dupiol et al., 2013; Wong et al., 2011).

The genus *Platynereis* comprises non-calcifying worms that are abundant off Ischia Island (Italy) in the Mediterranean Sea, including natural CO<sub>2</sub> vents (Ricevuto et al., 2014; Gambi et al., 2016). Based on reciprocal transplant experiments and genetic examinations, Calosi et al. (2013) concluded that the vent-

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