

Impact of Ocean Acidification on Gene Expression Dynamics during Development of the Sea Urchin Species *Heliocidaris erythrogramma*

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Abstract : Marine invertebrate species with calcifying larvae are especially vulnerable to ocean acidification (OA) caused by rising atmospheric CO₂ levels. Acidic conditions can delay development, suppress metabolism, and decrease the availability of carbonate ions in the ocean environment for skeletogenesis. These stresses often result in increased larval mortality, which may lead to significant ecological consequences including alterations to the larval settlement, population distribution, and genetic connectivity. Importantly, many of these physiological and developmental effects are caused by genetic and molecular level changes. Although many studies have examined the effect of near-future oceanic pH levels on gene expression in marine invertebrates, little is known about the impact of OA on gene expression in a developmental context. Here, we performed mRNA-sequencing to investigate the impact of environmental acidity on gene expression across three developmental stages in the sea urchin *Heliocidaris erythrogramma*. We collected RNA from gastrula, early larva, and 1-day post-metamorphic juvenile sea urchins cultured at present-day and predicted future oceanic pH levels (pH 8.1 and 7.7, respectively). We assembled an annotated reference transcriptome encompassing development from egg to ten days post-metamorphosis by combining these data with datasets from two previous developmental transcriptomic studies of *H. erythrogramma*. Differential gene expression and time course analyses between pH conditions revealed significant alterations to developmental transcription that are potentially associated with pH stress. Consistent with previous investigations, genes involved in biomineralization and ion transport were significantly upregulated under acidic conditions. Differences in gene expression between the two pH conditions became more pronounced post-metamorphosis, suggesting a development-dependent effect of OA on gene expression. Furthermore, many differences in gene expression later in development appeared to be a result of broad downregulation at pH 7.7: of 539 genes differentially expressed at the juvenile stage, 519 of these were lower in the acidic condition. Time course comparisons between pH 8.1 and 7.7 samples also demonstrated over 500 genes were more lowly expressed in pH 7.7 samples throughout development. Of the genes exhibiting stage-dependent expression level changes, over 15% of these diverged from the expected temporal pattern of expression in the acidic condition. Through these analyses, we identify novel candidate genes involved in development, metabolism, and transcriptional regulation that are possibly affected by pH stress. Our results demonstrate that pH stress significantly alters gene expression dynamics throughout development. A large number of genes differentially expressed between pH conditions in juveniles relative to earlier stages may be attributed to the effects of acidity on transcriptional regulation, as a greater proportion of mRNA at this later stage has been nascent transcribed rather than maternally loaded. Also, the overall downregulation of many genes in the acidic condition suggests that OA-induced developmental delay manifests as suppressed mRNA expression, possibly from lower transcription rates or increased mRNA degradation in the acidic environment. Further studies will be necessary to determine in greater detail the extent of OA effects on early developing marine invertebrates.

Keywords : development, gene expression, ocean acidification, RNA-sequencing, sea urchins

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