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Survivors of the Sea: Investigating the genomics and survival strategies of the diatom *Skeletonema marinoi*

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Akademisk avhandling för filosofie doktorsexamen i Naturvetenskap med inriktningen marina vetenskaper, som med tillstånd från Naturvetenskapliga fakulteten kommer att offentligt försvaras fredagen den 13e oktober 2023 kl. 10:00 i Korallrevet, rumsnummer 3401, Natrium, Institutionen för marina vetenskaper, Medicinaregatan 7B, 413 90 Göteborg.

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Abstract

Diatoms are an ecologically important group of phytoplankton, responsible for around 20% of global primary production. One of the features contributing to their success is their ability to form resting stages, a response to adverse conditions in which they enter a dormant state and sink to the sediment. While some resting stages may germinate upon the return of favourable conditions and become resuspended, others can become buried in the dark, anoxic sediment for long periods, retaining their viability. Resting stages of some species have been revived in the lab after over a century, yet the precise mechanisms behind this ability to survive so long in this state is not well understood. One such species capable of forming long-lived resting stages is *Skeletonema marinoi*. The draft reference genome of this marine diatom was recently assembled and annotated, and thus my thesis has involved using this genome as a tool to explore the species and investigate its resting stage survival.

In **paper I**, the use of ancient diatom DNA (i.e. DNA obtained from revived diatom resting stages) in research was reviewed, with particular attention to *S. marinoi*. While much insight has already been gained using this resource, developments in several fields have led to exciting prospects for future research.

In **papers II** and **III**, I compared gene expression between vegetative cells of *S. marinoi*, and resting stages induced by exposure to darkness, anoxia, and nitrogen starvation. After re-exposure to nitrate, the resting stages' gene expression was measured using RNA sequencing over the course of six months. In **paper II**, I performed a differential expression analysis to determine which processes were active in this 'dormant' life stage. Much of the resting stages' central metabolism underwent shifts in expression, and genes involved in protein synthesis were upregulated throughout the resting stage. In **paper III**, I examined the RNA sequencing results from the perspective of allelic expression bias, and identified a handful of genes showing significant shifts in such bias between vegetative and resting cells. Of these, I noted several that formed complete pathways related to the formation of diatoms' silica cell walls.

Lastly, in **paper IV**, I developed a bioinformatic tool – Bamboozle – to identify novel, intraspecific genomic barcoding loci, capable of tracking the relative abundance of multiple strains of *S. marinoi* in co-culture over time. This tool was initially applied to 54 strains of *S. marinoi*, revealing loci that enabled subsequent tracking of strains during an artificial evolution experiment. Bamboozle has been further developed to accommodate haploid organisms, and was used to identify intraspecific barcoding loci in the model green alga *Chlamydomonas reinhardtii*.

The results of this thesis provide insights and highlight further questions regarding diatom resting stages, as well as providing a novel tool for studying these fascinating phytoplankton. Application of additional omics methods to diatom resting stages in future, and testing of Bamboozle on natural diatom populations, should both lead to further understanding of this taxon.