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Whole Transcriptomic Analysis Provides Insights into Molecular Mechanisms for Toxin Biosynthesis in a Toxic Dinoflagellate *Alexandrium catenella* (ACHK-T)

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Abstract: Paralytic shellfish toxins (PSTs), a group of neurotoxic alkaloids, are the most potent biotoxins for aquatic ecosystems and human health. Marine dinoflagellates and freshwater cyanobacteria are two producers of PSTs. The biosynthesis mechanism of PSTs has been well elucidated in cyanobacteria; however, it remains ambiguous in dinoflagellates. Here, we compared the transcriptome profiles of a toxin-producing dinoflagellate *Alexandrium catenella* (ACHK-T) at different toxin biosynthesis stages within the cell cycle using RNA-seq. The intracellular toxin content increased gradually in the middle G1 phase and rapidly in the late G1 phase, and then remained relatively stable in other phases. Samples from four toxin biosynthesis stages were selected for sequencing, and finally yielded 110,370 unigenes, of which 66,141 were successfully annotated in the known databases. An analysis of differentially expressed genes revealed that 2866 genes altered significantly and 297 were co-expressed throughout the four stages. These genes participated mainly in protein metabolism, carbohydrate metabolism, and the oxidation-reduction process. A total of 138 homologues of toxin genes were identified, but they altered insignificantly among different stages, indicating that toxin biosynthesis might be regulated translationally or post-translationally. Our results will serve as an important transcriptomic resource to characterize key molecular processes underlying dinoflagellate toxin biosynthesis.

Keywords: dinoflagellate; *Alexandrium catenella*; paralytic shellfish toxins; toxin biosynthesis; cell cycle; RNA-seq

1. Introduction

Dinoflagellates are not only important primary producers in marine ecosystems, but also major causative agents producing paralytic shellfish toxins (PSTs) [1]. Comprising saxitoxin (STX) and its recorded 57 analogs, PSTs act as the most potent and selective voltage-gated sodium channel blockers [2,3]. They can cause human neural system syndromes, occasionally even death, and lead to approximately 2000 toxicosis cases annually worldwide [4]. Owing to their severe threat to human health and serious damage to the marine ecosystem, STX and its biosynthesis mechanism have aroused wide public concern.

Recently, the gene cluster *sxt* for STX synthesis has been elucidated in cyanobacteria, another well-known organism producing STX, followed by the identification of *sxt* clusters in several other cyanobacterial species [5–8]. With a length ranging from 25.7 kb to 36 kb, the clusters encode 24 to 32 proteins that participate in toxin biosynthesis [9]. Integrating their functions and the intermediates produced by each reaction, the biosynthesis pathway is proposed and refined in cyanobacteria [5,7,9].