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Review

Marine dinoflagellate proteomics: Current status and future perspectives☆

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ABSTRACT

Dinoflagellates are not only the important primary producers and an essential component of the food chain in the marine ecosystem, but also the major causative species resulting in harmful algal blooms (HABs) and various shellfish poisonings. Although much work has been devoted to the dinoflagellates, our understanding of them is still extremely limited owing to their unusual features. Proteomics, a large-scale study of the structure and function of proteins in complex biological samples, has been introduced to the study of marine dinoflagellates and has shown its powerful potential with regard to revealing their physiological and metabolic characteristics. However, the application of proteomic approaches to unsequenced dinoflagellates is still in its infancy and faces considerable challenges. This review summarizes recent progress in marine dinoflagellate proteomics and discusses the limitations and prospects for this approach to their study.

Scientific question: The dinoflagellates are the major causative agent responsible for harmful algal blooms and paralytic shellfish poisoning around the world. However, our understanding of them is still extremely limited owing to their unusual features, such as large genome size and permanently condensed chromosomes, which impedes the monitoring, mitigation and prevention of HABs.

Technical significance: Proteomics, a large-scale study of the structure and function of proteins in complex biological samples, has been introduced to the study of marine dinoflagellates and has shown its powerful potential with regard to revealing their physiological and metabolic characteristics.

Scientific significance: This review summarizes recent progress in marine dinoflagellate proteomics with regard to methodology, cell growth, toxin biosynthesis, environmental stress, cell wall and surface, and symbiosis, and discusses the limitations and prospects for this approach to dinoflagellate study.

This article is part of a Special Issue entitled: Proteomics of non-model organisms.

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Contents

1. Introduction

The dinoflagellates, a large group of unicellular protists, are prominent members of the plankton in marine ecosystems and freshwater habitats, as well as in the benthic and sea ice environment [1]. They are not only the key primary producers and an important component of the food chain and of coral reef building, but also the major causative species of harmful algal blooms (HABs) in the ocean [2]. Moreover, many dinoflagellate species are able to produce natural toxins which result in various types of shellfish poisoning and cause adverse impacts on the environment, the economy and human health [3]. Recently, dinoflagellate-causing HABs have attracted considerable attention because of the global increase of HABs in terms of their frequency, intensity and geographic distribution [4].

The most prominent feature of dinoflagellates is their unusual genome size and organization. Estimates of dinoflagellate DNA content range from 3 to 250 pg·cell^{-1}, corresponding to approximately 3000–245,000 Mb [5]. Moreover, the chromosomes of most dinoflagellates are permanently condensed throughout the cell cycle, and attached to the nuclear envelope during cell division. These unique features have brought serious challenges to the study of dinoflagellates [6], resulting in the lack of dinoflagellate genetic information, which impedes our understanding of them and, consequently, the monitoring, mitigation and prevention of HABs.

Proteins are vital parts of living organisms, since they participate in essentially every structure and activity of life, *i.e.* cell growth, proliferation and homeostasis. Therefore, it is logical that the study of proteins should help uncover the various physiological metabolic pathways of cells. Proteomics involves a large-scale study of the structure and function of proteins in a complex biological sample [7] and, in contrast to conventional biochemical approaches, provides effective strategies and tools for profiling and identifying a number of proteins at a time, allowing simultaneous isolation and identification of hundreds to thousands of proteins in one sample. In the past few years, the proteomic approach has been applied to the study of dinoflagellates, and has shown its powerful potential with regard to revealing their essential physiological and metabolic characteristics. In this paper, we review the advancement of proteomics in the study of marine dinoflagellates (see Table 1), and discuss the challenges and opportunities of this approach for their future study.

2. Proteomics of marine dinoflagellates

2.1. Strategies for dinoflagellate proteomic study

In the past few years, the gel-based proteomic approach is the main work flow applied to the study of marine dinoflagellates (Fig. 1). In this work flow, one or two-dimensional electrophoresis (1-DE or 2-DE) is used to separate proteins, followed by matrix-assisted laser desorption–ionization time-of-flight mass spectrometry (MALDI-TOF MS) analysis or electrospray ionization mass spectrometry (ESI MS) analysis. The protein samples are separated either by molecular mass (MW) directly, or by isoelectric focusing (IEF), followed by gel separation based on MW. However, there are several technical drawbacks of the current 2-DE method, such as poor identification of hydrophobic, extreme acidic/alkaline side, and high-molecular-weight (~200 kDa) proteins, which limit the application and intensive exploration of the whole-cell proteome [8]. Recently, a quantitative proteomic approach, two dimensional fluorescence difference gel electrophoresis (2-D DIGE) has been introduced to dinoflagellate study [9–11]. Compared to the traditional 2-DE approach, a direct labeling of the lysine groups on proteins with cyanine dyes is involved before IEF proceeds to 2-D DIGE, and the use of a pooled internal standard increases the robustness of statistical analysis by reducing biological and experimental variations.

Usually, the differentially expressed or targeted proteins are subjected to digestion into peptides using trypsin or other enzymes. After that, peptide analysis can be conducted through two operational approaches. A unique peptide mass fingerprint (PMF), measured as mass over charge (m/z), is provided by each peptide after MS analysis. Next, tandem mass spectrometry (MS/MS) analysis obtains a peptide fragment fingerprint (PFF)