Broad Spectrum and Mode of Action of an Antibiotic Produced by *Scytonema* sp. TISTR 8208 in a Seaweed-Type Bioreactor

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**ABSTRACT**

A photobioreactor was constructed using anchored polyurethane foam strips (1 × 1 × 40 cm) fixed onto a stainless-steel ring to prevent flotation, as a biomass support material (BSM). This type of reactor was named a seaweed-type bioreactor. A filamentous cyanobacterium, *Scytonema* sp. TISTR 8208, which produces a novel cyclic dodecapeptide antibiotic, was immobilized in seaweed-type photobioreactor and cultivated with air containing 5% CO$_2$ sparged at a gas flow rate of 250 mL/min under illumination at a light intensity of 200 μmol photon m$^{-2}$s$^{-1}$. The antibiotic produced in the seaweed-type photobioreactor was purified by HPLC and examined regarding its spectrum and mode of action. The antibiotic effectively inhibited the growth of Gram-positive bacteria, pathogenic yeasts, and filamentous fungi, but it had only a weak effect on Gram-negative bacteria. Scanning electron micrograph analysis showed that the most characteristic change was swelling of the cells after exposure to the antibiotic. The antibiotic seems to alter the conformation of the microbial cell membrane, thereby changing its permeability, leading to osmotic shock.

**Index Entries:** Cyanobacteria; *Scytonema*; photobioreactor, antibiotic; polyurethane foam.

**INTRODUCTION**

Prokaryotic cyanobacteria can be grown photoautotrophically using light energy and CO$_2$, which is a major greenhouse gas partially responsible for global warming. Recently developed technology enables CO$_2$ to be
recovered from the emission gases of steam power plants. It is environmentally and economically important to produce valuable substances photoautotrophically from CO₂ by cyanobacteria. Various strains of cyanobacteria are known to produce intracellular and extracellular metabolites, with diverse biological activities, which are classified into two groups (1). The first group contains lactones, phenols, and acids such as cyanobacterin, a γ-lactone antialgal from *Scytonema hofmanni* (2), an antibacterial, brominated phenol from *Calothrix brevissima* (3), and an antimicrobial, O-methyl acid from various shallow-water varieties (4). The second group, which is the major one, consists of nitrogen-containing substances, such as a cytotoxin, malyngamide D from *Lyngbya majuscula* (5), and an antialgal and antimycotic, hapalindole A from *Hapalosiphon fontinalis* (6). The authors previously screened nine strains and five genera of cyanobacteria for antibiotic production, and a filamentous cyanobacterium, *Scytonema* sp. TISTR 8208, had the strongest activity against the bacteria tested (7). Stable immobilization of the cyanobacterium, which was shown to produce a cyclic peptide antibiotic, could be established by utilizing a fibrous biomass support material. The optimal medium composition for the production of the antibiotic was determined (8), and a seaweed-type photobioreactor was then constructed for the continuous cultivation of the cyanobacterium (9). Stable production of the antibiotic was achieved in the bioreactor for 16 d.

The aim of the present study was to characterize the antibiotic produced by immobilized *Scytonema* sp. TISTR 8208, to determine its spectrum and mode of action toward susceptible microorganisms.

**MATERIALS AND METHODS**

**Cultivation of cyanobacterium**

*Scytonema* sp. TISTR 8208, which was obtained from the culture collection of the Thailand Institute of Scientific and Technological Research Center (TISTR), was cultivated in modified BGA medium (MBGA) (7). The seaweed-type photobioreactor used is illustrated in Fig. 1. Details of the bioreactor dimensions were given previously (9). Cyanobacterial cells of about 1.6 g dry wt were inoculated into the 2.3-L bioreactor containing 2.0 L of MBGA medium. The bioreactor was incubated in a 30 ± 1°C incubation room. The basal conditions of light illumination and gas (air containing 5% CO₂) flow rate were 200 µmol photon m⁻²s⁻¹ and 250 mL/min, respectively, unless otherwise stated. A linear bank of fluorescent lamps was used on one side of the bioreactor.

**Purification of Antibiotic**

The culture supernatant was concentrated under reduced pressure at 30°C, and the antibiotic was extracted with methanol. After vacuum drying, the sample was then partitioned into a solvent system of CHCl₃: