The Characterization of a New Metabolite from a *Trichodesmium* Bloom

**ABSTRACT**

Our laboratory has been investigating blooms of *Trichodesmium*, a genus of ecologically important, nitrogen-fixing cyanobacteria, collected from Padre Island in the Gulf of Mexico. *Trichodesmium* species are an underexplored biological source of cyanobacteria—a taxa that has been shown to produce chemically diverse secondary metabolites. With our focus on the isolation and structure characterization of new bioactive marine natural products, our research group has discovered over 25 new-to-science compounds over the past three years from these blooms. UV and mass spectrometry-guided isolation of *Trichodesmium* chromatography fractions were utilized to isolate a new metabolite. Isolation of this metabolite was carried out by means of silica gel vacuum liquid chromatography, solid phase extraction chromatography, and high-performance liquid chromatography. The structure of this molecule was determined using high-resolution mass spectrometry along with 1D and 2D NMR analysis. The molecule contains interesting structural features such as a lactone ring and an aldehyde functional group. This molecule is similar to 11 others isolated by the Bertin laboratory in a group named the trichophycins. However, this new metabolite does not feature a chlorovinylindene moiety, which is a hallmark of the trichophycins. The isolation and characterization of this molecule adds to our laboratory's pure compound library and provides a new entity to screen for biological activity. Furthermore, inspection of the structure of this new metabolite provokes questions as to its biosynthesis.

**Extraction and Fractionation**

We obtained this fraction through silica vacuum liquid chromatography of the total extract, where “F” was the most polar fraction. Fraction “I” was then loaded over a C18 column using solid phase extraction chromatography to obtain fractions I1-I4. Fraction I1 was further fractionated over C18 to give five fractions (I1A-I1E). While previous analysis used bioassay-guided isolation (neuro-2A cytotoxicity), our study used UV and MS-based isolation.

**HPLC of Fraction I-1-DE**

**1H NMR (500 MHz, DMSO-d6)**

**UV/Vis Spectra**

**Mass Spectrometry**

**STRUCTURE OF PI-2014-I-1-DE-4-A-1**

In order to determine this structure, we also used further 2D NMR experiments including COSY and TOCSY (correlations shown in bold). In addition to this we compared the NMR experiments to those from trichophycin F. In total, we isolated less than 0.5 mg of this metabolite. HRMS and UV measurements are consistent with proposed structure. Additional NMR experiments planned to address questions of configuration.

**STRUCTURE OF TRICHOPHYCIN F**

Our new molecule is very similar to trichophycin F (pictured above). However, in the new structure the chlorovinylindene group is replaced with an aldehyde, and there is one additional unsaturation in the new molecule’s carbon backbone.

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**Trichodesmium Images**

- Bloom in Gulf of Mexico
- Microscopic filaments

**Trichodesmium biomass shipped to M. Bertin**

Bioassay-guided isolation

**New metabolite**

**Trichophycin F**

MS/MS of purified molecule compared to similar molecule trichophycin F also discovered by The Bertin Lab. Many fragment peaks from the new compound are two fewer than trichophycin F fragment peaks which is evidence of the similarity of the two compounds, and suggests one extra unsaturation in the new metabolite’s core structure.