Cellular Location of Amphidinol Implicates a Role in Guanine Crystal Formation in *Amphidnium carterae Hulburt*

Oral

Toxins & Other Microalgal Metabolites

Allen R Place [1], Peter Mojzes [2], Avital Wagner [3], Benjamin A. Palmer [4].

(1) University of Maryland Center for Environmental Science,, IMET, Professor, 701 East Pratt Street, Baltimore, US 21202.
(2) Charles University, Institute of Physics, Faculty of Mathematics and Physics,, Prague, Czechia.
(3) Ben-Gurion University of the Negev, Department of Chemistry, Beer-Sheb, Israel.
(4) Ben-Gurion University of the Negev, Department of Chemistry, Beer-Sheb, Israel.

place@umces.edu

Since the original isolation of the polyhydroxypolyene molecule, amphidinol 1, from *Ampidinium carterae Hulburt*, its hemolytic and antifungal activity was apparent. The subsequent seven congeners all exhibited antifungal, hemolytic, cytotoxic and ichthyotoxic activities and a strong surfactant property with the ability to perturb membranes. The sterol dependency of this membrane disruption was further shown with calcein leakage from liposomes containing cholesterol and that the two tetrahydropyran rings (which are antipodal) take a hairpin conformation permitting insertion into the bilayer. Further, amphidinol interacts with membrane sterols through the strict molecular recognition of the stereochemistry of the sterol 3-OH group. Amphidinol is produced through a polyketide synthase (PKS) pathways starting with the photorespiration product glycolate. Finally, recent lipid bilayer studies have shown amphidinol 3 forms different types of sterol-aided channels in a concentration dependent manner. Amphidinol is produced through a polyketide synthase (PKS) pathways starting with the photorespiration product glycolate. Amphidinol has a unique Raman spectrum, which we used to localize it within living cells near the vacuole where guanine crystals form. The identification was unambiguously supported by confocal Raman and transmission electron microscopy as well as stable isotope

labeling. Using surface plasmon resonance with surfaces coated with amphidinol and karmitoxin we were able to show reversible binding of guanine and xanthine at pH 7.4 but no binding at pH 4.0. We would like to propose that the production of these polyhydroxypolyene molecules *in situ* may assist in the crystallization of guanine, an important nitrogen storage source.

Palabras Clave

(1) Keywords: Polyketides, (2) Toxins, (3) Guanine.

Financiamiento:

Funding: Czech Science Foundation No. 21-26115S.; ERC Starting Grant (grant no. 852948, “CRYSTA-LEYES”) awarded to BAP.