UNIVERSITY OF CALIFORNIA, SAN DIEGO

The Natural Products Chemistry of Marine Ascidians: Structural Elucidation and Molecular Modeling Studies of Novel Secondary Metabolites

A dissertation submitted in partial satisfaction of the requirements for the degree

Doctor of Philosophy in Oceanography

by

Heonjoong Kang

1994

ABSTRACT OF DISSERTATION

The Natural Products Chemistry of Marine Ascidians:
Structural Elucidation and Molecular Modeling Studies of Novel Secondary Metabolites

by

Heonjoong Kang
Doctor of Philosophy in Oceanography
Scripps Institution of Oceanography
University of California, San Diego, 1994

Professor William H. Fenical, Chairman

Ascidians are well-known producers of amino acid-derived secondary metabolites. Recent chemical and biological investigations of taxonomically diverse ascidians have revealed a variety of pharmacologically potent natural products and the importance of these metabolites in the survival of physically vulnerable ascidians in predator-rich habitats. However, at issue are the real sources of novel secondary metabolites isolated from marine ascidians, which are related to symbiosis due to complex associations of the ascidians with microbial symbionts. Furthermore, rapidly developing resistance of infectious diseases and cancers toward traditional antibiotics and anti-cancer drug needs unprecedented novel natural products. Despite great advance in

separation science, marine natural products research in ascidians was relatively limited to nonpolar compounds. Furthermore, the aquatic environment implicates the significance of hydrophilic molecules in defensive adaptations and symbiosis. Therefore, the main objective of this research was to investigate biosynthetic diversity and limitation of ascidian secondary metabolism with emphasis of identification of hydrophilic bioactive molecules and unprecedented carbon skeletons. In this way, a broader understanding of ascidian secondary metabolism, and a chemical data base for studies of chemical ecology, could be established.

A variety of chromatographic methods, such as high performance liquid chromatography and high speed countercurrent chromatography, were used to isolate polar compounds. Extensive spectroscopic techniques, including a variety of two-dimensional NMR experiments and mass spectrometric methods as well as computer-molecular modeling, were utilized to elucidate their structures.

This research resulted in the structural elucidation of 25 new secondary metabolites in 10 different structural classes, including 8 new carbon skeletons from marine sources. A high diversity of chemical structures along with various bioactivities, such as cytotoxicity against colon cancer cells, antimicrobial activity, and anti inflammatory activity, were demonstrated. Several natural products showed cytotoxic activities directed against a drug-resistant human colon cell line, HCT 116. Some of these compounds are known to inhibit an enzyme which is frequently used as a chemotherapeutic target. In addition to them, some had antibacterial and antiinflammatory activities. However, not every compound was bioactive. Perhaps some of them may explain the chemical defense mechanism of marine ascidians against predators.

ACKNOWLEDGEMENTS

I would like to express my gratitude to several people for their help in this work. First of all, I thank my advisor, Dr. Fenical, for his guidance and mentorship during Ph. D. research. He made me open my eyes as a scientist about how to approach problems and how to conduct research. Most importantly he taught me human relationship. I strongly feel that I am lucky to meet him. Dr. Faulkner, he gave invaluable advice and discussion about marine natural products. He also taught me how to conduct molecular modeling studies. I greatly appreciated his teaching. I thank all of my committee members for their reading my manuscript and advice.

Mr. Jensen collected all of my sample and conducted microbial isolation and fermentation. I really appreciate his help. Dr. Robert Jacobs at UC, Santa Barbara kindly performed antiinflammatory tests in his lab. Mr. Cummings conducted the rest of biotesting. I thank them so much. Without their help this research could not be possible. I am also extremely grateful to Dr. Monniot for her work on identification of many chemically interesting ascidians I investigated.

I also thank the mass spectrometry center at UC, Riverside, especially Dr. New. Without his help and suggestion, many projects could not be accomplished.

I am grateful to Drs. Wright, Trischman, Pathirana, and Mr. Brueggeman for NMR maintenance, good friendship, teaching me 2-D NMR and structure searching. Dr. Trischman helped me so much in many ways. At the time of my arrival, she kindly explained everything. Also she gave encouragement and wonderful friendship for several years at SIO. I thank everybody in Dr. Fenical and Dr. Faulkner's Lab. for their suggestion through the group meeting and joyable relationship.

I also thank all present and past members of Korean Scientific Seminar members in San Diego for their invaluable discussion and suggestion. I am also grateful to my exadvisor, Dr. Kim. His guidance and training of me at the previous graduate school made me everything easier during my Ph.D. study in all aspects. Dr. Park gave me encouragement, guidance, and support. His door was widely open anytime for me. I really thank him for everything. Special thanks go to Dr. Shin and Dr. Lee for their advice and encouragement. In the most difficult first two years, they helped me to adapt SIO as quickly as possible. Whenever I had difficult time, they encouraged me and

suggested some ways. I also thank Drs. D.-K. Lee, Y. Lee, Y.-H. Lee, H. Jeong, and Mr. D. Kim and D. Min for wonderful friendship.

Finally, I greatly thank my parents, parents-in-law, brothers, and sister for their endless love, support, and encouragement. Specially, I thank my wife Sooyun for her love and support. I would like to give all of joy from this research to my wife and daughter, Nah-Young.

Finally, I would like to acknowledge Korean Government and California Sea Grant for financial support.

CALIFORNIA SEA GRANT
Fenical; R/MP-54