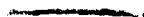


Marine Biotechnology

Volume 1 Pharmaceutical and Bioactive Natural Products

Edited by

David H. Attaway

National Oceanic and Atmospheric Administration 
Washington, D.C.

and

Oskar R. Zaborsky

*National Research Council
National Academy of Sciences
Washington, D.C.*

PLENUM PRESS • NEW YORK AND LONDON

Library of Congress Cataloging-in-Publication Data

Marine biotechnology / edited by David H. Attaway and Oskar R. Zaborsky.

p. cm.

Includes bibliographical references.

Contents: v. 1. Pharmaceutical and bioactive natural products

ISBN 0-306-44174-8 (v. 1)

1. Marine biotechnology. 2. Marine pharmacology. 3. Natural products. I. Attaway, David H. II. Zaborsky, Oskar R.

TP248.27.M37M365 1993

660'.6--dc20

92-43270

CIP

ISBN 0-306-44174-8

©1993 Plenum Press, New York
A Division of Plenum Publishing Corporation
233 Spring Street, New York, N.Y. 10013

All rights reserved

No part of this book may be reproduced, stored in a retrieval system, or transmitted in any form or by any means, electronic, mechanical, photocopying, microfilming, recording, or otherwise, without written permission from the Publisher

Printed in the United States of America

Contributors

Ami Ben-Amotz, National Institute of Oceanography, Israel Oceanographic and Limnological Research, Tel-Shikmona, Haifa 31080, Israel

Matthew W. Bernart, College of Pharmacy, Oregon State University, Corvallis, Oregon 97331

Mary A. Bober, Marine Science Institute, and Department of Biological Sciences, University of California, Santa Barbara, California 93106

Bruce F. Bowden, Department of Chemistry and Biochemistry, James Cook University, Townsville, Queensland 4811, Australia

Martha Cohen-Parsons, Department of Chemistry, University of Illinois, Urbana, Illinois 61801

Brent R. Copp, Department of Medicinal Chemistry, University of Utah, Salt Lake City, Utah 84112

Phil Crews, Department of Chemistry and Biochemistry, and Institute of Marine Sciences, University of California, Santa Cruz, California 95064

Marianne S. de Carvalho, Marine Science Institute, and Department of Biological Sciences, University of California, Santa Barbara, California 93106

D. John Faulkner, Scripps Institution of Oceanography, University of California at San Diego, La Jolla, California 92093-0212

William Fenical, Marine Research Division, Scripps Institution of Oceanography, University of California at San Diego, La Jolla, California 92093-0236

Mark P. Foster, Department of Medicinal Chemistry, University of Utah, Salt Lake City, Utah, 84112

William H. Gerwick, College of Pharmacy, Oregon State University, Corvallis, Oregon 97331

Lisa M. Hunter, Department of Chemistry and Biochemistry, and Institute of Marine Sciences, University of California, Santa Cruz, California 95064

Chris M. Ireland, Department of Medicinal Chemistry, University of Utah, Salt Lake City, Utah, 84112

Robert S. Jacobs, Marine Science Institute, and Department of Biological Sciences, University of California, Santa Barbara, California 93106

Peer B. Jacobson, Marine Science Institute, and Department of Biological Sciences, University of California, Santa Barbara, California 93106

Paul R. Jensen, Marine Research Division, Scripps Institution of Oceanography, University of California at San Diego, La Jolla, California 92093-0236

Leonard A. McDonald, Department of Medicinal Chemistry, University of Utah, Salt Lake City, Utah, 84112

Koji Nakanishi, Suntory Institute for Bioorganic Research, Shimamoto, Mishima-gun, Osaka, Japan, and Department of Chemistry, Columbia University, New York, New York 10027

Yoko Naya, Suntory Institute for Bioorganic Research, Shimamoto, Mishima-gun, Osaka, Japan

Isabel Pinto, Marine Science Institute, and Department of Biological Sciences, University of California, Santa Barbara, California 93106

Derek C. Radisky, Department of Medicinal Chemistry, University of Utah, Salt Lake City, Utah 84112

Donald W. Renn, FMC Corporation, Rockland, Maine 04841

Kenneth L. Rinehart, Department of Chemistry, University of Illinois, Urbana, Illinois 61801

Mukesh K. Sahni, Department of Chemistry, William Paterson College, Wayne, New Jersey 07470

Francis J. Schmitz, Department of Chemistry and Biochemistry, University of Oklahoma, Norman, Oklahoma 73019

Gurdial M. Sharma, Department of Chemistry, William Paterson College, Wayne, New Jersey 07470

Lois S. Shield, Department of Chemistry, University of Illinois, Urbana, Illinois 61801

Yuzuru Shimizu, Department of Pharmacognosy and Environmental Sciences, College of Pharmacy, The University of Rhode Island, Kingston, Rhode Island 02881

J. Christopher Swersey, Department of Medicinal Chemistry, University of Utah, Salt Lake City, Utah 84112

Kazuo Tachibana, Suntory Institute for Bioorganic Research, Shimamoto, Mishima-gun, Osaka, Japan; *current address*: Department of Chemistry, University of Tokyo, Hongo, Bunkyo, Japan

Stephen I. Toth, Department of Chemistry and Biochemistry, University of Oklahoma, Norman, Oklahoma 73019

Allen B. Williams, Marine Science Institute, and Department of Biological Sciences, University of California, Santa Barbara, California 93106

Preface

Biotechnology may be defined as the application of scientific and engineering principles to the processing of materials by biological agents to provide goods and services (Bull *et al.*, 1982, p. 21) or as any technique that uses living organisms (or parts of organisms) to make or modify products, to improve plants or animals, or to develop microorganisms for specific use (OTC, 1988). In line with these broad definitions we can consider marine biotechnology as the use of marine organisms or their constituents for useful purposes in a controlled fashion. This series will explore a range of scientific advances in support of marine biotechnology. It will provide information on advances in three categories: (1) basic knowledge, (2) applied research and development, and (3) commercial and institutional issues. We hope the presentation of the topics will generate interest and interaction among readers in the academic world, government, and industry. This first volume examines chemical and biological properties of some natural products that are useful or potentially useful in research and in the chemical and pharmaceutical industries. One chapter describes a system for producing such substances on a large scale.

Biotechnology incorporates molecular biology in order to go beyond traditional biochemical technology such as the production of antibiotic drugs from bacterial cultures in bioreactors. Development of the technology for production of antibiotics in this way resulted from fundamental advances in chemistry, pharmacology, microbiology, and biochemical engineering. It is likely that molecular biology will be used to improve the efficiency of this technology. One of the objectives of this series is to emphasize the importance of interdisciplinary science and to encourage improvement of the environment for it in academic institutions. Another objective is to demonstrate the rich array of materials and processes in the marine world that have no terrestrial counterparts and to suggest that the workings

of this diverse world should be explored aggressively. The majority of plant and invertebrate phyla on earth are either exclusively or predominantly marine. Many of these marine organisms are so poorly understood that they have yet to be fully described and named. Developing a thorough understanding of the ocean's biological materials and processes will provide new information for technological development. The authors who have contributed to this volume make this point eloquently.

We acknowledge the enthusiasm and cooperation with which the advisory board and authors have endorsed this series and its first volume.

David H. Attaway
Oskar R. Zaborsky

REFERENCES

- Bull, A. T., Holt, G., and Lilly, M. D., 1982, *Biotechnology: International Trends and Perspectives*, Organisations for Economic Co-operation and Development, Paris.
- OTC (Office of Technology Assessment), 1988, *New Developments in Biotechnology 4: U.S. Investment in Biotechnology*, Office of Technology Assessment, Congress of the United States, Washington, D.C.

Contents

Chapter 1

<i>Biomedical Potential of Marine Natural Products</i>	1
---	---

*Chris M. Ireland, Brent R. Copp, Mark P. Foster,
Leonard A. McDonald, Derek C. Radisky, and J. Christopher Swersey*

1. Introduction	1
2. Marine Natural Products That Act at Membrane Receptors	4
3. Antitumor Compounds	12
4. Tumor Promoters	16
5. Anti-inflammatory/Analgesic Compounds	23
6. Antiviral Agents	25
7. Metabolites Which Affect Microfilament-Mediated Processes	28
8. The Future	30
References	30

Chapter 2

<i>Isolation, Structural and Mode-of-Action Studies on Bioactive Marine Natural Products</i>	45
---	----

Yoko Naya, Kazuo Tachibana, and Koji Nakanishi

1. Introduction	45
2. Crustacean Molt-Inhibiting Regulators	47
2.1. General	47

2.2. Materials and EBI Assay	49
2.3. Isolation of EBI	50
2.4. Enzymatic Transformation of 3-OHK into XA	51
2.5. Physiological Change Induced by Eyestalk Ablation	52
2.6. <i>In Vivo</i> Effects of L-3OHK and XA on Crayfish Molting	53
2.7. Hormonal Characteristics of EBI; Titer Changes of MH and EBI	53
2.8. Inhibition Mechanism	56
2.9. Conclusion	57
3. Shark Repellants in the Defense Secretion of Pardachirid Soles	59
4. Tunichromes, the Blood Pigments of Tunicates (Sea Squirts)	66
4.1. General	66
4.2. Isolation and Structure of Tunichromes	66
4.3. Synthesis of Tunichromes	68
4.4. Assays for Metal Ions and Tunichrome	68
4.5. Intracellular Vanadium Environment	70
4.6. Mechanistic Considerations of Vanadium Assimilation	71
4.7. Biological Roles of Tunichrome and Vanadium	72
References	72

Chapter 3

Pharmacological Studies of Novel Marine Metabolites 77

*Robert S. Jacobs, Mary A. Bober, Isabel Pinto, Allen B. Williams,
Peer B. Jacobson, and Marianne S. de Carvalho*

1. Introductory Remarks	77
2. Pharmacological Models of Cellular and Molecular Processes	78
2.1. Eicosanoid Biosynthesis in Tunicates	79
2.2. Eicosanoid Biosynthesis in Coralline Red Algae	86
3. Site and Mechanism of Action of Marine Natural Products	87
3.1. Fuscoside: A Novel Inhibitor of Leukotriene Biosynthesis Isolated from the Caribbean Soft Coral <i>Eunicea fusca</i>	87
3.2. Mechanism of Action of Scalaradial	90
3.3. Multidrug Resistance in CH ^R -C5 Cells	92
4. Summary	96
References	97

Chapter 4*Eicosanoids and Related Compounds from Marine Algae* 101*William H. Gerwick and Matthew W. Bernart*

1. Introduction	101
2. Eicosanoids and Related Compounds from Marine Algae	102
2.1. Historical	102
2.2. Cyanobacteria	103
2.3. Rhodophyta	107
2.4. Chlorophyta	134
2.5. Phaeophyta	139
3. Conclusion	146
3.1. Metabolic Themes	146
3.2. Role of Eicosanoids in Marine Plants	147
3.3. Marine Eicosanoids and Related Compounds in Medicine and Physiological Research	148
References	149

Chapter 5*Marine Proteins in Clinical Chemistry* 153*Gurdial M. Sharma and Mukesh K. Sahni*

1. Introduction	153
2. Vitamin B ₁₂ -Binding Proteins of Marine Organisms	154
2.1. Vitamin B ₁₂ -Binding Proteins of the Horseshoe Crab	157
2.2. Vitamin B ₁₂ -Binding Proteins of Marine Phytoplankton	168
3. Limulus Proteins for the Detection of Endotoxins	169
4. Marine Lectins	172
5. Conclusion	176
References	177

Chapter 6*Medical and Biotechnological Applications of Marine Macroalgal
Polysaccharides* 181*Donald W. Renn*

1. Introduction	181
2. Physiological Activities	182

2.1. Antiviral	183
2.2. Anticoagulant, Antithrombic	183
2.3. Antitumor, Antimetastatic	184
2.4. Anti-Ulcer	184
2.5. Cholesterol-Lowering, Antilipemic	184
2.6. Immunoregulator, Cellular Response Modifier	184
2.7. Hemostats, Wound Dressings	185
2.8. Cervical Dilator	185
3. Model Systems for New Drug Screening	185
3.1. Use of Physiological Activities to Develop Model Systems for New Drug Screening	185
4. Biomedical Assay Applications	186
4.1. Serum Protein Electrophoresis	187
4.2. Immunological Assays	187
4.3. Microbial Growth	188
4.4. Genetic Disorder Detection by Restriction Fragment Analysis	188
5. Biotechnology-Oriented Applications	189
5.1. Media for Gene Fragment Separations	189
5.2. Media for Gene Mapping	190
5.3. Cell Immobilization	191
6. Projections for the Future	191
References	192

Chapter 7

Antitumor and Cytotoxic Compounds from Marine Organisms 197

Francis J. Schmitz, Bruce F. Bowden, and Stephen I. Toth

1. Introduction	197
2. Polyketides	199
2.1. Fatty Acid Metabolites	199
2.2. Long-Chain Acetylenes	199
2.3. Aliphatic Ester Peroxides	201
2.4. Prostanoids	202
2.5. Complex Polyketides	203
2.6. Macrolides	207
3. Terpenes	220
3.1. MonoterpeneS	220
3.2. Sesquiterpenes	221
3.3. Diterpenes	226
3.4. Sesterterpenes	236

3.5. Triterpenes	238
3.6. Sterols	240
4. Nitrogen-Containing Compounds	248
4.1. Amides of Fatty Acids	248
4.2. Tyrosine-Based Metabolites	248
4.3. Other Amides	250
4.4. Pyrroles	254
4.5. Imidazoles	254
4.6. Indoles	256
4.7. Pyridines	260
4.8. Quinolines and Isoquinolines	262
4.9. Quinolizidines and Indolizidines	265
4.10. Prianosins/Discorhabdins	265
4.11. Polycyclic Aromatic Alkaloids: Acridine Alkaloids	267
4.12. Guanidines	270
4.13. Peptides and Depsipeptides	270
4.14. Nucleosides	284
4.15. Glycoproteins	284
4.16. Proteins	285
5. Polysaccharides	285
6. Summary	286
References	289
Review Articles on Cytotoxic/Antitumor Compounds from Marine Organisms	308

Chapter 8

Antiviral Substances	309
-----------------------------------	------------

Kenneth L. Rinehart, Lois S. Shield, and Martha Cohen-Parsons

1. Introduction	309
2. Marine-Derived Antiviral Program at the University of Illinois	310
3. Antiviral Assays	310
4. Very Active Antiviral Agents	311
4.1. Didemnins	312
4.2. Eudistomins	314
4.3. Mycalamides and Onnamide A	316
4.4. Avarol and Avarone	317
4.5. Ptilomyacin A and Crambescidins	317
4.6. Hennoxazoles	318
4.7. Thrysiferol and Related Triterpenes	319

4.8. Solenolides and Briantheins	320
4.9. Spongiadiol and Related Compounds	321
4.10. Ara-A	322
5. Active Antiviral Agents	323
5.1. Dercitin	323
5.2. Indolocarbazole	323
5.3. Topsentins	324
5.4. Variabilin	325
5.5. Reiswigins	325
5.6. Prostaglandins	326
5.7. Macrolactin A	326
6. Modestly Active Antiviral Agents	327
6.1. Misakinolide A and Bistheonellides	327
6.2. Sceptrins and Ageliferins	328
6.3. Halitunal	329
6.4. Sesquiterpenoid Isocyanide	330
6.5. Acarnidines and Polyandrocarpidines	330
6.6. Tubastrine	331
6.7. Saponins	331
6.8. BDS-I	332
6.9. Aplidiashpingosine	332
6.10. Cyclohexadienone	333
6.11. Reticulatines	333
6.12. Chamigrene Derivatives	333
6.13. Polysaccharides	334
7. Conclusions	334
References	335

Chapter 9

<i>The Search for Antiparasitic Agents from Marine Animals</i>	343
---	------------

Phil Crews and Lisa M. Hunter

1. Introduction	343
1.1. Terminology and Scope of Parasitic Diseases	345
1.2. General Approaches to Combating Parasitic Diseases	345
2. Screening Methods for Anthelmintics	348
2.1. The Disease Targets	348
2.2. Bioassay Strategies	348
3. Anthelmintic-Active Natural Products	349

3.1. Agents of Nonmarine Origin	349
3.2. Marine Sponge-Derived Compounds	352
3.3. Non-Sponge-Derived Compounds	376
4. Screening Methods for Antiprotozoal Agents	377
4.1. The Disease Targets	377
4.2. Bioassay Strategies	377
5. Antiprotozoal-Active Natural Products	378
5.1. Known Agents of Nonmarine Origin	378
5.2. Marine-Derived Compounds	380
6. Future Prospects	382
References	383

Chapter 10

Dinoflagellates as Sources of Bioactive Molecules 391

Yuzuru Shimizu

1. Introduction	391
2. Organisms	392
2.1. Classification of Dinoflagellates	392
2.2. Culturing of Dinoflagellates	393
3. Saxitoxin and Gonyautoxin Derivatives: Sodium Channel Blockers	395
3.1. Isolation and Chemistry	395
3.2. Pharmacology	396
4. Brevetoxins and Ciguatoxins: Sodium Channel Activators	397
4.1. Isolation and Chemistry	397
4.2. Pharmacology	398
5. Other Polyether Compounds Produced by Dinoflagellates	400
5.1. Okadaic Acid and Its Derivatives	400
5.2. Antitumor and Antimicrobial Macrolides	400
6. Miscellaneous: Maitotoxin	403
7. Biosynthesis of Dinoflagellate Products and Metabolism	404
7.1. Biosynthesis of Saxitoxin Derivatives	404
7.2. Biosynthesis of Brevetoxins	404
7.3. Characteristics of the Metabolic Pathways	404
8. Conclusion	406
References	407

Chapter 11

<i>Production of β-Carotene and Vitamins by the Halotolerant Alga Dunaliella</i>	411
---	-----

Ami Ben-Amotz

1. <i>Dunaliella</i> and Its Environment	411
2. β -Carotene Production	412
3. Cell Composition	413
4. Biotechnology of <i>Dunaliella</i>	413
5. Products of <i>Dunaliella</i> Cultivation	415
References	416

Chapter 12

<i>Marine Microorganisms: A New Biomedical Resource</i>	419
--	-----

William Fenical and Paul R. Jensen

1. Introduction	419
2. Microbial Diversity in Marine Environments	420
2.1. Archaeabacteria	423
2.2. Eubacteria	424
2.3. Eukaryotes	431
3. Bioactive Metabolites from Marine Microorganisms	434
3.1. Microalgae	435
3.2. Chemical Studies of Marine Bacteria	436
3.3. Symbiotic Marine Bacteria—Origins of Marine Toxins	444
3.4. Chemical Studies of Marine Fungi	446
4. Discussion and Prospects for the Future	449
4.1. Marine Microorganisms As a Resource for Bioactive Metabolites	449
4.2. Basic Research in Marine Microbiology	449
4.3. The Roles of Industry and Academia	450
References	450

Chapter 13

<i>Academic Chemistry and the Discovery of Bioactive Marine Natural Products</i>	459
<i>D. John Faulkner</i>	
1. Introduction	459
2. The Historical Record	460
3. The Current Status of Academic Research	465
4. Collection and Identification of Marine Organisms	466
5. Screening of Crude Extracts for Bioactivity	467
6. Isolation and Identification of Bioactive Natural Products	468
7. Pharmacological Screening of Pure Compounds	469
8. Commercial Development of Marine Natural Products	470
9. Conclusions	471
References	472
<i>Index</i>	475

Biomedical Potential of Marine Natural Products

*Chris M. Ireland, Brent R. Copp, Mark P. Foster,
Leonard A. McDonald, Derek C. Radisky,
and J. Christopher Swersey*

1. INTRODUCTION

Marine natural products, the secondary or nonprimary metabolites produced by organisms that live in the sea, have received increasing attention from chemists and pharmacologists during the last two decades. Interest on the part of chemists has been twofold: natural products chemists have probed marine organisms as sources of new and unusual organic molecules, while synthetic chemists have followed by targeting these novel structures for development of new analogs and new synthetic methodologies and strategies (Albizati *et al.*, 1990). The rationale for investigating the chemistry of marine organisms has changed over the past several decades. Early investigations were largely of a “phytochemical” nature, reporting detailed metabolite profiles similar to those reported for terrestrial plants in previous decades. However, analogous to investigations of terrestrial plants, more recent studies of marine organisms have focused on their potential applications, particularly to the treatment of human disease and control of agricultural

Chris M. Ireland, Brent R. Copp, Mark P. Foster, Leonard A. McDonald, Derek C. Radisky, and J. Christopher Swersey • Department of Medicinal Chemistry, University of Utah, Salt Lake City, Utah 84112.

Marine Biotechnology, Volume 1: Pharmaceutical and Bioactive Natural Products, edited by David H. Attaway and Oskar R. Zaborsky. Plenum Press, New York, 1993.