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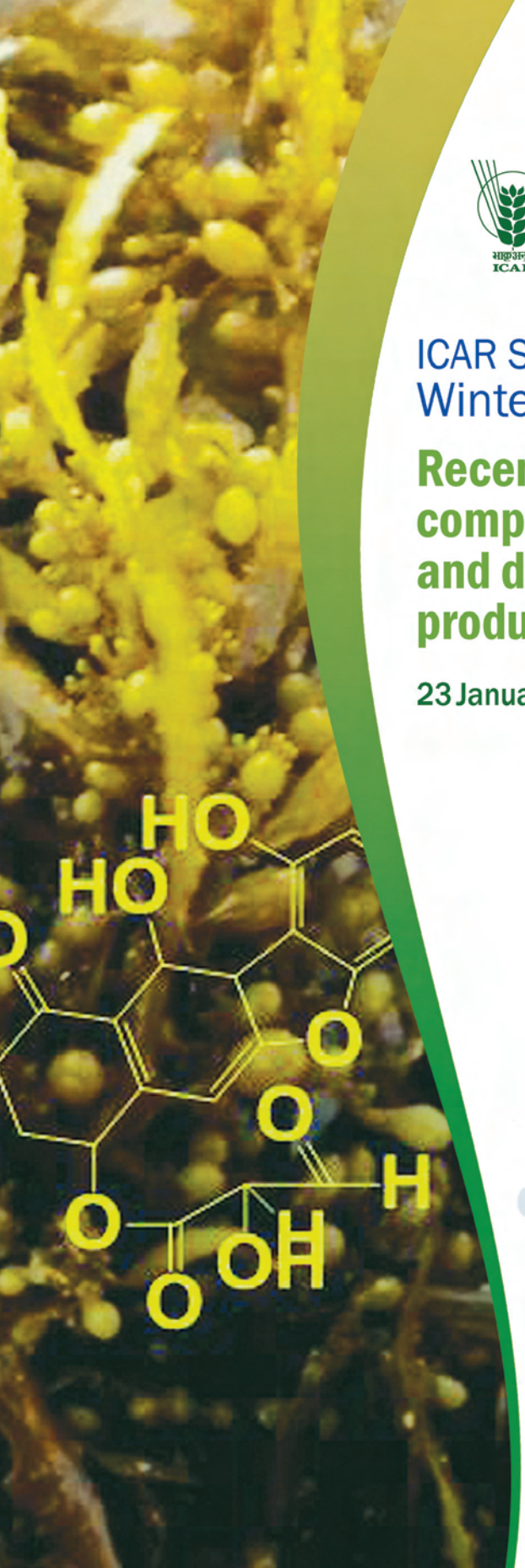
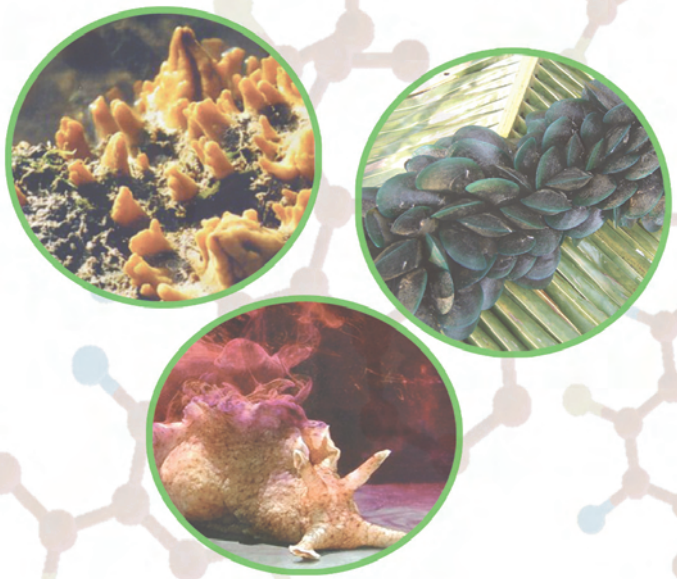



ICAR Sponsored
Winter School on

# Recent advances in bioactive compounds from marine organisms and development of high value products for health management

**23 January to 12 February 2018**



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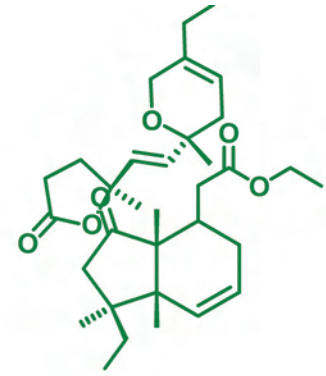

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## Course Manual

ICAR-Winter School on

**Recent advances in bioactive compounds from marine organisms and development of high-value products for health management**

CMFRI Training Manual Series No. 13/2018
ICAR-Central Marine Fisheries Research Institute,
23 January - 12 February, 2018

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Edited by Foxit Reader

# FOREWORD

There has been a growing interest in the marine derived bioactive compounds in the recent years, and the functional foods, enriched with natural ingredients have been proved to provide beneficial action for human health. Marine derived bioactive components and the functional food ingredients demonstrated to possess potential health benefits. High value secondary bioactive metabolites from the marine organisms are attracting attention because of the growing demand for new compounds of 'marine natural' origin, having potential applications in pharmaceutical fields, and concerns about the adverse effects by synthetic drugs and their derivatives. The pioneering R & D works at ICAR-Central Marine Fisheries Research Institute on marine bioprospecting envisaged a systematic approach involving chemical profiling of major species of marine organisms for bioactive pharmacophore leads for activity against various diseases, and a library of molecules with bioactive potential. The research work in this institute developed protocols to prepare various pharmaceutical leads, nutraceuticals/ functional food supplements enriched with lead molecules with different properties against various drug targets for use against various life-threatening diseases.

ICAR-Central Marine Fisheries Research Institute is the pioneering marine research institute in India to work in the frontier area of bioactive molecule discovery from marine organisms as promising therapeutic agents against various diseases, aquatic food product technology, and development of high value products for health management. This prestigious research institute of Indian Council of Agricultural Research is working in the broad national interest of producing high value bioactive leads from the marine organisms, which would provide promising therapeutic agents against various diseases. This institute has developed and commercialized the nutraceutical products Cadalmin™ Green Algal extract (Cadalmin™ GAe) and Antidiabetic extract (Cadalmin™ ADe) as green alternatives to synthetic drugs to combat rheumatic arthritic pains and type-2 diabetes, respectively to a leading biopharmaceutical company in India. The anti-inflammatory nutraceutical Cadalmin™ Green Mussel extract (Cadalmin™ GMe) from Asian green mussel Perna viridis has been commercialized with Amalgam Group of Companies. Cadalmin™ Antihypercholesterolemic extract (Cadalmin™ ACe) has been developed from seaweeds to combat dyslipidemia leading to obesity, and the product was out-licensed to a leading Indian MNC in wellness and obesity management. Antimicrobial therapeutic product from marine bacteria as oral applicant has been developed and the product is in pipeline for commercialization. Seaweed-derived natural template inspired synthetic derivatives as potential pharmacophores were designed and developed. Several nutraceutical and cosmeceutical products from marine organisms are in pipeline, and are being commercialized.

The objective of the National level ICAR Winter School on "Recent advances in bioactive compounds from marine organisms and development of high-value products for health management" is to provide up-to-date information and acquaint the participants with the latest technologies on isolation and characterization of marine natural products of pharmaceutical importance from marine organisms, general and advanced methods of isolation procedures by chromatography, classification of organic compounds and their characterization by advanced spectroscopic experiments. This program further aims to give exposure to the chemical perspectives of marine organisms, primary and secondary bioactive metabolites from fish and marine organisms to develop bioactive compounds and high-value functional food products. Theory and practical classes will be conducted in these areas to provide the participants a hands-on experience.

This ICAR Winter School is organized with the full funding support from ICAR, New Delhi, and the twenty-five participants from various parts of India who are attending this programme were selected after scrutiny of their applications based on their bio-data. They are serving as academicians, such as Professors/Scientists, and in similar posts. The faculties include the knowledgeable scientists and professors from various parts of India and abroad. This training will enable the participants to efficiently carry out their academic programmes, and to plan research on bioactive molecule discovery in their respective laboratories and institutes so that they can formulate the strategies for research.

The Winter School on "Recent advances in bioactive compounds from marine organisms and development of high value products for health management" is very ideal for the current scenario of increasing lifestyle diseases and human health. Understanding the importance of natural products in the health care system of India, ICAR-Central Marine Fisheries Research Institute has reasonably contributed in the various aspects. The Manual released on this occasion covers all aspects of marine natural products prepared by the experts in their respective fields. I congratulate the Course Director of this programme, Dr. Kajal Chakraborty and Head of the Marine Biotechnology Division, Dr. P. Vijayagopal, along with other staff members of Marine Biotechnology Division and Central Marine Fisheries Research Institute for their sincere efforts in bringing out the manual in time, and to arrange the programme in a befitting manner.

**A. Gopalakrishnan**
Director, ICAR-Central Marine Fisheries Research Institute
Kochi, Kerala

# PREFACE

Marine-derived bioactive components and the functional food ingredients with potential health benefits are an emerging area of research. The rich diversity of flora and fauna in the marine and coastal habitats of the Indian subcontinent represent an untapped reservoir of bioactive compounds with valuable pharmaceutical and biomedical use. Considering the underutilization of these groups of marine organisms, exploring bioactive compounds and development of any biologically useful products have benefits as health products. Comprehensive analyses demonstrated that during the last decade the average proportion of bioactive compounds among the new compounds is declining, though there are a large number of marine natural products yet to be explored. This may indicate that the research level of bioactivity is not keeping up with the discovery of new compounds. Thus, the research tools and methods for finding bioactivity need to be improved. The first improvement is about methods of spectral and bioactivity-guided separation and purification of marine-derived secondary metabolites, which combine the discovery of new compounds. These improvements in technology are dependent upon the automation in spectroscopy, which also allows the study of the functions of new compounds extracted from the target marine organisms. Second, for the discovery of new lead compounds and artificial intelligence for drug development evolved to a more mechanistic approach that targets specific molecular lesions. Combined with high-throughput screening through a large number of drug targets, bioactivity research against various life-threatening diseases will be effective in revealing the potentially useful biological properties of marine natural products. Furthermore, the discovery of new bioactive compounds from marine metabolites will form the basis for new drug leads. Thus, the new compounds will absolutely compose an abundant resource for future bioactivity research and drug development. Various medicinal and biomedical products from marine flora and fauna provide a myriad of benefits for human health and multiple life-threatening diseases, and therefore, are the attractive options for the food and pharmaceutical industry. The increasing interest in marine-based functional food ingredients and nutraceutical formulations in the last decade along with increased number of patents filed/granted have appropriately demonstrated the possibilities of bioactive from marine organisms to maintain and improve human health and well-being.

The present ICAR Winter School on "Recent advances in bioactive compounds from marine organisms and development of high-value products for health management" is designed to acquaint the participants with the advances in marine bioactive compounds with emphasis on the latest technologies on isolation and characterization of marine natural products of pharmaceutical importance. The course is planned in such a way that it covers both theoretical and practical aspect of recent advances in bioactive compounds from marine organisms. This programme will strengthen the knowledge of participants with regard to

the general and advanced methods of isolation procedures by chromatography, and their characterization by advanced spectroscopic experiments aspects.

I wish to thank the Education Division of Indian Council of Agricultural Research for giving us an opportunity to organize this ICAR Winter School. We are grateful to Dr. A. Gopalakrishnan, Director, ICAR-Central Marine Fisheries Research Institute, for his guidance, continuous interest in the course and providing all necessary facilities. I am highly obliged to Dr. P. Vijayagopal, Head, Marine Biotechnology Division for his guidance and support for the programme. All the scientists of Marine Biotechnology Division, technical staff, supporting staff and research scholars supported us in organizing the ICAR Winter School. I recall with gratitude the marvellous effort and help in preparing this manual by Minju Joy, Research Scholar of Marine Biotechnology Division. I take this opportunity to thank all the faculty members who have devoted their valuable time and contributed material for the preparation of the manual. I am confident that the Course Manual would aid the participants to enhance their knowledge and competence in the area of marine bioactive compounds and their applications for the development of high-value products for health management.

**Kajal Chakraborty**
Course Director

January, 2018

# CONTENTS

CHAPTER

01

# MARINE ORGANISMS: THE UNDEREXPLORED RESOURCES TO DEVELOP HIGH VALUE COMPOUNDS AND THERAPEUTIC PRODUCTS

**A. Gopalakrishnan**

*Director, ICAR-Central Marine Fisheries Research Institute, Kochi*

There has been a growing interest in functional foods (or nutraceuticals) in recent years, and the functional foods, enriched with natural ingredients have been proved to provide beneficial action for human health. Marine derived bioactive components and the functional food ingredients demonstrated to possess health benefits. High value secondary bioactive metabolites from the marine organisms are attracting attention because of the growing demand for new compounds of 'marine natural' origin, having potential applications in pharmaceutical fields, and concerns about the toxic effects by synthetic drugs and their derivatives. The R & D works at ICAR- Central Marine Fisheries Research Institute on marine bioprospecting envisaged a systematic approach involving chemical profiling of major species of marine organisms for bioactive pharmacophore leads for activity against various oxidative stress-induced diseases with a focus on hypercholesterolmia, hypertension, diabetes, inflammation, hypothyroidism, and a library of molecules with bioactive potential. This research work also developed protocols to prepare various nutraceuticals/functional food supplements enriched with lead molecules with different properties against various drug targets for use against various lifestyle disorders. Combined with high throughput screening through a large number of drug targets, bioactivity research against these diseases will be effective in revealing the potentially useful biological properties of marine natural products. Furthermore, the discovery of new bioactive compounds from marine metabolites will form the basis for new drug leads. Thus, the new compounds will absolutely compose an abundant resource for future bioactivity research and drug development.

## MIRACLE CURE OF SEVERAL LIFE THREATENING DISEASES: NUTRACEUTICALS FROM MARINE ORGANISMS

The rich diversity of marine organisms represents an untapped reservoir of bioactive compounds with valuable pharmaceutical and biomedical use. The pioneering research work at ICAR-Central Marine Fisheries Research Institute envisages a systematic approach involving chemical profiling of major species of mollusks, seaweeds and seaweed-associated heterotrophs for lead pharmacophores coupled with evaluation of target biological activities against different disease models, for example, 3-hydroxy-3-methylglutaryl coenzyme A reductase, type-2 diabetes modulators (dipeptidyl peptidase-4, protein tyrosine phosphatase 1B), angiotensin-I, inflammatory cyclooxygenases and lipoxygenases. Optimized physical/chromatographic procedures have been developed by this institute to isolate and purify the molecules with target bioactivities.


*Nutraceuticals developed by ICAR-Central Marine Fisheries Research Institute for use against type-2 diabetes, obesity/dyslipidemia and rheumatoid arthritis*

The research works at ICAR- Central Marine Fisheries Research Institute developed a hitherto unraveled database of marine organisms with small molecular weight bioactive molecules responsible to combat various life-threatening diseases. ICAR-Central Marine Fisheries Research Institute is pioneering marine research institute in India to work in the frontier area of marine bioprospecting/bioactive molecule discovery from seaweeds/ mollusks/marine heterotrophs and development of high value nutraceutical products as dietary supplements and health management. This prestigious marine fisheries research institute of Indian Council of Agricultural Research (ICAR) has developed and commercialized the nutraceutical products Cadalmin™ Green Algal extract (Cadalmin™ GAe) and Antidiabetic extract (Cadalmin™ ADe) as green alternatives to synthetic drugs to combat rheumatic arthritic pains and type-2 diabetes, respectively to **Celestial Biolabs Limited, Hyderabad, a leading biopharmaceutical company in India with** Rs. 5,00,000/- license fee along with 5% royalty. The anti-inflammatory nutraceutical Cadalmin™ Green Mussel extract (Cadalmin™ GMe) from Asian green mussel *Perna viridis* has been commercialized with Amalgam Group of Companies with a royalty of 5% on annual sale basis. The active principles in Cadalmin™ GMe isolated from *P. viridis* exhibited potential capacities to inhibit experimentally induced inflammation, and can act as dual inhibitors of membrane arachidonate oxygenation by cyclooxygenase-2 (COX-2) and lipoxygenase (5-LOX) pathways, thus decreasing pro-inflammatory prostaglandin ($PGE_2$ and $PGF_{2\acute{a}}$)/leukotriene synthesis and down-regulating the inflammatory sequence. Cadalmin™ Antihypercholesterolemic extract (Cadalmin™ ACe) has been developed from seaweeds to combat dyslipidemia and obesity, and the product was out-licensed to **VLCC Personal Care Limited**, a leading Indian MNC in wellness and obesity management in particular, with manufacturing facilities in India and Singapore). Antimicrobial therapeutic product from seaweed-associated bacterium as oral applicant has been developed and the toxicity studies are being carried out. The lead molecules with action against angiotensin converting enzyme-I, from seaweeds were isolated, and added to a nutraceutical product that is being out-licensed. Seaweed-derived natural template

inspired synthetic derivatives as potential pharmacophores with potential antibacterial activities against methicillin-resistant *Staphylococcus aureus* and anti-angiotensin-I inhibitory activities were designed and developed. Several cosmeceutical products from seaweeds are in pipeline, and are being commercialized.

## ANTIARTHRITIC NUTRACEUTICAL: CADALMIN™ GREEN MUSSEL EXTRACT (CADALMIN™ GME)

Cadalmin™ Green Mussel extract (Cadalmin™ GMe) contains 100% natural marine bioactive anti-inflammatory ingredients extracted from Asian green mussel *P. viridis*. The nutraceutical product is effective to combat chronic joint pain, arthritis/ inflammatory diseases. The active principles in Cadalmin™ GMe isolated from *P. viridis* exhibited potential capacities to inhibit experimentally induced inflammation, and can act as dual inhibitors of membrane arachidonate oxygenation by cyclooxygenase-2 (COX-2) and lipoxygenase pathways, thus decreasing pro-inflammatory prostaglandin ($PGE_2$ and $PGF_{2á}$)/leukotriene synthesis and down-regulating the inflammatory sequence. *In vivo* mammalian model studies revealed that this nutraceutical product showed considerable inhibition on inflammation after sixth hour, illustrating that Cadalmin™ GMe is potent but relatively slow-acting anti-inflammatory agent. This product is free from deleterious ***trans*** fatty acids, free radicals/ free radical adducts/low molecular weight carbonyl compounds, and has been proved to be safe from a long term preclinical/toxicity studies on the experimental subjects.


*Cadalmin™ Green Mussel extract (Cadalmin™ GMe) for use against joint pain and arthritis*

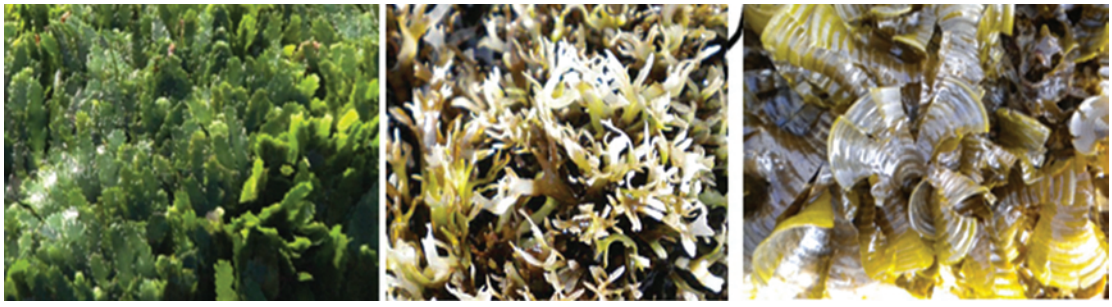
Cadalmin™ Green Mussel extract (Cadalmin™ GMe) contains 100% natural marine bioactive anti-inflammatory ingredients extracted from green mussel *Perna viridis*. The product is effective to combat arthritic pain and inflammatory diseases, and effective green

alternative to synthetic non steroidal anti-inflammatory drugs and other products available in the market. The active principles in Cadalmin™ GMe isolated from *P. viridis* were found to competitively inhibit inflammatory cyclooxygenases (COX-1,2) and lipoxygenase (5-LOX) in inflammation and oxidative stress reaction, resulting in decreased production of pro-inflammatory prostaglandins and leukotrienes. *In vivo* animal model studies revealed that active principles effectively suppressed the edema produced by the histamine, which indicates that they exhibit its anti-inflammatory action by means of either inhibiting the synthesis, release or action of anti-inflammatory mediators. Cadalmin™ GMe is an indigenous product, and is highly cost effective and more effective in terms of its activity with that of the imported products available in the market. Till date no medications are available to combat arthritic pain. The available options available are knee replacement or painkillers, which have multiple adverse effects on human health.

## STATUS OF COMMERCIALIZATION

Cadalmin™ GMe has been commercialized during 2012 with Amalgam Group of Companies. This is the first nutraceutical produced by an institute of Indian Council of Agricultural Research (ICAR). This product is commercially produced and marketed by Accelerated Freeze Drying Company Pvt. Ltd. under their brand name "Musseltone®".

## NUTRACEUTICAL PRODUCTS FROM SEAWEEDS



*The seaweeds contain a large assemblage of species that predominate in the coastal shelf areas of Indian subcontinent*

## MIRACLE CURE OF SEVERAL LIFE THREATENING DISEASES: NUTRACEUTICALS FROM SEAWEEDS

The rich diversity of seaweeds represents an untapped reservoir of bioactive compounds with valuable pharmaceutical and biomedical use. The pioneering research work at ICAR-Central Marine Fisheries Research Institute envisages a systematic approach involving chemical profiling of major species of seaweeds for lead pharmacophores coupled with evaluation of target biological activities against different disease models, for example, 3-hydroxy-3-methylglutaryl coenzyme A reductase, type-2 diabetes modulators (dipeptidyl

peptidase-4, protein tyrosine phosphatase-1B), angiotensin-I, inflammatory cyclooxygenases and lipoxygenases. Optimized physical/chromatographic procedures have been developed by this institute to isolate and purify the molecules with target bioactivities.


*Nutraceuticals developed by ICAR-Central Marine Fisheries Research Institute for use against type-2 diabetes, obesity/dyslipidemia and rheumatoid arthritis*

The research works at ICAR-Central Marine Fisheries Research Institute developed a hitherto unraveled database of seaweeds with small molecular weight bioactive molecules responsible to combat various life-threatening diseases. This research work also relates to develop protocols to prepare nutraceutical products enriched with lead pharmacophores with different properties against various drug targets for use against obesity/dyslipidemia, hypertension, diabetes, and inflammatory disorders. The research works carried out in the institute developed natural anti-inflammatory supplements enriched with lead molecules as neutraceutical Cadalmin™ Green Algal extract (Cadalmin™ GAe) from seaweeds as effective green alternative to the synthetic drugs available in the market to combat **rheumatic arthritic pains**. This product has been out-licensed to the biopharmaceutical company for commercial production and marketing in India and abroad. The research efforts to isolate the lead molecules with action against type-2 diabetes led to the development of a nutraceutical product Cadalmin™ Antidiabetic extract (Cadalmin™ ADe) from marine algae that has been out-licensed to leading Biopharma Company based at Hyderabad. Cadalmin™ Antihypercholesterolemic extract (Cadalmin™ ACe) developed from seaweeds to combat dyslipidemia and obesity, and the product is being out-licensed to a pharmaceutical company.

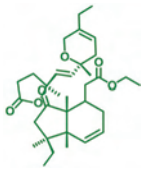
## A HEALING TOUCH OF MOTHER NATURE

Seaweed derived bioactive components and the functional food ingredients with potential health benefits are an emerging area of research. Various nutraceutical or functional

food supplements and biomedical products from seaweeds provide a myriad of benefits for human health and multiple life threatening diseases, and therefore, are the attractive options for the pharmaceutical and healthcare industry. Novel secondary bioactive metabolites from these ocean herbs are attracting attention because of the growing demand for new compounds of 'marine natural' origin, having potential applications in pharmaceutical fields, and concerns about the toxic effects by synthetic drugs and their derivatives.

Comprehensive analyses demonstrated that during the last decade the average proportion of bioactive compounds among the new compounds is declining, though there are a large number of marine natural products yet to be explored. This may indicate that the research level of bioactivity is not keeping up with the discovery of new compounds. Thus, the research tools and methods for finding bioactivity need to be improved. The first improvement is about methods of nuclear magnetic resonance/mass spectroscopy and bioactivity-guided separation and purification of marine-derived small molecular weight secondary metabolites, which combine the discovery of new compounds.

These improvements in technology are dependent upon the automation in spectroscopy, which also allow the study of the functions of new compounds extracted from the target organisms. Second, for the discovery of new lead compounds for drug development, screening for quantitative structure-activity relationship analyses and artificial intelligence evolved to a more mechanistic approach that targets specific molecular lesions. Combined with high throughput screening through a large number of drug targets, bioactivity research against hypertension, type-2 diabetes, hypercholesterolmia, obesity/dyslipidemia, type-2 diabetes, and inflammation will be effective in revealing the potentially useful biological properties of seaweed-derived marine natural products. Furthermore, the discovery of new bioactive compounds from seaweeds will form the basis for new drug leads. Thus, the new compounds will absolutely compose an abundant resource for future bioactivity research and drug development. Recently, efforts are underway to exploit the bioactive leads from the natural origin for use as food supplements in the food and pharmaceutical industry. These naturally derived antioxidant leads are the preferred alternatives to the synthetic derivatives due to their safety, sustainability and effectiveness. The long history of the usage



seaweeds in the part of their staple diet of the people from Korea, Japan, China etc. demonstrates their nutritional and health supplementing qualities. There are reports that different genera of seaweeds mainly belonging to *Sargassum, Porphyra, Ecklonia, Laminaria* etc have been used in Chinese medicine to treat against different diseases. Seaweeds, especially *Laminaraia* spp were used to treat against goitre because of its richness in iodine in China and Europe. These studies demonstrate the beneficial effect of seaweeds or seaweed derived compounds in pharmaceutical industry.

## ICAR-CENTRAL MARINE FISHERIES RESEARCH INSTITUTE: A PIONEERING INSTITUTE IN INDIA TO DEVELOP A SERIES OF NUTRACEUTICAL PRODUCTS FROM SEAWEEDS

ICAR-Central Marine Fisheries Research Institute is the pioneering marine research institute in India to work in the frontier area of marine bioprospecting/bioactive molecule discovery from seaweeds and development of high value nutraceutical products as dietary supplements and health management. This prestigious marine fisheries research institute of Indian Council of Agricultural Research (ICAR) has developed and commercialized the nutraceutical products Cadalmin™ Green Algal extract (Cadalmin™ GAe) and Antidiabetic extract (Cadalmin™ ADe) as green alternatives to synthetic drugs to combat rheumatic arthritic pains and type-2 diabetes, respectively. Cadalmin™ Antihypercholesterolemic extract (Cadalmin™ ACe) has been developed from seaweeds to combat dyslipidemia and obesity, and the product is being out-licensed to a pharmaceutical company. Semisynthetic C-4/C-6 methylene-polycarboxylate cross-linked hybrid drug delivery system and a topical antibacterial formulation have been developed from seaweeds, and were found to be comparable with commercially available products. The lead molecules with action against angiotensin converting enzyme-I, from seaweeds were isolated, and added to a nutraceutical product that is being out-licensed. Seaweed-derived natural template inspired synthetic derivatives as potential pharmacophores with potential antibacterial activities against methicillin-resistant *Staphylococcus aureus* and anti-angiotensin-I inhibitory activities were designed and developed. Several cosmeceutical products from seaweeds are in pipeline, and are being commercialized.

## A GREEN REMEDY NUTRACEUTICAL FOR TYPE-2 DIABETES FROM SEA: ICAR-CMFRI'S CADALMIN™ ANTIDIABETIC EXTRACT (CADALMIN™ ADE)

Diabetes mellitus is a metabolic disorder which affects the endocrine system of the body, characterized by defects in carbohydrate, lipid, and protein metabolism. Globally the frequency of disorder is rising gradually; patients suffering from this disorder are unable to turn out or respond properly to insulin produced in the body. It was estimated that 246 million persons in the world suffered from type 2 diabetes in 2015 and that this number will reach at least 380 million in 2025. The number of diabetic patients is rapidly rising in most


parts of the world. People with type 2 diabetes are often sedentary, obese, middle-aged adults with an increased risk of macrovascular disease, retinopathy, nephropathy, neuropathy, and hypertension. These health complications lead to increased morbidity and premature death. Type-2 diabetes receives more attention than type-1 diabetes because it is considered to be a preventable disease. Type 2 diabetes is caused by an imbalance between blood sugar absorption and insulin secretion. Post-prandial hyperglycemia plays an important role in development of type-2 diabetes.

Controlling plasma glucose levels are essential for delaying or preventing type-2 diabetes. While efficient in attenuating the rise in blood glucose levels in many patients, the continuous use of the synthetic drugs is often associated with undesirable side effects, such as liver toxicity and adverse gastrointestinal symptoms. It is for this reason that there is a need for natural alternatives which have no adverse or unwanted secondary effects. Dietary ingestion of seaweeds has been shown to decrease diabetic complications in human and also have strong antioxidant properties. It is, therefore, that a search of safer and effective alternatives to synthetic drugs to combat type-2 diabetes led the scientists of ICAR-Central Marine Fisheries Research Institute to investigate into the seaweeds, which are natural bounty of sea, for valuable secondary metabolites, which are anti-diabetic in nature, and can offer relief to type-2 diabetes. A concerted research in this area led them to develop a 100% vegetarian nutraceutical Cadalmin™ Antidiabetic extract (ADe) from selected seaweeds. Cadalmin™ ADe contains 100% natural marine bioactive ingredients from selected seaweeds by a patented technology. The bioactive ingredients in Cadalmin™ ADe competitively inhibit dipeptidyl peptidase-4 and tyrosine phosphatase 1B thereby hindering the occurrence of type-2 diabetes. Type-2 diabetes and obesity are characterized by resistance to hormones insulin, possibly due to attenuated or diminished signaling from the receptors. A large body of data have identified protein tyrosine phosphatase 1B (PTP1B) as a major negative regulator of insulin signaling. Pharmacological agents capable of inhibiting the negative regulator(s) of the signaling pathways like PTP 1B are expected to potentiate the action of insulin and therefore be beneficial for the treatment of Type 2 diabetes. Antidiabetic extract inhibits tyrosine phosphatase 1B (PTP1B) thereby hindering the occurrence of type-2 diabetes. Another mode of action of Antidiabetic extract is inhibition of dipeptidyl peptidase-4 (DPP-4), which is an antigenic enzyme expressed on the surface of most cell types and is associated with immune regulation and signal transduction. DPP-4 inactivates the incertins GLP-1 and

GIP by removing amino acids from these peptide hormones. GLP-1 and GIP are essentially required for insulin secretion from the â-cells of pancreas. Cadalmin™ Antidiabetic extract has no side effects ($LD_{50}$ > 5000 mg/kg BW) as proved from the preclinical and acute/long term chronic toxicity studies on experimental subjects. Preclinical trials showed no toxicity related significant changes in renal or hepatic function, hematological indices and serum biochemical parameters in the experimental subjects. The results also demonstrated a lack of test substance-related general organ or systemic toxicity and hypoglycaemic disorders following oral administration at a dose as high as 2000 mg/kg/d. The active ingredients in the product were packed in plant-based capsules to meet the dietary needs of vegetarians. Cadalmin™ ADe has a promising consumer appeal, and market potential especially for the large vegetarian population in India and abroad. The unique biochemical engineering techniques adopted to retain antidiabetic activities in preparation of Cadalmin™ ADe assures higher shelf life. Cadalmin™ ADe has been officially released by the Hon'ble Union Minister of Agriculture, Shri Radha Mohan Singh on 26th July 2015 at a function in Patna during the ICAR Foundation Day. This product has been commercialized with a pharmaceutical company and ICAR-CMFRI is in search of more commercial partners for wider dissemination of the product in the marketplace.

## CADALMIN™ GREEN ALGAL EXTRACT (CADALMIN™ GAE): A VEGETARIAN NUTRACEUTICAL DEVELOPED FROM SEAWEEDS FOR USE AGAINST JOINT PAIN AND ARTHRITIS


Arthritis represents a major health problem and its global burden is rising at an alarming rate with millions of people suffering from this disease due to various reasons. Many of the allopathic prescriptions include Non Steroidal Anti-Inflammatory Drugs (NSAIDs) and cyclooxygenase inhibitors used in controlling arthritic conditions have known side effects, especially with long term usage. About 25% of the users experience some kind of side effect and 5% develop serious health consequences such as stomach bleeding, stroke, and acute renal failure. The New England Journal of Medicine reports that "anti-inflammatory drugs (prescription and over-the-counter, which include Advil®, Motrin®, Aleve®, Ordus®, Aspirin, and over 20 others) alone cause over 16,500 deaths and over 103,000 hospitalizations per year in the US", according to a review article published in the New England Journal of Medicine.

The side-effects of synthetic drugs led the scientists of ICAR-Central Marine Fisheries Research Institute to investigate into natural products for safer and effective alternatives.



Scientists at ICAR-Central Marine Fisheries Research Institute identified the seaweeds, which are natural bounty of sea, to possess valuable compounds that can offer relief to arthritis and associated joint pain.

Cadalmin™ Green Algal extract (Cadalmin™ GAe) contains a unique blend of 100% natural marine bioactive anti-inflammatory ingredients extracted from selected seaweeds with a patented ecofriendly "green" technology. The product is effective to combat arthritic pain and inflammatory diseases in human beings. The active principles in Cadalmin™ GAe competitively inhibit pro-inflammatory mediators, resulting in decreased production of inflammatory prostaglandins and leukotrienes, and its activity was found to be superior to some of the synthetic non steroidal anti-inflammatory drugs available in the market. A lower cycloxygenase$_I$/lipoxygenase$_V$ and cycloxygenase$_{I/II}$ (<1.0), simultaneous inhibition of cycloxygenase$_{II}$ and lipoxygenase$_V$ enzymes and significant *in vivo* activity indicate higher selectivity and lower side-effect profiles of Cadalmin™ GAe as compared to the synthetic non-steroidal anti-inflammatory drugs. Long term animal model experiments proved the efficiency and safety of this nutraceutical. Cadalmin™ GAe suppresses the edema produced by histamine, and exhibits its anti-inflammatory action by means of either inhibiting the synthesis, release or action of anti-inflammatory mediators. The mean lethal dose ($LD_{50}$) of Cadalmin™ GAe was found to be greater than 4000 mg/kg body weight of the mammalian subjects that indicate the safety of the product. As part of the preclinical assay of the product, feeding of Cadalmin™ GAe even at a dose upto 2500 mg/kg body weight did not induce significant change in body weights, hematological indices, histopathological, and serum biochemical parameters between the control and treated groups indicating that it has no toxicity to the mammalian subjects.

Cadalmin™ GAe is an indigenous product, and is highly cost effective and more effective in terms of its activity with that of the imported products available in the market. Till date no medications are available to combat arthritic pain. The available options available are knee replacement or painkillers, which have multiple adverse effects on human health. Also there are technologies like creams/aurvedic medicines that are either not effective or not scientifically documented. Cadalmin™ GAe is a solution to these problems. This nutraceutical has been developed to concentrate the anti-inflammatory bioactive compounds/principles in a capsule (400 mg), whereas the active ingredients therein competitively inhibit pro-inflammatory mediators, resulting in decreased production of inflammatory prostaglandins and leukotrienes. Cadalmin™ GAe was distributed to the patients suffering with chronic joint pain and arthritis, and questionnaire and clinical trial-based studies revealed that more than 98% of the respondents were satisfied with the product with about 70-85% relief in joint pain and arthritis. None of the respondents reported any side effects. The diagnostically useful autoantibody termed as Rheumatoid Factors (RFs), which are the most useful

prognostic marker for rheumatoid arthritis, significantly reduced from more than 300 IU/mL to less than 20-35 IU/mL within a period of two months of consuming the product. The mean lethal dose ($LD_{50}$) of Cadalmin™ GAe was found to be greater than 4000 mg/kg body weight of the mammalian subjects that indicate the safety of the product unlike the painkillers available in the market. This product has been commercialized with a pharmaceutical company and ICAR-CMFRI is in search of more commercial partners for wider dissemination of the product in the marketplace.

## CADALMIN™ ANTIHYPERCHOLESTEROLEMIC EXTRACT (CADALMIN™ ACE): ANTIOBESITY NUTRACEUTICAL FROM SEAWEEDS


Obesity is a medical condition in which excess body fat has accumulated to the extent that it may have a negative effect on health. Obesity is one of the leading preventable causes of death worldwide. The prevalence of obesity has significantly increased over the last several decades. Increased abdominal (visceral) fat accumulation and obesity is associated with type-2 diabetes, abnormalities in lipid metabolism and coronary artery disease. Approximately 70% of obese patients are dyslipidemic, which include elevated serum triglyceride (hypertriglyceridemia), reduced high-density lipoprotein, and increased small, dense low-density lipoprotein (LDL) particle number. The treatment of the dyslipidemia is focused on lipid-lowering therapy by statin treatment, which has proved adverse effects including drug-induced insulin resistance, chronic inflammation, reduced immunity (by depletion of CoQ10) and increased cancer risk.

The side-effects of synthetic drugs led the scientists of ICAR-Central Marine Fisheries Research Institute to investigate into natural alternatives from seaweeds for safer and effective alternatives. Scientists at this institute identified seaweeds, which possess valuable compounds that can serve as natural remedy for obesity and dyslipidemia. CadalminTM Antihypercholesterolemic extract (CadalminTM ACe) is nutraceutical product developed by ICAR-Central Marine Fisheries Research Institute, as natural remedy for obesity and dyslipidemia from seaweeds, which are commonly available in the Indian coastal waters.

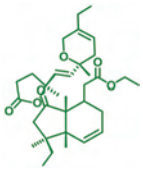
Bioactive pharmacophore leads from seaweeds were used to develop the nutraceutical product, and were found to inhibit hydroxymethyl glutaryl coenzyme A reductase, various target receptors and other rate limiting enzymes, which are responsible to cause obesity



and dyslipidemia. Cadalmin™ ACe can be administered to regulate clinical indicators leading to dyslipidemia or obesity, total adipose tissue and visceral fat, triglycerides, cholesterol, both good and bad, known as HDL, VLDL and LDL. Cadalmin™ ACe contains 100% natural marine bioactive ingredients from selected seaweeds by a patented technology, and would be made available in 400 mg capsules. The nutraceutical does not have any side effects as established by detailed preclinical trials. Cadalmin™ ACe is the only product made by 100% natural marine bioactive ingredients from seaweeds as a natural remedy of obesity and dyslipidemia. The active ingredients in the product would be packed in plant-based capsules to meet the dietary needs of the large vegetarian population in India and abroad. Large scale extraction of the active principles from the raw material was optimized in a factory unit. The total yield of the active principles from the raw material in the factory unit was found to be greater than 20%, which demonstrated the commercial feasibility of the nutraceutical product. The product is being out-licensed to the pharmaceutical company. Shri Justice P. Sathasivam, Hon'ble Governor of Kerala, released the product during the inaugural ceremony of the Platinum Jubilee celebrations of the ICAR-Central Marine Fisheries Research Institute at the Headquarters in Kochi on 18th February 2017. This product is commercialized with VLCC during 23rd may 2017.

## CONCLUSIONS

Marine derived bioactive components with potential health benefits are an emerging area of research. Considering their underutilization, exploring bioactive compounds and development of any biologically useful products has duel benefits-as health products and their commercial farming of mollusks and seaweeds in coastal habitats, resulting in C-sequestration and C-budgeting in a scenario where climate change may pose a serious threat in future. Development of value-added products from these underutilized species will also promote their farming in coastal habitats, which has not been seriously explored earlier due to the lack of knowledge about their commercial importance. ICAR-Central Marine Fisheries Research Institute has devoted research program to develop various health products from marine organisms towards their utilization based on the National Policy to harness the potential of this natural wealth of Indian coastal waters. ICAR-Central Marine Fisheries Research Institute is also in the process of developing more health products from the underutilized seaweeds for treatment against thyroid disorder, hypertension and other lifestyle diseases. Several cosmeceutical products, such as seaweed-based antimicrobial ointment, hybrid drug delivery system are also in the pipeline, and are being commercialized. This prestigious marine fisheries research institute of Indian Council of Agricultural Research (ICAR) is working in the broad national interest of producing high value nutraceutical products/pharmaceutical leads from the marine organisms, which will provide promising therapeutic agents against various diseases.



## SUGGESTED READINGS

A process to concentrate anti-inflammatory principles from green mussel *Perna viridis* L. and a product incorporating these ingredients. 2065/CHE/2010 (Indian patent application).

A product containing anti-inflammatory principles from green mussel *Perna viridis* L. and a process thereof 2066/CHE/2010 (Indian patent application).

A process to prepare antioxidant and anti-inflammatory concentrates from brown and red seaweeds and a product thereof 2064/CHE/2010 (Indian patent application).

A Process to isolate anti-inflammatory principles from green mussel *Perna viridis* L. to prepare a stabilized nutraceutical supplement against inflammatory disorders and a product thereof 5198/CHE/2012 (Indian patent application).

A product containing anti-inflammatory principles from brown seaweeds and a process thereof 5199/CHE/2012 (Indian patent application).

Anti-inflammatory principles in a preparation of brown seaweeds 4254/DEL/2015 (Indian patent application).

A process to prepare antidiabetic concentrates from seaweeds and a product thereof 3366/DEL/2015 (Indian patent application).

A process to prepare anti-dyslipidemic concentrate from seaweed and a product therof Application No 201711013741 (Indian patent application).


Inauguration of winter school 2018 by Padma Bhushan Dr. Manju Sharma

Photo with Dr. K. Gopakumar, Formerly DDG ICAR (Fy)

Field visit to India Sea Foods

Field visit to BOS Naturals

Field visit to Accelerated Freeze Drying Co. Ltd

**Photo with Dr. Meledath Govindan**

**Lectures and Interactive Sessions**

**Practical Sessions**


ICAR-CENTRAL MARINE FISHERIES RESEARCH INSTITUTE
Ernakulam North P.O., Kochi-682018, Kerala
ICAR Sponsored Winter School on Recent Advances in Bioactive Compounds from Marine Organisms and Development of High Value Products for Health Management
January 23 to February 12, 2018
CMFRI
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Central Marine Fisheries Research Institute
കേന്ദ്ര സമുദ്ര മത്സ്യ ഗവേഷണ സ്ഥാപനം
WELCOME
Sitting (L to R) Minimol K.C., Grace Thomas, Kajal Chakraborty (Course Director), P. Vijayagopal (Head, Marine Biotechnology Division), A. Gopalakrishnan (Director), Paulson Mathew, Sathu T., Radhakrishnan E.K.
Standing (L to R) Aswathy Elizabeth Mani, Sreemol C.K., Prima Francis, Soumya Krishnan, Minju Joy, V. Rani, Seeja Thomachan Panjikkaran, Sheneya Festus, Drishya K., Anie Y., Suja Rani S., Sindhu Issac, Teena P. Varghese, Magna Thomas, Santwana Palai, Norma Xavier Chelat, Naheef K., Satya Narayan Sahoo, Jaimin Hareshbhai Bhatt, Ajoy Saha, Senthil Kuppusamy, Kedar Shashikant Damle, Shubhajit Dhara, Midhun Dominic C.D., Manukuttan K.S., Suji Chandru, Tima Antony, Soumya Salas

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