

# SYSTEMATIC REVIEW OF MARINE SPONGE-ASSOCIATED MICROBES PRODUCING LIPASE ENZYMES FOR INDUSTRIAL APPLICATIONS

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## ABSTRACT

Actinomycetes are a remarkably diverse group of filamentous, Gram-positive bacteria with extensive genetic and functional capabilities, particularly noted for synthesizing a broad spectrum of secondary metabolites that have significant therapeutic, environmental, and industrial applications. These metabolites form a prolific reservoir of bioactive compounds, including antibiotics, antitumor agents, antifungals, and anthelmintics, many of which have already been developed into clinically valuable drugs and continue to serve as templates for novel therapeutics. Actinomycetes contribute an estimated 70–75% of all known microbial secondary metabolites, highlighting their central role in natural product discovery and drug development.

Marine-derived actinomycetes, including genera such as *Streptomyces*, *Salinispora*, *Micromonospora*, and *Nocardiosis*, are especially rich sources of structurally unique bioactive molecules with potent antimicrobial, anticancer, antioxidant, and antiviral properties. These organisms have adapted to extreme and nutrient-limited marine environments, producing secondary metabolites with novel modes of action that frequently differ from those isolated from terrestrial microbes.

Symbiotic associations between marine sponges and actinomycetes represent a particularly valuable ecological niche, as sponge-associated bacteria often account for a substantial proportion of the host biomass and are responsible for many sponge-linked bioactive compounds. These symbionts have been shown to generate molecules exhibiting diverse pharmacological activities, including novel antimicrobial and antioxidant compounds, and are increasingly being explored for drug discovery. In addition to pharmaceuticals, marine actinomycetes are being investigated for their roles in environmental biotechnology, such as the biodegradation of pesticides, toxic metals, and other pollutants, as well as their suitability for applications in bioremediation, biofouling prevention, and waste management. This expands their utility beyond therapeutics to environmental sustainability.

The enzymes produced by marine actinomycetes—including lipases, proteases, cellulases, chitinases, and other hydrolytic and oxidative enzymes—have substantial biocatalytic potential for industrial processes, such as food processing, biofuel production, detergents, agriculture, and textile manufacturing. Their ability to function under extreme conditions (e.g., high salinity, pressure, and variable temperatures) positions them as eco-friendly alternatives to traditional chemical catalysts. Emerging techniques such as genome mining, heterologous expression of biosynthetic gene clusters, and advanced cultivation strategies are enhancing our ability to discover previously "silent" secondary metabolites, thereby unveiling new chemical entities with high therapeutic and commercial value.

This paper reviews the current state of knowledge regarding the applications of marine actinomycetes in applied research—with an emphasis on their bioactive metabolites, enzyme technology, ecological roles, and industrial prospects—and outlines future directions for exploration, including advanced molecular approaches and sustainable exploitation of these versatile microorganisms.

**KEYWORDS:** Actinomycetes, Marine Biotechnological Applications, Lipase, Marine Sponges, Marine Enzymes

## 1. INTRODUCTION

Marine ecosystems constitute one of the most extensive and biologically diverse environments on Earth and serve as an abundant source of novel bioactive molecules, secondary metabolites, enzymes, polysaccharides, pigments, and other functional biomaterials. The exploration of marine biodiversity for biotechnological innovation has gained increasing importance due to the structural novelty and functional diversity of compounds produced by marine organisms. These bioresources provide immense opportunities for the development of value-added biotechnological products and services. In particular, marine-derived enzymes exhibit remarkable properties such as high catalytic efficiency, stability under extreme conditions, and tolerance to wide ranges of temperature, salinity, and pH, making them highly desirable for industrial, domestic, commercial, and healthcare applications.

Oceans occupy nearly 75% of the Earth's surface and represent the largest habitat for life, hosting some of the most ancient, complex, and evolutionarily diverse organisms. The marine environment is characterized by

highly heterogeneous and often extreme conditions, including permafrost zones, hydrothermal vents, cold seeps, abyssal plains, deep-sea trenches, and hypersaline regions. These habitats experience dramatic variations in temperature (ranging from subzero to over 350°C), hydrostatic pressure (1–100 atm or more), light availability (from highly photic surface waters to completely aphotic deep-sea zones), and nutrient concentrations (from oligotrophic to eutrophic conditions). Such environmental extremes have driven marine organisms to evolve unique physiological, biochemical, and genetic adaptations, resulting in the synthesis of structurally distinct and biologically potent metabolites.

Marine biodiversity comprises an estimated 300,000 species of flora and fauna, yet only a small fraction has been systematically explored for its biotechnological potential. The scientific pursuit of marine bioresources began in earnest during the 1960s, when researchers recognized the extraordinary chemical diversity and pharmaceutical promise of marine natural products. Since then, marine organisms have been identified as prolific producers of metabolites exhibiting a wide range of biological activities, including antitumor, antiviral, antibacterial, antifungal, anti-inflammatory, antioxidant, and enzyme-inhibitory properties. Many of these compounds interact with critical physiological systems, such as the central and autonomic nervous systems, as well as cardiovascular, respiratory, neuromuscular, and gastrointestinal systems, highlighting their relevance in therapeutic and drug discovery research.

Bioactive compounds and secondary metabolites isolated from marine ecosystems originate from diverse taxonomic groups, with contributions from algae and microalgae, including cyanobacteria and diatoms (~9%), marine bacteria (~18%), and sponges (~37%), along with tunicates, mollusks, corals, and fungi. Marine microorganisms, in particular, have gained prominence due to their rapid growth, metabolic versatility, and ability to produce novel compounds through unique biosynthetic pathways. Advances in metagenomics and microbial cultivation techniques have further expanded access to previously unculturable marine microbes, revealing new opportunities for enzyme discovery and natural product development.

Marine biotechnology is a highly interdisciplinary field that integrates marine biology, molecular biology, biochemistry, genetics, bioengineering, and computational sciences to harness marine organisms for the benefit of humankind. Its applications span several domains, including biomedical, agricultural, environmental, and industrial biotechnology. Marine-derived products have contributed to the development of anticancer drugs, antibiotics, vaccines, hormones, biofertilizers, functional foods, nutraceuticals, and bioactive coatings. Compared to terrestrial biotechnology, marine biotechnology remains relatively underexplored, yet it offers unparalleled chemical diversity and innovation potential.

The scope of marine biotechnology extends across numerous application areas such as human and environmental health, bioremediation of oil spills and heavy metals, biomining, biofouling control, aquaculture enhancement, fisheries management, biomass and biofuel production, biomedical materials, tissue engineering, genomics, proteomics, metabolomics, bioinformatics, nanotechnology, and bioprocess engineering. Marine biomaterials, including biopolymers, chitosan, collagen, and bio-ceramics, are increasingly used in medical implants, wound dressings, and drug delivery systems. Additionally, marine-derived enzymes play a crucial role in the production of fine chemicals, agrichemicals, cosmetics, pharmaceuticals, food additives, nutritional supplements, and molecular probes.

Despite its immense promise, marine biotechnology faces several challenges that limit its full-scale industrial application. These include difficulties in sampling extreme marine environments, limited access to deep-sea ecosystems, challenges in organism identification and high-throughput screening, low metabolite yields, and complexities associated with large-scale cultivation and sustainable supply. Furthermore, an insufficient understanding of the cellular, molecular, and genetic mechanisms governing the biosynthesis and regulation of marine metabolites hampers efficient production and optimization strategies. Emerging tools such as synthetic biology, metabolic engineering, genome mining, and artificial intelligence-driven drug discovery are expected to address these limitations and accelerate innovation in the field.

In conclusion, marine biotechnology holds extraordinary potential to contribute significantly to the global biotechnology market and to address pressing challenges in healthcare, food security, environmental sustainability, and industrial development. Comprehensive exploration, conservation, and sustainable utilization of marine bioresources, supported by advanced molecular and engineering technologies, are essential to unlock the full potential of marine ecosystems for future biotechnological advancements.

## **2. MATERIALS AND METHODS**

This systematic review was conducted using multiple methodological approaches, and specialized software tools were employed to identify and organize all relevant studies included in the analysis. An extensive literature survey was conducted through direct searches of major electronic databases, namely Google Scholar, PubMed/MEDLINE, Scopus, and Web of Science. In addition to the primary search, a secondary screening process was performed by examining the reference lists of selected articles, thereby ensuring comprehensive coverage of all relevant and significant publications.

### **3. Applications**

#### **3.1 Lipase**

Microbial lipases are industrially valuable biocatalysts extensively applied in the dairy, bakery, oil, meat, fish processing, and beverage industries, where they play a critical role in improving food quality, flavor

development, and processing efficiency. Beyond food applications, lipases are widely utilized in the detergent and cosmetic industries due to their efficiency in lipid degradation under diverse conditions.

Notably, substantial lipase activity has been reported in yeast species belonging to the genera *Cryptococcus*, *Leucosporidium*, and *Metschnikowia*, isolated from Antarctic marine environments, highlighting their ecological and industrial relevance. Several Actinomycetes strains have also demonstrated a pronounced ability to hydrolyze oils and fats. Lipases and esterases together constitute a heterogeneous group of hydrolytic enzymes responsible for catalyzing the breakdown of lipid substrates, particularly triglycerides.

Among Actinomycetes (Table 1), species such as *Streptomyces exfoliates* and *Nocardiopsis alba* are recognized for producing lipases capable of cleaving ester bonds in triglycerides, yielding glycerol and free fatty acids. Owing to these catalytic properties, lipases exhibit significant potential in the processing of fats and oils, cosmetic formulations, clinical diagnostics, and detergent manufacturing.

**Table 1: Marine sponge-associated actinomycetes for biotechnological applications**

| Actinomycetes Source/Genus                     | Bioactive Compound/Product      | Biotechnological/Industrial Application     |
|------------------------------------------------|---------------------------------|---------------------------------------------|
| <i>Streptomyces</i> ,<br><i>Micromonospora</i> | Novel Secondary Metabolites     | Antibiotics, Antitumour agents, Antifungals |
| <i>Nocardiopsis</i> , <i>Salinispora</i>       | Alkaloids, Peptides, Terpenes   | Antimicrobial, Anti-inflammatory drugs      |
| Marine Rare Actinomycetes                      | Polyketides, Unique metabolites | Drug discovery, Enzyme inhibitors           |
| Sponge-associated Actinos                      | Lipases, Esterases              | Biofuels, Fine chemicals, Bioremediation    |
| <i>Verrucosipora</i>                           | Novel Marine Natural Products   | Therapeutic agents                          |

#### 4. Types of Microbial Lipases

##### 4.1 Bacterial Lipases

Bacterial lipases are broadly categorized into intracellular and extracellular forms. Intracellular lipases are typically glycoproteins, whereas extracellular lipases are predominantly lipoproteins. The production of intracellular lipases is influenced by various physicochemical and nutritional factors, including temperature, pH, oxygen availability, carbon and nitrogen sources, lipid substrates, and inorganic salts. The modulatory effect of polysaccharides on lipase production was first reported in 1979.

These enzymes are generally constitutive in nature and are susceptible to inhibition by serine hydrolase inhibitors. Well-established bacterial lipase producers include species of *Streptomyces*, *Alcaligenes*, *Arthrobacter*, *Pseudomonas*, *Chromobacterium*, and *Achromobacter*.

##### 4.2 Fungal Lipases

Fungal lipases have been the subject of extensive investigation since the 1950s and are particularly valued for their thermal and pH stability, high substrate specificity, and functional activity in organic solvents. Their production via batch fermentation processes, coupled with economical extraction techniques, renders fungal lipases more advantageous than their bacterial counterparts for large-scale applications.

Prominent fungal lipase-producing genera include *Aspergillus*, *Candida*, *Humicola*, *Pichia*, *Rhizopus*, *Mucor*, *Saccharomyces*, *Geotrichum*, and *Penicillium*. Among these, *Rhizopus* spp. are widely recognized for their efficiency in converting triglycerides into monoglycerides and catalyzing interesterification reactions of fats and oils. *Mucor* spp. are distinguished by their ability to produce thermostable extracellular lipases, making them suitable for industrial processes requiring elevated temperatures.

#### 5. Marine Sponge Symbiosis

Symbiosis refers to a long-term biological association between two or more organisms living in close physical proximity. In marine ecosystems, such relationships are highly diverse and ecologically significant, with numerous invertebrates and marine animals maintaining symbiotic interactions with complex microbial communities.

These associations provide mutual benefits, including nutritional support, protection, and enhanced survival for both the host and microbial symbionts. For example, sulfur-oxidizing mutualistic bacteria supply fixed carbon to invertebrates inhabiting deep-sea hydrothermal vents, while receiving oxygen and reduced inorganic compounds from their hosts.

Symbiotic microorganisms also contribute to host defense mechanisms through the production of bioactive compounds that deter pathogens and predators. Notable examples include *Alteromonas*-mediated protection of the crustacean *Palaemon macrodactylus* against *Lagenidium callinectes* via the antifungal compound 2,3-indolinedione, as well as the production of the antibacterial enzyme phospholipase A2 (PLA2) by *Streptomyces dendra* sp. nov. MSI051, isolated from the marine sponge *Dendrilla nigra*. These defensive metabolites play a crucial role in protecting the sponge from predation and biofouling.

## 6. Optimization of Lipase Production from Marine Sponge Symbiotic Actinomycetes

The Plackett-Burman experimental design has been utilized to screen for the potential impact of multiple conditions on lipase production. In this experiment, low (-) and high (+) values of every variable were examined. Eleven independent variables were analyzed in twelve different combinations arranged in accordance with the Plackett-Burman matrix. Each variable's major influence was estimated using a standard equation where  $E_{xi}$  represents the variable's primary impact,  $M_i^+$  and  $M_i^-$  represent the trials' respective lipase activity (U/ml) when the independent variables were present in high and low concentrations, and  $N$  is half the total number of trials. Statistical significance and t-values for equal unpaired samples were calculated, and a verification test was run to ensure the improved medium was valid.

As a vital part of the food processing sector, oil and fat modification necessitates environmentally friendly solutions. Lipases enable the modification of lipid properties within glycerides by replacing one or more lipids through the relocation of fatty acid chains. This process has the potential to convert low-quality, low-cost lipids into high-quality, high-value fats.

Lipases act as catalysts for fatty acid esterification, hydrolysis, and interesterification. Fats and oils undergo esterification and interesterification to create specialty fats and partial glycerides, employing positional and fatty acid-specific lipases, respectively. Industrially, these methods show more potential than hydrolysis for mass-producing fatty acids. Hydrolysis of fats and oils using an immobilized lipase membrane reactor has led to a reduction in downstream processing, thereby decreasing total processing costs. A recently developed, environmentally friendly system containing highly selective microbial phospholipases removes phospholipids from vegetable oils (de-gumming).

Research efforts aim to produce food-grade, economically viable, immobilized 1,3-regioselective lipases (e.g., Lipozyme TL 1M) intended for creating frying fats and interesterifying common vegetable oils and lard components. It is not possible to obtain modified acylglycerols by predictable chemical acylglycerol interesterification of fats and oils alone. Therefore, immobilized lipases from *C. cylindracea* Ay30, *C. antarctica* (CAL-B), *Pseudomonas* sp., *G. candidum*, and *H. lanuginosa* are utilized to esterify functionalized phenols and create lipophilic antioxidants for application in sunflower oil.

For the hydrolysis of oils and fats, lipases have been utilized in immobilized, pure, or cell-bound forms across several investigations. Triacylglycerol lipase created by genetically modified *A. oryzae* was scientifically approved for usage in the food sector in 2002 by the ANZFA (Australia New Zealand Food Authority) as a processing aid in the production of fats and oils for oil de-gumming. Furthermore, a new technique for immobilizing lipases based on silica granulation has drastically reduced the time and expense of operations. These creative technologies are currently widely used for the production of commodity oils and fats free of trans-fatty acids.

## 7. Marine Sponge Symbiotic Actinomycetes in Bioremediation and Industrial Applications

In light of environmental activists' growing worries about pollution in industrial soil and water systems, research into the use of actinobacteria in bioremediation has become an active topic of study. For the removal of metals, pesticides, and mixed pollutants, better methods are required. Existing techniques for removing hazardous chemicals are not always efficient, especially with inorganic compounds. While inorganic chemicals are typically only partly degraded (forming intermediates that can be more dangerous than the original pesticide), organic compounds can be entirely decomposed into safe products by microbial action.

Since actinobacteria are common in soil and water and naturally maintain ecological balance by disintegrating organic and inorganic substances, they are practical candidates for bioremediation. Certain species of Actinobacteria utilize pesticides as carbon sources, fully degrading them into harmless base elements. For instance, strains like *Streptomyces spinosus* generate tyrosinase enzymes essential for eliminating phenols, a contaminant present in many water-polluting pesticides. Studies indicate that tyrosinase from these microorganisms is often more efficient than previously identified tyrosinases from mushrooms. Furthermore, *Mycobacterium* and *Streptomyces*, two genera of marine actinomycetes, are highly effective degraders of organic pollutants in marine environments. Some marine species, such as *S. albus chlorinus*, exhibit pesticide/herbicide degradation activity without negatively impacting the surrounding ecology.

In addition to pesticide rehabilitation, actinomycetes are prospective candidates for removing toxic metals, which are notoriously difficult to extract using conventional procedures. Because Actinobacteria is one of the few bacterial phyla with high metal tolerance, it is ideal for metal bioremediation. Many *Streptomyces* species have the unusual capacity to precipitate, bioaccumulate, and adsorb hazardous metals, enduring high concentrations safely. For example, *S. lividans* has demonstrated the ability to absorb metals like Cd(II) and Cu(II), while other *Streptomyces* and *Amycolatopsis* species can bioaccumulate Pb, Cd, Cr, and Zn. The metabolic processing of heavy metals by the order Actinomycetales is a robust but under-studied subject that calls for further investigation.

*Streptomyces* is also a top choice for treating mixed pollution—the presence of combined harmful organic and inorganic substances. This complex pollution necessitates a wide range of cleaning strategies to prevent inadvertently creating novel dangerous chemicals during treatment. Mixed microbial cultures, incorporating several species, are often utilized to address this. Additionally, plastics—a growing environmental crisis—can be broken down by several actinomycetes, including species of *Rhodococcus* and *Streptomyces*.

The use of actinomycetes for the removal of radioactive waste is another emerging area of bioremediation research. In a study of marine actinobacteria near an Indian nuclear power plant, a species of *Nocardiopsis* was discovered to be capable of adsorbing substantial amounts of radioactive cesium through passive extracellular binding processes. In another investigation, marine *Streptomyces* created an Extracellular Polymeric Substance (EPS) that increased  $\text{Sr}^{2+}$  biosorption. Similarly, *Streptomyces sporoverrucosus* was found to adsorb uranium by collecting large amounts on its cell walls. The identification of actinomycetes capable of radioactive waste removal represents a much safer, more affordable, and environmentally friendly alternative to current cleanup methods.

## 8. Future Directions and Industrial Potential

- Recent studies have highlighted the importance of marine sponge-associated actinomycetes and their lipases for potential biotechnological applications. However, there are still several areas that require more research:
- **Biogeography and Phylogeny:** Exploring the biogeography and phylogeny of sponge-associated actinomycetes and their lipases can provide insights into their potential role in the sponge-microbe symbiosis and their evolutionary relationships.
- **Functional Characterization:** Further investigation is needed into the functional characterization of these actinomycetes and their lipases. This entails identifying their enzymatic characteristics and investigating their prospective uses in sectors including bioremediation, medicines, and biofuels.
- **Isolation and Cultivation Techniques:** For widespread biotechnological use, the development of effective and sustainable techniques for the isolation and cultivation of sponge-associated actinomycetes is required. This demands an understanding of the complex and dynamic interactions between the sponge host, the actinomycetes, and other associated microbes.
- Overall, further research on marine sponge symbiotic actinomycetes and their lipases has the potential to uncover new biotechnological applications and advance our understanding of the complex relationships between sponges and their microbial communities.

## 9. CONCLUSION

In conclusion, marine sponges represent a rich source of actinomycetes that produce biologically active compounds. The symbiotic association between marine sponges and actinomycetes indicates that there is still much to learn about this distinctive ecosystem. Additional research is expected to provide a great deal of novel bioactive substances with significant potential uses in biotechnology, medicine, and industrial enzyme production.

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