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REVIEW

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Fatty acid-heterocyclic hybrids from green algae as novel therapeutic prospects for diabetes management

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Abstract

Diabetes mellitus is a major global health concern, driven by insulin dysfunction, oxidative stress, and chronic inflammation. Current treatments often have side effects, prompting interest in natural alternatives. Green algae have emerged as a promising source of fatty acid-heterocyclic hybrids—bioactive compounds with improved bioavailability, cell permeability, and multi-target antidiabetic activity. This review highlights their structural features, biosynthesis, extraction, and analysis, along with key mechanisms of action, including enzyme inhibition (α-glucosidase, α-amylase), modulation of insulin signaling (AMPK, PI3K/Akt), antioxidant and anti-inflammatory effects, and beta-cell protection. While preclinical results are promising, further research is needed to standardize activity, enhance bioavailability, and validate efficacy through clinical studies. Fatty acid-heterocyclic hybrids from green algae offer a novel and promising approach to diabetes management.

Keywords Green algae, Fatty acid, Diabetes, Antidiabetic agents, Insulin resistance, Bioactive marine compounds, Oxidative stress

1 Introduction

Diabetes mellitus is characterized by high blood sugar for a long period of time and is one of the modern health challenges because of the escalating trend worldwide related to obesity, inactivity, and age [1]. The incidence of diabetes is in the hundreds of millions and is likely to increase in the next two decades [2, 3]. It has been projected that there will be 9.50 million sufferers of type 1 diabetes [4], while the prevalence of type 2 diabetes is on the rise, with an estimated 462 million individuals affected globally, corresponding to 6.28% of the world's population [5]. In Indonesia, according to the International Diabetes Federation, the prevalence of diabetes among adults in 2024 is estimated to be 11.3%, which translates to approximately 20.4 million people [6]. Therapeutic strategies involving synthetic hypoglycemic agents and insulin could successfully lower the blood glucose in the circulation, but these interventions exhibit significant side effects, little success in attacking the root cause of the progression of the disease, and failure to

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manage in the long term [7, 8]. This scenario paves the way for the use of natural products as potential alternatives for managing diabetes, especially from plant-derived compounds [9, 10].

Marine algae are great resources of diverse bioactive metabolites, including polysaccharides, polyphenols, and fatty acids, which can ameliorate the events associated with oxidative stress, inflammation, and metabolism [11, 12]. Among these, green algae possess unique biochemical complexity, which makes them more efficient in synthesizing fatty acid-heterocyclic hybrid molecules [13]. These hybrid molecules share an amphiphilic nature of the fatty acid and a specific biological property exhibited by the heterocyclic rings, such as enzymatic inhibition, anti-oxidation, and cellular regulation. The inhibition of oxidative and inflammatory events could control the deterioration of the beta-pancreatic cells and insulin resistance [14].

This review addressed the role of green algae-based fatty acid-heterocyclic hybrids in regulating diabetes by exploring their mechanism of action and comparing them with other synthetic and natural resources for managing diabetes. This review is based on the available experimental and conceptual advancements to summarize the updated information concerning the discovery, extraction, characterization, and the mechanism of action of green algae-derived fatty acid-heterocyclic hybrid molecules for managing diabetes. This review includes limitations and future studies.

To effectively present the latest advancement of fatty acid-heterocyclic hybrids, this study evaluated data and information extracted from in vitro and in vivo experiments, comparing the use of synthetic hypoglycemic agents and natural resources for managing diabetes, and exploring research and development history. In all three approaches, reliability and relevance were appraised with all aspects of the literature, including methodological challenges. Overall, this is a comprehensive review and provides insight into current advances on green algae-based fatty acid-heterocyclic hybrids in managing diabetes, including its limitations and future prospects.

Few reports concerning green algae-based fatty acid-heterocyclic hybrids have been published related to diabetes, which include alpha-amylase and alpha-glucosidase enzyme inhibition, cellular glucose uptake, and cytoprotective effects on beta-pancreatic cells [15, 16]. However, these findings are preliminary results, and no clinical trials have been conducted. Moreover, the extraction and characterization of different compounds in green algae-derived hybrids require sophisticated protocols in order to extract each component, as the components share similar solubility.

Therefore, the aim of this review is to comprehensively explore the therapeutic potential of fatty acid-heterocyclic hybrid compounds derived from green algae in the context of type 2 diabetes management. This paper provides an integrative synthesis of current knowledge surrounding their extraction methods, structural characteristics, pharmacological activities, and molecular mechanisms of antidiabetic action, particularly focusing on enzyme inhibition, insulin signaling modulation, beta-cell preservation, and anti-inflammatory and antioxidant effects.

The significance of this review lies in its systematic consolidation of preclinical evidence that supports the multifunctionality of these compounds, which may offer a promising natural alternative to conventional synthetic antidiabetic agents that are often limited by adverse effects and mono-target actions. Moreover, by highlighting the molecular duality of fatty acid-heterocyclic hybrids—combining membrane-permeable

fatty acid chains and biologically active heterocyclic moieties—this review underlines their potential as next-generation multitarget therapeutic agents.

The novelty of this work stems from its focused discussion on the untapped potential of green algae as a sustainable and underexplored source of fatty acid-heterocyclic hybrids, a compound class rarely highlighted in marine pharmacology for diabetes management. To our knowledge, this is the first review that specifically unifies the chemical rationale, extraction technology, structural-functional relationships, and therapeutic implications of green algae-derived fatty acid-heterocyclic hybrids in type 2 diabetes, while also outlining current research gaps and proposing future directions, including structure–activity optimization, standardization, and clinical translation.

1.1 Search strategy and literature

The literature search strategy employed for this review involved systematic queries in reputable scientific databases, including PubMed, Scopus, Web of Science, ScienceDirect, and Google Scholar. The search was carefully structured using specific terms grouped into three main concepts: green algae ("Green algae," "Chlorophyta," "Ulva," "Chlorella," "Marine algae"), fatty acid-heterocyclic hybrids ("Fatty acid-heterocyclic hybrid," "Fatty acid hybrid compounds," "Fatty acid conjugates," "Hybrid bioactive compounds," "Heterocyclic fatty acids"), and diabetes management ("Diabetes," "Type 2 diabetes," "Insulin resistance," "Glycemic control," "Antidiabetic"). Boolean operators were strategically applied to combine these concepts effectively, ensuring comprehensive coverage of relevant scientific literature. Inclusion criteria were limited to peer-reviewed studies published in English since 2000, focusing specifically on bioactive compounds from green algae and their antidiabetic properties, including both in vitro and in vivo models. Exclusion criteria omitted studies on non-green algae or those without explicit relevance to diabetes treatment. To enrich the review further, references cited within key retrieved articles, recent review papers on marine bioactives, and pertinent book chapters on marine pharmacology and natural product chemistry were also consulted. This structured approach facilitated a robust analysis, beginning with an introduction to diabetes mellitus and the rationale for marine-derived bioactives, followed by an exploration of the structural properties, synthesis, and pharmacological effects of fatty acid-heterocyclic hybrids from green algae. Subsequent sections detailed extraction and characterization methods, elucidated potential antidiabetic mechanisms—including enzyme inhibition, modulation of insulin signaling, anti-inflammatory, antioxidant effects, and pancreatic beta-cell protection—and summarized key pharmacological findings. Finally, the literature review highlighted limitations, such as challenges in bioavailability, standardization, and the scarcity of clinical trials, proposing future research directions for advancing these promising hybrid compounds toward therapeutic application.

2 Fatty acid-heterocyclic hybrids: emerging bioactive compounds

In the realm of innovative bioactive compounds, this section describes fatty acid-heterocyclic hybrids, their structural aspects, synthesis methods, and pharmacological benefits with reference to diabetes. These compounds are being mined out from green algae as well as synthesized in laboratories. The mechanism of action of these compounds is discussed in this chapter, along with their significant benefits and integration as an innovative treatment method for diabetes as multifunctional bioactive compounds.

2.1 Definition and structural features

A fatty acid–heterocyclic hybrid is a chemical entity in which a heterocyclic core — an organic ring system containing at least one non-carbon atom such as nitrogen, oxygen, or sulfur — is covalently linked to a fatty acid or fatty-acyl moiety (Fig. 1). The fatty acid component typically consists of a long hydrocarbon chain (usually C8–C22) terminated with a carboxylic acid or its derivatives (esters, amides, hydrazides), while the heterocyclic component can range from simple aromatic heterocycles (e.g., pyridine, imidazole, triazole, coumarin) to complex fused systems (e.g., quinoline, thiazolidinedione). This hybridization strategy combines the lipophilicity and membrane-affinity of fatty acids with the diverse biological recognition and reactivity of heterocycles, often enhancing pharmacokinetic properties and bioactivity.

Fatty acid-heterocyclic hybrid compounds, in which a fatty acid is covalently linked to a heterocyclic ring, have attracted much attention in the last decades. The addition of a fatty acid may enhance the solubility of natural products in hydrophobic lipid membranes and thereby improve their oral bioavailability and systemic distribution [17], often limited by the poor solubility of many natural products in either aqueous or lipid environments. In addition, the covalent linkage between the fatty acid and the heterocyclic moieties is likely to improve their stability in vivo and transport them across cellular membranes, which would be advantageous because diabetes pathophysiology mostly occurs in membrane environments.

A comprehensive look at the chemical structures of fatty acid-heterocyclic hybrid compounds reveals a wide range of molecular diversity that underpins their varied biological activities. These hybrids, formed by covalently linking a fatty acid to a heterocyclic ring, have been a focus of extensive research, particularly in the synthesis and evaluation of their medicinal properties [18]. The structural variations often involve the type of heterocyclic ring and the length and saturation of the fatty acid tail. For instance, five-membered heterocycles such as pyrazole, pyrrole, and oxadiazole have been synthesized with various fatty acid chains. In one study, a series of 2,5-dimethyl pyrrole derivatives were created by reacting fatty acid hydrazides with acetonyl acetone, demonstrating how a fatty acid chain can be integrated into a nitrogen-containing heterocyclic system [19]. Other research has focused on six-membered heterocycles like quinoline and isoquinoline, where fatty acid chains were attached to form hybrid molecules [20]. The specific chemical linkages, such as amide bonds, can be crucial in creating stable and effective compounds for therapeutic applications. These studies collectively highlight how the precise chemical structure, including the type of heterocyclic scaffold and the characteristics of the fatty acid, can be systematically altered to produce a library of compounds with diverse biological profiles.

This structural diversity is a key determinant of the compounds' biological and pharmacological properties. For example, the anticancer activity of these hybrids has been shown to be dependent on both the nature of the heterocyclic ring and the length of the fatty acid chain [13]. Some studies have found that a carbon chain length in the range of

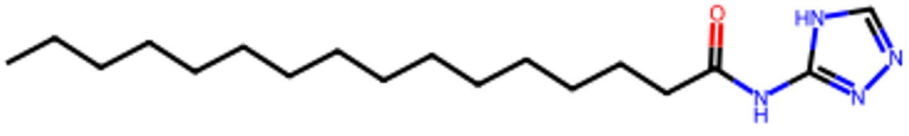


Fig. 1 Example of C16 Fatty Acid (1,2,3-triazole Hybrid)

14–18 atoms is optimal for inhibiting cancer cell growth, while the addition of double bonds can also enhance potency and selectivity against certain cell lines [21]. Beyond cancer, these fatty acid-heterocyclic hybrids have also shown promise as antimicrobial and antidepressant agents, with their activity influenced by the specific fatty acid chain length and the presence of hydroxyl groups on the chain. The strategic combination of fatty acids, which can enhance lipophilicity and membrane permeability, with heterocyclic rings, which provide specific bioactivity, offers a powerful platform for designing next-generation therapeutics. This approach allows researchers to create compounds that are not only potent but also have improved bioavailability and reduced toxicity by potentially targeting multiple disease pathways simultaneously.

Combining two bioactive domains with complementary functions is the rationale behind hybrid natural products in drug discovery. Regarding fatty acid-heterocyclic hybrid compounds, the typical features are medium- to long-chain fatty acid residues and nitrogen, oxygen, or sulfur heterocycles. Nitrogen heterocycles such as pyrroles and imidazoles can target specific enzymes and receptors in the cell that are important for glucose homeostasis and insulin signaling in type 2 diabetes [22]. The modulation of the length and saturation of fatty acids influences the affinity of fatty acid-heterocyclic hybrid compounds toward membrane environments, enhancing their ability to selectively interact with lipid bilayers. The incorporation of oxygen or sulfur atoms into the heterocyclic portion increases redox potential, enhancing their antioxidant activity.

This strategy can be used to obtain hybrid compounds tailored for the treatment of diverse aspects of type 2 diabetes, ranging from glycemic control to prevention or delay of diabetic complications (Fig. 2). Fatty acid-heterocyclic hybrid compounds reduce postprandial hyperglycemia by inhibiting α-glucosidase and α-amylase activity in the small intestine, both of which are very important for complex carbohydrate metabolism [23]. The inhibition occurs by blocking the enzyme's active site for substrate binding by competitive or non-competitive mechanisms.

By combining dual targets in the treatment of diabetes – inhibition of carbohydrate metabolizing enzymes and oxidative stress-related factors – fatty acid-heterocyclic hybrid compounds control both the source and utilization of glucose. The fatty acid portion anchors the hybrid compound to cell membranes or to the surface of digestive enzymes, and the heterocyclic portion inhibits or modulates enzyme activity [24]. This can lead to a more gradual release of glucose into the bloodstream, thus lowering postprandial glycemia and mitigating diabetic complications. This is a dual-target drug strategy for which the hybrid is better able to provide its pharmacological effect than using a single compound alone.

Some studies have focused on the multiple diabetes-relevant molecular functions of green algae-derived fatty acid-heterocyclic hybrid compounds, and there is also evidence that they improve insulin signaling, reduce inflammation, and protect pancreatic beta cells from stress-induced cytotoxicity [25, 26]. It can be said that green algae-derived fatty acid-heterocyclic hybrid compounds combine multiple molecular and metabolic functions. In these types of hybrids, the modulation of adiponectin secretion and the activation of AMP-activated protein kinase (AMPK) have been demonstrated, two molecular targets directly associated with enhanced glucose homeostasis and metabolism [27]. Also, the antioxidant and anti-inflammatory activities enhance beta-cell survival, which further lowers the risk of late diabetic complications as the patients are able

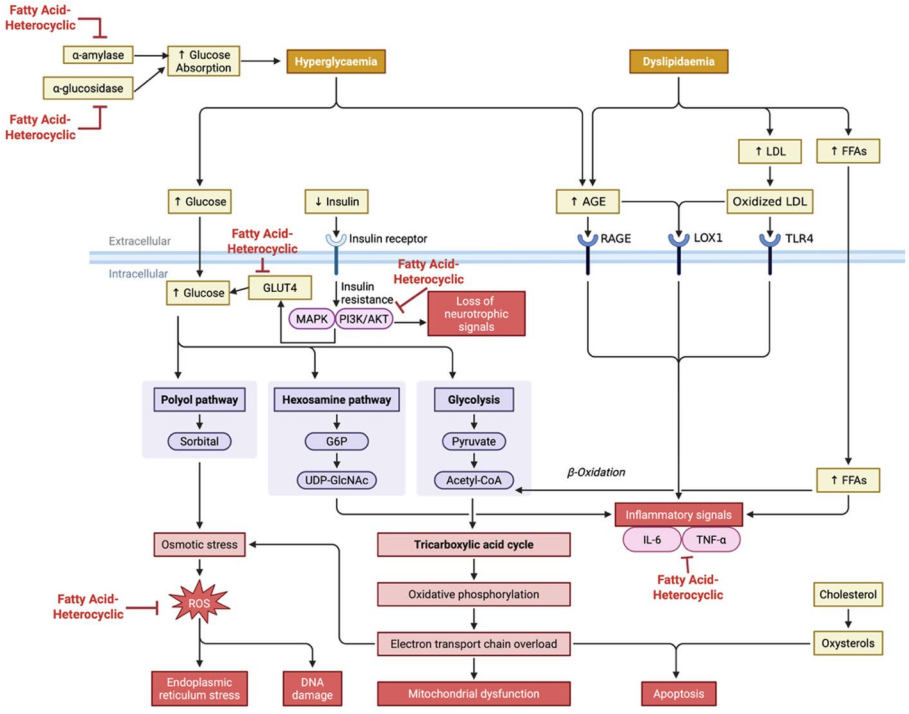


Fig. 2 Pharmacological Mechanisms of Fatty Acid-Heterocyclic Hybrids Against Type 2 Diabetes. The figure summarizes the main antidiabetic mechanisms of fatty acid-heterocyclic hybrids: inhibition of α-glucosidase and α-amylase, modulation of insulin signaling (via PI3K/Akt, AMPK), GLUT4 translocation, oxidative stress reduction, and anti-inflammatory effects through cytokine regulation. Each mechanism is linked to hybrid structural domains. Created using BioRender

to produce their own insulin. Thus, they cover multiple parameters and molecular processes altered in type 2 diabetes.

The dual function of green algae-derived fatty acid-heterocyclic hybrid compounds has the potential to link membrane-related metabolic regulation with intracellular signaling pathways and thus to improve their antidiabetic activity [13]. The amphiphilic feature can facilitate the binding of hybrid molecules to cell membranes and therefore influence membrane-related processes, such as the action of glucose transporters and localized enzymatic activities, and the heterocyclic portion impacts insulin signaling, inducing a cascade of metabolic events inside the cell, such as improved glucose transport and AMPK phosphorylation. In addition, the anti-oxidative activity protects pancreatic beta-cells against high-glucose- and high-cholesterol-induced cellular oxidative stress in vitro.

It is likely that the spatial arrangements and/or the precise chemistries of the hybrid linkages in fatty acid-heterocyclic hybrid compounds are important to ensure their specificity and potency toward distinct diabetic-relevant molecular targets, which would further contribute to the rational bioengineering of these natural products with improved drug candidates. Consistent with this hypothesis, in vivo bioactivity studies of total green algae extracts showed significant improvements in several diabetic metabolic markers. And further structural-bioactivity association analysis proved that these beneficial activities are exclusively attributed to the presence of fatty acid-heterocyclic hybrid compounds [13]. This opens the door for green algae to become a valuable resource of

multifunctional natural drug candidates with the potential of becoming innovative therapeutics against type 2 diabetes.

In conclusion, the structural features, bioactivity, and multi-target mechanism suggest that fatty acid-heterocyclic hybrid compounds may be potent lead candidates for the treatment of diabetes.

2.2 Synthetic approaches and biosynthetic pathways

Fatty acid-heterocyclic hybrids from green algae can be produced by biosynthetic mechanisms that occur naturally in algal cells, or through synthetic chemical routes in the laboratory [28]. Each method offers several advantages but is equally limited by others, rendering a unified approach more efficient and productive. In green algae, fatty acid-heterocyclic hybrids are naturally biosynthesized by enzymatic processes involving esterification or amidation between fatty acid precursors and heterocyclic moieties [18]. Biosynthetic capacity in green algae permits the formation of structurally diverse hybrids. It also provides an environmentally friendly process using renewable resources. However, yields are often low and subject to algal species and growth conditions. Chemical synthesis methods can offer more scalable procedures for the production of fatty acid-heterocyclic hybrids as well as allow for a more precise structural modification and synthesis of analog libraries for a better understanding of the structure-activity relationship.

However, one cannot have access to a diverse array of bioactive hybrids made naturally by algae using solely synthetic routes. To extract fatty acid-heterocyclic hybrids from green algae, solvent-based techniques such as methanolic or ethanolic extraction have been applied. In these extractions, solvent polarity, temperature, and duration of extraction are often optimized to isolate fatty acid-heterocyclic hybrids [29]. To isolate pure compounds from algal extracts, liquid-liquid partitioning and high-performance liquid chromatography (HPLC) have been used to separate fatty acid-heterocyclic hybrids from complex algal biomass. Linking these isolation protocols to bioassay-guided screening is important for correlating antidiabetic effects to isolated compounds. There are also significant hurdles for consistency between laboratories. The difference in algal biomass from year to year and in different locations may hinder the reproducibility of antidiabetic activity between laboratories.

Because the chemical structure of many fatty acid-heterocyclic hybrids found in green algae is similar to several metabolites commonly found in algal extracts, gas chromatography-mass spectrometry (GC-MS), liquid chromatography-mass spectrometry (LC-MS), and nuclear magnetic resonance (NMR) spectroscopy have been implemented for metabolite characterization and identification to provide structural characterization and identification of hybrid fatty acid-heterocycles and to ensure their structural distinction from any non-hybrid substances with similar structure [30, 31]. In the identification and characterization of fatty acid-heterocyclic hybrids, GC-MS, LC-MS, and NMR can be used to distinguish hybrid metabolites with varied alkyl chain length, degree of unsaturation, and diverse heterocyclic ring systems for assessment of their ability to inhibit enzymes involved in the pathogenesis of T2D.

Green algae present significant structural diversity in fatty acid-heterocyclic hybrid synthesis, a result of combining the structural and physicochemical variety inherent to fatty acid tails with various heterocyclic moieties. Structural variations include fatty acid

chains that vary in length and degree of unsaturation as well as several combinations of five- and six-membered heterocyclic systems [18]. Fatty acid tail length and degree of unsaturation are key determinants for the bioavailability of the hybrid in terms of the kinetics of drug absorption and metabolism, plasma levels of the drug, distribution of the drug throughout the organism, and bioavailability. The degree of saturation directly determines the pharmacokinetics, lipophilicity, and capacity for membrane-targeting ability [13]. Heterocycles are useful structures in modern therapeutics because of the high degree of biological activity that they contribute to their conjugates. Additionally, the functional groups on the heterocyclic systems serve as binding sites and increase cell and tissue specificity through different mechanisms [32]. In contrast to fatty acids, the role of the heterocyclic part of hybrid fatty acid-heterocycles in in vivo potency and bioavailability is generally less understood. It is believed that diverse heterocyclic substituents lead to variable and diverse enzyme inhibition, bioactivity, cellular uptake, and specificity.

Although algae have the capacity to synthesize a vast diversity of fatty acid-heterocyclic hybrids, the exploitation of biosynthetic pathways to drug discovery requires careful considerations of the biosynthesis and potential biosynthetic regulation of these complex hybrid molecules within the algal cell. Biosynthetic processes in algae can aid in the isolation of hybrids and identification of lead structures for the development of antidiabetic therapies [11]. Optimization of extraction strategies and protocols for algal lipids must be coupled to characterization of their genetic regulation. Also, co-culture strategies can be exploited for enhancing specific active compounds. It has been shown that the production of select compounds is greater in the presence of bacteria co-cultured with microalgae compared to pure algal culture [33].

Genetic alterations in the metabolic pathways of algal biosynthesis also hold significant potential for altering the lipid content of algae and potentially enhancing production of hybrid fatty acid-heterocycles with higher activity against diabetes-related enzymes [34]. These efforts can be complemented with the analysis of "omics" data, where, in some cases, increased levels of transcripts involved in fatty acid biosynthesis have been recorded after exposure to nitrogen depletion and enhanced light intensities in algal biomass. Combining multiple biochemical analyses, such as genome and proteome information, with other cellular characteristics, such as lipid and biomass content, leads to optimized algal biomass production conditions as well as extraction protocols for hybrid lipids with optimal health benefits that can be used in downstream clinical studies. Therefore, genetic information will be instrumental in the sustainable harvesting and biosynthesis of hybrid fatty acid-heterocyclic metabolites for antidiabetic therapeutic development.

In summary, both biosynthetic and synthetic routes offer important paths for discovering fatty acid-heterocyclic lead compounds to aid in antidiabetic therapy development. While the algal biosynthetic routes show many potential advantages in drug discovery in regard to yield, diversity, and environmentally conscious chemistry, a hybrid approach combining laboratory chemistry with algal biosynthesis presents an opportunity to circumvent some of the inherent limitations in algal culture variability and low yield, as is generally the case in biosynthetic pathways.

2.3 Pharmacological advantages of hybrids

Fatty acid-heterocyclic hybrids synthesized from green algae have good pharmacological advantages due to their structural duality, which improves their potential usefulness in antidiabetic therapy. The structural duality involves the membrane targeting ability of the fatty acid moiety with enzyme and receptor selectivity by heterocyclic scaffolds [35]. This helps to improve their bioavailability and efficiency of multi-target modulation, thus showing better effectiveness compared to monofunctional antidiabetic drugs. Thus, increasing their ability to be properly delivered and dispersed in cells to achieve their therapeutic potential. This characteristic gives it an edge over other natural products and places it as a potential candidate for further development of drugs with antidiabetic ability because of its ability to penetrate membrane/aqueous interfaces.

One of the pharmacological advantages of fatty acid-heterocyclic hybrids is their strong ability to selectively bind to enzymes and receptors that are linked with the pathology of type 2 diabetes, e.g., α-glucosidase, α-amylase, and PTP1B, and thus inhibiting them [36, 37]. Inhibition of these enzymes and receptors with high selectivity is critical for diabetes management, because they are involved in pivotal steps in glucose metabolism. This ability to selectively bind to enzymes and receptors is because of their heterocyclic scaffolds, which fit the enzyme and receptor binding domains. This gives it more advantages than the non-hybrid compounds, which possess a single target inhibitory activity. Due to the multiple processes affected, they are effective in reducing the long-term metabolic impairment in people with T2D. Therefore, further study of this ability is needed.

In order to avoid chronic postprandial hyperglycemia, there is a need to retard the absorption and metabolism of sugars and other complex carbohydrates, and therefore, fatty acid-heterocyclic hybrids help in managing blood sugar and long-term complications associated with type 2 diabetes [25]. In addition to the inhibition of α-glucosidase and α-amylase, they have been found to reduce the secretion of inflammatory cytokines like TNF-α and IL-6. This anti-inflammatory activity helps in reducing the onset of insulin resistance. The ability to reduce inflammation is important in diabetes mellitus since it induces insulin resistance. Also, they reduce the impact of oxidative stress. In addition to improving the activity of antioxidant enzymes, which is critical for combating reactive oxygen species (ROS), they also have a protective role in preserving the health and functionality of pancreatic beta cells, which is important for insulin synthesis. With these findings, it is clear that fatty acid-heterocyclic hybrids improve insulin sensitivity, reduce inflammation, and oxidative stress. Hence, fatty acid-heterocyclic hybrids have an advantage over the synthetic drugs and thus should be further studied.

The fact that fatty acid-heterocyclic hybrids activate AMPK in vitro and improve glycemic control in vivo makes them superior candidates for diabetes mellitus prevention and treatment when compared to other monofunctional natural products [38]. Therefore, to enhance the potential pharmacological activity of fatty acid-heterocyclic hybrids as anti-diabetic agents, further studies on their pharmacological activities should be explored to prevent, manage, and reduce the risk of developing type 2 diabetes mellitus and other metabolic syndrome-related ailments. In addition, fatty acid-heterocyclic hybrids also enhance adiponectin secretion, which induces insulin sensitivity. Insulin sensitivity is affected by inflammation and is usually present in individuals suffering from T2D; since the fatty acid-heterocyclic hybrids exhibit anti-inflammatory activities,

these hybrids can improve glucose uptake in vivo by decreasing oxidative stress in cells and boosting cell metabolism by restoring insulin sensitivity.

The lipid chains present in these hybrids contribute to enhancing their oral absorption and cell permeation, while the heterocyclic domains are mainly involved in inhibiting glucose metabolism enzymes such as PTP1B and DPP-4, insulin signaling pathways, and improving insulin sensitivity. A study by [39] showed that extracts of seaweeds Ulva and Chlorella, which are mainly made up of fatty acid-heterocyclic hybrids, improve glucose tolerance and reduce the levels of fasting blood glucose and plasma insulin compared to the other non-hybrid compounds extracted. This indicates that hybridization helps to achieve better PK and PD properties. Thus, studies exploring the optimal configuration of these chains and scaffolds can yield potential therapeutic molecules against T2D.

Based on the current evidence, it can be proposed that fatty acid-heterocyclic hybrids help to bridge the gap between transmembrane processes, such as glucose uptake, and intracellular pathways, such as insulin sensitivity enhancement. This hypothesis indicates that these hybrids possess a dual anchoring and enzyme-regulating mechanism of action and hence are potential candidates for antidiabetic therapy. They affect the GLUT4 translocation on the cell surface by inhibiting insulin receptors. They have also been proposed to stimulate metabolic pathways (AMPK) and improve signaling pathways (PI3K/Akt) in insulin-responsive tissues [40]. More research to modulate and understand the role of these hybrids on insulin receptor distribution within lipid rafts and their clustering could reveal more evidence of their ability to improve insulin receptor sensitivity.

Moreover, fatty acid-heterocyclic hybrids possess health-promoting properties such as antioxidative and hepatoprotective activity. It has been established that they possess strong antioxidant ability in vivo by suppressing the oxidative stress levels [21]. Antioxidants and ROS scavengers (e.g., superoxide dismutase, catalase) have been shown to decrease the levels of oxidative stress markers in various experimental T2D models, and, therefore, it is of great importance that more extensive evaluations are done to better understand the anti-oxidative mechanisms of fatty acid-heterocyclic hybrids in vitro, as well as to examine the effectiveness of this anti-oxidative activity. With the antioxidant abilities of these hybrids, they may protect cells from hyperglycemia-induced toxicity in diabetic subjects and other metabolic diseases characterized by enhanced oxidative stress levels. On the other hand, fatty acid-heterocyclic hybrids are a very attractive choice as potential hepatoprotective candidates due to their ability to inhibit lipid peroxidation, lipid accumulation, and inflammatory responses in liver tissue. They not only help to reduce fat accumulation, thereby preventing fatty liver, but they also help in reducing lipid peroxidation. With their hepatoprotective and antioxidant characteristics, the natural sources of fatty acid-heterocyclic hybrids will definitely make an ideal therapeutic resource for improving the liver's health and help avoid fatty liver associated with diabetes mellitus. Besides, these fatty acid-heterocyclic hybrids are found to possess great hepatoprotective effects, which were reported by [41], and ursolic acid, a derivative of them, has been further studied to promote its solubility and bioavailability by undergoing esterification into oleanolic acid-3-O-oleate ester. They show great efficiency as a potential therapeutic agent for the treatment of metabolic syndrome-related complications and hepatopathy in vivo. In addition, their anti-angiogenic effect against

the abnormal growth of blood vessels, leading to damage in diabetic retinopathy, contributes to their ability to treat more complications resulting from T2D.

Based on the various roles they play, such as improvement of insulin sensitivity, the fact that these hybrids have multi-target characteristics on enzymes and receptors, and the activation of the molecular pathways of glucose uptake and secretion (adiponectin secretion), fatty acid-heterocyclic hybrids offer the potential for improved therapy in the treatment and prevention of diabetes. AMPK also induces skeletal muscle and adipose glucose uptake and metabolism via phosphorylation-dependent mechanisms that enhance glucose transporter translocation and glucose clearance. Therefore, a promising strategy to restore insulin receptor function and signaling activity would be to inhibit protein kinases involved in these processes, as well as to improve glucose uptake through regulating PPAR and PI3K/Akt. Based on their potent anti-diabetic properties, these fatty acid-heterocyclic hybrids are very appealing for the development of new therapeutics, especially for the prevention, management, and reduction in the risk of developing T2DM and related metabolic diseases, such as dyslipidemia and cardiovascular ailments, both in humans and in animal models.

3 Green algae as source of fatty acid-heterocyclic hybrids

The sustainable production of fatty acid-heterocyclic hybrids, which possess antidiabetic properties that hold promise, can be achieved through green algae, a natural reservoir. The complexity and richness of these bioactive compounds are revealed through exploring their identification, extraction, and characterization techniques. For the purpose of harnessing the full potential of algae in the development of innovative and effective diabetes therapeutics, an understanding of these processes is essential, particularly within the broader scope of natural product research.

3.1 Identification and extraction techniques

Extraction of fatty acid-heterocyclic hybrids from green algae is essential in developing them as potential antidiabetic drugs (Fig. 3). Methanolic and ethanolic-based solvents have been widely used to extract the hybrids due to higher yields and minimized degradation. Methanol is most efficient when extracting polar heterocycle-bearing hybrids, while mixed solvent systems are used for the extraction of polar and non-polar hybrids. However, using these conventional solvents as the major method indicates a knowledge gap to use more benign solvents for hybrid extraction.

Temperature regulation can minimize the degradation of hybrids during extraction. Cold extraction or room-temperature extraction preserves the native structure of enzymatically synthesized hybrid molecules. For instance, heated conditions and oxidative processes cause the degradation of several hybrid structures of fatty acids and heterocycles [42]. It has to be considered that it takes long extraction times to prevent any thermal or oxidative denaturation of bioactive molecules when using lower-temperature extraction conditions.

Prior to extraction, pretreatment of algal biomasses to remove impurities such as other organic residues, cellular organelles, cellular debris, and excess of water, may improve the efficiency of hybrid extraction [43]. During pretreatment, algal biomass is homogenized, and then the resulting homogenate is filtered for the removal of insoluble particles. This procedure promotes solvent penetration into algal cells, increasing the yields

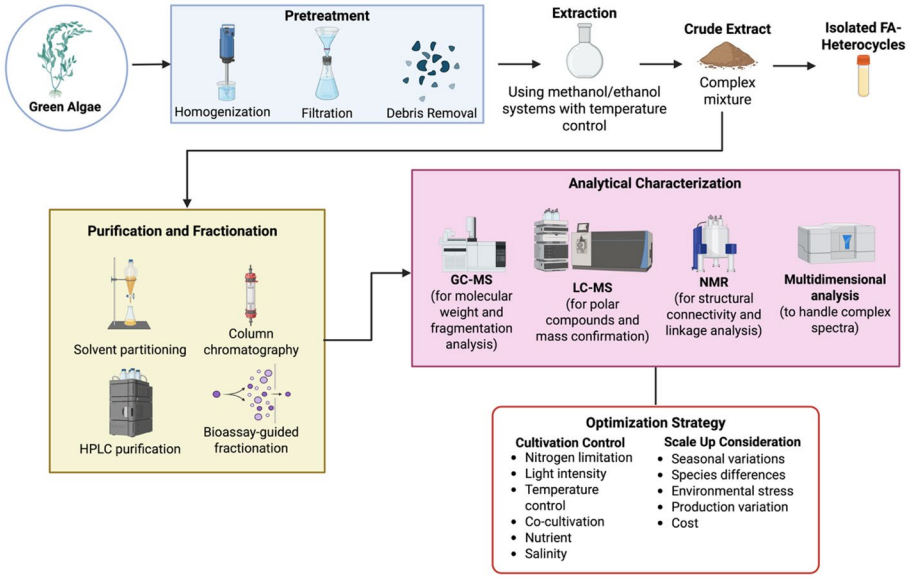


Fig. 3 Extraction and Characterization Workflow of Fatty Acid-Heterocyclic Hybrids. Overview of the extraction, purification, and identification steps of fatty acid-heterocyclic hybrids from green algae. Techniques include methanol/ethanol extraction, solvent partitioning, HPLC, GC-MS, LC-MS, and NMR for structural elucidation. Optimized protocols enhance hybrid yield and enable bioactivity correlation. Created using BioRender

of desired hybrids, by making the procedure more reproducible. The reproducibility, though, can depend on factors such as the source and composition of green algal biomass, solvent type, sample preparation, and operational parameters.

Several analytical tools are useful for chemical characterization, structural identification, and quantification of these complex hybrid molecules. GC-MS, LC-MS, and NMR are widely utilized to identify the presence of fatty acid-heterocyclic hybrids in green algae. GC-MS and LC-MS techniques can be utilized to detect minor hybrid constituents present within algal extracts, even at trace concentrations [44]. In addition to the quantitative analyses, mass spectroscopy is an important tool for the qualitative identification and structural confirmation of the fatty acid-heterocyclic hybrids. NMR spectroscopy is used to analyze the connectivity of fatty acids and heterocycles and provides detailed structural information to identify their identities. NMR can distinguish a particular hybrid from many similar compounds present in the crude extract by determining their specific connections, which are used to verify structural assignment. Due to the structural diversity and similarities of the compounds extracted from algal matrices, challenges like overlapping spectra may lead to ambiguous results. Therefore, multidimensional spectral analysis and sequential purification techniques are used to improve structural identification accuracy.

Isolation of fatty acid-heterocyclic hybrids is challenging due to the simultaneous extraction of multiple algal constituents with varying compositions. Crude algal extracts often show the presence of mixtures of hybrids and other organic metabolites. A single isolation step for these hybrid molecules is hard to achieve due to the abundance of many compounds bearing the same chemical composition; thus, isolation needs to be accomplished by a combination of several extraction and isolation steps. A standard isolation scheme comprises extraction, purification, isolation, and analysis. Due to low yields of natural hybrid products, multistep isolation, which requires a large number of

solvents, and tedious and time-consuming steps, are required to achieve purified fractions for activity evaluation [45]. Most chromatographic techniques for hybrid isolation involve partitioning based on relative lipophilic/hydrophilic ratios among solvents, and/or column and high-performance liquid chromatography (HPLC) that utilizes varying resin and eluent combinations to facilitate the separation. Bioassay-guided fractionation further confirms the hybrid's potential antidiabetic activity, which can be further assessed for other therapeutic and biomedical purposes.

Scale-up of hybrid extraction is difficult to manage because of the compositional variation within natural resources during different phases of development. Algal cultivation may be limited because of species-specific seasonal variations in biomass. Production variation is one of the greatest challenges when attempting to scale up the extraction of hybrids because natural product production by algal metabolism varies depending on climate change and other environmental stress factors [46]. Due to species differences, different biomass availability, variations in laboratory-scale extraction, and changes in the environment, extraction and scale-up are often difficult to compare for different algal extracts.

Although extraction techniques have been developed to isolate a wide variety of hybrids, there is an increased desire to manipulate algae for higher yields of these important molecules. It has been reported that, due to the environmental stresses, algae accumulate a diversity of secondary metabolites and other chemical constituents as adaptive measures to combat the stress effects. Therefore, it is possible to affect the secondary metabolites through changing a wide variety of physical parameters, such as light exposure, temperature, nutrients, and stress stimuli. Nutrient limitation increases the yield of fatty acid-heterocyclic hybrids in some algae. For instance, depriving of nitrogen supply or increasing irradiance induced increased hybrid biosynthesis in several algae [47, 48]. Nutrient limitation or environmental stress stimuli such as high salinity could improve the hybrid content per cell in algae and, therefore, can make algae more effective in production. Similarly, co-cultivation of algae with other organisms, such as bacteria or yeasts, can improve the yields of hybrids. Additionally, altering the levels of enzymes involved in the biosynthesis of fatty acid-heterocyclic hybrids is one of the promising ways to enhance the synthesis of target products by metabolic pathway regulation.

In order to produce target compounds in large quantities, algal cultivation needs to be performed with high reliability and reproducibility. Bioreactor culture techniques have been developed to regulate environmental factors and to grow algae on a large scale for the production of biomass [49, 50]. In controlled, artificial environmental conditions within bioreactors, algal strains may produce a higher amount of desired fatty acid-heterocyclic hybrids and can yield reproducible results. Moreover, monitoring algal strains within bioreactors using omics technology can provide insights that could lead to further increased accumulation of hybrids with therapeutic importance. Despite the potential for higher biomass productivity and regulated biochemical quality, cultivation in bioreactors can be extremely expensive to build and requires significant investment in human resources and expertise.

Most cultivation methods are usually designed according to the species-specific properties, and environmental conditions or stresses. Fatty acid-heterocyclic hybrids in algae are also influenced by the availability of nutrients, environmental factors, and light intensity. Therefore, different factors can greatly impact the yield, production, or properties

of algae. Based on such analyses and optimization, a selective medium and the precise method of nutrient supplement can be decided. With careful control of environmental variables, higher biomasses can be cultivated with limited resources.

3.2 Characterization of compounds derived from green algae

Characterization of fatty acid-heterocyclic hybrids from green algae is important for their development as antidiabetic agents. Analytical methods such as GC-MS, LC-MS, and NMR are necessary to determine the molecular weight, fragmentation pattern, and proton and carbon environment, and to confirm their hybrid structures from those of non-hybrid metabolites. This guarantees that observed antidiabetic activities are attributed to correctly identified hybrids. Algal extract complexity means that the methods have to be integrated to yield unambiguous results. Application of the methods demands reliable, robust protocols for identification to benefit from their potential as therapeutics.

Spectroscopic methods, particularly 2D NMR spectroscopy and MS/MS techniques [51], are valuable in determining the linkage between fatty acids and heterocycles by probing the structural relationship between heterocyclic rings and fatty acid chains. Such knowledge is critical to linking structure to the activity of different combinations. Even minor changes in the linkage can alter pharmacological outcomes. The downside of the methods is that they require a well-trained specialist and are expensive to operate and maintain, limiting their use to well-equipped laboratories.

The complex metabolite composition in green algae requires the use of sequential and complementary purification protocols to isolate fatty acid-heterocyclic hybrids. Sequential solvent partitioning to fractionate crude algal extracts using solvents of differing polarities can reduce non-hybrid constituents in fractions containing hybrids. This ensures less interference from metabolites in bioassays to measure antidiabetic activity. Fractionation and purification techniques provide fractions for HPLC-based purification for the structural isolation of closely related fatty acid-heterocyclic hybrids from extracts and algal matrices [52]. Combining these techniques increases chances for proper identification and ensures that bioactivity measured is attributable to specific hybrids rather than co-extracted non-hybrids, despite the difficulty and labor of these methods. The application of HPLC is useful for hybrid isolation from algal matrix constituents to ensure adequate compound amounts for pharmacology testing. Combined or orthogonal purification steps ensure that hybrid structural characterization is associated with the observed antidiabetic effects. This minimizes any possibilities of improper bioactivity assessment owing to the presence of non-hybrids and increases chances for more reliable attribution of bioactivity.

The lipid composition of green algae, such as Ulva lactuca, includes various saturated, monounsaturated, and polyunsaturated fatty acids and their derivatives. Unsaturated fatty acids have been found to improve cytoprotective effects such as the activation of Nrf2-ARE [53]. Therefore, different structural fatty acid variants from green algae are worth exploring for their pharmacological benefits as antidiabetic agents. The limitation is the diversity of fatty acids in algae among the algal species. All green algal fatty acids can act as precursors for fatty acid-heterocyclic hybrids [13].

Another potential approach is to culture algae under controlled conditions in an attempt to elicit and increase the expression of certain biosynthetic pathways in green algae. Some of the cultivation conditions tested for their influence on metabolic

production were light intensity, nutrients, temperature, and salinity. It was observed that nitrogen-deprived and high light intensity cultures, in particular, had increased total biomass, metabolite yield, and the expression of targeted biosynthetic pathways [54, 55]. This is advantageous in that it is cost-effective and eco-friendly. This also addresses some of the limitations of traditional methods that rely on complex and labor-intensive chromatographic protocols that separate only based on chemical attributes and not by biological activity [13].

In conclusion, characterization of fatty acid-heterocyclic hybrids from green algae requires in-depth scientific exploration and advancement to enhance the possibility of their development as antidiabetic therapeutics.

3.3 Antidiabetic activities of algae-derived hybrids

A number of fatty acid-heterocyclic hybrids from green algae have been determined as inhibitors of both α-glucosidase and α-amylase, which prevents the absorption of glucose (see Table 1), therefore avoiding hyperglycemia after meals. This ability to reduce postprandial glucose levels and blood sugar is especially important for diabetic patients [56]. In green algae, most hybrids inhibit both carbohydrate-digesting enzymes; this suggests that their structures can occupy the binding sites of both enzymes. These properties suggest that fatty acid-heterocyclic hybrids can be further developed as anti-diabetic drugs [23]. However, these assumptions need to be further studied in order to elucidate how the molecular diversity of the hybrids affects the inhibition mechanisms and how to obtain improved hybrids for glucose absorption control.

In crude algae extracts, it has been suggested that polyphenols and polysaccharides might exert synergy effects with the fatty acid-heterocyclic hybrids. They could synergistically exert glucose level-regulating activity and improve other related health benefits [56]. Since the results regarding hybrid activity are commonly based on the activity of the crude extract or fractionations, this limits the possibility to clearly assign such activity solely to the isolated hybrid; instead it is recommended to assess it by isolating and identifying such compound utilizing bioassay-guided fractionation of these complex mixtures. Furthermore, the interactions between hybrids with other algae bioactive compounds need to be clearly investigated.

Based on the above results, the following hypothesis is proposed. The fact that fatty acid-heterocyclic hybrids appear in numerous different species and display a high number of variations suggests that this molecular diversity enables a widespread mechanism of action. In this way, their capability to inhibit the intestinal uptake of glucose might derive from the nature of their structure. They may be able to insert into multiple binding sites from the intestine enzymes and transporters involved in glucose absorption and control postprandial glucose levels. This hypothesis needs to be validated with a more comprehensive assessment in the form of molecular docking and enzymatic tests to provide substantial data regarding enzyme bindings and activity [57].

Studies in Ulva and Chlorella species have also shown that fatty acid-heterocyclic hybrids improve glucose uptake by cells, leading to increased insulin sensitivity [13]. This is a crucial feature in treating insulin resistance, a hallmark of type 2 diabetes. Fatty acid-heterocyclic hybrids can enhance the translocation of glucose transporters (GLUT) in the skeletal muscle and adipose tissues, which could further lead to improved peripheral uptake of glucose by the body. Several in vitro experiments have also identified an

Table 1 Antidiabetic data for fatty acid heterocyclic-hybrid molecules

Authors (Year)	Hybrid Type (Short)	Model (In Vitro / In Vivo)	Reported IC_{50} or Dose (Units As In Paper)	Comparative Activity / Note
Boudebbous et al. (2022) [64]	1,3,4-Thiadiazole – 1,2,4-triazine hybrids (series where heterocyclic cores were acylated/modified; paper presents acyl-type substitutions)	In-vitro α-glucosidase assay	IC_{50} range 14.36 ± 0.70 → 743.15 ± 0.86 μM; best = 14.36 μM	Best analogs ≈ are significantly more potent than the acarbose control reported in that study (acarbose ≈ 844.8 μM in the same assay)
Khan et al. (2023) [65]	1,2,3-triazole containing bis-hydrazone hybrids (lipophilic acyl groups introduced on heterocycle scaffold)	In-vitro α-glucosidase & α-amylase	IC_{50} range reported ≈ 0.70 ± 0.05 → 35.70 ± 0.80 μM for the most potent series	Several compounds showed sub-10 μM α-glucosidase inhibition and dual α-amylase/α-glucosidase activity — suggested as lead molecules (paper compares with acarbose)
Pedrood et al. (2022) [66]	Diphenyl-quinoxaline-6-carbohydrazide hybrids (acyl-hydrazone motifs = acylated/hydrophobic substituents on heterocycle)	In-vitro α-glucosidase assay	IC_{50} range 110.6 ± 6.0 → >750 μM; most potent ~ 110.6 μM	Authors compared to acarbose (acarbose reported ~ 750 μM in their assay); some hybrids were better than acarbose in that assay
Esmaili et al. (2023) [67]	Pyrano[3,2-c]quinoline-1,2,3-triazole hybrids — hydrophobic substituents/alkyl/acyl side chains used to tune lipophilicity	In-vitro α-glucosidase & α-amylase.	Example best: compound 5 g IC_{50} = 7.54 ± 1.10 μM (others in low-μM range)	Several hybrids showed sub-10 μM α-glucosidase inhibition; docking and kinetics included
Nkoana et al. (2024) [68]	Various heterocycle-acyl hybrids (e.g., benzothiazine, chalcone hybrids bearing lipophilic chains)	In-vitro α-glucosidase & α-amylase (some studies also report cell-based glucose uptake assays)	Reported IC_{50}s vary widely — sub-micromolar → hundreds of μM depending on scaffold; many best analogs in the single-digit to low-tens μM range	Multiple series included acyl/fatty-like residues to improve potency and membrane interactions; compared with acarbose or standard inhibitors
Kaya et al. (2024) [69]	Aromatic carbohydrazide / isobenzofuranone / imidazole-based hybrids with acyl groups	In-vitro α-glucosidase & α-amylase; some in-vivo hypoglycemic screening reported for lead(s)	Examples: many IC_{50}s reported across the series; e.g., representative analog 8i IC_{50} ≈ 153.5 μM in that paper for one end of the series	Most active analogs were advanced to kinetic/docking work; a few tested in rodent models (where reported) showed oral antihyperglycemic effects at mg/kg doses
Singh et al. (2024) [70]	Triazole-tethered thymol/carvacrol-coumarin hybrids (natural-product + heterocycle + lipophilic terpenyl/fatty residues — hybrid design).	In-vitro α-glucosidase and in-vivo antihyperglycemic test (rodent)	Lead AS14 IC_{50} = 4.32 ± 0.11 μM (α-glucosidase); in-vivo led to significant antihyperglycemic activity in mouse model	Hybridization of lipophilic (terpenyl) and heterocyclic/coumarin moieties produced selective α-glucosidase inhibitors with in-vivo efficacy

increased level of the major GLUT receptor GLUT4 in adipose tissues when compared to untreated controls. In vitro experiments suggest this mechanism is due to the regulation of GLUT-dependent signaling pathways, which promote the entrance of glucose into the cells by translocation and exocytosis in the cell surface. However, it has not been proven if the process is induced by insulin signaling pathways.

The activation of adiponectin in tissues can further prevent and reduce the incidence of obesity-related diseases. Adiponectin is an adipocyte-secreted protein hormone that enhances insulin sensitivity by acting upon muscle, liver, and adipose tissues, where its receptors are expressed [56]. Also, this protein regulates glucose, lipid metabolism, and

inflammation in mice. Adiponectin improves insulin sensitivity, reducing metabolic and atherosclerotic syndromes [58]. However, it is also necessary to assess the molecular mechanism involved in the stimulation of adiponectin secretions by these hybrids.

The presence of both polar and non-polar regions in this hybrid structure, or, in other words, the amphiphilic properties, may make them easily inserted into lipid membranes and could affect intracellular signaling events that regulate glucose metabolism. The coexistence of a fat molecule linked to a cyclic system might determine the way the molecule can be introduced inside the membrane and how the signaling pathways are subsequently affected [59]. The activity of these hybrids in influencing glucose control can have a more pronounced effect than in the isolated chemical entities. However, the biophysical role of this structural property in their function at the cellular level still needs to be validated through detailed characterization in different biological settings.

Numerous studies also indicated that several algae species exert anti-inflammatory activity, which can have synergistic effects on glucose homeostasis [58]. Inflammation is the primary source that affects the pancreatic β-cell, the one responsible for insulin secretion, in patients suffering from type 2 diabetes [60]. Oxidative stress can reduce the synthesis of insulin by the pancreatic β-cell. Thus, oxidative stress and pro-inflammatory cytokines, together with advanced glycation end-products and other elements associated with cellular metabolism, are all contributors to β-cell dysfunction and the progression of diabetes [61]. In in vitro studies, these hybrids have been proven to act at low doses on TNF-α and IL-6 production. Furthermore, other studies show the upregulation of key antioxidant enzymes, such as superoxide dismutase and glutathione peroxidase, after hybrid treatment [60]. It is thus possible that hybrids exert antioxidant benefits through intracellular signaling, which, in turn, may attenuate the inflammation caused by these events. In the above-mentioned study, levels of TNF-α and IL-6 were significantly decreased by the algal hybrids in vivo, as was malondialdehyde content, an index of lipid peroxidation; antioxidant markers were augmented [62]. Therefore, future studies are required in order to establish more concrete evidence that might help prove this hypothesis.

Due to the fact that the pancreatic β-cell plays the central role in the production of insulin, the ability of these hybrids to reduce the number of cytokines present might also allow them to protect themselves from death due to their cytotoxicity [58]. Since it has been identified that hybrids could protect the pancreatic β-cells, the next step is to confirm this activity at the level of cells, with a more specific study targeting intracellular molecules [57]. Some animal studies have found a protective effect in β-cells where they are still capable of insulin production, and pancreatic islet integrity, thus preventing diabetes progression.

Fatty acid-heterocyclic hybrids might be even more effective than other potential therapeutic compounds, such as polyphenols, since the former seem to be beneficial at multiple levels (see Table 1). These hybrids could serve as insulin-sensitizing agents that function by stabilizing cellular membranes, enzyme inhibitors, and immune modulators, providing multi-target actions that can be useful in treating the multifactorial conditions of diabetes [63]. The primary challenge consists of isolating hybrids by performing bioassay-guided fractionation. The ultimate objective remains the identification of each bioactive compound, not only to establish structure-activity relationships and perform

biological studies for testing specific mechanisms, but also to understand each contribution to the overall activities (see Table 1).

In conclusion, algae-derived hybrids show promising results against various complications associated with insulin resistance. Their anti-inflammatory and anti-oxidant activities could further contribute to their therapeutic use in diabetes and obesity by protecting the pancreatic β-cell from destruction. Further evidence is necessary to prove these possibilities.

4 Molecular mechanisms of action for diabetes

Comprehending how fatty acid-heterocyclic hybrids exert influence on critical molecular pathways is essential for uncovering their therapeutic potential for diabetes. Specifically, this section focuses on the molecular mechanisms involved in insulin signaling, pancreatic beta-cell function, inflammation, and gut microbiota modulation. The mechanisms unveiled provide crucial understanding to translate their experimental efficacy to promising clinical interventions in the development of new strategies for diabetes treatment.

4.1 Modulation of insulin signaling pathways

Fatty acid-heterocyclic hybrids from green algae modulate the insulin signaling pathways and therefore seem interesting compounds to fight the complex nature of type 2 diabetes by improving glucose homeostasis. They improve the phosphorylation and activation of the insulin receptor and substrates. It is suggested that these compounds improve this pathway via adiponectin by increasing its serum levels. Adiponectin is a potent insulin-sensitizing hormone, improving the response of target tissues to this hormone (Fig. 4). The exact mechanism of action by which the fatty acid-heterocyclic hybrids are able to induce the expression of this hormone is still to be clarified. It would be necessary to conduct further studies to understand whether the expression of this

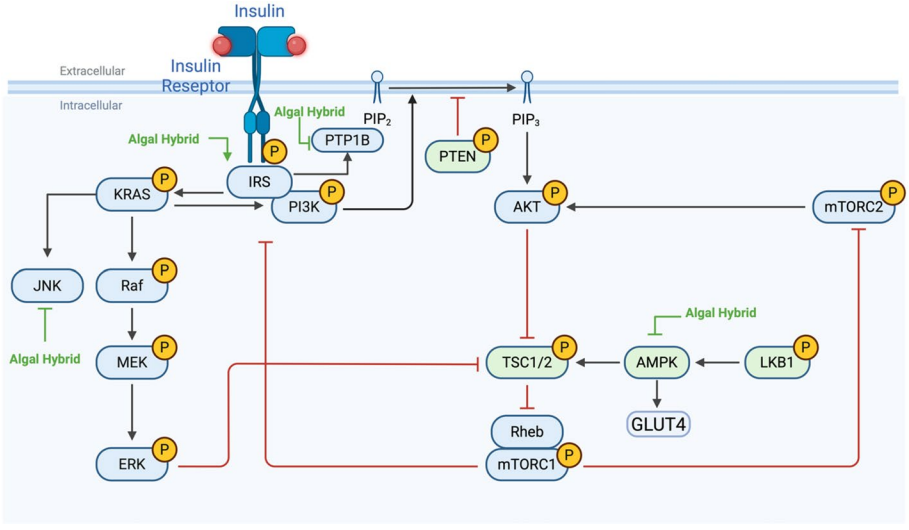


Fig. 4 Insulin Signaling Modulation by Algal Hybrids. A mechanistic diagram showing how fatty acid-heterocyclic hybrids modulate insulin signaling pathways. Key interactions include enhancement of insulin receptor substrate (IRS) phosphorylation, activation of PI3K/Akt, suppression of PTP1B and inflammatory kinases (e.g., JNK), and AMPK activation leading to GLUT4 translocation. Created using BioRender

hormone is improved by direct interaction of the algal compounds with the adiponectin receptors or by indirect actions of their metabolites.

Fatty acid-heterocyclic hybrids from green algae exert their hypoglycemic effects through a sophisticated, multi-target approach that addresses the core pathologies of diabetes. At the cellular level, these compounds actively modulate insulin signaling pathways by enhancing the phosphorylation of the insulin receptor and its substrates (IRS), thereby propagating the signal downstream [23]. This activation promotes the recruitment of PI3K, which in turn activates Akt. The PI3K/Akt pathway is critical for driving the translocation of GLUT4 to the cell membrane, facilitating glucose uptake in insulin-responsive tissues such as muscle and fat. In parallel, these hybrids activate AMPK, a key energy sensor that stimulates glucose uptake and metabolism independent of insulin signaling. The compounds also counter insulin resistance by inhibiting negative regulators of these pathways, such as PTP1B, which dephosphorylates and inactivates the insulin receptor. Furthermore, they suppress inflammatory kinases like c-Jun N-terminal kinase (JNK), which typically impair insulin signaling by phosphorylating IRS on serine residues. This comprehensive modulation of multiple signaling cascades, along with the suppression of pro-inflammatory cytokines such as TNF-α and IL-6, collectively restores cellular glucose homeostasis and improves overall glycemic control.

The fact that these compounds improve the levels of adiponectin suggests that they contribute to the improvement of the activation of insulin receptor substrates [71]. This process leads to the propagation of the signal along the insulin pathway, allowing for downstream insulin-dependent effects, such as glucose transport. This involves the phosphorylation of the substrates, which allows for the recruitment and activation of multiple enzymes responsible for transmitting the insulin-induced signal, such as phosphoinositide 3-kinase (PI3K) and Akt. Further studies should be performed to analyze whether the increase in substrate phosphorylation occurs due to their direct binding to the algal compounds or due to the improved activation of the insulin receptor.

The presence of both fatty acids and heterocyclic parts in the structure of algal hybrids endows them with amphiphilic properties. This might contribute to their association with membranes, allowing the compound to get closer to membrane-bound receptors, improving the interactions with this system. Consequently, this might improve the receptor binding and autophosphorylation. By bringing the compounds close to the insulin receptor, they may be able to enhance the receptor's binding to insulin, improving autophosphorylation and the propagation of the signal through the pathway [72]. To understand whether the presence of this structural amphiphilicity really confers advantages regarding the receptor engagement of fatty acid-heterocyclic hybrids, further studies regarding the synthesis and effect of the precursors used in their formation must be performed.

These compounds appear to be able to counteract the insulin signaling defects characteristic of type 2 diabetes by multi-target modulation of different key enzymes of insulin-mediated glucose transport. As stated, they increase the levels of IRS phosphorylation, but, additionally, they can activate other key players, such as the PI3K/Akt and the AMPK, which are important for the process of glucose transport into the cell [73]. The activation of this kinase promotes glucose uptake by improving GLUT4 expression and translocation to the cell membrane, thus being an important event in the process of insulin-mediated glucose transport [74]. Since this kinase improves the process

by directly enhancing GLUT4 translocation independently of insulin, this seems to be a significant event that needs further investigation. In future studies, the participation of each pathway in the improved insulin signaling should be further determined, and whether their contribution is also observed in the skeletal muscle or is limited to other organs.

There are also studies that reveal the ability of fatty acid-heterocyclic hybrids to inhibit the activity of enzymes that can downregulate the insulin signaling cascade, such as the protein tyrosine phosphatase 1B (PTP1B) [75]. This enzyme dephosphorylates the insulin receptor (IR), thereby abolishing its activation, leading to the end of the insulin signaling pathway. By suppressing the activity of this enzyme, the hybrid can amplify the duration of IR activity and enhance the phosphorylation of substrates. PTP1B plays a critical role in regulating insulin sensitivity since, often, it is seen as highly activated in insulin resistance and diabetes, and it is an important cause of the attenuation of the effects of insulin. Understanding the structural aspects of fatty acid-heterocyclic hybrids that are important for their activity to counteract PTP1B activity can improve their therapeutic potential and increase selectivity for their targets.

Due to the importance of inflammatory processes in the development of insulin resistance, several studies address the anti-inflammatory properties of algal compounds. It is known that, during chronic inflammation, cells activate stress kinases, such as c-Jun N-terminal kinase (JNK), that are able to impair insulin signaling by phosphorylating IRS on serine residues, an event that abolishes their effect as insulin effectors [76]. Fatty acid-heterocyclic hybrids have been shown to reverse the inhibitory phosphorylation of IRS-1 by suppressing inflammatory pathways and, in turn, JNK signaling. By abolishing this negative phosphorylation event, these compounds contribute to the regeneration of insulin signaling cascade integrity.

Inflammation also abolishes the proper activity of the insulin signaling pathway by promoting the activation of some cytokines, which can hinder the pathway's activity. Fatty acid-heterocyclic hybrids were shown to reduce the expression and activity of some pro-inflammatory cytokines, such as tumor necrosis factor-alpha (TNF-α), contributing to improved activating phosphorylation profiles within the insulin pathway [77]. Moreover, by modulating inflammatory pathways, the fatty acid-heterocyclic hybrids can improve other metabolic alterations associated with type 2 diabetes. However, these studies need more characterization by identifying the exact intracellular targets that are able to counteract inflammatory responses and improve insulin signaling activity.

The overall results found in these studies are in favor of the antidiabetic properties of algal compounds. By the analysis of blood glucose and other parameters from experimental models, a marked improvement in glucose control and sustained insulin action is evident in samples that are treated with fatty acid-heterocyclic hybrids [25, 78]. Moreover, biochemical assays performed on different intermediates of insulin signaling suggest an improvement of the activity in the presence of these hybrids, promoting an increased phosphorylation of the insulin receptor and its substrates. These preliminary results of preclinical models suggest that algae contain novel fatty acid-heterocyclic hybrids with insulin signaling regulation and glucose metabolism that may be applied to type 2 diabetes. While promising results from animal and cell models are seen with the use of fatty acid-heterocyclic hybrids derived from green algae, to address the multifactorial complications associated with type 2 diabetes, human studies are also critical.

Unfortunately, often this process is much more difficult since human models require extensive studies in order to allow proper data collection and interpretation.

The effectiveness of new therapies against type 2 diabetes depends on multiple variables. While animal models are a relevant means of study, they still lack some key information that must be obtained from humans, such as potential side effects, drug bioavailability, and efficacy, among other relevant pharmacokinetic features [79]. This often reflects a certain complexity in designing and interpreting human studies since we are not genetically identical and are often treated with different drugs. Additionally, the heterogeneity observed within a population with type 2 diabetes must be considered. Multiple comorbidities, such as hypertension, hypercholesterolemia, and obesity, greatly affect the responses of patients during a clinical trial, resulting in variations in glucose levels, metabolic parameters, or even the extent and duration of action of algal hybrids [80]. In this case, a great sample size must be studied in order to gain sufficient insights into the impact of algal hybrids in type 2 diabetes patients.

The most important issue that must be solved for these novel bioactive compounds is the poor data regarding their pharmacokinetic fate. No standardized protocol for absorption, distribution, metabolism, and excretion assays in humans is found for algal compounds. It is critical to perform studies to understand how these fatty acid-heterocyclic hybrids interact with the complex physiology of the human gastrointestinal tract. It also needs to be further determined which organs are responsible for its metabolism. The bioavailability and activity must also be determined for each metabolite in humans to see how this impacts its therapeutic potential. Delivery systems designed for increased intestinal bioavailability or that target specific organs or cells responsible for insulin production/utilization may be necessary to fully understand their potential benefits in the management of type 2 diabetes.

Another important study regarding fatty acid-heterocyclic hybrids needs to address the structure and activity relationships. There is a report concerning novel fatty acids that have been shown to improve insulin sensitivity in animals and humans. These fatty acids also present a wide structural variability depending on chain length and the number and positioning of saturation [81]. This is also the case with algal fatty acid-heterocyclic hybrids. Different algae present diverse fatty acid chains and heterocyclic structures, each of which may have different activities that regulate different enzymes or transporters responsible for improving the insulin signaling pathway.

Future research would benefit by evaluating fatty acid-heterocyclic hybrids, varying only their fatty acid lengths, saturation points, or heterocyclic substitutions, to understand which characteristics account for certain effects. This would allow us to better define the mechanism behind the observed benefits of insulin action or any undesirable effects. The structure-activity relationship studies of the algae-derived fatty acid-heterocyclic hybrids should include high-resolution mass spectrometry, and also 2D-nuclear magnetic resonance, as well as in silico molecular docking experiments. These studies would improve the understanding of the mechanism of action for these algal compounds as well as the identification of the most efficient structures for use in preclinical trials and the subsequent potential clinical use. In addition, the knowledge obtained from the structural and activity characterization would allow further improvement and optimization by performing computer-based analyses.

In summary, these algae-derived fatty acid-heterocyclic hybrids improve several of the defects characteristic of the complex nature of type 2 diabetes. To obtain more specific and detailed information about the effects of these novel molecules, studies need to be carried out regarding the following:

- Structure-activity relationship assessment;
- Bioavailability and bioavailability studies in humans;
- Standardized absorption, distribution, metabolism, and excretion (ADME) protocols.

4.2 Regulation of pancreatic beta-cell function

Fatty acid-heterocyclic hybrids have shown significant potential in regulating pancreatic beta-cell function and preventing beta-cell damage for diabetic care. By reducing oxidative stress and inflammatory cytokines, these hybrids may be able to restore beta-cell function. In vivo and ex vivo tests show improved islet functionality after application (Fig. 5). This alteration is thought to play a significant role in the prevention of beta-cell dysfunction.

It has been revealed that green algae-derived hybrids have significant anti-inflammatory properties by lowering the levels of inflammatory cytokines such as TNF-α and IL-6. Moreover, it was discovered that algae may also promote anti-inflammatory cytokine responses in vivo [82]. In the research by [83], hybrids displayed an increase in

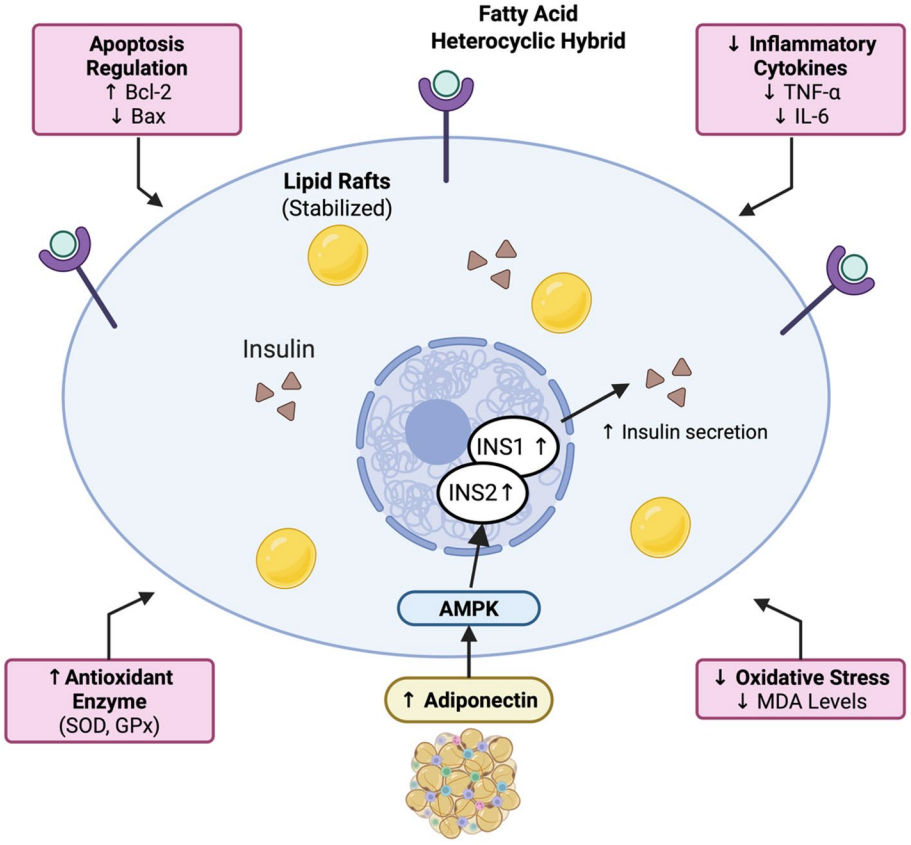


Fig. 5 Impact of Fatty Acid-Heterocyclic Hybrids on Pancreatic Beta-Cell Function. Illustration of protective effects of algal hybrids on pancreatic beta-cells. Mechanisms include antioxidative enzyme upregulation, reduction of TNF-α and IL-6, stabilization of lipid rafts, inhibition of apoptosis (via Bcl-2/Bax modulation), and stimulation of insulin gene expression (INS1, INS2) through AMPK activation. Created using BioRender

the production of anti-inflammatory mediators that are vital for maintaining beta-cell viability.

Fatty acid-heterocyclic hybrids have also exhibited significant antioxidant effects to upregulate hepatic antioxidant enzyme activity and reduce oxidative stress [84]. In addition, it was revealed that laminarin and fucoidan may not only reduce liver oxidative stress but may also effectively prevent the exacerbation of oxidative damage in pancreatic tissue as a result of their positive impacts in other organs [85]. This may occur because a reduction in liver oxidative stress results in lower systemic levels of oxidative stress. This finding is consistent with the importance of lowering beta-cell oxidative stress because this will promote protection against beta-cell dysfunction and death.

According to research, some hybrid fractions are able to effectively decrease fasting blood glucose, restore insulin secretion from pancreatic beta-cells, and enhance insulin sensitivity in diabetic animal models. In addition to their previously demonstrated ability to enhance beta-cell function and protect against beta-cell damage, there is evidence that fucoidan supplementation significantly elevates the levels of adiponectin and other pro-insulin mediators in both animal models and human cells [84].

It has been reported that several algae-derived hybrids could effectively alter beta-cell membrane fatty acid composition and therefore improve beta-cell function. For example, a higher n-3 and n-6 long-chain PUFA ratio has shown potential to effectively preserve beta-cell function and enhance insulin sensitivity [86]. In individuals who are potentially at risk of latent autoimmune diabetes or type 2 diabetes who exhibit autoimmune characteristics, it is possible that supplementation with n-3 PUFAs derived from green algae has the potential to act as a unique and tailored therapeutic modality due to its specific effects on the cell [87, 88].

In order for the cellular signaling pathways to be activated, signaling protein receptors and the insulin granule release machinery are required. However, the lipid raft is important to support these signal transducers and, therefore, initiate signal transmission. Once the algae-derived hybrid inserts into the beta-cell's phospholipid bilayer, the phospholipid structure within the cell membrane would likely undergo changes in its biophysical state and composition [86]. These adaptations can influence the activity and regulation of insulin granule docking proteins and signaling receptors in the cell. Furthermore, the change in phospholipid composition influences the function of cellular membrane signaling and its transduction. This suggests that incorporating algae-derived hybrids into beta-cell phospholipid bilayers enhances beta-cell function and survival. Nonetheless, more detailed studies are needed to investigate the biophysical mechanism of algae-derived hybrids that directly affect cellular membranes.

The activation of energy sensors, such as AMPK, may result in the metabolic and insulin-stimulating effects of fatty acid-heterocyclic hybrids. Additionally, green algae were able to activate AMPK to elicit elevated gene transcription of INS1 and INS2 to elicit beta-cell protective effects in an in vitro setting and in a mouse model [89]. The activation of the aforementioned energy sensor resulted in a significantly elevated expression of the insulin gene and, therefore, was demonstrated as an important protective factor for beta-cell survival and function. This may potentially prove to be extremely beneficial, as the function of most of these processes is suppressed in response to hyperglycemia.

In recent years, it has been revealed that the secretion of adiponectin not only improves peripheral insulin sensitivity, but also that it may directly support beta-cell

health [90]. Adiponectin promotes activation of prosurvival kinases in beta-cells and protects islet cells from apoptosis. As such, further research into the relationship between algae-derived hybrids and adiponectin is important to promote positive health impacts for beta-cell function. Additionally, recent research shows that hybrids can prevent beta-cell apoptosis by lowering pro-apoptotic proteins such as Bax and activating anti-apoptotic markers such as Bcl-2. The enhancement of cell viability and lowering of apoptotic signal activation can result in greater islet cell mass and overall improved insulin secretion, which is therefore important for diabetic individuals, as both forms of diabetes can ultimately contribute to diminished insulin-producing capacity.

Overall, algae extracts comprise diverse biochemical constituents. However, whether their effect results from a synergistic action of individual hybrids or the presence of just one predominant active hybrid is still unknown. To identify certain molecules as effective hybrids requires more effort. These effects and biochemical analyses of purified and crude extracts for biological activities can then be utilized to elucidate the precise structures. Identification and characterization would greatly aid in establishing definitive structure-activity relationships. The elucidation of this relationship will assist in gaining approvals for these promising hybrids as nutraceuticals or even medicines.

Moreover, the crude extract analysis and biological and biochemical analysis-driven bioactivity fractionation assay will allow us to isolate the highly bioactive hybrids and determine their structure. Such a bioactivity fractionation assay is highly recommended for evaluating crude extracts with multiple compounds. This information would also allow for direct comparison of the in vivo effects of crude extracts with bioactive hybrids. Furthermore, with the advancement of purification techniques, hybrid-to-hybrid differences could be fully distinguished to explore whether these subtle changes have differential effects on islet cells. By utilizing this combination of extraction and bioactivity-driven purification methods, a broader spectrum of bioactive molecules can be identified, and a structure-activity relationship can be characterized, which facilitates regulatory authorization of new drug sources, nutraceuticals, and medicines for beta-cell regeneration.

In this area of research, recent advancements in molecular and analytical technologies have led to improvements. Although hybrid analyses are possible, they still create a hurdle in the elucidation and development of effective hybrids due to the fact that most of the algal extracts have shown various hybridized structures [84]. Moreover, due to the lack of individual hybrid information, it has proven very difficult to measure algal hybrid effects and apply them as medicinal treatments. However, through advanced methodologies, like high-throughput cell assay screening paired with the LC-MS and NMR database mapping system, these algal species, along with many other species in the kingdom of algae, will become easier to isolate in order to allow for exploration and future biomedical advancements.

In conclusion, these fatty acid-heterocyclic hybrids were selected because they appear to have great health and disease-modifying benefits, which is also supported by experimental evidence demonstrating that various compounds from this class have antioxidant and anti-inflammatory activities, stimulate insulin production, and inhibit pancreatic cell death.

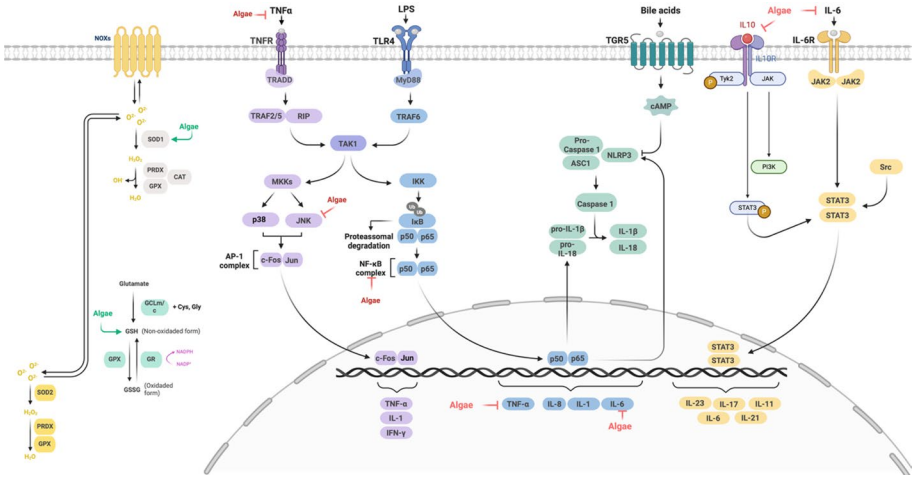


Fig. 6 Anti-Inflammatory and Antioxidant Roles of Algae-Derived Hybrids in Diabetes. This figure presents dual roles of fatty acid-heterocyclic hybrids in suppressing diabetes-related inflammation and oxidative stress. The compounds downregulate pro-inflammatory cytokines (TNF-α, IL-6), upregulate IL-10 and antioxidant enzymes (SOD, GSH-Px), and reduce lipid peroxidation, thereby protecting tissues and improving insulin sensitivity. Created using BioRender

4.3 Anti-inflammatory and antioxidant effects

Fatty acid-heterocyclic hybrids from green algae have been shown to have great anti-inflammatory capacity in diabetic models by regulating the production of pro-inflammatory cytokines such as tumor necrosis factor-alpha (TNF-α) and interleukin-6 (IL-6) (Fig. 6). Both of them enhance insulin resistance and beta-cell impairment, thus decreasing the production of inflammatory cytokines, reducing chronic inflammation, and promoting beta-cell protection and glucose utilization. On the other hand, more research is needed to understand the pathways involved in the selective inhibition of inflammatory cytokines like TNF-α and IL-6 (Fig. 5).

The reduction in the production of the pro-inflammatory cytokines TNF-α and IL-6 is an attractive therapeutic strategy because, when their levels are elevated, they enhance inflammatory conditions, leading to insulin resistance and, thus, to diabetes progression [91]. Therefore, reducing these inflammatory mediators could contribute to restoring glucose homeostasis in diabetic patients and promoting beta-cell health. Chronic inflammation is directly related to insulin resistance and is associated with the elevated levels of TNF-α and IL-6, which can activate stress kinases such as c-Jun N-terminal kinase (JNK) and in this manner inhibit insulin signaling by the activation of insulin receptor substrates. In this way, this natural compound suppresses inflammatory stress, promoting an increase of insulin sensitivity in peripheral tissues and pancreas beta-cells. Conventional anti-inflammatory drugs have also the capability to inhibit inflammation, however, unlike algae-derived hybrids, they are not directly associated with the beginning of inflammatory stress. Conventional drugs modulate the production of pro-inflammatory cytokines by the activation of pathways such as nuclear factor kappa B (NF-κB) and JNK, therefore, they act later on the inflammatory cascade, while green algae hybrids, in turn, act in the first steps, targeting the inflammatory response and therefore preventing diabetes progression. Moreover, the ability of these hybrids to regulate cellular inflammation also decreases lipid peroxidation levels. The overall anti-inflammatory and antioxidant activity provides benefits in metabolic regulation and protection of target tissues [92]. Nonetheless, more research is needed in order to prove the

hypothesis that inflammation reduction and inflammatory-related gene suppression is a novel strategy for effective glycemic management in the clinical treatment of diabetes.

Besides the downregulation of the pro-inflammatory mediators TNF-α and IL-6, the fatty acid-heterocyclic hybrids also enhanced the production of the anti-inflammatory cytokine interleukin-10 (IL-10), which is necessary to stimulate anti-inflammatory pathways as well as to promote tissue repair [93]. This mechanism is very relevant to re-establish homeostasis and restore immunity response and regulate systemic inflammation in chronic inflammatory diseases, such as diabetes. Furthermore, the mechanisms by which the levels of IL-10 are enhanced by the algal hybrids were not explored, and therefore need further investigation in order to identify those specific compounds responsible for the upregulation of this cytokine. These effects are not totally explored, and therefore, more research is needed, analyzing, for example, the correlation between structural changes among different hybrids, with their bioactivity in relation to specific cytokines.

Finally, the fatty acid-heterocyclic hybrids, derived from green algae, displayed potent antioxidant activity that is associated with their anti-inflammatory capacity. These hybrids enhance the activity of several hepatic antioxidant enzymes such as superoxide dismutase and glutathione peroxidase, thus decreasing the oxidative damage of beta-cells and peripheral tissues, improving glycemic control as well as cytoprotection. These effects seem relevant in diabetes management because the reduction of oxidative stress can lead to better glycemic control. Oxidative stress plays a critical role in the development of diabetes-related complications [94, 95]. Thus, these algal-derived compounds show a promising potential against these events by enhancing endogenous antioxidant defense mechanisms. Further studies are needed to identify which of these algae-derived hybrids are more effective antioxidants, thus preventing oxidative damage in vivo.

As oxidative stress is an important contributor in the development of diabetes, the prevention or reversal of ROS-mediated damage, by scavenging free radicals and by enhancing the gene expression of antioxidant enzymes, is are major therapeutic approach. In fact, the hybrid, described by [39], proved that lipid peroxidation was reduced by decreasing malondialdehyde levels in pancreas tissue as well as by restoring plasma membrane integrity in liver cells from diabetic rats. It is also proven that green algae compounds were capable of suppressing the increase in lipid peroxidation in plasma and liver cells caused by diabetes induction. These promising results indicate that algal compounds have a dual scavenging action of ROS that may provide enhanced antioxidant protection compared with single antioxidants. However, more in vitro and in vivo research needs to be performed in order to understand how the combination of different fatty acid chains and heterocyclic scaffold in the hybrid molecules affects antioxidant mechanisms, contributing to additive or synergistic actions of ROS scavenging and suppression of oxidative damage by means of upregulating antioxidant genes.

In summary, algae-derived fatty acid-heterocyclic hybrids may offer superior benefits and mechanisms of action for antioxidant prevention and treatment of diabetic diseases compared with conventional polyphenols or other synthetic antioxidants that target ROS. While most of the available antioxidants only scavenge free radicals, algae-derived compounds modulate enzymatic as well as non-enzymatic oxidative stress, also affecting metabolic signaling, like inflammation. Their amphiphilic nature allows them to localize inside cells and associate with enzymatic and non-enzymatic cellular membranes related

to ROS production and signaling. These characteristics make algae-derived antioxidants able to intervene in metabolic and inflammatory pathways, providing protection against oxidative damage, insulin resistance, and impaired glucose transport. Moreover, the capability of algal-derived hybrids to suppress pro-inflammatory transcription factors such as NF-κB, which regulates the expression of genes involved in inflammation, is another advantage of these natural compounds as antioxidant drug candidates. While conventional antioxidants can only target specific pathways of ROS, these algal-derived hybrids also target ROS production, which may provide broader metabolic control and anti-inflammatory action by reducing stress signaling. Overall, these benefits offer the rationale to use algae-derived antioxidant hybrids in diabetes prevention or as coadjuvants with conventional antidiabetic therapies for improved therapeutic efficacy and to decrease the dosage or adverse effects in long-term medical treatment.

Animal experiments have also shown the impact of algal-derived fatty acid-heterocyclic hybrids in oxidative stress and blood glucose and/or lipid parameters reduction. For example, algae-derived hybrids decreased fasting blood glucose, inflammatory markers, and plasma triglycerides in diabetic rat models. More recently, it was also confirmed that chronic treatment with this hybrid improved metabolic parameters, as the elevated levels of fasting blood glucose and plasma triglycerides were reversed in the diabetic animal model. The levels of oxidative stress in rat models were reduced, as it was observed decreased levels of two important oxidative stress markers in plasma. Plasma 8-isoprostane and protein carbonyl content (PCC), which are the major oxidized end products of cellular lipid peroxidation and protein oxidation, were reduced, revealing the physiological beneficial effects of the chronic treatment with this natural compound. Due to the complexity and chronic nature of diabetes, there is an emerging need to evaluate the involvement of energy regulators such as AMPK in the antioxidant action of algae-derived hybrids. AMPK, a sensor of cell energy, promotes glucose uptake, fatty acid oxidation, and reduction of oxidative stress in metabolic tissues and also inhibits inflammation. Interestingly, metformin is an established treatment used for Type 2 diabetic patients, which acts by improving glucose utilization, decreasing triglyceride accumulation, reducing lipid peroxidation, and enhancing cellular antioxidant defenses [96]. This agent promotes the activation of AMPK-mediated processes that decrease oxidative stress through the enhancement of antioxidant defense mechanisms. Further research is necessary in order to clarify how the AMPK activation mechanism relates to the cellular processes activated by the intake of green algae-derived hybrids. In addition, the identification of potential synergistic effects between algae-derived hybrids and conventional antidiabetic treatments would also pave the way for more efficient and lower-dose regimens.

Besides the promising results mentioned before, the development of these compounds is made difficult due to the complex mixture of many different algal molecules contained in extracts that possess antioxidant and anti-inflammatory action. With this challenge in mind, bioassay-guided fractionation has proven to be a powerful tool for the identification of structure-activity relationships between chemical structures and antioxidant or anti-inflammatory activity [97]. It also allows for the determination of a single active molecule within the algal extracts using a combination of chemical isolation and bioassay evaluation, followed by structural characterization with highly effective analytical technologies. The use of next-generation technologies such as liquid chromatography

coupled with high-resolution mass spectrometry and nuclear magnetic resonance has brought great success for the determination of algal molecules with antioxidant activity. However, the vast majority of these algae-based studies identify mixtures as a bioactive source rather than the characterization of the individual active compounds. Therefore, to ensure the development and clinical use of algae-derived hybrids, it is necessary to isolate single hybrid molecules to identify those possessing high biological activity. Further studies, designed to explore the antioxidant and anti-inflammatory effects of different green algae compounds containing fatty acid chains and heterocyclic scaffolds, are recommended. These investigations may correlate particularities of the chains with particular effects, thus optimizing algal hybrid structures for anti-inflammatory and antioxidant activity.

The enhancement of biosynthesis of promising anti-inflammatory and antioxidant fatty acid-heterocyclic hybrids by altering algal culture conditions would be a highly attractive feature for clinical use. Techniques like metabolic engineering or stress induction could potentially enhance the production of the most promising hybrids. These approaches remain an under-explored avenue of study. Thus, more research needs to be performed in order to find ways to alter cultivation conditions to improve or induce fatty acid-heterocyclic hybrid biosynthesis of those with higher biological activity.

In sum, there is significant evidence supporting the effectiveness of the anti-inflammatory and antioxidant activity of green algae-derived fatty acid-heterocyclic hybrids, thus making it an interesting approach in diabetes prevention. The ability to regulate the production of pro-inflammatory cytokines, to up-regulate IL-10, and to enhance the activity of hepatic antioxidant enzymes is one of the reasons why these natural compounds hold promise for the treatment and prevention of diabetic events. Nevertheless, more research is needed regarding the exact characterization of bioactive single molecules, the relationship between the structural features of algae-derived compounds and their biological action, the exploration of algal metabolism, and the potential of algal hybrids as drug candidates to target mechanisms involved in chronic inflammation.

4.4 Modulation of gut microbiota

Fatty acid-heterocyclic hybrids obtained from green algae have shown the ability to modulate the gut microbiota, impacting several factors related to metabolic health and the development of diabetes. This effect enhances the abundance of beneficial bacterial taxa, particularly Bifidobacterium and Lactobacillus [39]. Their effect on the gut microbiota is related to their prebiotic function and their bioactive properties. The selective stimulation of beneficial gut microbial taxa has been postulated as being dependent on the structural differences within these hybrids, even though the mechanisms by which they induce this stimulation are unclear.

Ulvan and sulfated polysaccharides obtained from green algae show the potential to selectively stimulate short-chain fatty acids (SCFAs), such as butyrate and propionate [56]. Both SCFAs are fundamental in maintaining the integrity of the intestinal barrier and the sensitivity of insulin, contributing to the reduction of fasting glucose levels in vivo, emphasizing the effects on the host's metabolism. Ulvan's prebiotic function suggests that, together with the gut microbiota, these algae hybrids may control metabolic outcomes in antidiabetic therapies [98]. However, further studies are needed to determine whether these effects are observed similarly across various human and animal

models, where variables, such as different diets, microbiota compositions, or genetic factors, play a role.

Experimental evidence has demonstrated that fatty acid-heterocyclic hybrids may diminish harmful bacterial groups that often cause endotoxemia, and that lead to pro-inflammatory cascades and further inducing insulin resistance. Certain species of Clostridium and Enterobacteriaceae are frequently targeted in microbial therapies for such complications. Even though the effect of the hybrids on microbial taxa is evident in the current data, the molecular pathways, or what exactly determines their specific targeting, are not. It is also unknown if the same hybrid will exert the same influence over time, since the structure and interactions of gut ecosystems are dynamically regulated.

Based on this evidence, one hypothesis states that fatty acid-heterocyclic hybrids could be specially designed to stimulate the growth of particular bacteria to promote metabolic effects. Each hybrid may have specificity for a distinct bacterial type based on its molecular differences: fatty acid chains, saturation, and nature of the heterocyclic ring [99]. The ability to induce positive health outcomes will depend on the selective interaction between the hybrid structure and a specific microbial population. The modulation of gut microbiota is a promising strategy, and tailored approaches through diets, drug combinations, or novel pharmaceuticals may become a reality. Further research in structure-activity relationships is necessary to evaluate the selective interaction between hybrids and gut microbial taxa.

Certain green algae metabolites, such as ulvan extracted from Ulva lactuca, increase the number of SCFA-producing bacterial taxa, such as Faecalibacterium prausnitzii and Roseburia spp [56]. They are inversely associated with markers of insulin resistance and systemic inflammation, further indicating the capacity of algae-derived hybrids in improving metabolism by directly affecting the gut. This leads to higher fecal concentrations of SCFAs that improve insulin sensitivity and glucose control. However, current studies in this area provide broad taxonomies and limited specificity down to species and strains that are influenced by the compounds' structures. Thus, information on the differential responses of specific microorganisms to supplementation with fatty acid-heterocyclic hybrids is lacking.

Algae hybrids may also impact host metabolism through their modulation of the gut microbiota through the effect of SCFAs on G-protein coupled receptors (GPR41 and GPR43) that control the metabolism through insulin secretion, energy expenditure, and adipogenesis [100, 101]. The gut is a rich signaling hub controlled by numerous ligands and receptors with far-reaching hormonal effects. Algae hybrids exert a dual effect on microbial composition and the receptor-ligand axis. However, more work will be needed to understand which outcomes are due to the hybrids or their gut microbial metabolites, thus impacting host metabolism.

Algae-derived hybrids have demonstrated benefits for the improvement of the metabolic profile in experimental animals, particularly by affecting the lipid and bile acid pathways [11]. Since bile acids influence both lipid and glucose levels in the body, these changes in the microbiota may improve both of the metabolic pathways associated with T2D. More investigation is needed to understand how specific hybrids can be used alone or in combination with probiotics and prebiotics. In future work, algae-derived hybrids may also be used in combination with other microbial metabolites that mimic

endogenous compounds, since these novel combinations can promote synergistic effects and optimize metabolic health.

Some green algae-derived hybrids improve glucose homeostasis by promoting the growth of gut microbial species that strengthen the intestinal mucosal barrier, leading to reduced gut permeability. It appears that Akkermansia muciniphila, a bacterium associated with the intestinal lining and gut health [39], could be stimulated by these algae hybrids. The reduction of gut permeability induced by the increase of A. muciniphila may also decrease the leakage of lipopolysaccharides (LPS) across the gut, a main stimulus of inflammation that often leads to chronic disorders [102, 103]. Improvements in glucose control also contribute to lower levels of systemic inflammatory cytokines and oxidative stress markers, highlighting that gut permeability may also be altered by other bacterial species, and that other signaling pathways might contribute to the overall anti-inflammatory effects induced by the hybrids. Although these observations indicate a decrease in endotoxemia that reduces pro-inflammatory cytokines, further studies will be needed to directly assess the mechanism by which algae-derived hybrids reduce pro-inflammatory bacterial taxa to improve gut barrier integrity and its permeability.

The observed reductions in inflammatory and oxidative stress induced by the algae hybrid treatment seem to occur as a result of improvements in gut microbial taxa. However, this may depend on the specific properties of the algal metabolites, and may indicate that they directly influence anti-inflammatory and antioxidant activities. A thorough analysis using microbiomic, metabolomic, and hormonal tools may identify which particular hybrids and specific microbial products or changes in the host endocrine or immune systems account for the alleviation of systemic inflammation [104], thus guiding future applications of dietary intervention.

Algae hybrids might also influence the enteroendocrine system. The impact of algae hybrids on glucose control could involve the modulation of incretin hormones. These hormones (glucagon-like peptide-1 (GLP-1), glucose-dependent insulinotropic polypeptide (GIP), etc.) are often related to increased insulin secretion and a reduction of the rise in blood glucose postprandially. Some animal models of T2D that have been treated with algae-derived hybrids show enhanced GLP-1 hormone in blood [83]. This can provide an additional explanation by which algal hybrids improve T2D-related complications. Further studies are required to address the specific signals that lead to altered release of the incretin hormones induced by the gut microbes or other changes in the metabolism of the body.

The diversity and tunability of the hybrid structures represent an avenue for selectively targeting the host, and could promote tailored treatments in combination with current drug regimens or as supplements. Systematically, metagenomic and metabolomic data should be merged with data for hormone-responsive cells of the pancreas to understand how specific gut microbial metabolic outputs can lead to improvements in systemic health and hormone responses. If particular compounds induce preferential responses that target certain microbial metabolic pathways, we may be able to predict the most efficacious treatment regimen on a patient-to-patient basis based on the particular structure of a specific hybrid compound.

Unfortunately, the interaction between hybrid structures and gut microbial taxa remains to be investigated. In vitro and in vivo models should be used to explore the ability of a specific chemical structure to affect different gut microbial communities and

their related products. This can be assessed using germ-free and humanized microbiota transplants. High-throughput screening methods may allow for a comprehensive examination of interactions in specific human and animal microbiota. Furthermore, these studies should combine multi-omics (e.g., transcriptomics, proteomics, metabolomics, etc.) to identify molecular signatures of microbiota-mediated antidiabetic effects.

In conclusion, several fatty acid-heterocyclic hybrids derived from green algae show great potential as modulators of the gut microbiota and could, therefore, be explored in metabolic diseases such as T2D. There remain significant challenges to overcoming the design, as well as the mechanisms that impact a single or multiple microbial species. Studies are needed to clarify the structure-activity relationship and to assess the mechanism by which hybrid structures exert differential effects. New studies that merge microbial composition analyses with metabolomics data and host hormone responses are imperative. These multi-dimensional datasets should provide valuable insights into how to achieve an optimized and personalized gut microbiota community that impacts metabolism and is specific to an individual's metabolic profile.

5 Current limitations and challenges

Several challenges and limitations exist that hinder the realization of the therapeutic potential of fatty acid-heterocyclic hybrids from green algae as a future drug for diabetes treatment. For instance, limited research has been conducted regarding their bioavailability and the extent of their absorption. In most in vitro studies, the effects of intestinal absorption, the food matrix, and metabolic degradation of the hybrids are not well considered, influencing their stability, efficacy, and bioavailability [105, 106].

Current studies investigating the antidiabetic properties of these fatty acid-heterocyclic hybrids typically rely on in vitro experiments or short-term animal models, which have limitations in predicting human metabolic responses. Furthermore, there have been no large, multicentric, long-term, or clinically relevant trials conducted, and there is a lack of controlled clinical trials to evaluate their true antidiabetic efficacy. The complex mechanism of human metabolism, involving multiple organ interactions and hormonal feedback, requires more realistic long-term research to fully understand the therapeutic effectiveness of these hybrids [107].

Even though the chemical structures of fatty acid-heterocyclic hybrids make them ideal for cellular entry, their susceptibility to metabolism in the gastrointestinal tract and liver should be investigated. Do the hybrids or their metabolites have bioactivity? What is their metabolic fate in the body? Answering these questions and determining their pharmacodynamic activity is an essential step in assessing the true antidiabetic efficacy of fatty acid-heterocyclic hybrids from algae.

Dietary fibers, proteins, and other bioactive compounds in algae may alter the rate of solubilization and release of these hybrids in the digestive tract [108]. This may hinder or promote their bioactivity. Therefore, more efforts are required to study the influence of the food matrix on these molecules. The absence of a standard analytical method for estimating the absorption, distribution, metabolism, and excretion (ADME) of the hybrids would pose significant challenges in calculating the risk-benefit ratio of using these compounds in the clinical setting. Accurate in vivo pharmacokinetic data are difficult to obtain because of the lack of highly advanced techniques such as stable isotope labeling in mass spectrometry.

The extraction of the desired fatty acid-heterocyclic hybrid from green algae is not always straightforward. The challenge arises in part from the complexity of the extraction processes and a lack of knowledge on how to standardize the methodology. Factors such as algal species, culture methods, and extraction techniques can significantly influence the yields, purity, and bioactivity of the extracts. Therefore, more efforts are required to standardize the extraction procedure for each specific type of algae. Algal metabolism is strongly dependent on extrinsic factors, such as the availability of nutrients, light intensity, temperature, season, pH, and salt concentration [109]. These factors can lead to variability from batch to batch. To avoid batch-to-batch variability, it would be necessary to optimize culture conditions, either by precisely controlling environmental conditions, for example, by utilizing closed-loop bioreactors, or through genetic manipulation. Differences in the extraction solvent used can affect the amount and purity of fatty acid-heterocyclic hybrids that are isolated, directly influencing the biological activity and comparability of studies in different laboratories.

As there are currently no established protocols for isolating pure fatty acid-heterocyclic hybrids from algal species, some pharmacological studies do not accurately reflect the intrinsic antidiabetic activity of these compounds. For instance, some studies report on the effects of algal extracts containing a mixture of other natural metabolites as well [110]. Future research should focus on a bioassay-guided isolation of active algal components. The cost of purification and production by spectroscopic methods, such as chromatographic techniques, at the industrial scale is very high and would limit their large-scale production for treating millions of people. Therefore, bioengineering and bioprocess optimization efforts are greatly required.

The structure of fatty acid-heterocyclic hybrids derived from green algae is complex and difficult to characterize using current analytical methods. As a result, the biological (antidiabetic) activity has not been correlated with certain functional moieties within the structure of these molecules [109]. The development of more advanced and high-throughput spectrometric techniques would facilitate their identification. Furthermore, because fatty acids and heterocyclic moieties are also commonly found in algae, their chromatographic and spectrometric signatures are superimposed, complicating the detection and quantification of real hybrids [109]. This requires further improvement of techniques such as HPLC-MS and GC-MS that can differentiate structurally similar substances and discriminate bioactive molecules from the algal extract in a reproducible manner.

In order to reliably correlate chemical structure with antidiabetic activity, further analyses using bioassay-guided fractionation approaches linked with cutting-edge techniques, such as NMR-based metabolite identification, are required to separate true fatty acid-heterocyclic hybrids from non-hybrid compounds, thereby determining which chemical substructures contribute to a desired biological property. Crude extracts of algae contain several chemical entities that need to be fractionated further and purified extensively to obtain the desired target molecules. The complexity of the algal matrix may lead to the degradation of fatty acid-heterocyclic hybrids or the removal of synergistic co-factors [109].

A crucial gap in the current research is the lack of a robust clinical perspective, which is essential for translating promising preclinical findings into viable therapies. While our review acknowledges the absence of clinical trials, a deeper elaboration on

key translational issues is necessary. This includes addressing the critical need for comprehensive data on toxicity, pharmacokinetics (PK), bioavailability, and the significant regulatory hurdles that any new therapeutic candidate must overcome. For fatty acid-heterocyclic hybrids, their unique structure, combining a lipid tail with a heterocyclic head, presents a complex challenge for PK studies. This requires sophisticated analytical techniques, such as stable isotope labeling, to accurately track their absorption, distribution, metabolism, and excretion (ADME) in the body. Without this fundamental information, determining a safe and effective dosage for humans is impossible, and any serious clinical consideration remains out of reach.

Furthermore, the path to regulatory approval for these compounds is fraught with challenges that must be explicitly discussed. Beyond efficacy, a thorough toxicological profile is non-negotiable. This includes assessing potential genotoxicity, carcinogenicity, and long-term safety, especially since these compounds are being considered for chronic conditions like diabetes. The potential for off-target effects and systemic toxicity must be evaluated, as well as any possible interactions with conventional antidiabetic medications. As these hybrids are derived from natural sources, their interaction with the food matrix and the gut microbiota also needs to be characterized, as these factors can significantly alter their bioavailability and bioactivity. Addressing these issues requires a transition from basic in vitro and animal studies to well-designed, placebo-controlled clinical trials that rigorously test for efficacy, safety, and optimal dosage in diverse human populations.

There is currently a limited amount of in vivo studies to test and confirm that algal extracts or pure fatty acid-heterocyclic hybrids exert their therapeutic benefits through the improvement of insulin sensitivity and/or pancreatic beta-cell survival [111, 112]. Most in vivo experiments fail to account for all complex and interacting elements found in the human body, such as the immune system, the cardiovascular system, different organ systems, and hormonal regulations, since in vitro and in vivo experiments are conducted under extremely simplified conditions. Therefore, experiments on human subjects with multiple, coexisting illnesses and with multi-drug use would be required. As no trials have been conducted to determine the appropriate dose of fatty acid-heterocyclic hybrids to use on human subjects, additional controlled studies in large, multicentric, long-term, or clinically relevant trials are required to validate and clarify their real antidiabetic efficacy, safety, and optimal dosage for therapeutic use in patients with T2D. Due to the lack of research, it is difficult to ascertain how the fatty acid-heterocyclic hybrids affect the immune system. Studies should be performed to evaluate potential immunomodulatory effects.

Even though earlier reports have shown that no apparent cytotoxicity was detected for some cell lines, further studies are required to evaluate the effects of toxicity in vivo on mammals, including teratogenicity and carcinogenicity [109]. Potential off-target effects and systemic toxicity of most algae-derived bioactive metabolites are largely unknown and require further investigation. Genotoxicity, carcinogenicity, and toxicity to reproduction are all important factors in evaluating the potential of using fatty acid-heterocyclic hybrids as a medicine.

Fatty acid-heterocyclic hybrids are comprised of very structurally heterogeneous molecules. It remains to be determined if and how these algal metabolites may be interacting with host metabolic pathways or the gut microbiota when consumed long-term. The

potential accumulation and metabolic transformation of these hybrids in tissue over time and the possible interactions with common antidiabetic agents are also completely uncharacterized. Many different organs and/or tissues are being targeted in the body for treatment using algae-derived fatty acid-heterocyclic hybrids. Therefore, long-term safety studies are imperative to determine the impact of these new drug candidates on non-target tissues.

Even though the algae-derived fatty acid-heterocyclic hybrids look promising as an antidiabetic treatment for T2D, some challenges should be addressed before they can be used therapeutically. Additional research is needed for the proper identification and extraction of effective hybrids.

6 Future directions and research recommendations

The development and standardization of extraction and isolation procedures for fatty acid-heterocyclic hybrids from green algae is critical to overcoming reproducibility and yield barriers. Poor recovery rates and the labile nature of the hybrids require optimized solvent-based extraction strategies in which parameters such as polarity, temperature, and time must be carefully adjusted to minimize decomposition and recover bioactive hybrids with high antidiabetic bioactivity. Purification of highly complex algal matrix can be achieved through HPLC fractionation or multidimensional liquid chromatography to isolate hybrids with high purity and remove structurally similar molecules that can potentially compromise the bioactivity analyses. A combination of these techniques with real-time analytical methods such as MS and NMR may be required to ensure proper structure elucidation and reproducibility of extraction efficiency among different algal species. Development of bioassay-guided isolation protocols is essential to isolate individual hybrids linked to specific pharmacological activities. Finally, bioengineering strategies involving modified culture conditions such as nutrient availability, light exposure, or co-cultivation with microorganisms may elicit the production of a certain number of specific hybrids. Although this approach has the potential to facilitate the scale-up of particular hybrids, there are several important points that will need to be addressed before it can become a widespread strategy.

Further in vivo studies are required to address the lack of comprehensive evidence regarding the antidiabetic effectiveness of these compounds. Preclinical animal models are considered to be important predictors of the effects of a molecule on human physiology and physiology. These are well-established tools for evaluating drug effectiveness, mechanisms of action, and efficacy of pharmaceuticals. In vitro and in vivo animal models may often over-simplify complex physiological conditions, such as diabetes, and may lack hormonal feedback systems and multi-organ interactions [113]. Systematic in vivo testing using diabetic animal models that accurately mimic the multifaceted pathophysiology of the human disease, with varying degrees of insulin resistance, comorbidities, and exposure to chronic medications, is essential for translating in vitro findings to clinical trials [114]. Once hybrids have been found to be effective in preclinical animal models, clinical trials will be needed to determine the optimal dosing regime, treatment duration, and safety. Randomized, placebo-controlled clinical trials that employ adequate endpoints such as reduction of HbA1c levels, improved glucose tolerance, and overall safety and efficacy are required before these algal fatty acid-heterocyclic hybrids can be considered appropriate candidates for clinical use. In addition to determining

optimal dosage strategies, randomized placebo-controlled clinical trials with an adequate duration are also required to determine the long-term efficacy and safety of algal-derived fatty acid-heterocyclic hybrids. These trials will ideally incorporate multiple endpoints in a comprehensive effort to determine the effectiveness of the compounds in a diabetic human population and to determine their role in regulating diabetes complications such as nephropathy, neuropathy, or retinopathy. Mechanistic biomarker studies focusing on DPP-4 and PPAR-γ may also aid in achieving the regulatory approval of this algal-derived novel class of drugs [114].

The lack of definitive data on bioavailability, pharmacokinetics, and metabolism in animal and human systems limits the progress of this promising therapeutic class. The ADME properties of algae-derived fatty acid-heterocyclic hybrids, along with potential biotransformation by the gut microbiota, are critical to determine the dosage, clinical activity, safety, and drug interactions. To identify suitable molecules that are well-absorbed, distributed, metabolized, and excreted, advanced pharmacokinetic evaluations will need to be conducted. For example, labeled hybrids can be administered to animals to examine the organ-specific concentration, absorption, distribution, metabolic clearance rate, and half-life, and to evaluate potential target organs for the drug. In vitro metabolism studies using human liver microsomes can identify the nature of the metabolites that can be potentially active and enhance or reduce the antidiabetic and toxicity potential of the hybrid [115]. Information gained on the absorption and drug metabolism of these hybrid molecules may provide important feedback that can be used in the development of novel drug delivery systems that enhance hybrid bioactivity and drug-targeting potential. For example, encapsulation and conjugation systems of these hybrids can improve their solubility and prevent their degradation in the body.

Further investigation into the mechanism of action will be crucial in bringing algae-derived fatty acid-heterocyclic hybrids to clinical trials. The multi-target action of these hybrids raises considerable potential, but to translate the biological activity into medicinal use, it is important to elucidate the molecular mechanisms and the precise target site. Although crude algal extracts are readily available for mechanistic studies, researchers must use pure algal-derived hybrid components in combination with bioassay-guided fractionation to isolate individual hybrid molecules and to confirm which structures are driving the mechanistic response. Future work should aim to understand how the algal-derived fatty acid-heterocyclic hybrids regulate gut microbiota. This may be addressed using combined metagenomics and metabolomics and may lead to insights into the changes to the microbiota as a result of exposure to these algal molecules. Furthermore, more extensive studies are needed to assess whether algal-derived hybrids modulate the release of incretin hormones like GLP-1 and to assess the cytoprotective role of these algal-derived hybrids on beta cells, since these actions of algal fatty acid-heterocyclic hybrids will aid in improving diabetic conditions as well as protecting beta-cell health. Further advancement toward the development of potent therapeutics may involve the use of emerging in silico tools to aid drug design and discovery and the employment of high-throughput assays to discover hybrid molecules that have the desired mechanism(s) of action. With respect to the former, this may include in silico docking, which is designed to estimate binding energies of a variety of hybrid molecules based on the geometry of the ligand and the structure of a target receptor/enzyme. In

terms of the latter, this may require employing combinatorial bioassay strategies for the de novo discovery of hybrid leads that exhibit the desired biological activities.

Proper toxicological evaluation, as described for any new pharmaceutical candidates, is required to ensure the safety of algal fatty acid-heterocyclic hybrid drugs for the treatment of diabetes. Initial work performed on the literature showed very low cytotoxicity of algal extracts, but more rigorous investigations are necessary. Initial in vitro testing should be performed with primary human cell lines that are diabetic vs. non-diabetic, allowing the examination of selective cytotoxicity or metabolic imbalances [116]. Both acute and chronic in vivo testing in animals is required and will provide critical information regarding hepatic and renal function, histopathology, and immunogenicity in treated animals as well as the potential for bioaccumulation of these molecules. Additional screening for allergenicity and hemocompatibility is recommended, as it may impact whether the hybrids are administered parenterally or via the oral route. Testing of specific hybrids for oral delivery will require in vitro testing to examine their capacity to diffuse across the gut barrier, their interactions with intestinal enterocytes and metabolism, and the absorption by these cells [117]. All aspects of toxicology are greatly enhanced by the use of in silico screening that enables the prediction of biological activity and toxicity. Computer-based methods are available that predict the toxicity of new chemicals. All chemical substances have a complex structure, with a number of substituents and bonds, each contributing in some way to its overall properties, including its toxicology. In silico toxicology techniques identify structural alerts in new chemicals, and this allows the synthesis and assessment of safer molecules, which can also be incorporated into libraries of molecules to accelerate drug design and discovery [115]. These methods are beneficial to identify compounds that have a higher degree of safety, decreasing the burden on animal testing. After the drugs are in widespread clinical use, a robust post-marketing surveillance program that ensures continuous vigilance against unexpected adverse reactions is also required.

Overall, the progress of algal fatty acid-heterocyclic hybrids as therapeutics for the treatment of diabetes will require optimized extraction and isolation protocols, comprehensive in vivo validation, improved knowledge of bioavailability and metabolism, deeper mechanistic studies, and proper toxicological and safety assessments.

7 Conclusion

Fatty acid-heterocyclic hybrids from green algae represent a novel and promising class of bioactive compounds with significant antidiabetic potential. Their multifactorial mechanisms—ranging from inhibition of carbohydrate-digesting enzymes, modulation of insulin signaling pathways, to antioxidative and anti-inflammatory effects—highlight their therapeutic relevance in addressing the complex pathophysiology of diabetes. Supported by emerging preclinical evidence, these hybrids offer an innovative strategy for drug development. Nevertheless, comprehensive pharmacokinetic profiling, safety validation, and well-designed clinical trials are urgently needed to translate their potential into clinical practice.

Abbreviations

ADME	Absorption, distribution, metabolism, and excretion
AMPK	AMP-activated protein kinase
Bcl-2	B-cell lymphoma 2
DPP-4	Dipeptidyl peptidase-4

ERK	Extracellular signal-regulated kinase
FFA	Free fatty acids
GC–MS	Gas chromatography–mass spectrometry
GIP	Glucose-dependent insulinotropic polypeptide
GLP-1	Glucagon-like peptide-1
GLUT	Glucose transport protein
GPR	G-protein coupled receptors
HPLC	High-performance liquid chromatography
IRS	Insulin receptor substrate
JNK	c-Jun N-terminal kinase
LC–MS	Liquid chromatography–mass spectrometry
LDL	Low-density lipoprotein
LPS	Lipopolysaccharides
mTORC1	Mammalian target of rapamycin complex 1
NF-κB	Nuclear factor kappa B
NMR	Nuclear magnetic resonance
PCC	Protein carbonyl content
PI3K/Akt	Phosphoinositide 3-kinase/Akt pathway
PK	Pharmacokinetics
PD	Pharmacodynamics
PTP1B	Protein tyrosine phosphatase 1B
PUFA	Polyunsaturated fatty acids
ROS	Reactive oxygen species
SAR	Structure–activity relationship
SCFAs	Short-chain fatty acids
SOD	Superoxide dismutase
T2D	Type 2 diabetes
TNF-α	Tumor necrosis factor-alpha

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References

1. Verma AK, Goyal Y, Bhatt D, Dev K, Alsahli MA, Rahmani AH, et al. A compendium of perspectives on diabetes: A challenge for sustainable health in the modern era. Diabetes Metab Syndr Obes. 2021;14:2775–87. https://doi.org/10.2147/DMSO.S304751.
2. Singer ME, Dorrance KA, Oxenreiter MM, Yan KR, Close KL. The type 2 diabetes modern preventable pandemic and replicable lessons from the COVID-19 crisis. Prev Med Rep. 2022;25(101636):101636. https://doi.org/10.1016/j.pmedr.2021.101636.
3. Gregory GA, Robinson TIG, Linklater SE, Wang F, Colagiuri S, de Beaufort C, et al. Global incidence, prevalence, and mortality of type 1 diabetes in 2021 with projection to 2040: a modelling study. Lancet Diabetes Endocrinol. 2022;10(10):741–60. https://doi.org/10.1016/S2213-8587(22)00218-2.
4. Ogle GD, Wang F, Haynes A, Gregory GA, King TW, Deng K, et al. Global type 1 diabetes prevalence, incidence, and mortality estimates 2025: results from the international diabetes federation atlas, 11th edition, and the T1D index version 3.0. Diabetes Res Clin Pract. 2025;225(112277):112277. https://doi.org/10.1016/j.diabres.2025.112277.
5. Huang X, Wu Y, Ni Y, Xu H, He Y. Global, regional, and National burden of type 2 diabetes mellitus caused by high BMI from 1990 to 2021, and forecasts to 2045: analysis from the global burden of disease study 2021. Front Public Health. 2025;13:1515797. https://doi.org/10.3389/fpubh.2025.1515797.
6. International Diabetes Federation. Indonesia [Internet]. International Diabetes Federation. 2023. https://idf.org/our-network/regions-and-members/western-pacific/members/indonesia/. Accessed 9 Aug 2025.
7. Bradley MC, Chillarige Y, Lee H, Wu X, Parulekar S, Muthuri S, et al. Severe hypoglycemia risk with long-acting insulin analogs vs neutral Protamine Hagedorn insulin. JAMA Intern Med. 2021;181(5):598–607. https://doi.org/10.1001/jamainternmed.2020.9176.
8. D'Souza MS, Brady RE, Ray SD. Side effects of insulin and other antihyperglycemic drugs. Side effects of drugs annual. Elsevier; 2021. pp. 451–61. https://doi.org/10.1016/bs.seda.2021.08.007.
9. Rasouli H, Hosseini-Ghazvini SM-B, Adibi H, Khodarahmi R. Differential α-amylase/α-glucosidase inhibitory activities of plant-derived phenolic compounds: a virtual screening perspective for the treatment of obesity and diabetes. Food Funct. 2017;8(5):1942–54. https://doi.org/10.1039/c7fo00220c.
10. Rasouli H, Yarani R, Pociot F, Popović-Djordjević J. Anti-diabetic potential of plant alkaloids: revisiting current findings and future perspectives. Pharmacol Res. 2020;155(104723):104723. https://doi.org/10.1016/j.phrs.2020.104723.
11. Menaa F, Wijesinghe U, Thiripuranathar G, Althobaiti NA, Albalawi AE, Khan BA, et al. Marine algae-derived bioactive compounds: A new wave of nanodrugs? Mar Drugs. 2021;19(9):484. https://doi.org/10.3390/md19090484.
12. Choudhary B, Chauhan OP, Mishra A. Edible seaweeds: A potential novel source of bioactive metabolites and nutraceuticals with human health benefits. Front Mar Sci. 2021;8. https://doi.org/10.3389/fmars.2021.740054.
13. Taher N, Mentang F, Montolalu RI, Gunawan WB, Taslim NA, Mayulu N, et al. Green seaweeds fatty acids and heterocyclic derivatives against cancer: opinion on future nutraceutical application. Front Oncol. 2023;13:1145919. https://doi.org/10.3389/fonc.2023.1145919.
14. Aedh AI, Alshahrani MS, Huneif MA, Pryme IF, Oruch R. A glimpse into milestones of insulin resistance and an updated review of its management. Nutrients. 2023;15(4). https://doi.org/10.3390/nu15040921.
15. Cha S-H, Zhang C, Heo S-J, Jun H-S. 5-Bromoprotocatechualdehyde combats against palmitate toxicity by inhibiting parkin degradation and reducing ROS-induced mitochondrial damage in pancreatic β-cells. Antioxid (Basel). 2021;10(2):264. https://doi.org/10.3390/antiox10020264.
16. Dembitsky VM. Hydrobiological aspects of fatty acids: unique, rare, and unusual fatty acids incorporated into linear and Cyclic lipopeptides and their biological activity. Hydrobiology. 2022;1(3):331–432. https://doi.org/10.3390/hydrobiology1030024.
17. Kesharwani R, Jaiswal P, Patel DK, Yadav PK. Lipid-based drug delivery system (LBDDS): an emerging paradigm to enhance oral bioavailability of poorly soluble drugs. Biomed Mater Devices. 2022. https://doi.org/10.1007/s44174-022-00041-0.
18. Majrashi TA, Sabt A, Abd El Salam HA, Al-Ansary GH, Hamissa MF, Eldehna WM. An updated review of fatty acid residue-tethered heterocyclic compounds: synthetic strategies and biological significance. RSC Adv. 2023;13(20):13655–82. https://doi.org/10.1039/d3ra01368e.
19. Varshney H, Ahmad A, Rauf A, Husain FM, Ahmad I. Synthesis and antimicrobial evaluation of fatty chain substituted 2,5-dimethyl pyrrole and 1,3-benzoxazin-4-one derivatives. J Saudi Chem Soc. 2017;21:S394–402. https://doi.org/10.1016/j.jscs.2014.04.008.
20. Venepally V, Prasad RBN, Poornachandra Y, Kumar CG, Jala RCR. Synthesis of novel Ethyl 1-ethyl-6-fluoro-7-(fatty amido)-1,4-dihydro-4-oxoquinoline-3-carboxylate derivatives and their biological evaluation. Bioorg Med Chem Lett. 2016;26(2):613–7. https://doi.org/10.1016/j.bmcl.2015.11.063.
21. Venepally V, Reddy Jala RC. An insight into the biological activities of heterocyclic-fatty acid hybrid molecules. Eur J Med Chem. 2017;141:113–37. https://doi.org/10.1016/j.ejmech.2017.09.069.
22. Egbujor MC, Tucci P, Onyeije UC, Emeruwa CN, Saso L. NRF2 activation by nitrogen heterocycles: A review. Molecules. 2023;28(6). https://doi.org/10.3390/molecules28062751.
23. Farwa U, Raza MA. Heterocyclic compounds as a magic bullet for diabetes mellitus: a review. RSC Adv. 2022;12(35):22951–73. https://doi.org/10.1039/d2ra02697j.
24. Ali O, Szabó A. Review of eukaryote cellular membrane lipid composition, with special attention to the fatty acids. Int J Mol Sci. 2023;24(21). https://doi.org/10.3390/ijms242115693.
25. Galal-Khallaf A, Mousa D, Atyah A, El-Bahnsawye M, Hussein MKA, El Sayed IET, et al. In vivo antidiabetic and antilipidemic effect of thiazolidine-2,4-Dione linked heterocyclic scaffolds in obesity-induced zebrafish model. Pharmaceuticals (Basel). 2025;18(7):1023. https://doi.org/10.3390/ph18071023.
26. Hamad ARA, Sadasivam M, Rabb H. Hybrid lipids, peptides, and lymphocytes: [internet]ew [internet]ra in [internet]ype 1 diabetes [internet]esearch [Internet]. J Clin Invest. 2019;9. https://doi.org/10.1172/JCI130313.

27. Begum M, Choubey M, Tirumalasetty MB, Arbee S, Mohib MM, Wahiduzzaman M, et al. Adiponectin: A promising target for the treatment of diabetes and its complications. Life (Basel). 2023;13(11):2213. https://doi.org/10.3390/life13112213.
28. Blasio M, Balzano S. Fatty acids derivatives from eukaryotic microalgae, pathways and potential applications. Front Microbiol. 2021;12:718933. https://doi.org/10.3389/fmicb.2021.718933.
29. Lima-Pereira Y, de Souza EMO, dos Reis DS, Barcellos-Silva IGC, Miki KSL, Veiga-Júnior VF, et al. Current perspectives on the extraction, isolation, and identification of fats and fatty acids using conventional and green methods. Separations. 2025;12(6):160. https://doi.org/10.3390/separations12060160.
30. Saber AA, Rashedy SH, Rushdi MI, Saber H, Attia EZ, Abdel-Rahman IAM, et al. Insights into the phytochemical and Pharmacological natural products of the green macroalga *Halimeda* (Chlorophyta). Nat Prod Commun. 2025;20(3). https://doi.org/10.1177/1934578x251331137.
31. Bustamam MSA, Pantami HA, Azizan A, Shaari K, Min CC, Abas F, et al. Complementary analytical platforms of NMR spectroscopy and LCMS analysis in the metabolite profiling of isochrysis Galbana. Mar Drugs. 2021;19(3):139. https://doi.org/10.3390/md19030139.
32. Karthikeyan S, Grishina M, Kandasamy S, Mangaiyarkarasi R, Ramamoorthi A, Chinnathambi S, et al. A review on medicinally important heterocyclic compounds and importance of biophysical approach of underlying the insight mechanism in biological environment. J Biomol Struct Dyn. 2023;41(23):14599–619. https://doi.org/10.1080/07391102.2023.2187640.
33. Tong CY, Honda K, Derek CJC. A review on microalgal-bacterial co-culture: the multifaceted role of beneficial bacteria towards enhancement of microalgal metabolite production. Environ Res. 2023;228(115872):115872. https://doi.org/10.1016/j.envres.2023.115872.
34. Kothari R, Singh HM, Azam R, Goria K, Bharti A, Singh A, et al. Potential avenue of genetic engineered algal derived bioactive compounds: influencing parameters, challenges and future prospects. Phytochem Rev. 2023;22(4):935–68. https://doi.org/10.1007/s11101-023-09859-y.
35. Proschak E, Heitel P, Kalinowsky L, Merk D. Opportunities and challenges for fatty acid mimetics in drug discovery. J Med Chem. 2017;60(13):5235–66. https://doi.org/10.1021/acs.jmedchem.6b01287.
36. Trifonov RE, Ostrovskii VA. Tetrazoles and related heterocycles as promising synthetic antidiabetic agents. Int J Mol Sci. 2023;24(24). https://doi.org/10.3390/ijms242417190.
37. Ngoc Toan V, Son Hai D, Thi Kim Van H, Minh Tri N, Ngoc Toan D, Thi Thanh Mai N, et al. Design, synthesis, inhibitory activity, and molecular simulations study for d-glucose-conjugated thioureas containing pyrimidine ring as multitarget inhibitors against α-amylase, α-glucosidase, DDP-4, and PTP1B in type 2 diabetes mellitus. RSC Med Chem. 2024. https://doi.org/10.1039/d4md00334a.
38. Li M, Ding L, Cao L, Zhang Z, Li X, Li Z, et al. Natural products targeting AMPK signaling pathway therapy, diabetes mellitus and its complications. Front Pharmacol. 2025;16:1534634. https://doi.org/10.3389/fphar.2025.1534634.
39. Pereira L, Valado A. Algae-derived natural products in diabetes and its complications-current advances and future prospects. Life (Basel). 2023;13(9). https://doi.org/10.3390/life13091831.
40. Pradhan T, Gupta O, Kumar V, Sristi, Chawla G. A comprehensive review on the antidiabetic attributes of thiazolidine-4-ones: synthetic strategies and structure-activity relationships. Arch Pharm (Weinheim). 2023;356(2):e2200452. https://doi.org/10.1002/ardp.202200452.
41. Mlala S, Oyedeji AO, Gondwe M, Oyedeji OO. Ursolic acid and its derivatives as bioactive agents. Molecules. 2019;24(15):2751. https://doi.org/10.3390/molecules24152751.
42. Tamel Selvan K, Goon JA, Makpol S, Tan JK. Therapeutic potentials of microalgae and their bioactive compounds on diabetes mellitus. Mar Drugs. 2023;21(9):462. https://doi.org/10.3390/md21090462.
43. Gautam R, Patial SK, Singh S. Algae biomass: importance, harvesting techniques, extraction methods, and associated challenges. Value added products from bioalgae based biorefineries: opportunities and challenges. Singapore: Springer Nature Singapore; 2024. pp. 67–94. https://doi.org/10.1007/978-981-97-1662-3_4.
44. Pantami HA, Ahamad Bustamam MS, Lee SY, Ismail IS, Mohd Faudzi SM, Nakakuni M, et al. Comprehensive GCMS and LC-MS/MS metabolite profiling of chlorella vulgaris. Mar Drugs. 2020;18(7):367. https://doi.org/10.3390/md18070367.
45. Zhang Q-W, Lin L-G, Ye W-C. Techniques for extraction and isolation of natural products: a comprehensive review. Chin Med. 2018;13:20. https://doi.org/10.1186/s13020-018-0177-x.
46. Chen H, Qi H, Xiong P. Phycobiliproteins-A family of algae-derived biliproteins: productions, characterization and pharmaceutical potentials. Mar Drugs. 2022;20(7):450. https://doi.org/10.3390/md20070450.
47. Li T, Jiang Y, Lin S, Zhong S, Jian S, Chen W, et al. Environmental irradiance and nitrogen source variability trigger the intracellular carbon and nitrogen reallocation of Gracilariopsis lemaneiformis (Rhodophyta). Aquaculture. 2023;575(739802):739802. https://doi.org/10.1016/j.aquaculture.2023.739802.
48. Cui L, Yang B, Tan L, Xu J, Xie L, Jiang W, et al. High light and nitrogen deficiency synergistically lead to biofuel and biomass accumulation in diatoms. Chem Eng J. 2024;498(155818):155818. https://doi.org/10.1016/j.cej.2024.155818.
49. Zhao R, Wang L, Wang Q, Dai S, Han Z, Lei X, et al. Integrated model for design, optimization, scale-up, culture scheme, and production evaluation of a bioreactor and its application for improving algal biomass and carbon fixation in bioreactors. ACS Sustain Chem Eng. 2022;10(48):15939–49. https://doi.org/10.1021/acssuschemeng.2c05332.
50. Udayan A, Sirohi R, Sreekumar N, Sang B-I, Sim SJ. Mass cultivation and harvesting of microalgal biomass: current trends and future perspectives. Bioresour Technol. 2022;344(Pt B):126406. https://doi.org/10.1016/j.biortech.2021.126406.
51. Mahrous E, Chen R, Zhao C, Farag MA. Lipidomics in food quality and authentication: A comprehensive review of novel trends and applications using chromatographic and spectroscopic techniques. Crit Rev Food Sci Nutr. 2024;64(25):9058–81. https://doi.org/10.1080/10408398.2023.2207659.
52. Corrêa PS, Morais Júnior WG, Martins AA, Caetano NS, Mata TM. Microalgae biomolecules: extraction, separation and purification methods. Processes (Basel). 2020;9(1):10. https://doi.org/10.3390/pr9010010.
53. Egbujor MC, Buttari B, Profumo E, Telkoparan-Akillilar P, Saso L. An overview of NRF2-activating compounds bearing α,β-unsaturated moiety and their antioxidant effects. Int J Mol Sci. 2022;23(15):8466. https://doi.org/10.3390/ijms23158466.
54. Ran X, Shen Y, Jiang D, Wang C, Li X, Zhang H, et al. Nutrient deprivation coupled with high light exposure for bioactive chrysolaminarin production in the marine microalga isochrysis zhangjiangensis. Mar Drugs. 2022;20(6):351. https://doi.org/10.3390/md20060351.
55. Cui H, Yang F, Li Y. Exogenous Methyl jasmonate enhances lipid production in isochrysis Galbana under nitrogen deprivation and high light. Algal Res. 2021;58(102406):102406. https://doi.org/10.1016/j.algal.2021.102406.

56. Vijaya D. Innovations in algal drug research, with a focus on diabetes mellitus (DM) inhibitors of enzymes. Int J Multidisciplinary Res. 2023;5(5). https://doi.org/10.36948/ijfmr.2023.v05i05.6694.
57. Rayapu L, Chakraborty K, Valluru L. Marine algae as a potential source for anti-diabetic compounds - A brief review. Curr Pharm Des. 2021;27(6):789–801. https://doi.org/10.2174/1381612826666200909124526.
58. Agarwal S, Singh V, Chauhan K. Antidiabetic potential of seaweed and their bioactive compounds: a review of developments in last decade. Crit Rev Food Sci Nutr. 2023;63(22):5739–70. https://doi.org/10.1080/10408398.2021.2024130.
59. Samovski D, Jacome-Sosa M, Abumrad NA. Fatty acid transport and signaling: mechanisms and physiological implications. Annu Rev Physiol. 2023;85(1):317–37. https://doi.org/10.1146/annurev-physiol-032122-030352.
60. Popović-Djordjević JB, Katanić Stanković JS, Mihailović V, Pereira AG, Garcia-Oliveira P, Prieto MA, et al. Algae as a source of bioactive compounds to prevent the development of type 2 diabetes mellitus. Curr Med Chem. 2021;28(23):4592–615. https://doi.org/10.2174/0929867328666210325100654.
61. Khalid M, Petroianu G, Adem A. Advanced glycation end products and diabetes mellitus: mechanisms and perspectives. Biomolecules. 2022;12(4):542. https://doi.org/10.3390/biom12040542.
62. Ávila-Román J, García-Gil S, Rodríguez-Luna A, Motilva V, Talero E. Anti-inflammatory and anticancer effects of microalgal carotenoids. Mar Drugs. 2021;19(10):531. https://doi.org/10.3390/md19100531.
63. Rocha S, Lucas M, Ribeiro D, Corvo ML, Fernandes E, Freitas M. Nano-based drug delivery systems used as vehicles to enhance polyphenols therapeutic effect for diabetes mellitus treatment. Pharmacol Res. 2021;169(105604):105604. https://doi.org/10.1016/j.phrs.2021.105604.
64. Boudebbous K, Boulebd H, Derabli C, Bendjeddou L, Bensouici C, Harakat D, et al. Synthesis, in vitro and in Silico studies of naphto-1,3-oxazin-3(2H)-one derivatives as promising inhibitors of cholinesterase and α-glucosidase. J Mol Struct. 2021;1225(129103):129103. https://doi.org/10.1016/j.molstruc.2020.129103.
65. Khan I, Rehman W, Rahim F, Hussain R, Khan S, Rasheed L, et al. Synthesis and in vitro α-amylase and α-glucosidase dual inhibitory activities of 1,2,4-triazole-bearing bis-hydrazone derivatives and their molecular Docking study. ACS Omega. 2023;8(25):22508–22. https://doi.org/10.1021/acsomega.3c00702.
66. Pedrood K, Rezaei Z, Khavaninzadeh K, Larijani B, Iraji A, Hosseini S, et al. Design, synthesis, and molecular Docking studies of diphenylquinoxaline-6-carbohydrazide hybrids as potent α-glucosidase inhibitors. BMC Chem. 2022;16(1):57. https://doi.org/10.1186/s13065-022-00848-4.
67. Esmaili S, Ebadi A, Khazaei A, Ghorbani H, Faramarzi MA, Mojtabavi S, et al. Novel pyrano[3,2-c]quinoline-1,2,3-triazole hybrids as potential anti-diabetic agents: in vitro α-glucosidase inhibition, kinetic, and molecular dynamics simulation. ACS Omega. 2023;8(26):23412–24. https://doi.org/10.1021/acsomega.3c00133.
68. Nkoana JK, Mphahlele MJ, More GK, Choong YS. Exploring the 3,5-dibromo-4,6-dimethoxychalcones and their flavone derivatives as dual α-glucosidase and α-amylase inhibitors with antioxidant and anticancer potential. Antioxid (Basel). 2024;13(10):1255. https://doi.org/10.3390/antiox13101255.
69. Kaya B, Acar Çevik U, Çiftçi B, Duran HE, Türkeş C, Işık M, et al. Synthesis, α-glucosidase, α-amylase, and aldol reductase inhibitory activity with molecular Docking study of novel imidazo[1,2-a]pyridine derivatives. ACS Omega. 2024;9(42):42905–14. https://doi.org/10.1021/acsomega.4c05619.
70. Singh A, Singh K, Kaur K, Sharma A, Mohana P, Prajapati J, et al. Discovery of Triazole tethered thymol/carvacrol-coumarin hybrids as new class of α-glucosidase inhibitors with potent in vivo antihyperglycemic activities. Eur J Med Chem. 2024;263(115948):115948. https://doi.org/10.1016/j.ejmech.2023.115948.
71. Mourad AAE, Mourad MAE. Enhancing insulin sensitivity by dual PPARγ partial agonist, β-catenin inhibitor: design, synthesis of new αphthalimido-o-toluoyl2-aminothiazole hybrids. Life Sci. 2020;259(118270):118270. https://doi.org/10.1016/j.lfs.2020.118270.
72. Shanak S, Bassalat N, Barghash A, Kadan S, Ardah M, Zaid H. Drug discovery of plausible lead natural compounds that target the insulin signaling pathway: bioinformatics approaches. Evid Based Complement Alternat Med. 2022;2022:2832889. https://doi.org/10.1155/2022/2832889.
73. Martínez Báez A, Ayala G, Pedroza-Saavedra A, González-Sánchez HM, Chihu Amparan L. Phosphorylation codes in IRS-1 and IRS-2 are associated with the activation/inhibition of insulin canonical signaling pathways. Curr Issues Mol Biol. 2024;46(1):634–49. https://doi.org/10.3390/cimb46010041.
74. van Gerwen J, Shun-Shion AS, Fazakerley DJ. Insulin signalling and GLUT4 trafficking in insulin resistance. Biochem Soc Trans. 2023;51(3):1057–69. https://doi.org/10.1042/BST20221066.
75. Teimouri M, Hosseini H, ArabSadeghabadi Z, Babaei-Khorzoughi R, Gorgani-Firuzjaee S, Meshkani R. The role of protein tyrosine phosphatase 1B (PTP1B) in the pathogenesis of type 2 diabetes mellitus and its complications. J Physiol Biochem. 2022;78(2):307–22. https://doi.org/10.1007/s13105-021-00860-7.
76. Yan H, He L, Lv D, Yang J, Yuan Z. The role of the dysregulated JNK signaling pathway in the pathogenesis of human diseases and its potential therapeutic strategies: A comprehensive review. Biomolecules. 2024;14(2):243. https://doi.org/10.3390/biom14020243.
77. Al-Wahaibi LH, Abdel-Rahman MH, El-Adl K, Youssif BGM, Bräse S, Abdel-Aziz SA. New diaryl-1,2,4-triazolo[3,4-a]pyrimidine hybrids as selective COX-2/sEH dual inhibitors with potent analgesic/anti-inflammatory and cardioprotective properties. ACS Omega. 2024;9(31):33494–509. https://doi.org/10.1021/acsomega.4c00870.
78. Li Z, Qiu Q, Xu X, Wang X, Jiao L, Su X, et al. Design, synthesis and Structure-activity relationship studies of new thiazole-based free fatty acid receptor 1 agonists for the treatment of type 2 diabetes. Eur J Med Chem. 2016;113:246–57. https://doi.org/10.1016/j.ejmech.2016.02.040.
79. Singh R, Gholipourmalekabadi M, Shafikhani SH. Animal models for type 1 and type 2 diabetes: advantages and limitations. Front Endocrinol (Lausanne). 2024;15:1359685. https://doi.org/10.3389/fendo.2024.1359685.
80. Guerra JVS, Dias MMG, Brilhante AJVC, Terra MF, García-Arévalo M, Figueira ACM. Multifactorial basis and therapeutic strategies in metabolism-related diseases. Nutrients. 2021;13(8):2830. https://doi.org/10.3390/nu13082830.
81. Dyall SC, Balas L, Bazan NG, Brenna JT, Chiang N, da Costa Souza F, et al. Polyunsaturated fatty acids and fatty acid-derived lipid mediators: recent advances in the Understanding of their biosynthesis, structures, and functions. Prog Lipid Res. 2022;86(101165):101165. https://doi.org/10.1016/j.plipres.2022.101165.
82. Matin M, Koszarska M, Atanasov AG, Król-Szmajda K, Jóźwik A, Stelmasiak A, et al. Bioactive potential of algae and algae-derived compounds: focus on anti-inflammatory, antimicrobial, and antioxidant effects. Molecules. 2024;29(19). https://doi.org/10.3390/molecules29194695.

83. Sharifuddin Y, Chin Y-X, Lim P-E, Phang S-M. Potential bioactive compounds from seaweed for diabetes management. Mar Drugs. 2015;13(8):5447–91. https://doi.org/10.3390/md13085447.
84. Ramya CRC, Sowmya R, Kumari A, Mariappan GMG, Pethe A, Ingle R. Bioactive compounds from marine algae: dual benefits for diabetes and renal health. J Neonatal Surg. 2025;14(14S):134–42. https://doi.org/10.52783/jns.v14.3539.
85. Sharma PP, Baskaran V. Polysaccharide (laminaran and fucoidan), Fucoxanthin and lipids as functional components from brown algae (Padina tetrastromatica) modulates adipogenesis and thermogenesis in diet-induced obesity in C57BL6 mice. Algal Res. 2021;54(102187):102187. https://doi.org/10.1016/j.algal.2021.102187.
86. Tian H, Wang S, Deng Y, Xing Y, Zhao L, Zhang X, et al. Fatty acid profiles and their association with autoimmunity, insulin sensitivity and β cell function in latent autoimmune diabetes in adults. Front Endocrinol (Lausanne). 2022;13:916981. https://doi.org/10.3389/fendo.2022.916981.
87. Li X, Bi X, Wang S, Zhang Z, Li F, Zhao AZ. Therapeutic potential of ω-3 polyunsaturated fatty acids in human autoimmune diseases. Front Immunol. 2019;10:2241. https://doi.org/10.3389/fimmu.2019.02241.
88. Liu Y, Ren X, Fan C, Wu W, Zhang W, Wang Y. Health benefits, food applications, and sustainability of microalgae-derived N-3 PUFA. Foods. 2022;11(13):1883. https://doi.org/10.3390/foods11131883.
89. Melloni M, Sergi D, Simioni C, Passaro A, Neri LM. Microalgae as a nutraceutical tool to antagonize the impairment of redox status induced by snps: implications on insulin resistance. Biology (Basel). 2023;12(3):449. https://doi.org/10.3390/biology12030449.
90. Biondi G, Marrano N, Borrelli A, Rella M, Palma G, Calderoni I, et al. Adipose tissue secretion pattern influences β-cell wellness in the transition from obesity to type 2 diabetes. Int J Mol Sci. 2022;23(10):5522. https://doi.org/10.3390/ijms23105522.
91. Velikova TV, Kabakchieva PP, Assyov YS, Georgiev TA. Targeting inflammatory cytokines to improve type 2 diabetes control. Biomed Res Int. 2021;2021(1):7297419. https://doi.org/10.1155/2021/7297419.
92. Grosso G, Laudisio D, Frias-Toral E, Barrea L, Muscogiuri G, Savastano S, et al. Anti-inflammatory nutrients and obesity-associated metabolic-inflammation: state of the Art and future direction. Nutrients. 2022;14(6):1137. https://doi.org/10.3390/nu14061137.
93. Milusheva M, Todorova M, Gledacheva V, Stefanova I, Feizi-Dehnayebi M, Pencheva M, et al. Novel anthranilic acid hybrids-an alternative weapon against inflammatory diseases. Pharmaceuticals (Basel). 2023;16(12). https://doi.org/10.3390/ph16121660.
94. Papachristoforou E, Lambadiari V, Maratou E, Makrilakis K. Association of glycemic indices (hyperglycemia, glucose variability, and hypoglycemia) with oxidative stress and diabetic complications. J Diabetes Res. 2020;2020:7489795. https://doi.org/10.1155/2020/7489795.
95. Iacobini C, Vitale M, Pesce C, Pugliese G, Menini S. Diabetic complications and oxidative stress: A 20-year voyage back in time and back to the future. Antioxid (Basel). 2021;10(5):727. https://doi.org/10.3390/antiox10050727.
96. Kim D-S, Scherer PE. Obesity, diabetes, and increased cancer progression. Diabetes Metab J. 2021;45(6):799–812. https://doi.org/10.4093/dmj.2021.0077.
97. Gadallah AS, Mujeeb-Ur-Rehman, Atta-Ur-Rahman, Yousuf S, Atia-Tul-Wahab, Jabeen A, et al. Anti-inflammatory principles from tamarix aphylla L.: A bioassay-guided fractionation study. Molecules. 2020;25(13):2994. https://doi.org/10.3390/molecules25132994.
98. Chen Y, Ouyang Y, Chen X, Chen R, Ruan Q, Farag MA, et al. Hypoglycaemic and anti-ageing activities of green Alga Ulva lactuca polysaccharide via gut microbiota in ageing-associated diabetic mice. Int J Biol Macromol. 2022;212:97–110. https://doi.org/10.1016/j.ijbiomac.2022.05.109.
99. Yosief HO, Sarker MI. Naturally derived fatty acid based antibacterial agents. ACS Symposium Series. Washington, DC: American Chemical Society; 2021. pp. 91–117. https://doi.org/10.1021/bk-2021-1392.ch006
100. Cheong K-L, Yu B, Chen J, Zhong S. A comprehensive review of the cardioprotective effect of marine algae polysaccharide on the gut microbiota. Foods. 2022;11(22):3550. https://doi.org/10.3390/foods11223550.
101. Li J, Zhao J, Tian C, Dong L, Kang Z, Wang J, et al. Mechanisms of regulation of glycolipid metabolism by natural compounds in plants: effects on short-chain fatty acids. Nutr Metab (Lond). 2024;21(1):49. https://doi.org/10.1186/s12986-024-00829-5.
102. Di Vincenzo F, Del Gaudio A, Petito V, Lopetuso LR, Scaldaferri F. Gut microbiota, intestinal permeability, and systemic inflammation: a narrative review. Intern Emerg Med. 2024;19(2):275–93. https://doi.org/10.1007/s11739-023-03374-w.
103. Candelli M, Franza L, Pignataro G, Ojetti V, Covino M, Piccioni A, et al. Interaction between lipopolysaccharide and gut microbiota in inflammatory bowel diseases. Int J Mol Sci. 2021;22(12):6242. https://doi.org/10.3390/ijms22126242.
104. Clemente-Suárez VJ, Redondo-Flórez L, Rubio-Zarapuz A, Martín-Rodríguez A, Tornero-Aguilera JF. Microbiota implications in endocrine-related diseases: from development to novel therapeutic approaches. Biomedicines. 2024;12(1). https://doi.org/10.3390/biomedicines12010221.
105. Mahapatra MK, Kumar R, Kumar M. Synthesis, biological evaluation and in Silico studies of 5-(3-methoxybenzylidene)thiazolidine-2,4-dione analogues as PTP1B inhibitors. Bioorg Chem. 2017;71:1–9. https://doi.org/10.1016/j.bioorg.2017.01.007.
106. Aisha, Raza MA, Sumrra SH, Javed K, Saqib Z, Maurin JK, et al. Synthesis, characterization and molecular modeling of amino derived Thiazolidinones as esterase and glucosidase inhibitors. J Mol Struct. 2020;1219(128609):128609. https://doi.org/10.1016/j.molstruc.2020.128609.
107. Grattoni A, Korbutt G, Tomei AA, García AJ, Pepper AR, Stabler C, et al. Harnessing cellular therapeutics for type 1 diabetes mellitus: progress, challenges, and the road ahead. Nat Rev Endocrinol. 2025;21(1):14–30. https://doi.org/10.1038/s41574-024-01029-0.
108. Demarco M, Oliveira de Moraes J, Matos ÂP, Derner RB, de Farias Neves F, Tribuzi G. Digestibility, bioaccessibility and bioactivity of compounds from algae. Trends Food Sci Technol. 2022;121:114–28. https://doi.org/10.1016/j.tifs.2022.02.004.
109. Diab AS, Elsayed KNM, El-Sherbeeny AM, Al Zoubi W, Bellucci S, Abukhadra MR. Synthesis and characterization of turbinaria Ornata mediated zn/zno green nanoparticles as potential antioxidant and anti-diabetic agent of enhanced activity. Front Mar Sci. 2024;11. https://doi.org/10.3389/fmars.2024.1444618.
110. Dimopoulou M, Kolonas A, Stagos D, Gortzi O. A review of the sustainability, chemical composition, bioactive compounds, antioxidant and antidiabetic activity, neuroprotective properties, and health benefits of microalgae. Biomass (Basel). 2025;5(1):11. https://doi.org/10.3390/biomass5010011.

111. Naim MJ, Alam O, Alam MJ, Hassan MQ, Siddiqui N, Naidu VGM, et al. Design, synthesis and molecular Docking of Thiazolidinedione based benzene sulphonamide derivatives containing pyrazole core as potential anti-diabetic agents. Bioorg Chem. 2018;76:98–112. https://doi.org/10.1016/j.bioorg.2017.11.010.
112. Mohamed MAA, Abd Allah OA, Bekhit AA, Kadry AM, El-Saghier AMM. Synthesis and antidiabetic activity of novel Triazole derivatives containing amino acids. J Heterocycl Chem. 2020;57(6):2365–78. https://doi.org/10.1002/jhet.3951.
113. Green G. Characterizing arguments about endocrine disruptors and human health [Internet]. University of British Columbia; 2023. https://doi.org/10.14288/1.0423802
114. Ahmad S, Akhtar S, Kamal MA, Farooqui A. Diabetes mellitus: exploring biomarkers, complications, and therapeutic strategies with a spotlight on cyanobacterial bioactive compounds - A comprehensive review. Curr Diabetes Rev. 2024. https://doi.org/10.2174/0115733998307764240909114007.
115. Animish A, Jayasri MA. A retrospective review of marine algae and the strategies employed for prospective diabetes management. Algal Res. 2023;74(103209):103209. https://doi.org/10.1016/j.algal.2023.103209.
116. Gavanji S, Bakhtari A, Abdel-Latif R, Bencurova E, Othman EM. Experimental approaches for induction of diabetes mellitus and assessment of antidiabetic activity: an in vitro and in vivo methodological review. Fundam Clin Pharmacol. 2024;38(5):842–61. https://doi.org/10.1111/fcp.13009.
117. Xu Y, Shrestha N, Préat V, Beloqui A. An overview of in vitro, ex vivo and in vivo models for studying the transport of drugs across intestinal barriers. Adv Drug Deliv Rev. 2021;175(113795):113795. https://doi.org/10.1016/j.addr.2021.05.005.

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