Story of Young Innovators in Health Sciences

"Fucoxanthin derivative": A novel candidate to treat the hyperglycemic condition in patients with Type 2 Diabetes Mellitus

Gunathilaka MDTL[1], Samarakoon KW[3], Ranasinghe P[2], Peiris LDC[1*]

[1]Department of Zoology / Genetics & Molecular Biology Unit (Center for Biotechnology), Faculty of Applied Sciences, University of Sri Jayewardenepura, Sri Lanka, [2]Industrial Technology Institute, Sri Lanka, [3]KDU Institute for Combinatorial Advanced Research and Education (KDU- CARE), General Sir John Kotelawala Defence University, Sri Lanka.

**Corresponding author (email: dinithi@sjp.ac.lk)*

Overview: Diabetes is the third leading cause of death worldwide associated with significant complications with a considerable impact on public health and poses a major threat to health and development worldwide. Type 2 Diabetes Mellitus (DM) is by far the most common type of diabetes (>90%), and it is characterized by hyperglycemia associated with insulin deficiency and insulin resistance. The prevalence of type 2 diabetes and obesity have increased globally due to rapid urbanization and unhealthy diets. More than 90% of patients with diabetes mellitus are either overweight or obese. The World Health Organization (WHO) has estimated that by 2035, the incidence of diabetes mellitus and impaired glucose tolerance will increase by up to 592 million and 471 million people, respectively. Type 2 diabetes is most common in Africa and South Asian countries; type 2 diabetes is prevalent among Sri Lankans. According to recent statistics, one in every five Sri Lankan adults has diabetes or is in the pre-diabetes stage.

Higher incidences of diabetes cases are reported annually in developed and developing countries. It is increasingly evident that the available therapeutic interventions such as insulin administration and oral antidiabetic drugs have either limited efficacy or detrimental side effects. Therefore, it is essential to keep searching for an effective drug that may benefit patients suffering from type 2 DM, leading to the curing of these patients. Marine algae are known for their beneficial effects. Phenolic, flavonoids, carotenoids, carbohydrates, lipids, proteins, and peptides found in algae are the key pharmacological constituents against diabetes, cancer, inflammation, and allergic conditions. Further, marine algae-derived accessory pigments are important as they possess beneficial biological activities. Fucoxanthin is the most abundant accessory pigment found in brown seaweeds and reported to have potent biological activities such as antioxidant, antidiabetic, anticancer activities.

Choonospora minima is a brown alga that belongs to the family Scytosiphonaceae. Polyphenols purified from brown algae are considered as a rich source of antioxidants with significant health-promoting properties. However, no one has been reported about the hypoglycemic potential

of *Chnoonospora minima* nationally and internationally. The present study focused on biologically guided fractions for *Chnoonospora minima* to identify inhibitory activities of carbohydrate hydrolysis enzymes and isolate a novel therapeutic active molecule against hypoglycemia.

Objectives of the research: The present study aimed to determine *the in-vitro* hypoglycemic activity of the crude methanol extract of *Chnoonospora minima* and its fractions using the inhibitory activity of carbohydrate hydrolysis enzymes. Further, it aimed to isolate a novel therapeutic compound with hypoglycemic activities.

Method: De-polysaccharide methanol extract of *C. minima* was partitioned with hexane, chloroform, and ethyl acetate, and *In-vitro* hypoglycemic activity was evaluated using α-amylase and α-glucosidase inhibitory activities. The α-amylase inhibitory activity was determined using the 3,5-dinitrosalicylic acid method, and α-glucosidase inhibitory activity was determined using α-glucosidase enzyme and p-nitrophenyl-a-D-glucopyranoside substrate. The chloroform fraction was selected for compound isolation using silica gel 60 and Sephadex LH20 column chromatography through bioactivity guided fractionation. Once an active sub-fraction of chromatography was established, it was further purified using reverse-phase HPLC purification and liquid chromatography-mass spectrometry (LC-MS) analysis. Finally, the structural elucidation was carried out using nuclear magnetic resonance (NMR) analysis.

Findings: In-vitro hypoglycemic *assays* - According to the results shown in Table 1, the most potent alpha-amylase inhibitory activity was exhibited by the chloroform fraction (IC_{50}: 3.17±0.09µg/mL) compared to the standard acarbose (87.43±0.59µg/mL). Similar inhibitory activity was observed in alpha-glucosidase inhibitory activity ($IC_{50\ chloroform\ fraction}$: 1.99±0.01 µg/mL; $IC50_{\ acarbose}$: 0.38±0.06µg/mL).

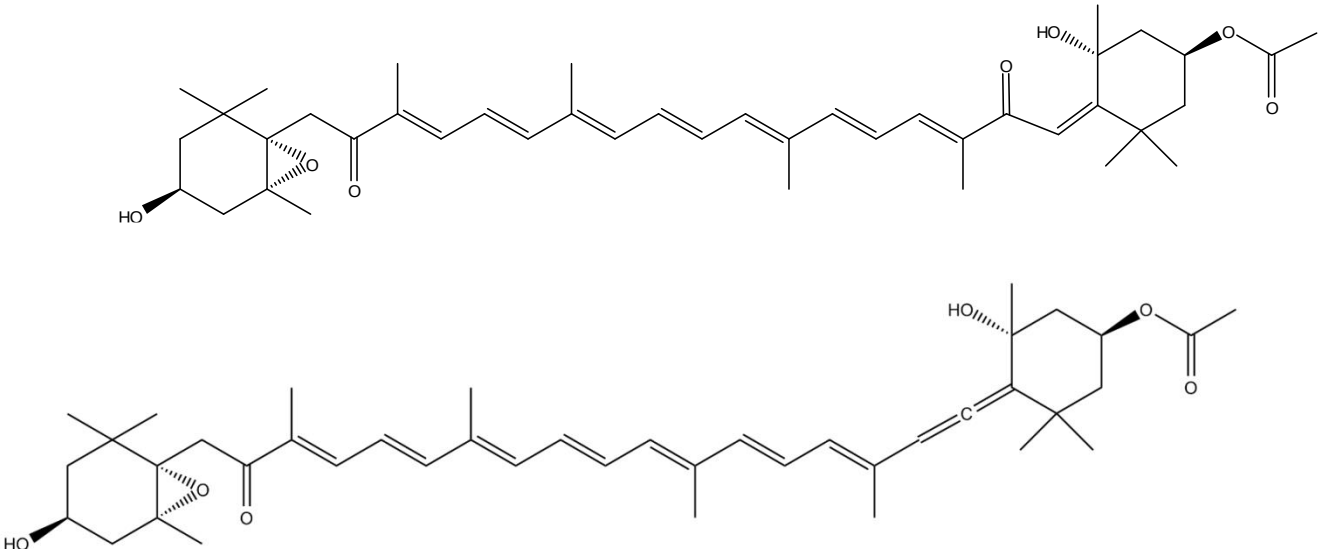
Table 1. α-amylase and α-glucosidase inhibitory activity (IC_{50-} µg/mL) of *C.minima* methanol extract and its fractions.

Extract/fraction	Anti-amylase activity	Anti-glucosidase activity
Crude methanol extract	18.25±0.13	3.83 ±0.05
Hexane fraction	305.49±5.76	38.86 ±0.71
Chloroform fraction	**3.17±0.02**	**1.99 ±0.01**
Ethyl acetate fraction	7.05±0.03	2.36±0.02
Aqueous fraction	80.85±3.91	3.89±0.03
Standard acarbose	87.43±0.59	0.38±0.06

Results are expressed as mean±SD; n = 4. *p<0.05 as compared with the standard acarbose.

Compound isolation: Upon purification of active sub-fraction by reverse-phase HPLC and liquid chromatography-mass spectrometry (LC-MS), the structure was elucidated using NMR spectroscopy. The 1D and 2D NMR data confirmed the structure as a "**fucoxanthin derivative**,"

a novel compound as shown in Figure 1. The newly isolated fucoxanthin derivative exhibited an extra unsaturated ketone at the C8` position (Figure 1), making the new compound unique.



(a)Newly isolated compound "fucoxanthin derivative"; Chemical Formula: $C_{42}H_{58}0_7$

(b). Fucoxanthin; Chemical formula: $C_{42}H_{58}0_6$

Figure 1. Structure of the (a) Newly isolated compound "fucoxanthin derivative"; (b) fucoxanthin.

Conclusion: The results confirm the hypoglycemic efficacy of *C. minima*. The study was also able to isolate a novel compound from *C. minima*, which could be used in developing a novel drug or supplement as a potential hypoglycemic agent.

Practical Implications: It is essential to evaluate the hypoglycemic potential of the isolated compound via several modes. Furthermore, *in vivo* trials are required to support therapeutic use and design a suitable dosage form to produce a new drug or supplement from *C. minima* extract to help manage blood glucose levels and complications.

Novelty: To the best of the authors' knowledge, this study is the first to isolate the novel active compound "fucoxanthin derivative" from *C. minima*. No research has been reported nor isolated the newly isolated compound "fucoxanthin derivative". The compound nomenclature is undergoing.

Benefit to the society: Further *in-vitro* and *in-vivo* studies of the newly isolated "fucoxanthin derivative" can lead to developing a novel drug to treat patients with Type 2 DM. The potential must be sought in the Sri Lankan community or their health promotion and well-being over the treating hyperglycemic conditions/ metabolic diseases. Moreover, the process development based on the extraction of *Chnoonospora minima* in the functional food industry will be considered.