Original Article

ANTIOXIDANT AND ANTI-INFLAMMATION ACTIVITIES OF ETHANOL EXTRACT FROM SEABLITE (*SUAEDA MARITIMA*) ROOT

Kanittada Thongkao[1], Narin Kakatum[2], Yuttana Sudjaroen[3]

[1,3]Department of Applied Science, Faculty of Science and Technology, Suan Sunandha Rajabhat University, Bangkok- 10300, Thailand

[2]Applied Thai Traditional Medicine Program, College of Allied Health Sciences, Suan Sunandha Rajabhat University, Samut Songkhram- 75000, Thailand

Abstract

Seablite or *Suaeda maritima* (L.) Dumort, a salt marsh plant growing in mangrove forest widely distributed on the landward margin of mangrove habitats of Thai Gulf and Southern of Thailand. Thai traditional medicine is using *S. maritima* root for abscess healing, allergic symptom relieves and treatment of skin diseases. However, biological properties of *S. maritima* root are very sparsely reported. In this study was attempt to evaluate antioxidant and anti-inflammation properties besides presence of phytochemicals in *S. maritima* root. Roots were extracted with 95% ethanol by maceration. Total phenolic content (TPC) and total flavonoid content (TFC) of *S. maritima* root extract was determined by colorimetric absorbance of reaction mixture measurements. Antioxidant property of *S. maritima* root extract was evaluated by DPPH, NO radical scavenging and lipid peroxidation inhibition. In vitro anti-inflammation of S. maritima root extract was evaluated by inhibition of NO production of LPS-induced macrophages. TPC and TFC were 37.39±0.46 mg of GAE/g and 67.41±0.27 mg of QE/g, respectively. Extract was strongly inhibited DPPH radical and lipid peroxidation (IC_{50} = 0.22 ±0.2 and 2.75±0.5 mg/ml). However, *S. maritima* root extract was poorly inhibited NO radical (IC_{50} > 1,000 mg/ml). *S. maritima* root extract was reduced NO production from LPS-induced macrophage cells (20.5±2.8%). There was 0.6 time of anti-inflammation activity from triamcinolone acetonide at same concentration (0.1 mg/ml). Ethanol extract of *S. maritima* root was contained flavonoids as most of bioactive compounds and possessed preferable in vitro antioxidant and anti-inflammation properties.

Keywords: antioxidant, anti-inflammation, seablite root, *Suaeda maritima*, traditional medicine.

INTRODUCTION

Halophytes are habitat about one percentage of land plants, which can be survive and breed under salinity of wetland as extreme condition [1]. Morphological, physiological and biochemical adaptations of halophytes both at a cellular and molecular level in the whole plant or in particular tissues are reduced harmful effects from excessive salinity [1]. There have been applied in ethno-pharmaceutical uses due to nutritional and biological benefits.

Access this article online	
Quick Response Code:	Website: www.pnrjournal.com
	DOI: 10.47750/pnr.2022.13.03.015

Address for correspondence: Kanittada Thongkao,
Faculty of Science and Technology, Suan Sunandha Rajabhat University, Bangkok- 10300, Thailand

E-mail address: kanittada.th@ssru.ac.th

How to cite this article: Kanittada Thongkao, Narin Kakatum, Yuttana Sudjaroen, ANTIOXIDANT AND ANTI-INFLAMMATION ACTIVITIES OF ETHANOL EXTRACT FROM SEABLITE (*SUAEDA MARITIMA*) ROOT, J Pharm Negative Results 2022;13(3):101-104.

Active metabolites are often occurrence in halophytes, which is protected oxidative damage from salty and UV-irradiate environment such as phenolic compounds and/or some vitamins with antioxidant properties [2]. Thus, halophytes are food sources with health promoting properties by high nutritive value plus antioxidant metabolites [3]. Seablite or *Suaeda maritima* (L.) Dumort, a salt marsh plant growing in mangrove forest widely distributed on the landward margin of mangrove habitats of Thai Gulf and Southern of Thailand [4, 5]. Young leaves can be edible both fresh or cooked vegetable, which are quite salty taste. Local Thai cuisine has different preparing processes for reduce salty taste and providing various menu including traditional *S. maritima* salad, *S. maritima* curry with crabs, or scalded *S. maritima* with chili paste [5]. There is also use as animal feeding [6]. Leaf and stem extracts from *S. maritima* had possessed antioxidant properties assayed with total antioxidant capacity, DPPH radical scavenging activity, metal chelating, ferric reducing power and nitric oxide scavenging activity; and phenolic and ascorbic acid contents had contained [7]. Phytochemicals of leaf and stem extracts had contained higher amount of carbohydrates, protein, tannins, alkaloids and flavonoids [7]. Hepatoprotective effect with antioxidant property of ethanol extract S. maritima has been also report in animal model [8]. Thai traditional medicine is using *S. maritima* root for bone enrichment, hair loss prevention, abscess healing, allergic symptom relieves and treatment of skin diseases. However, biological properties of *S. maritima* root are very sparsely reported. In this study was attempt to evaluate antioxidant and anti-inflammation properties besides presence of phytochemicals in *S. maritima* root. Finding is providing bioefficiency of *S. maritima* root for its pharmaceutical applications.

Materials and methods

Plant Collection and Preparation

S. maritima were collected from salt pan area, which located nearby College of Allied Health Sciences, Suan Sunandha Rajabhat University, Samut Songkhram, Thailand. Botanical characteristics of herbs were authenticated [4] by senior botanist. *S. maritima* root (1 kg) was washed, cutting in small pieces, dried in an oven at 45°C and grinding. Ground of *S. maritima* root is macerated with 95% ethanol (3 days) and concentrated under by evaporator under vacuum.

Evaluation of Bioactive Compounds

Total phenolic content (TPC) of S. maritima root extract was determined by Folin-Ciocalteu method. Colorimetric absorbance of reaction mixture was measured at 760 nm. Range of gallic acid concentrations were used as standard curve. TPC was represented as mg of gallic acid equivalent (GAE) per g [9, 10, 11]. Total flavonoid content (TFC) of S. maritima root extract was determined by aluminium chloride colorimetric method. Colorimetric absorbance of reaction mixture was measured at 420 nm. Range of quercetin concentrations were used as standard curve. Flavonoid content was represented mg of quercetin equivalent (QE) per g [9, 12].

In Vitro Antioxidant Assays

DPPH radical scavenging activity: 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical in present of *S. maritima* root extract (0.01-1 mg/ml) was reduced DPPH absorbance. DPPH radical (6×10^{-5} M) and ascorbic acid were used as negative and positive controls, respectively [7, 8, 13]. Lipid peroxidation inhibition (LPI) activity: lipid peroxidation in present of *S. maritima* root extract (0.001-10 mg/ml) was reduced absorbance of ferric iron-thiocyanate complex. α-tocopherol was used as positive control [7, 8, 14]. NO radical scavenging activity: nitric oxide (NO) radical in present of *S. maritima* root extract (0.01-1 mg/ml) was reduced absorbance of Griess reagent reaction. Ascorbic acid was used as positive control [7, 8, 15]. Absorbance reaction mixture of all assays were monitored by micro-titer plate reader at maximum absorbance wavelength (λmax). Results was calculated from triplicate measurements and interpreted as 50% inhibitory concentration (IC_{50}) of *S. maritima* root extract.

In Vitro Anti-Inflammatory Test

Monitoring of NO inhibition produced from lipopolysaccharide (LPS)-induced macrophage cells was used to evaluate anti-inflammatory activity of ethanol extract from S. maritima root. Briefly, mouse macrophage cell (RAW264.7) was obtained from Thailand Institute of Scientific and Technological Research (TISTR), Pathum Thani, Thailand. Cells were seeded in high-glucose Dulbecco's modified Eagle's medium (containing 10% fetal bovine serum, 100 U/mL penicillin, and 100 µg/mL streptomycin) and incubated at 37 °C (containing 5% CO_2) in humidified incubator. After overnight culture in a 24-well plate (1×10^5 cells/well, 500 µL medium/well), the cells were pre-treated with herbal mixture for 1 h and lipopolysaccharide (LPS) for an additional 24 h, the culture supernatant from each well was collected and used to measure NO production. 50 µl of culture medium and 100 µl of Griess reagents were added and incubated in a 96-well plate at room temperature (10 min). Absorbance of reaction mixture was then measured using a microplate reader [15, 16, 17]. Triamcinolone acetonide (0.1 mg/ml) was used as positive control. Results was calculated from triplicate measurements and interpreted as 50% inhibitory concentration. This study had exempted by Suan Sunandha Rajabhat University Ethic Committees, Thailand (COE.1-099/2022).

Statistical analysis

Bioactive compounds, antioxidant and anti-inflammatory activities of S. maritima root were represented by descriptive

statistics and compared with controls.

Results

After ethanol extraction, characteristics of *S. maritima* root extract was semi-solid and had dark brown color. TPC and TFC of S. maritima root extract were 37.39±0.46 mg of GAE/g and 67.41±0.27 mg of QE/g, respectively. Extract was strongly inhibited DPPH radical and lipid peroxidation (IC_{50} = 0.22 ±0.2 and 2.75±0.5 mg/ml) and can comparable with ascorbic acid and α-tocopherol, respectively. However, *S. maritima* root extract was poorly inhibited NO radical (IC_{50} > 1,000 mg/ml) (Table 1). *S. maritima* root extract was reduced NO production from LPS-induced macrophage cells (20.5±2.8%). There was 0.6 time of anti-inflammation activity from triamcinolone acetonide at same concentration (0.1 mg/ml). Therefore, anti-inflammation activity of extract was unevaluated in higher concentration due to color interference on absorbance measurement (Table 2).

Table 1: Bioactive compounds and antioxidant activity of *S. maritima* root extract

Sample / Assay	TPC	TFC	DPPH	NO	LPI
Ethanol extract	188.34±2.96	19.24±1.59	0.06±0.00	1.30±0.2	502.4±21.97
α-tocopherol	-	-	-	-	0.39±0.10
Ascorbic acid	-	-	0.03±0.01	0.25±0.05	-

Table 2: Anti-inflammation activity and cytotoxicity of *S. maritima* root extract a

Sample / Conc. (mg/ml)	Inhibition of NO production (%)					
	0.0001	0.001	0.01	0.1	1.0	10.0
Ethanol extract	ND	17.6±2.8	19.7±2.3	20.5±2.8	9.8±1.3 [b]	ND
Triamcinolone acetonide	27.8±4.9	34.3±3.3	31.6±4.9	30.0±3.3	36.1±3.0	ND

a Inhibition of NO production from lipopolysaccharide (LPS)-induced macrophage represented as percentage; b Sample color was interfered NO measurement; ND = Not determined

Discussion

There was first report on antioxidant and anti-inflammation of *S. maritima* root rather than leaf, which had been already reported [5-8]. We had confirmed antioxidant and anti-inflammation activities that may corresponded to Thai medicinal used such as, abscess healing, allergic symptom relieves and dermatitis treatment. In case of skin infection, *S. maritima* root is need to evaluate for anti-microbial activity against pathogenic bacteria including *Staphylococcus aureus* and *Cutibacterium acnes*, and pathogenic yeast including *Candida albicans* and *Malassezia furfur*. The use of *S. maritima* root on bone enrichment and hair loss prevention is still scientific unsupported. Some of halophytes were reported in uses of traditional medicine and confirmed on relating biological properties. Roots of the halophyte *Polygonum maritimum* L. are used in traditional medicine in Asia, Europe and Africa to treat inflammation and diabetes and scientific data on antioxidant, anti-inflammation and anti-diabetic properties had supported [18, 19]. *Limonium* spices are important halophytes containing a variety of bioactive compounds of medicinal interest. *Limonium* root extracts from some species including *L. bellidifolium*, *L. globuliferum*, *L. gmelinii*, *L. lilacinum*, *L. sinuatum* and *L. iconicum* were potent antioxidant properties along with various assays including DPPH and ABTS radical scavenging and metal chelating properties [20].

The *Limonium* root extracts had found as active inhibitors of key enzymes such as, acetylcholinesterase, butyrylcholinesterase, amylase, tyrosinase and glucosidase, which are play roles in various diseases [20]. In addition, L. gmelinii root extract had protective responded to oxidative stress in brain and can improve motor functions both in vitro and in vivo model [21]. Hence, we are suggested that further studies will be conduct on evaluation of key enzyme inhibition by S. maritima root and its bioactive compound characterizations.

Conclusion

Ethanol extract of *S. maritima* root was contained flavonoids as most of bioactive compounds and possessed preferable antioxidant and anti-inflammation properties.

Acknowledgements

We are sincerely thanking to Suan Sunandha Rajabhat University, Bangkok, Thailand for research funding and technical supports. We are thankful to Asst. Prof. Dr. Pimporn Thongmuang, College of Allied Health Science, Samut Songkhram Education Canter, Suan Sunandha Rajabhat University, Samut Songkhram, Thailand as senior pharmacist was provided herbal identification.

Conflict of interest

There is no conflict of interest in this study

References

1. Rozema J, Flowers T. Crops for a salinized world. Science 2008;322(5907):1478-1480.
2. Ventura Y, Sagi M. Halophyte crop cultivation: the case for Salicornia and Sarcocornia. Environ Exp Bot 2013;92:144-153.
3. Bernal J, Mendiola JA, Ibáñez E, Cifuentes A. Advanced analysis of

nutraceuticals. J Pharm Biomed Anal 2011;55(4):758-774.
4. Tanaka T. Tanaka's Cyclopedia of Edible Plants of the World. Keigaku Publishing Co.: Tokyo, 1976.
5. Pornpitakdamrong A, Sudjaroen Y. Seablite (Suaeda maritima) product for cooking, Samut Songkram province, Thailand. Food Nutri Sci 2014;5: 850-856.
6. Bandaranayake WM. Bioactivities, bioactive compounds and chemical constituents of mangrove plants. Wetland Ecol & Manage 2002;10:421-452.
7. Patra KJ, Dhal KN, Thatoi NH. In vitro bioactivity and phytochemical screening of Suaeda maritima (Dumort): A mangrove associate from Bhitarkanika, India. Asian Pac J of Trop Med 2011;4,:727-734.
8. Ravikumar S, Gnanadesigan M, Inbaneson JS, Kalaiarasi A. Hepatoprotective and antioxidant properties of Suaeda maritima (L.) Dumort ethanolic extract on concanavalin-A induced hepatotoxicity in rats. Indian J Exp Biol 2011;49:455-460.
9. Thongkao K, Kakatum N, Thongmuang P, Sudjaroen Y. In vitro anti-inflammatory and anti-microbial activities of ethanol extract from Cnidoscolus chayamansa (Mc Vaugh) leaves. Int J Pharm Res 2020; 12(4): 4947-4950.
10. Singlenton VL, Orthoffer R, Raventos RM. Analysis of total phenols and other oxidation substrates and antioxidants by means of Folin-Ciocalteu reagent. Methods Enzymol 1999; 299: 152-178.
11. Biju J, Sulaiman CT, Satheesh G, Reddy VRK. Total phenolics and flavonoids in selected medicinal plants from Kerala. Int J Pharm Pharm Sci 2014, 6, 406-408.
12. Kelly EH, Anthony RT, Dennis JB. Flavonoid antioxidants: chemistry, metabolism and structure-activity relationships. J Nutri Biochem 2002;13:572-584.
13. Yen CG, Duh DP. Scavenging effect of methanolic extracts of peanut hulls on free-radical and active-oxygen species. J Agric Food Chem 1994;42:629-632.
14. Manosroi A, Kumguan K, Chankhampan C, Manosroi W, Manosroi J. Nanoscale gelatinase A (MMP-2) inhibition on human skin fibroblasts of Longkong (Lansium domesticum correa) leaf extracts for anti-aging. J Nanosci Nanotech 2012;12:7187-7197.
15. Aktas N, Genc Y, Gozcelioglu B, Konuklugil B, Harput US. Radical scavenging effect of different marine sponges from Mediterranean coasts. Rec Nat Prod 2013;7(2):96-104.
16. Cao Y, Chen J, Ren G, Zhang Y, Tan X, Yang L. Punicalagin prevents inflammation in LPS-induced RAW264.7 macrophages by inhibiting FoxO3a/autophagy signaling pathway. Nutrients 2019; 11(11): 2794.
17. Torres-Rodríguez ML, García-Chávez E, Berhow M, de Mejia EG. Anti-inflammatory and anti-oxidant effect of Calea urticifolia lyophilized aqueous extract on lipopolysaccharide-stimulated RAW 264.7 macrophages. J Ethnopharmacol 2016;188:266-274.
18. Rodrigues MJ, Custódio L, Lopes A, Oliveira M, Neng NR, Nogueira JM, et al. Unlocking the in vitro anti-inflammatory and antidiabetic potential of Polygonum maritimum. Pharm Biol 2017;55(1):1348-1357.
19. Mahmoudi M, Guemri I, Boughalleb F, Zaidi S, Hessini K, Abdellaoui R. Minerals, fatty acids, and antioxidant activity in sea knotgrass (Polygonum maritimum L.) seeds. Nat Prod Res 2021:1-5.
20. Senizza B, Zhang L, Rocchetti G, Zengin G, Ak G, Yıldıztugay E, et al. Metabolomic profiling and biological properties of six Limonium species: novel perspectives for nutraceutical purposes. Food Funct 2021;12(8):3443-3454.
21. Nurkenov T, Tsoy A, Olzhayev F, Abzhanova E, Turgambayeva A, Zhussupova A, et al. Plant extract of Limonium gmelinii attenuates oxidative responses in neurons, astrocytes, and cerebral endothelial cells in vitro and improves motor functions of rats after middle cerebral artery occlusion. Antioxidants (Basel). 2021;10(11):1814.