International Journal of Pharmaceutical Chemistry
ISSN: 2249-734X (Online)
Journal DOI: 10.7439/ijpc
CODEN: IJPCH3 (American Chemical Society)
Research Article

Antioxidant activity of the simple ascidian *Phallusia nigra* of Thoothukudi Coast

Devendiran Shanmuga Priya*, Henry Kohila Subathra Christy, Sankaran Sankaravadivu and Chandran Stella Packiam

Department of Chemistry, A.P.C. Mahalaxmi College for Women, Tuticorin 628 002, Tamilnadu, India

Abstract

Ascidians are a rich source of bioactive secondary metabolites. *Phallusia nigra* is a simple ascidian belonging to the family Ascidiidae found in plenty throughout the year along the Thoothukudi coast of India. Antioxidant activity was performed by DPPH (1, 1diphenyl-2-picryl hydrazyl) radical scavenging method for different extracts of *Phallusia nigra* which showed that the alcoholic extract of the animal on higher concentration possess better antioxidant potential when compared to that of the standard ascorbic acid. They exhibited strong antioxidant DPPH radical scavenging activity with absorbance of 0.1984 and 0.0553 for ascorbic acid and ethanolic extract respectively. The strongest antioxidant activity of ethanol extract may be due to the presence of flavonoids and phenols.

Keywords: *Phallusia nigra*, DPPH, Flavonoids..

1. Introduction

The ocean is considered to be an untapped source for many things including potential drugs. Ascidians are marine sedentary organisms. In some countries, mainly those of the Far East and certain parts of the Mediterranean, ascidians are eaten by man and are sufficiently important to merit an entry in the F.A.O yearbook of Fishery statistics [1]. *Microcosmus sulcatus*, *Styela plicata* and *Polycarpa pomaria* are taken as food in the Mediterranean [2]. *Halocynthia roretzi* in Japan, is even cultured in the North of Honsyu [3] for human consumption and *Pyura chilensis* is popular in South America [4] as a food source. Margalino and Destefano found that the flesh of *Microcosmus sulcatus* is almost as digestible as whole egg and the protein content higher [5]. Such is their abundance in some localities that ascidians have been considered as a possible source of cellulose, vanadium, protein and other chemicals [6,7]. Though the nutritive value of many natural resources of the sea has gained much attention, the importance of ascidians as food or drug has been totally neglected in our country. *Phallusia nigra* is a simple ascidian belonging to the family Ascidiidae occurring as the major component of fouling community on the hull of ships, piers, pilings, harbour installations and materials used for aquaculture operations in the Tuticorin Port Area. Previous studies show that the animal possesses antipyretic [8], analgesic anaesthetic [9], anti-inflammatory [10], wound healing [11] and antimicrobial activities [12, 13]. No reports are available on the antioxidant activity of different extracts of the simple ascidian *Phallusia nigra*. Hence, the present study focuses on the chemical investigation of antioxidant activity of the different extract of *Phallusia nigra* by DPPH method.

The DPPH assay method is based on the reduction of DPPH, a stable free radical [14]. The free radical DPPH with an odd electron gives a maximum absorption at 517 nm (purple colour). When Antioxidants react with DPPH, which is a stable free radical becomes paired off in the presence of a hydrogen donor (e.g., a free radical scavenging antioxidant) and is reduced to the DPPH and as consequence the absorbance's decreased from the DPPH [15]. Radical to the DPPH-H form, results in decolourization (yellow colour) with respect to the number of electrons captured [16]. More the decolourization more is the reducing ability. This test has been the most accepted model for evaluating the free radical scavenging activity of any new drug [17].

* Correspondence Info
Dr. D. Shanmuga Priya
Department of Chemistry,
A.P.C. Mahalaxmi College for Women,
Tuticorin 628 002, Tamilnadu, India
E mail: priyasaravanan12apcm@gmail.com

2. Materials and methods

2.1 *Collection and identification*

Phallusia nigra (Fig.1) was collected from Green Gate area (8˚48'N and 78˚11'E) of Thoothukudi Port, Tamil Nadu by SCUBA diving and identified using Key to identification of Indian ascidians [18]. A voucher specimen (AS 2083) was deposited in the Museum of the Department of Zoology, A.P.C. Mahalaxmi College for Women, Tuticorin 628002, Tamilnadu, India.

Fig. 1: *Phallusia nigra* Sav.

2.2 Preparation of extract

The whole animal was dried in shade and homogenized to get a coarse powder. The powder was successively extracted with various solvents such as petroleum ether (40^0-60^0 C), benzene, chloroform, ethanol, methanol and water.

2.3 DPPH Radical Scavenging Assay

The antioxidant activity of the animal extracts was measured on the basis of the scavenging activity of the stable 1, 1- diphenyl 2-picrylhydrazyl (DPPH) free radical according to the method described by Brand-Williams *et al* [19] with slight modifications. 1ml of 0.1mM DPPH solution in methanol was mixed with 1ml of animal extract solution of varying concentrations (50, 100, 150 and 200 µg/ml). Corresponding blank sample were prepared and L-Ascorbic acid was used as reference standard. Mixture of 1ml methanol and 1ml DPPH solution was used as control. The reaction was carried out in triplicate and the decrease in absorbance was measured at 517nm after 30 minutes in dark using UV-Vis spectrophotometer (UV-VIS Shimadzu). The inhibition % was calculated using the following formula, Inhibition % = Ac-As/Ac×100

Where Ac is the absorbance of the control

As is the absorbance of the sample [20]

3. Results and Discussion

The results of antioxidant activity of the different extracts at varying concentrations are presented in the Table 1.The ethanolic extract of the animal showed the significant antioxidant potential when compared to that of the standard ascorbic acid by DPPH scavenging assay method.

Table 1: Absorbance of different extract of *Phallusia nigra* at varying concentrations

Concentration (µg/ml)	Petroleum ether	Benzene	Chloroform	Ethanol	Methanol	Water	Standard Ascorbic acid
50	0.4944	0.5255	0.2124	0.1998	0.1139	0.1122	0.3126
100	0.4876	0.4879	0.2118	0.0921	0.1059	0.1080	0.2989
150	0.3922	0.4225	0.1989	0.0726	0.0976	0.0767	0.2126
200	0.2345	0.4100	0.0976	0.0553	0.0645	0.0621	0.1984

Absorbance of control at 517 nm 0.3846

Radical scavenging method for different extracts of *Phallusia nigra* showed that the chloroform, ethanol, methanol and aqueous extract of the animal on higher concentration possess better antioxidant potential when compared to that of the standard ascorbic acid. They exhibited strong antioxidant DPPH radical scavenging activity with absorbance of 0.1984 and 0.0553 for ascorbic acid and ethanolic extract respectively. Generally, the antioxidant properties of these extracts were found to be concentration dependent. Based on the results obtained, highly significant antioxidant potential was observed in the ethanol, methanol and aqueous extracts which are more polar in DPPH assay [21]. A preliminary chemical screening of the ethanolic extract of *Phallusia nigra* showed the presence

of alkaloids, terpenoids, flavonoids, glycosides, phenolic compounds and tannins [22]. The strongest antioxidant activity of ethanol extract may be due to the presence of any of these chemical constituents.

4. Conclusion

The results of the present study suggest that the alcoholic and aqueous extract of *Phallusia nigra* illustrates highly significant antidiabetic activity, which may be due to the presence of flavonoids and phenols.

Acknowledgement

The authors express their sincere gratitude to University Grants Commission, New Delhi for Financial assistance and our Secretary Tmt. C. Subbulakshmi and our Principal Dr. R. Vasuki for providing facilities to carry out the work.

References

[1] Food and Agriculture Organisation. Catches and landings. Yearbook of Fishery statistics. No.18; 1964.
[2] Harant H,. Les Tuniciers comestibies *Atti del 11 congresso Inter. Nazionvale d' Igiene di Medicina Mediterranca Palermo* 1951; 1-3
[3] Tokioka T,. Ascidians of Sagami Bay: Iwanami Bhoten, Tokyo, 1953.
[4] Van Name, WG. The North and South American ascidians: *Bull. Am. Mus. Nat. Hist*. 1945; 84: 1-476.
[5] Margalino GA, Destefano. M. Contributo alla conoscenza della digeribilita delle Ascidie Eduli. *Thalassia jonica*.1960; 3: 69-82. .
[6] Elori D, Komarovsky R. On the possible use of the fouling ascidian *Ciona intestinalis* as a source of vanadium, cellulose and other products. *Proc. tech. pap. gen. fish. Mediterr* 1951; 6: 261-267.
[7] Hebant-Joder AM, Etude sur les ciones du basin do Thau. *Revue. Trav. Inst. Pech. Marit* 1965; 29(4): 413-420.
[8] Gopalakrishnan S, Meenakshi VK, Shanmugapriya D. Antipyretic and Analgesic activity of *Phallusia nigra* Savigny, 1816. *Annals of Biological Research* 2011; 20, 2(4): 192-196.
[9] Gopalakrishnan S, Meenakshi VK, Shanmugapriya D, Anaesthetic activity of *Phallusia nigra* Savigny. *Annals of Biological Research* 2012; 3(4): 1863-1865.
[10] Gopalakrishnan S, Meenakshi VK, Shanmugapriya D, Anti-Inflammatory activity of Simple Ascidian, *Phallusia nigra* Savigny. *International Journal of Pharmaceutical sciences Review and Research*.2013; 22(2): 162-167.
[11] Gopalakrishnan S, Meenakshi VK, Shanmugapriya D. Wound healing activity of the methanolic extract of *Phallusia nigra* Savigny. *International Journal of Chemical and Pharmaceutical Sciences* 2013:45-51.
[12] Gopalakrishnan S, Meenakshi VK, Shanmugapriya D, Antimicrobial activity of the methanolic extract of *Phallusia nigra* Savigny. *Journal of Natural Product and Plant Resources* 2012; 2(5): 579-583.
[13] Shanmuga priya D, Kohila Subathra Christy H, S.Sankaravadivu. Antimicrobial activity of a simple ascidian, *Phallusia nigra*. *World journal of pharmacy and Pharmaceutical sciences*. 2015;
[14] Warrier, PK, Nambier, VPK, Raman Kutty C, Indian medicinal plants- A compendium of 500 species. Orient longman Ltd, Madras, 1994; Vol-I: 95-97.
[15] Harborne, J.B, Phytochemical methods- A guide to modern techniques of plant analysis, 3rd Edn, Springer (India) Pvt. Ltd, New delhi,1998: 5-32.
[16] Ghosh, M.N, Fundamentals of Experimental Pharmacology, 2nd Edn., Scientific Book Agency, Calcutta,1998: 174-179.
[17] Wagner, H, Bladet, S, et.al, Plant Drug Analysis-A TLC Atlas, 1st Edn, Springer verlag Berlin, Heidel berg, New York, 1996: 195-214.
[18] Meenakshi, V.K. (1997) Biology of a few chosen ascidians. Ph. D Thesis, Manonmaniam Sundaranar University, Tirunelveli, 157 -173.
[19] Brand-williams W, Cuvelier ME and Berset C.Use of free radical method to evaluate antioxidant activity .*Lebensmittel Wissenschaft and Technologie* 1995; 28(1): 25-30.
[20] Achola, KJ, *et al*, Bronchodilating and ut'erine activities of *Ageratum conyzoides* extract. *Pharmaceutical Biology* 1998; 36(2): 93-96.
[21] Zakaria ZA, Rofiee MS, The LK, Salleh MZ, Sulaiman MR, Somchit MN. *Bauhinia purpurea* leaves extracts exhibited *in vitro* antiproliferative and antioxidant activities. *African journal of Biotechnology* 2011; 10(1): 65-74.
[22] Gopalakrishnan S, Meenakshi VK, Shanmugapriya D, Pharmacological and preliminary *phallusia nigra* Sav ., *Global Journal of pharmacology* 2013; 7: 39-44.