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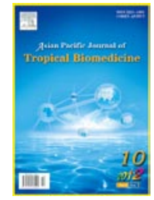

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Studies on the seasonal variations in the proximate composition of ascidians from the Palk Bay, Southeast coast of India

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ABSTRACT

Objective: To investigate the seasonal fluctuations of the proximate composition of the ascidians muscle. **Methods:** The moisture content was estimated by drying 1 g of fresh tissue at a constant temperature at 105 ℃ for 24 h.The loss of weight was taken as moisture content. The total protein was estimated using the Biuret method. The total carbohydrate in dried sample was estimated spectrophotometrically following the phenol- sulphuric acid method. The lipid in the dried sample tissue was gravimetrically estimated following the chloroform-methanol mixture method. Ash content was determined gravimetrically by incinerating 1 g dried sample in muffle furnace at about 550 ℃ for 6 h and results are expressed in percentage. **Results:** It was found very difficult to compare the monthly variations, as all the ten species, exhibited wide fluctuations in their proximate compositions. For the sake of convenience, average seasonal values were calculated by summing the monthly values. **Conclusions:** The proximate composition of the 10 commonly available ascidians showed high nutritive value and hence these groups especially solitary ascidians can be recommended for human consumption in terms of pickles, soup, curry and others after ensuring the safety of consumers.

1. Introduction

Ocean has potent bioactive compounds isolated from marine organisms which are currently used as food. A large proportion of natural compounds have been extracted from marine invertebrates[1]. More than 70% of our planet's surface is covered by oceans, and life on Earth has its origin in the sea. In certain marine ecosystems, such as coral reefs or the deep sea floor, experts estimate that the biological diversity is even higher than in tropical rain forests. Many marine invertebrates such as sponges, soft corals or shell-fewer mollusks are soft-bodied animals that are either sessile or slow moving and usually lack physical defenses like protective shells or spines, thus necessitating chemical defense mechanisms such as the ability to synthesize toxic and/or deterrent compounds[2]. The number of natural products isolated from marine organisms increases rapidly and now exceeds with hundreds of new compounds being discovered every year[3]. An important aspect of biodiversity conservation and sustainability of marine resources is the mitigation of non-indigenous species (NIS). To-date, there have been no reported extinctions of native marine species caused by exotic invaders[4]. Nonetheless, they pose a serious threat to the sustainability of aquaculture concerns[5] and they can alter the structure and composition of benthic communities[6], thereby threatening global marine biodiversity and resource sustainability. With only 16% of the worlds' marine eco regions free from NIS[7], invasive species are challenging pre- and post-border bio security strategies, and threatening biodiversity and ecosystem services around the world[8,9]. However, invasions can also provide insight into community ecology dynamics[10]; competitive interactions[11] and the resilience of native assemblages as NIS make their way into new ecosystems.

The combined availability of high-throughput molecular techniques and analyses of the resulting data based on explicit evolutionary models has caused a recent surge in the number of studies seeking to use genetic patterns to assess invasion pathways and the evolution of invasiveness[12,13]. Recent reviews of these molecular studies show that a wide array of taxa, geographic scales, and molecular markers have been covered over the past decade[14,15], with a range of results reported across both

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aquatic and terrestrial NIS[13]. A comprehensive analysis of molecular studies showed that conventional expectations of bottlenecks and reduced genetic variability for introduced populations do not always hold true for aquatic NIS, with only around 37% of studies reporting a significant loss of genetic variation in introduced populations[15]. However, most of these studies have sampled populations many years after the initial introductions and global spread. Marine taxa, also primarily sampled many years after introduction, exhibit a particularly wide range of molecular patterns. For example, high genetic diversity has been observed for the mitochondrial DNA gene cytochrome oxidase I (COI) in native and introduced populations of the ascidian *Microcosmus squamiger*. The first introduction of this species was recorded in1983 and it appears that extensive sharing of haplotypes has since occurred among populations[16].

The need for discovery of new and novel antibiotics is imperative because evidence suggests that development and spreads of resistance to any new antimicrobial agents is inevitable. Advances in molecular methods have shown that some symbiotic micro organisms are responsible for the production of secondary metabolites that serve as chemical defense for their hosts. Antibacterial activities have previously reported from extracts of some ascidians[17]. Marine organisms such as ascidians, sponges and soft corals, containing symbiotic microorganisms are a rich source of bioactive compounds. The ascidians are a rich source of compounds with cytotoxic proprieties; these marine natural products can be used for the discovery and development of novel chemotherapeutic agents[18]. Ascidians are conspicuous and important members of shallow benthic communities[19]. Therefore, information on their growth and biochemical and energetic composition[20] are important for evaluating their nutritional value and also in modeling the flow of materials and energy within marine benthos. Moreover, low nutrient levels could make some ascidians as unattractive prey while chemical defenses (acids, heavy metals, secondary metabolites *etc*) may make tissues distasteful to predators[21] and inhibit the settlement of fouling microorganisms[22]. To meet out the demand, marine products are playing an excellent role as they have high protein content. Marine invertebrates are being widely used as food around the world[23]. Seafood products are currently in high demand as they are considered healthy and nutritional[24]. Ascidians are also gaining importance as a source of animal protein. This creates the need for biochemical analysis of available native ascidians. The amount of carbohydrate, protein, lipid and minerals such phosphorous and calcium contents in few ascidians are previously reported by various authors[25].

2. Materials and methods

2.1. Collection and preparation of sample

Ten species of ascidians *Herdmania pallid* (*H. pallida*), *Microcosmus exasperates* (*M. exasperates*), *Microcosmus squamiger* (*M. squamiger*), *Microcosmus helleri* (*M. helleri*), *Didemnum psammathode* (*D. psammathode*), *Didemnum moseleyi* (*D. moseleyi*), *Lissoclinum fragile* (*L. fragile*), *Polyclinum madrasensis* (*P. madrasensis*), *Polyclinum indicum* (*P. indicum*), and *Polyclinum constellatum* (*P. constellatum*) were collected regularly during four seasons (Pre monsoon, Monsoon, Post monsoon and summer) in Palk Bay region, Southeast coast of India, from April 2010 to March 2011. The collected samples were immediately brought to laboratory in ice-cold condition. Species were identified as per the standard protocol[26-28]. In the laboratory, the collected ascidians were thoroughly washed with sea water to remove all the epiphytes and rinsed with distilled water to remove the salt and extraneous materials. Ascidians muscle samples were air dried for few days. The dried muscles were made in to powder for biochemical analysis such as protein, carbohydrate and lipids.

2.2. Moisture

To calculate the moisture content, 1 g of fresh tissue was oven dried at a constant temperature of 105 ℃ for 24 h[26]. The loss of weight was taken as moisture content.

2.3. Protein

The total protein was estimated using the Biuret method of Raymont *et al*[29]. The total carbohydrate in dried sample was estimated spectrophotometrically following the phenol-sulphuric acid method of Dubois *et al*[30].

2.4. Lipid

The lipid in the dried sample tissue was gravimetrically estimated following the chloroform-methanol mixture method of Tarjuelo *et al*[31].

2.5. Ash

Ash content was determined gravimetrically by incinerating 1 g dried sample in muffle furnace at about 550 ℃ for 6 h[32] and results are expressed in percentage.

2.6. Statistical analysis

Analysis of variance was performed to detect significant differences between the means of the aggregate and ascidians. Using the program Sigma-Stat, a Two-way ANOVA was calculated for multiple comparisons.

3. Results

The proximate composition of muscle tissue varied from species to species. In the present study, protein, carbohydrate and lipid contents of each ascidian species varied considerably. Seasonal variations in levels of proximate compositions in all the species were studied in this society. Each value in graph represents the mean of triplicate samples for each species. It was found very