Green mussel (*Perna viridis* L.) as healthy food, and as a nutraceutical supplement

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Ocean harbors a large biodiversity of marine fauna and flora with about 66,535 species of mollusks, among which 15,000 species of bivalves have been reported to occur. Bivalve mollusks are aquatic mollusks (Phylum: *Mollusca*; Class: *Bivalvia*), and comprise major marine fishery resources, mussels being a prominent member. Taxonomically, mussels belong to the *Mytilidue* family, the genus *Perna* being one of the most important from a commercial point of view.

Now-a-days mussels are consumed as seafood, especially in Belgium, the Netherlands, and France (called *Moules marinières*). Many countries practice mussel farming, and according to recent estimates, the global production has increased by 22.8 times from 0.07 million tonnes in 1950 to 1.58 million tones in 2003 (Food and Agriculture Organization, 2003). World wide, *Perna viridis* is known to be native to the coastal marine waters of the Indo-Pacific region, extending from the Arabian Gulf to the southern province of Guangdong and Fujian in China and southern Japan. In India, two species of marine mussels (green mussel - *Perna viridis* and brown mussel - *P. indica*) support a traditional sustenance fishery in Kerala and Goa. *P. viridis* occurs from the intertidal zone to a depth of 15 m. India has a mussel fishery based on wild stocks of green mussels. **India has risen to one among the top 10 mussel producing nations in Asia, with an annual production of 10,060 tonnes, worth $US1.79 million (Kripa and Mohamed, 2008).** In India, the technology for farming the green mussel, *Perna viridis*, was developed during 1970s and was subsequently tested for feasibility at various locations along the country's southeast and southwest coasts by Central Marine Fisheries Research Institute (Kuriakose, 1980). The Malabar Coast of Kerala is popularly called as the 'Mussel fishery zone of India', which accounts for the bulk of the mussel production in the country. However, in recent years, the increasing demand for mussels has enabled farmers in north and central Kerala to adopt commercial-scale technologies for mussel farming. Subsistence cropping of green mussels is common in those countries where they occur naturally. The consumption of bivalve molluscs in India, particularly in south Malabar area has

increased in the recent years in response to the higher availability under wild and cultured conditions. Proteins, lipids, minerals and fatty acids, contribute to

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Mytilidae is a family of small to large saltwater mussels, marine bivalve molliusks in the order Mytiloida. It is the only family in the order.... the nutritional value and organoleptic characteristics of *P. viridis*. Bivalve culture is recognized as a valuable food resource in many parts of Asia. Throughout the world, bivalve exploitation plays an important role in the national economy of many countries (Asokan et al 2001). Only recently, after the importance of mussel culture in India had been realised, studies on biochemical composition of the green mussel, *P. viridis* began to receive considerable attention.

***P. viridis*: A potential source of nutritional and pharmaceutical components**

The potential health benefits related to marine mussel consumption are due to the presence of inexpensive source of protein with high biological value and polyunsaturated fatty acids (PUFAs). In addition, this food group contributes to the intake of essential minerals and trace metals and certain vitamins with valuable pharmaceutical and biomedical potential. This is unlike most terrestrial organisms, which are rich in n-6 PUFA. In recent years, the n-3 PUFA from bivalve mollusks provide an almost unlimited variety of long chain PUFA, especially n-3 PUFA with beneficial roles in human health and reducing potential thrombosis and for the alleviation of inflammatory conditions viz., rheumatoid and osteoarthritis and asthma. While saturated and monounsaturated fatty acids may be synthesized in the body, PUFAs cannot be synthesized *de-novo* due to the lack of essential enzymes required to synthesize PUFAs in adequate levels from precursor fatty acids, and must be obtained from the diet (Chakraborty et al. 2010c, 2010d). These PUFas found only in fish and seafoods, have extremely beneficial properties for, in particular, the prevention of inflammation and CHAs. The extract from *Perna viridis* has earlier been found to be active against influenza, herpes and hepatitis viral strains (Patent No. RU 2043109). There are reports of Indian green mussel (*Perna viridis)* as a source of anti-HIV activity in vitro (Mitra et al. United States Patent 6770302 Appl. No: 10/112081: 03/29/2002). There is evidence that bivalve mollusks are useful in the treatment of inflammatory joint diseases (Miller et al., 1993). There are reports of dried flesh of the New Zealand mussel *Perna canaliculus* possessig PUFAs with anti-inflammatory and anti-prostaglandin effects (Zwar, 1994). In addition to the reported anti-inflammatory effects of n-3 and n-6, these compounds have been shown to have immune enhancing benefits, improving resistance to infection, inhibiting platelet function and reducing thrombosis. The integration with the nutritional composition and anti-inflammatory capacity provided a holistic assessment of the overall biological significance of *P. viridis* under different growth conditions (wild and farmed). The culture activity of this species in India was mainly confined to southern Malabar in 1980s. At present, culturing of green mussels has extended from Goa in the north to Alleppy in the south because of transplantation of mussel seeds. The coastal community of Malabar is involved in fishing and bivalve culture of *P. viridis*, to include consumption and sale of *P. viridis*. Screening

this important species grown under wild and cultured conditions for anti-inflammatory activities and nutritional composition will be valuable information of these highly consumed bivalve mollusks, *P. viridis*, in this location.

Biochemical composition of *P. viridis* samples from Kozhikode area of Malabar

Green mussel (*P. viridis*) samples were collected from southwestern Coast of India (Kozhikode, Elathur (Lat :11°54'11.6"N;75° 12'.21.8"E), located about 10 km north of Kozhikode city and is bounded by Arabian Sea on the west and Korapuzha River (Elathur River). Bivalve molluscs provide an inexpensive source of protein with high biological value. Maricultured green mussel exhibited higher protein content (141-236 mg/100 g) than that of wild (72-183 mg/100g) samples (Table 1). The wild *P. viridis* registered higher lipid content than wild samples. Significantly, wild *P. viridis* collected during winter exhibited higher cholesterol content (96.6 mg/100g) than those collected under farmed conditions (65.1 mg/100g) (Table 1). Since mollusks are known to have a limited capacity for sterol synthesis, the presence of cholesterol detected in their tissues may be ascribed to their exogenous origin, and is as an indication of the complex metabolic transformation undergone by exogenous sterols. Minerals are nutrients that are conserved by the body and play significant role in metabolism in human body. Minerals like Zn, Mn and Cu, which are known as essential elements, were found to be abundantly available either in maricultured and wild samples. In general, no significant differences were apparent between wild and cultured *P. viridis* samples excepting Fe, which was found to be significantly more in wild *P.viridis* samples (10.19 mg/100g). The levels of α-tocopherol, a vitamin with anti-oxidant properties, in *P. viridis* were low, and showed insignificant fluctuations among wild and cultured samples ($p > 0.05$) (0.12-0.13 IU). All *trans* retinol underwent no significant fluctuations, though a higher content of this vitamin was apparent in wild samples (8.2 IU) than in cultured (5.3 IU). Phylloquinone (K1) registered higher values in maricultured samples (2.2 µg/100g) than in wild (1.26 µg/100g), whereas vitamin C exhibited a reverse trend (9.89 vs 12.74 IU, respectively). Cholecalciferol (D_3) was found to be significantly higher in cultured *P. viridis* (412 IU), than in wild samples (352 IU).

Amino acid and fatty acid composition of *P. viridis*

Amino acids are one of the central chemicals needed by the body as building blocks of protein, and as intermediates in protein metabolism. Amino acids are of two general types, viz., essential and non-essential. Eight amino acids are generally regarded as essential for humans: phenylalanine, valine, threonine, tryptophan, isoleucine, methionine, leucine, and lysine. Additionally, cysteine (or sulphur-containing amino acids), tyrosine (or aromatic amino acids), histidine and arginine are essentially required by infants and growing children. In addition, the amino acids arginine, cysteine,

Table 1 Nutritional compositions of green mussels (*Perna viridis*) harvested under wild and cultured conditions from Kozhikode during the month of May-June.

	Wild	Cultured
Proximate compositions (% wet weight)		
Lipid (%)	2±0.13	1.7±0.16
Cholesterol (mg/100g)	96.6±0.78	65.1±1.32
Protein (mg/100g)	103.9±0.67	220.3±1.83
Vitamins (IU)		
Retinol (A)/IU	8.2±0.15	5.31±0.09
Cholecalciferol (D2)/IU	352.5±4.39	412.8±2.56
Tocopherol (E)/IU	0.13±0.01	0.12±0.01
Phylloquinone (K1) (μg/100g)	1.26±0.02	2.2±0.149
Ascorbic acid (C)/IU	12.74±0.15	9.89±0.09
Amino acids	**Amino acid (as mg/100g)**	
Σ Essential amino acids/E	1462.9	1631.8
ΣNon- essential amino acids/NE)	1320	2204.1
E/NE	1.107	0.7403
Fatty acid (as weight %)		
Σ Saturated fatty acids	30.28	34.41
Σ Monounsaturated fatty acids	27.29	27.26
Polyunsaturated fatty acids (% TFA)		
20:5n3 (EPA)	12.84 ± 1.04	12.68 ± 1.15
22:6n3 (DHA)	9.87 ± 0.96	9.60 ± 0.12
Σ PUFA	35.00	31.51

glycine, glutamine, histidine, proline, serine, and tyrosine are considered to be conditionally essential, signifying that they are not normally required in the diet, but must be supplied exogenously to specific populations that do not synthesize it in adequate amounts. A total of seventeen amino acids were identified and quantified in the samples of *P. viridis*. The ratio of essential (E, g/100 g protein)/nonessential (NE, g/100 g protein) amino acid was observed to be 0.7 (for cultured) to 1.1 (for wild). The results obtained from this study showed that *P. viridis* have well-balanced and high-quality protein source in the respect of E/NE ratio in all seasons. Fatty acids are very important biochemical indicators of bivalves contributing to their nutritional quality. Kozhikode samples have significantly lower SFA (30-34%), and no significant differences are apparent between wild and cultured samples. SFAs are used for energy storage, and therefore, their concentration increases during periods of enhanced feeding activity. No significant differences in MUFA content were apparent in wild and cultured samples (27.7% and 27.3%). Polyunsaturated fatty acids (PUFAs) are considered as single most important nutritional indicator dictating the quality of *P. viridis*. Kozhikode samples registered a significantly higher PUFA content (32-35%) in either growth

conditions. No significant differences were apparent in wild and cultured samples. The PUFA composition of marine mollusks is considered to be characterized by predominance of n-3 PUFAs, particularly eicosapentaenoic acid, EPA (20:5*n*-3, 12.7-12.8%) and docosahexaenoic acid, DHA (22:6*n*-3, 9.6-9.9%), which constitute usually near a half of total fatty acids. EPA and DHA are important *n*-3 PUFAs required essentially by human beings. No significant differences in the content of these important fatty acids between the samples harvested under wild and cultured conditions clearly indicated the optimum conditions maintained in mussel culture which resembles to that of wild conditions. These n-3 fatty acids, which are abundantly available in *P. viridis*, are an important source of anti-inflammatory activity.

Anti-inflammatory properties of *P. viridis*

Inflammation is the pathophysiological response of mammalian tissues to a variety of hostile agents, and the complex events and mediators involved in inflammation can induce, maintain and aggravate many disorders. Inflammation not only plays a role in the inflammatory diseases but also in the progression of arthritis, diabetes, and cancer. Current treatment options are mostly symptomatic and include Non Steroidal Anti-Inflammatory Drugs (NSAIDs) viz., aspirin, indomethacin, acemetacin, etodolac, tolmetin, ketorolac, oxaprozin, sulindac, etc and cyclooxygenase-2 COX_{II} inhibitors (rofecoxib) for pain relief but fail to block the progression of the disease. Moreover the NSAIDs were reported to have have undesirable side effects including damaging GI tract through both local and systemic effects. The reported side-effects and contra-indications of current AI drugs have lead to investigations into natural products for safer and more effective alternatives. Natural product-based anti-inflammatory agents with a transcriptional mode of action, good efficacy, and lower risk of side effects offer promising treatment and prevention of inflammation-related conditions. Many promising lead compounds have been reported from marine sources having anti-inflammatory activity. Mussels are commercially valuable species, easy to cultivate or collect in coastal areas, and are reported to cure arthritic inflammation. Gastroprotective and anti-inflammatory properties of green lipped mussel (*Perna canaliculus*) preparation was reported (Cobb and Ernst, 2006). A product containing anti-inflammatory principles (Seatone®) is being imported from New Zealand by Perma Healthcare, a Delhi-based dealer, and the same has major share in Indian market. However the major drawback associated with this product is high cost. Though Indian coastline is bestowed with rich sources of bivalve mollusks, there is no single indigenous product available in the Indian market as a treatment of inflammatory diseases and joint pain. A product (Cadalmin™GMe) containing 100% natural marine bioactive anti-inflammatory ingredients from green mussel *Perna viridis* to combat chronic joint pain, arthritis/ inflammatory diseases has been developed. The active principles in Cadalmin™GMe isolated from *P. viridis* were competitively inhibit inflammatory $COX_{I,II}$ and LOX_V in an inflammation and oxidative stress reaction, resulting in decreased production of inflammatory prostaglandins and leukotrienes, and its activity was found to be superior to the synthetic non steroidal anti-inflammatory drugs available in the market (Chakraborty et al., 2010a-b). It was found that the active principles isolated from P. viridis, and concentrated in the product registered higher COX_{II} and LOX_V inhibition (70-75%) than aspirin and indomethacin (55-66%, 5mg/ml). In vivo animal model studies revealed that the active principles effectively suppressed (64 and 77%, 2-4h) the edema produced by the histamine, which indicates

that they exhibit its anti-inflammatory action by means of either inhibiting the synthesis, release or action of anti-inflammatory mediators (Chakraborty et al., 2011a, b). Cadalmin GMe has techniques to identify the active components and to concentrate them to have higher activity; it has sustained activity, no toxicity, less leachability due to the unique biochemical engineering methodologies adopted to develop the product. This product is an effective green alternative to synthetic non steroidal anti-inflammatory drugs (*viz.*, aspirin containing drugs having undesirable side effects). Cadalmin™ GMe is designed to find a unique way to prevent the degradation by air, moisture, heat and light and to maximize the activity. The product is free from deleterious *trans* fatty acids, free radicals/ free radical adducts, and low molecular weight carbonyl compounds. This product is available as capsules and packaged in food grade polypropylene bottles. Cadalmin™ GMe is an indigenous product, and is highly cost effective with that of the imported products available in the market. The product know-how has been patented, and efforts are underway to commercialize this product.

Suggested readings

Asokan, P. K., V. P. Vipinkumar, K. K. Appukuttan, V. G. Surendranathan, and M. P. Sivadasan. 2001. Mussel culture in backwaters of Kasargod district, Kerala. *Marine Fisheries Information Service T & E Series 169*:9--11.

Chakraborty K., Vijayan, KK, Vijayagopal, P., Deepu J., Chakkalakal S, and Rao Syda (2010a). A process to concentrate anti-inflammatory principles from green mussel *Perna viridis* L. (Indian Patent IP 2065/CHE/2010)

Chakraborty K., Vijayan, KK, Vijayagopal, P., Deepu J., Chakkalakal S, and Rao Syda (2010b). A product containing anntiinflammatory principles from green mussel *Perna viridis* L. and a process thereof (Indian Patent IP 2066/CHE/2010)

Cobb, C.S & Ernst, E. Systematic review of a marine nutriceutical supplement in clinical trials for arthritis: the effectiveness of the New Zealand green-lipped mussel *Perna canaliculus*. *Clinical Rheumatology* (2006) 25: pp.275-284.

FAO (Food and Agriculture Organization). 2003. *FAO fisheries department. Fishery information data and statistics unit.* FISHSTAT Plus, universal software for fishery statistical time series version 2.30. FAO, Rome, Italy.

Kripa, V., and Mohamed, K.S. (2008). Green Mussel, *Perna viridis*, Farming in Kerala, India - Technology Diffusion Process and Socioeconomic Impacts. Journal of the World Aquaculture Society. 39: 612-624.

Kuriakose PS 1980. Mussels (Mytilidae: Genus *Perna*) of the Indian coast. *Bulletin Central Marine Fisheries Research Institute* **29**:1-5.

Miller, T.E., Dodd, J., Ormrod, D.J., Geddes, R., 1993. Anti-inflammatory activity of glycogen extracted from Perna canaliculus (NZ green-lipped mussel). Inflamm. Res. 38, C139–C142.

Zwar D. The magic mussel - arthritis another way? 2nd edn. Cairns, Australia: Ideas Unlimited. 1994:108.