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Green Tea Extract: A Natural Alternative to Sodium Metabisulfite for Melanosis Inhibition in Black Tiger Shrimp

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Abstract

Aquaculture, with a history spanning thousands of years, has seen significant growth since the 1960s. India plays a substantial role in seafood exports, with shrimp being a significant contributor to its total seafood exports and holding a leading share in key international markets. Among the various shrimp species, *Penaeus monodon* is also known as giant tiger shrimp or black tiger shrimp. A common post-harvest challenge in shrimp is melanosis, or black spot, which affects the sensory qualities, reduces shelf life and diminishes commercial value. This melanosis is a metabolic reaction caused by polyphenol oxidase (PPO) enzymes, which produce dark pigments called melanin. While chemical substances like sodium metabisulfite (SMBS) is often used to inhibit PPO activity, concerns regarding their safety have led to a search for natural alternatives. Research suggests that natural plant extracts, such as green tea, can effectively prevent black spots in shrimp during storage. Green tea is known for its antioxidant, antimicrobial, and antimutagenic properties. The study evaluated the effectiveness of green tea extract in controlling black spot formation in freshly harvested black tiger shrimp. Green tea extract proved to be an effective inhibitor of melanosis in black tiger shrimp, presenting a viable natural alternative to sodium metabisulfite (SMBS). Also found that higher concentrations (3%) of both treatments were more effective in preventing black spot formation. Furthermore, cold storage (<4°C) notably enhanced the preservation effects across all treatments.

Keywords: Shrimp, Melanosis, Natural Preservatives, Green Tea, SMBS

Introduction

The history of aquaculture can be dated back thousands of years and evidence can be found in ancient scriptures like Kautilya's Arthasastra and many more (FAO, 1999) [29]. However, the primary boom occurred in the 1960s.

130.9 million tons, or USD 312.8 billion, of aquaculture were produced worldwide in 2022, accounting for 59% of all fisheries and aquaculture production worldwide. Of the aquatic animals raised for food, 62.6 percent came from inland aquaculture, and 37.4 percent came from marine and coastal aquaculture. In aquatic animal production, aquaculture overtook capture fisheries for the first time with 94.4 million tonnes, accounting for 51 percent of global production and a record 57 percent of production intended for human consumption. (FAO 2024) [30]. In 2022-23, shrimp accounted for $5.6 billion of India's total seafood exports, which amounted to $8.09 billion (₹64,000 crore). In 2022-23, India held a dominant 40% share of seafood exports to the U.S. market, significantly surpassing competitors such as Thailand, China, Vietnam, and Ecuador (MPEDA 2024) [31].

The major species of shrimp cultured worldwide are *Litopenaeus vannamei* (white leg shrimp), *Penaeus monodon* (black tiger shrimp), *Penaeus semisulcatus* (green tiger/ flower shrimp), *Litopenaeus stylirostris* (western blue shrimp), *Fenneropenaeus indicus* (Chinese white shrimp), *Marsupenaeus japonicus* (kuruma shrimp), and *Fenneropenaeus merguensis* (banana shrimp). *Penaeus monodon* promised fast growth rate and suitability to farming condition among penaeid species. The giant tiger shrimp and black tiger shrimp are local names for *Penaeus monodon*. This shrimp has a wide natural range, extending from the east coast of Africa to the Indian Ocean, South Asia (http://www.cport.net/product/view/black-tiger-prawns).

Black Tiger shrimp exports increased by 24.91%, 11.33%, 8.28% in terms of quantity, ₹ value, and US$ terms (from US$ 321.23 million to US$ 347.84 million) respectively (MPEDA, 2024) [31].

The four districts of Gujarat namely Valsad, Navsari, Surat, and Bharuch are the major contributors for shrimp production having 95% of state shrimp farmer's population with the potential area of 69,583.91ha. (Jahnvi Tandel, 2018).

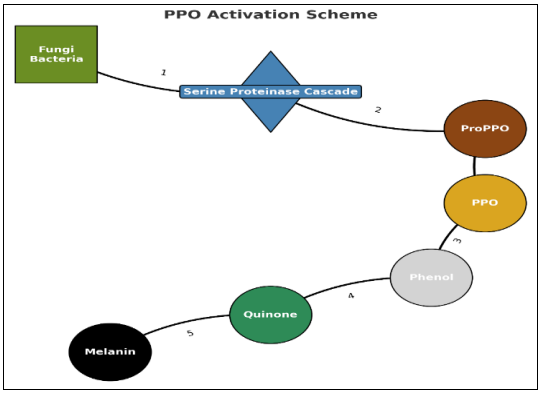
In crustaceans, melanosis, also referred to as "black spot," is a common problem during post-harvest storage. The production of dark pigments known as melanin, which mostly accumulate beneath the cephalothorax's carapace, is the result of metabolic reaction induced by polyphenol oxidases (PPO) enzymes (Nirmal & Benjakul, 2011a) [20]. Melanosis affects the sensory qualities of crustaceans, reducing their quality, shelf life, and ultimately their commercial value, even though it doesn't directly affect to consumers (Gomez-Guillen, Martínez-Alvarez, Llamas, & Montero, 2005 [12]; Kim, Marshall, & Wei, 2000).

Fig 1: Melanosis in shrimp

It is linked to more visual concerns than health hazards. One of the major issues facing the industry is melanosis, which affects the sensory qualities of crustaceans and quality of product, which minimizes consumer acceptability and causes financial loss (A.A. Gonçalves, A.R.M. de oliveira, 2015) [13].

Pathway

Polyphenoloxidase (PPO), also known as tyrosinase, catechol oxidase, and o-diphenol oxidase, is an enzyme found in shrimp and other crustaceans that catalyzes the browning reaction known as blackspot. It plays a critical role in the arthropod proPO system, being the key enzyme in melanin synthesis. PPO remains active until the shrimp are frozen or cooked, with activation triggered by microbial compounds like carbohydrates and lipopolysaccharides. There are two types of PPO activity: monophenol oxidase, which hydroxylates monohydroxyphenols, and o-diphenol oxidase, which oxidizes o-dihydroxyphenols to o-quinones (Zamorano *et al*., 2009) [27].



Source: A.A. Goncalves, A.R.M. de oliveira, 2016

Fig 2: PPO activation scheme

Prevention

A common practice is to use commercial products containing other substances (s), in addition to sodium metabisulfite which contribute to retard melanosis. To stop PPO activity, chemical substances have also been used. Over time, a number of methods and strategies have been created to stop PPO activity in seafood (Kim *et al*., 2000).

Allergy to sodium metabisulfite is known to induce asthma and contact dermatitis, and these reactions have been widely described in adulthood, nevertheless this kind of allergy is rare in childhood. Stringent regulations on the use of sulfites and increased awareness on food safety have necessitated the search for safe chemicals or natural additives for melanosis inhibition (Vitaliti G, *et al*, 2015) [26].

Research proves that natural plant extracts can prevent shrimp from developing black spots during storage (Jang *et al*., 2003 [15]; Gokoglu and Yerlikaya, 2008; Nirmal and Benjakul, 2011 [19]). Green tea has been reported to possess antioxidant, antimicrobial, antimutagenic and anticarcinogenic properties (Cabrera, Artacho, & Gimenez, 2006) [5].

Methodology

Extraction of Green Tea (*Camellia sinensis*)

To prepare the green tea extract, dried green tea (*Camellia sinensis*) leaves were first ground into a fine powder to maximize surface area for subsequent extraction using a high-speed blender. The powdered leaves were then passed through a stainless-steel sieve with an 80-mesh size to ensure uniform particle size and remove any coarse material. Distilled water was heated to a temperature range of 80–90 °C, a range known to be effective for extracting bioactive compounds while minimizing degradation. The sieved green tea powder was added to the hot water at a concentration of 25 g/L. The mixture was stirred continuously for a period of 15 to 30 minutes to enhance the extraction of polyphenols and other beneficial compounds. After stirring, the resulting mixture was filtered through Whatman No. 1 filter paper to remove solid residues and obtain a clear extract. The prepared extract was then collected and stored for further use in shrimp treatment and melanosis inhibition studies.

Treatment with extract and SMBS

Freshly harvested shrimp were used to evaluate the effectiveness of green tea extract and sodium metabisulfite (SMBS) in controlling black spot formation. The extract and SMBS were dissolved in water chilled to <4 °C at two different concentrations: 2%, and 3%. A control group, treated solely with chilled water (at <4 °C) without any additives, was also maintained to serve as a baseline for melanosis development. The shrimp were immersed in the prepared solutions to undergo a chill kill treatment, a process used to quickly lower body temperature and reduce enzymatic activity that contributes to melanosis. After treatment, the shrimp samples were divided and stored under two different conditions: one set at refrigeration temperature <4 °C and another at room temperature. The samples were stored for a maximum of 18 hours and visual observations were made every 3 hours to monitor the appearance and progression of black spots on the shrimp. The effectiveness of each concentration, as well as the impact of different storage temperatures, was evaluated based on melanosis development. All observations were carefully documented to determine which treatment best inhibited black spot

formation and maintained the visual and physical quality of the shrimp during short term storage.

Results

The control group (no treatment) was included to assess the natural progression of melanosis under both temperature conditions. The results were recorded and analysed to determine the effectiveness of each extract at each dose under both temperature conditions.

The chemical preservative SMBS is frequently used due to its ability to inhibit melanosis in shrimp. It functions as an antioxidant by reducing melanin activity, thus preventing black spots.

Green tea extract, abundant in catechins and polyphenols, has demonstrated various health benefits, such as antioxidant and anti-inflammatory effects.

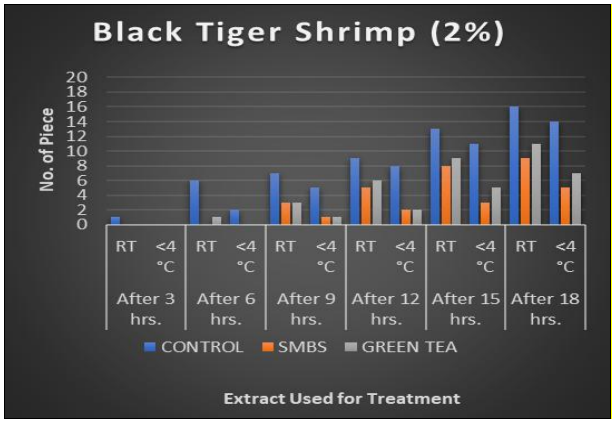


Fig 3: Black Tiger Shrimp treated with 2% dose

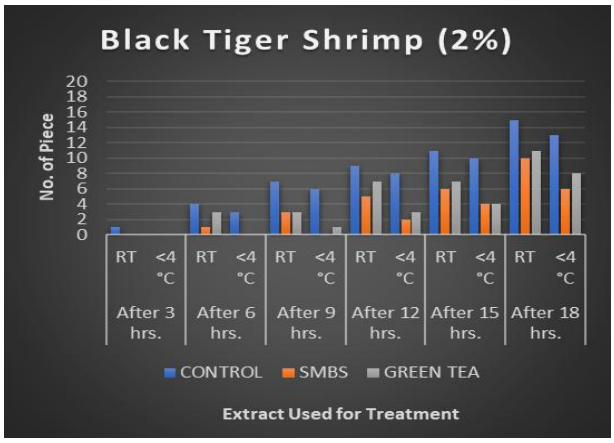


Fig 4: Black Tiger Shrimp treated with 2% dose

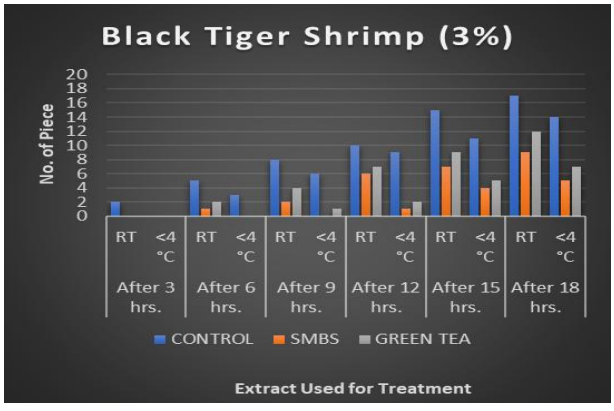


Fig 5: Black Tiger Shrimp treated with 3% dose

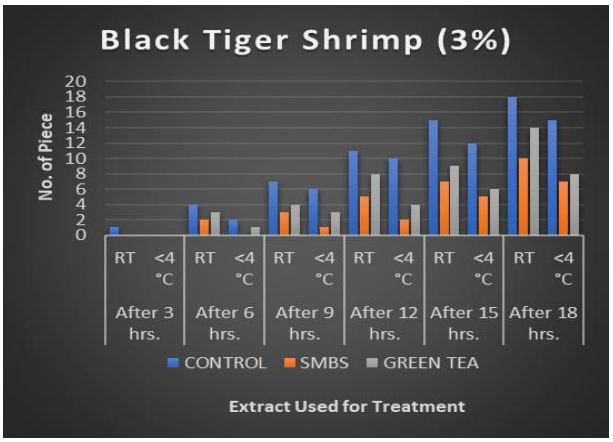


Fig 6: Black Tiger Shrimp treated with 3% dose

In both the 2% and 3% concentrations, the control group consistently shows the highest number of spoiled pieces over time, indicating its least effective preservation. As time progresses, the number of spoiled pieces in the control group increases significantly, particularly at room temperature (RT).

The SMBS (Sodium Metabisulfite) and green tea treatments generally demonstrate better preservation compared to the control. In most instances and at both concentrations, these treatments result in a lower number of spoiled pieces. The preservation effect is more pronounced at <4°C compared to RT for all treatments, highlighting the critical role of cold storage in shrimp preservation.

Comparing SMBS and green tea, SMBS often appears to be slightly more effective in reducing the number of spoiled pieces, especially at later time points and higher spoilage levels. However, green tea also shows a notable preservative effect, particularly in cold conditions. The 3% concentration of the treatments seems to provide a slightly more effective preservation than the 2% concentration, as evidenced by generally lower spoilage numbers at the 3% concentration compared to 2% at comparable time points and temperatures, especially for SMBS and green tea.

Discussion

Green tea leaves (*Camellia sinensis* L.) contain significant amounts of polyphenols, including catechins, flavonols, and phenolic acids, which are known for their strong antioxidant properties. Chen and Wan (1994) [6] found that tea polyphenols exhibit higher antioxidant activity than synthetic antioxidants, such as BHA (butylated hydroxyanisole) and BHT (butylated hydroxytoluene). In the present study, we found that the green tea extract was rich in catechins and demonstrated a moderate inhibitory effect on melanosis compared to SMBS in black tiger shrimp.

Conclusion

Green tea extract effectively inhibited melanosis in black tiger shrimp, offering a natural alternative to sodium metabisulfite (SMBS). Green tea, rich in catechins and polyphenols known for strong antioxidant properties, significantly reduced black spot formation compared to controls. While SMBS showed slightly superior efficacy, particularly at later stages, green tea extract still provided a moderate, significant reduction in melanosis. Higher concentrations (3%) of both treatments were more effective.

Cold storage (<4°C) further enhanced preservation for all treatments. This highlights green tea's potential as a safe, natural anti-melanosis agent for the aquaculture industry, given concerns about synthetic preservatives like SMBS.

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