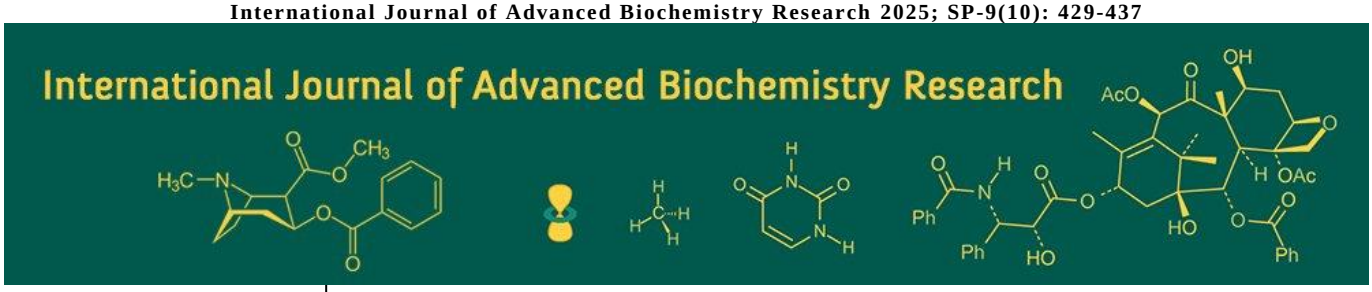

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Combined effects of green tea extract dip treatment and vacuum packaging on the shelf life extension of refrigerated stored streaked seer fish, *Scomberomorus lineolatus*, (Cuvier, 1829) steak

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Abstract
The present research was carried out to investigate the combined effects of Green Tea Extract (GTE) and Vacuum packaging (VP) on the quality and shelf-life extension of streaked seer fish (*Scomberomorus lineolatus*) steak, under refrigerated storage conditions. Treating the fish steak with GTE T1 (1%), T2 (2%) and T3 (3%) and VP, storing them at (4±1 °C) for 30 days, and assessing their quality biochemical, physical, microbiological, characteristics at five days interval. Biochemical analyses revealed significant changes in pH, TMA-N, TVB-N, and PV levels with storage duration, with the least changes observed in the GTE (2%) & VP treated sample. Microbiological analyses demonstrated lower levels of Total Plate Count (TPC) and Total Psychrotrophic Count in the treated sample. Moreover, the combined treatment of GTE & VP was found to enhance the shelf-life of streaked seer fish steak by inhibiting microbial growth, reducing lipid oxidation. In the conclusion, it is found that GTE (2%) with vacuum packaging is a highly effective preservative based on biochemical, microbiological analyses.

Keywords: Green tea extract, vacuzum packaging, streaked seer fish *Scomberomorus lineolatus*, improved quality, refrigerated storage, Shelf life

Introduction
Seafood, as suggested by nutritionists and health experts, is considered a valuable source of food, nourishment, livelihood and income (Rahmaniya and Sekharan, 2018) [27]. Fish are generally divided into two main categories: finfish, comprising lean and fatty varieties and shellfish which includes crustaceans and mollusks and among the edible crustaceans such as prawns, shrimp, crab and lobster hold considerable importance as valuable sources of nutrients substance for humans (Liu *et al*., 2013) [19]. Fish and fishery products are among the dietary items with a high value of commercial value, high quality protein content, vitamins, minerals and unsaturated fatty acids all of which are good for your health. (Kontominas *et al*., 2021) [15]. Fishery products on the other hand possess a perishable nature due to their elevated water content, abundant presence of small molecules and a neutral pH level, which create an ideal environment for bacterial and biochemical spoilage (Liu *et al*., 2013) [19]. Many dependable preservation techniques have been developed to provide premium quality fishery products with longer shelf lives, including Super chilling (Liu *et al*., 2013) [19], High hydrostatic pressure (HHP) (Roco *et al*., 2018) [29], Modified atmosphere packaging (MAP) and Vacuum Packaging (VP) (Czerwiński *et al*., 2021), Active packaging (AP) (Donget, 2018) Irradiation, and Edible coatings (Licciardello *et al*., 2018) [18].
India exported seafood totalling 13,69,264 metric tonnes (MT) at a value of Rs 57,720.98 crore (US$ 7.46 billion) in the years 2021-2022. The export of seafood during the year increased by 19.12% in terms of quantity, 31.71% in terms of rupee value, and 30.26% in terms of US dollar value. The United States, China, and the European Union import the majority of the seafood from India. With a share of 53.18% in terms of quantity and 75.11% in terms of total USD revenues, frozen fish remained the top export product. There was a remarkable increase of 31.68% in terms of USD value, 31.32% when measured in rupees,

and a 23.35% rise in volume during the given period. In the fiscal year 2021-22, India managed to export 7,28,123 metric tons (MT) of fish, amounting to a total value of USD 5,828.29 million. When it comes to import volumes, the United States stands out as the primary market, importing a substantial 342,572 MT of Indian seafood. Following closely, China takes the second spot with 125,667 MT, while the European Union ranks third with 90,549 MT. Japan follows with imports of 44,683 MT, South East Asia with 38,392 MT, the Middle East with 37,158 MT, and various other countries collectively importing 49,002 MT of Indian seafood (MPEDA, 2022) [22]. The per capita global consumption has shown a notable upward trend, escalating from 9 kg in 1961 to 20.2 kg in 2020, and it is projected to continue this trajectory, reaching an estimated 21.4 kg by the year 2030 (FAO, 2022) [8]. In India, the estimated per capita fish consumption is approximately 5-6 kg for the entire population. However, among the fish-eating population, this consumption is notably higher, typically ranging from 8-9 kg (Salim, 2016) [30].

Among fish the family Scombridae comprises commercially important species that contributes more percentage in fish production. The *Scomberomorus lineolatus*, commonly known as Streaked Spanish mackerel is widely distributed across India, especially along the Maharashtra and Gujarat coasts (Collette *et al*., 2011) [4]. The *Scomberomorus lineolatus* is predominantly a marine water pelagic species but is also found in coastal water. The peak season for fishing this fish is from October to December. It is found off Asian coasts from the west coast of India and Sri Lanka east to Java and does not extend east of Wallace's Line (Collette *et al*., 2011) [4]. Marine fish landings (tonnes) in India 2021 in seer fishes *Scomberomorus lineolatus* is 4 tonnes. The per capita global consumption has shown a notable upward trend, escalating from 9 kg in 1961 to 20.2 kg in 2020, and it is projected to continue this trajectory, reaching an estimated 21.4 kg by the year 2030.Seer fishes, constituting 1.7% of the total marine fish catch in the country, are highly regarded as high-value resources. They are prominently sourced from various regions, with Andhra Pradesh (14.3%) and Tamil Nadu (11.5%) along the east coast, and Gujarat (22.8%), Maharashtra (16.9%), and Kerala (16.1%) along the west coast emerging as the primary contributors to the seer fish catch (Tanna, 2020) [36]. These fishes are commonly marketed as fresh and dried-salted products.

Tea is one of the most widely consumed beverages worldwide and richer in antioxidants compared to other forms of tea. Tea is composed of polyphenols, caffeine, minerals, and trace amounts of vitamins, amino acids, and carbohydrates (Prasanth *et al*., 2019) [26]. According to the European Food Safety Authority (EFSA), 126 mg of catechins are present per 100 mL of green tea. However, according to the Food and Drug Administration (FDA), 71 mg of epigallocatechingallate will be present per 100 mL of green tea. (Rietveld and Wiseman 2003) [28]. Green tea contains polyphenolic compounds having antioxidant and antimicrobial properties that inhibit rancidity in fish and fishery products. Polyunsaturated fatty acids found in seafood are prone to oxidation-induced breakdown. The process making green tea extract is the most affordable, cost effective, and environmentally friendly (Mahapatra, 2022) [20]. Currently, one of the significant challenges in preserving the quality of seafood is preventing fat oxidation. In recent times, researchers have also been harnessing the power of natural antioxidants derived from plants and herbs. This approach is entirely natural and does not raise any health concerns when applied to seafood to maintain quality by preventing fat oxidation (Suyani *et al*., 2020) [35]. Green tea is a type of tea made from the leaves of Camellia sinensis, prepared by steeping or brewing them in hot water or alcohol. "Catechins include epigallocatechin-3-gallate (EGCG) (Leung *et al*., 2001) [17]. Green tea contains polyphenols, including epigallocatechingallate (EGCG), epicatechins, flavanols, and epigallocatechin-3-gallate (EGCG). Among these components, EGCG, which is a type of catechin, plays a crucial role in reducing fat oxidation and thereby helps preserve the quality of seafood to some extent when compared to untreated fish (Sang *et al*., 2011) [32]. The primary objective of this experiment is to preserve the quality of fish using antioxidants found in green tea. This study is conducted to enhance the quality and extend the shelf life of Seer fish steaks by incorporating green tea extract under refrigerated conditions (Patel *et al*., 2021) [34]. In affluent countries, refrigerated meat, fish, and other foodstuffs are frequently displayed using vacuum packaging (Islam *et al*., 2020) [13]. Comparing vacuum packaging to air packaging, the previous type method improves quality and extends shelf life. In the vacuum packaging process, the product is enclosed within a specialized plastic pouch designed with low oxygen permeability properties. The air inside the pouch is then removed, causing the bag to tightly conform to the product, after which it is sealed. This method effectively creates a vacuum environment within the packaging (Adams and Moss 2008) [1].

Considering all of these factors, the current study was with initiated with the following objectives:

A. To study the combined effects of Green tea extract treatment and vacuum packaging on quality attribute test of the Streaked Seer Fish (*Scomberomorus lineolatus*) steak.
B. To evaluate the changes in biochemical, microbiological characteristics of Streaked Seer Fish (*Scomberomorus lineolatus*) steak during refrigerated storage.

Material and Method

Raw Material

Fresh fish steak in experiment 11Kg Streaked Seer Fish (*Scomberomorus lineolatus*) with an average weight and length of about 685- 700 g and 42-48 cm were purchased from Veraval fish landing center (Longitude 20° 54.259 and Latitude 70° 22.439') and transported under iced condition in Styrofoam box with storage temperature range between 0-2 °C to Department of fish processing technology laboratory, College of Fisheries Science, K.U., Veraval. The raw material was thoroughly washed with chilled water to remove the dirt, slime and other foreign particle. The length and weight of the fish sample were taken and later stored according to the size of the material and afterwards beheading, Degutting, removing the fins and dressing fishes further washed with chilled water. Then dressed fish was cut into the steak and again washed with potable chilled water and it was done for further analysis.

Green tea extracts preparation (GTE)

Now, 1% w/v Green tea extract was prepared as follows. One gram of green tea powder was mixed with 100ml of distilled water, and this mixture was placed in a magnetic

stirrer at 30°C- 40°C for 45 minutes. Subsequently, the mixture was removed from the magnetic stirrer. The mixture was then filtered through Whatman filter paper no.1 to eliminate undissolved particles. The filtered solution, with soluble solid content, was applied as Green tea extract (GTE).

Procedure of Experiment

In this experiment, 1% w/v Green tea extract (GTE) was prepared from green tea powder and then dissolved in distilled water with a glass stirring rod for 5 minutes at room temperature. In treatment T0 (Control), the sample was kept without applying any treatment. In T1, the sample was dipped in 1% Green tea extract (w/v) for 10 minutes with vacuum packaging. In T2, the sample was dipped in 2% Green tea extract (w/v) for 10 minutes with vacuum packaging. In T3, the sample was dipped in 3% Green tea extract (w/v) for 10 minutes with vacuum packaging. After the treatments were applied, the samples were packed in 200 gauge plastic bags and stored under refrigerated conditions at 4±1°C. Physical, biochemical, microbial analyses were conducted at five-day intervals during storage at refrigerated temperatures.

Results

Raw Material Characteristics of Fish

The physical characteristics of fresh fish are presented in the fresh fish measured total length 47.18±1.34 cm, Standard length 42.91±1.31 cm weight of fish was 0.75±0.11 Kg (mean±SD) for experiment on an average. The average proximate composition of fresh fish (*Scomberomorus lineolatus*) as raw material presented in Table 1. The raw material of in experiment had the moisture content was about 73.58±0.55 , protein content was 18.59±0.05 , fat content was 3.33±0.04 and ash content was 1.44±0.02 (mean±SD). The fresh fish (*Scomberomorus lineolatus*) were assessed for quality using Biochemical, Physical, Microbiological, Sensory quality parameters and colour characteristics.The Biochemical characteristics for raw material such as pH, TMA-N, TVB-N, and PV was 6.57±0.13, 2.73±0.09 (mg/100g), 5.44±0.05 (mg/100g) and 1.92±0.03 (m.equ./kg of fat) respectively (mean±SD) was observed.The Microbiological characteristics for raw material such as Total plate count (TPC) was 3.21±0.48 log CFU/g (mean±SD) and Total psychrotrophic count was 2.66±0.32 log CFU/g for (mean±SD) was extremely low bacterial load much below than the critical limits indicating absolute freshness of raw material.

Table 1: Raw material characteristics of Streaked Seer Fish (*Scomberomorus lineolatus*) during experiment

A.	**Physical Characteristics**		**Mean±s.d**
	1	Total length (cm)	47.18±1.34
	2	Standard length (cm)	42.91±1.31
	3	Weight of fish (Kg)	0.75±0.11
B.	**Proximate Composition**		**Mean±S.D**
	1	Moisture (%)	73.58±0.55
	2	Crude Protein (%)	18.59±0.05
	3	Crude Fat (%)	3.33±0.04
	4	Total Ash (%)	1.44±0.02
C.	**Biochemical Characterisics**		**Mean±S.D**
	1	pH	6.57±0.13
	2	TVB-N (mg/100g)	5.44±0.05
	3	TMA-N (mg/100g)	2.73±0.09
	4	PV (milli equivalent/kg of fat)	1.92±0.03
E.	**Microbiological Characterisics**		**Mean±S.D**
	1	Total plate count (log cfu/g)	3.21±0.48
	2	E.coli(log cfu/g)	N.D
	3	S.aureus(log cfu/g)	N.D
	4	Total Psychrotrophic counts (log cfu/g)	2.66±0.32

4.2. Experiment For Finding the Individual and Combined Effects of Green Tea Extract and Vacuum Packaging on Fish Quality

4.2.1. Biochemical Characteristics

4.2.1.1. Changes in pH during refrigerated storage

The interface of treatments was also found significant difference during the complete storage period with a CV (%) of 2.61. Changes in pH content for initial pH value were as follows: on day 0, it was 6.37±0.15 for control, 6.34±0.13 for T1, 6.38±0.10 for T2 and 6.36±0.12 pH for T3 and the pH content in the samples increased as the refrigerated storage periods got longer (Table 2 and Figure 1). After 15 days of refrigerated storage at 4±1 °C, the pH value increased to 6.36±0.12 for T0 (control) and 6.77±0.14, 6.57±0.25 and 6.63±0.15 for T1, T2 and T3 samples respectively (mean±SD). After 15 days of storage, the control sample showed the highest increase in pH. The highest pH content was recorded for T0 (control) (7.71±0.13) followed by T1 (7.67±0.23) and T3 (7.43±0.12) and the lowest pH content was recorded for T2 (7.43±0.26) on the 30th day of refrigerated storage at 4±1 °C. Fish were not acceptable when the pH was greater than 7 or 8 (Poulter and Nicolaides, 1985). In the present study control (T0), T1 and crossed the critical limit between 20 days as against T2 and T3 was acceptable up to 20 days.

Table 2: Changes in pH of Streaked Seer fish (*Scomberomorus lineolatus*) during refrigerated storage

Storage Days	**T0(C)**	**T1**	**T2**	**T3**	**DX**
0	6.37±0.15	6.34±0.15	6.38±0.15	6.36±0.12	6.362
5	6.47±0.12	6.43±0.15	6.43±0.15	6.43±0.12	6.433
10	6.67±0.21	6.57±0.15	6.47±0.15	6.53±0.25	6.558
15	6.93±0.15	6.77±0.15	6.57±0.25	6.63±0.15	6.725
20	7.17±0.25	7.11±0.21	6.81±0.26	6.87±0.21	6.983
25	7.37±0.15	7.27±0.12	7.13±0.25	7.21±0.12	7.242
30	7.71±0.13	7.67±0.23	7.43±0.21	7.63±0.12	7.608
TX	6.952	6.881	6.738	6.810	

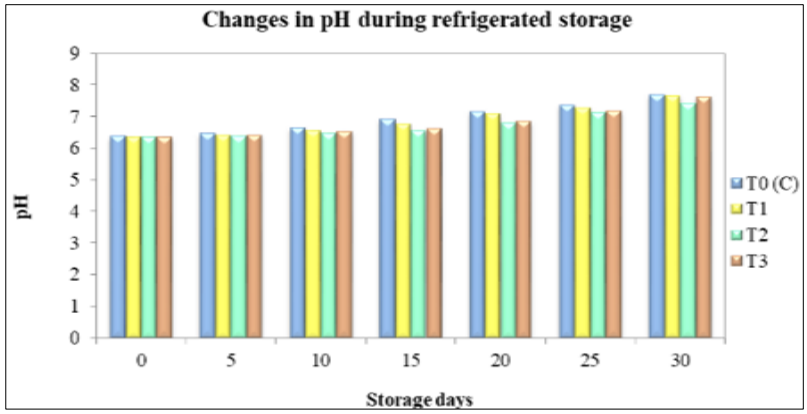

Fig 1: Changes in pH of Streaked Seer fish (*Scomberomorus lineolatus*) during refrigerated storage

4.2.1.2. Changes in Trimethylamine Nitrogen TMA-N during refrigerated storage

The interface of treatments was also found significantly different during the complete storage period with a CV (%) of 2.9. Changes in TMA-N content for initial TMA-N value were as follows: on day 0, it was 2.45±0.6 for control, 2.93±0.17 for T1, 2.92±0.18 for T2 and 2.92±0.47 TMA-N for T3 and the TMA-N content in the samples increased as the refrigerated storage periods got longer (Table 3 and Figure 2). After 15 days of refrigerated storage at 4±1 °C, the TMA-N value increased to 12.31±0.32 for T0 (control) and 11.02±0.3, 9.83±0.21 and 11.86±0.14 for T1, T2 and T3 samples respectively (mean±SD). After 15 days of storage, the control sample showed the highest increase in TMA-N. The highest TMA-N content was recorded for T0 (control) (23.78±0.21) followed by T3 (19.42±0.38) and T1 (19.39±0.38) and the lowest TMA-N content was recorded for T2 (18.41±0.4) on the 30th day of refrigerated storage at 4±1 °C. This is for the fresh fish, the limit was thought to be between 10-15 mg of TMA-N per 100 g of fresh fish (Connell, 1975). In the current study, T0, T1, and T3 exceeded the critical limit within 15 days, whereas T2 remained acceptable for up to 15 days.

Table 3: Changes in TMA-N of Streaked Seer fish (*Scomberomorus lineolatus*) during refrigerated storage

Storage Days	T0(C)	T1	T2	T3	DX
0	2.89±0.61	2.93±0.17	2.91±0.18	2.94±0.47	2.910
5	5.18±0.25	4.74±0.33	3.86±0.23	4.71±0.34	4.622
10	7.53±0.49	7.33±0.41	5.72±0.29	6.63±0.57	6.803
15	12.31±0.32	11.02±0.3	9.83±0.21	11.86±0.14	11.254
20	14.33±0.48	14.15±0.27	12.53±0.61	13.89±0.18	13.725
25	17.14±0.14	15.51±0.57	15.21±0.23	15.61±0.66	15.868
30	23.78±0.21	19.39±0.38	18.41±0.41	19.42±0.38	20.249
TX	11.818	10.724	9.781	10.720	

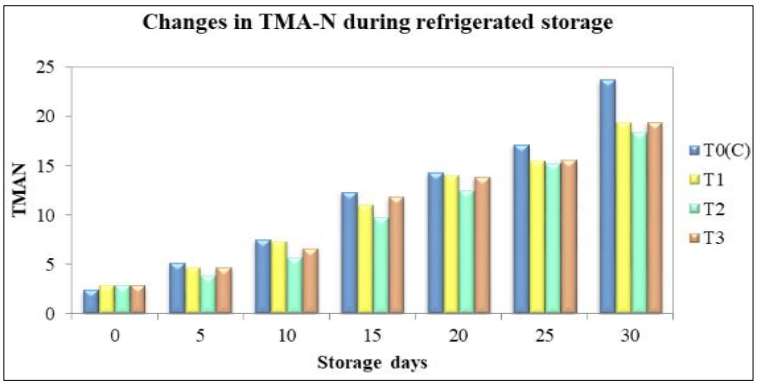

Fig 2: Changes in TMA-N of Streaked Seer fish (*Scomberomorus lineolatus*) during refrigerated storage

4.2.1.3. Changes in Total volatile base Nitrogen TVB-N during refrigerated storage

The interface of treatments was also found significant difference during the complete storage period with a CV (%) of 2.35. Changes in TVB-N content for initial TVB-N value were as follows: on day 0, it was 7.72±0.69 for control, 7.72±0.67 for T1, 7.73±0.66 for T2 and 7.73±0.66 TVB-N for T3 and the TVB-N content in the samples increased as the refrigerated storage periods got longer (Table 4 and Figure 3). After 15 days of refrigerated storage at 4±1 °C, the TVB-N value increased to 30.67±0.62for T0 (control) and 29.24±0.79, 27.15±0.77 and 28.57±0.55 for T1, T2 and T3 samples respectively (mean±SD). After 15 days of storage, the control sample showed the highest increase in TVB-N. The highest TVB-N content was recorded for T0 (control) (44.11±0.52) followed by T1 (43.62±0.55) and T3 (42.77±0.57) and the lowest TVB-N content was recorded for T2 (41.11±0.55) on the 30th day of refrigerated storage at 4±1 °C. Typically, when the level of total volatile basic nitrogen (TVB-N) in fish muscle reaches

the range of 35-40 mg per 100 g, the fish is generally considered to be spoiled. This level is often used as a threshold to assess the freshness and quality of fish (Lakshmanan, 2000). In the current study, T0 and T1 exceeded the critical limit within 15 days, whereas T2 and T3 remained acceptable for up to 15 days.

Table 4: Changes in TVB-N of Streaked Seer fish (*Scomberomorus lineolatus*) during refrigerated storage

Storage Days	T0(C)	T1	T2	T3	DX
0	7.75±0.69	7.72±0.67	7.70±0.66	7.73±0.66	7.725
5	16.21±0.71	14.75±0.77	11.95±0.12	14.68±0.62	14.398
10	25.38±0.59	23.41±0.58	20.43±0.55	22.86±0.76	23.019
15	30.67±0.62	29.24±0.79	27.15±0.77	28.57±0.55	28.907
20	34.31±0.67	33.46±0.59	31.22±0.73	32.81±0.74	32.950
25	41.12±0.79	40.14±0.54	34.58±0.39	39.92±0.57	38.939
30	44.11±0.52	43.62±0.55	41.11±0.55	42.77±0.57	42.902
TX	28.503	27.477	24.881	27.048	

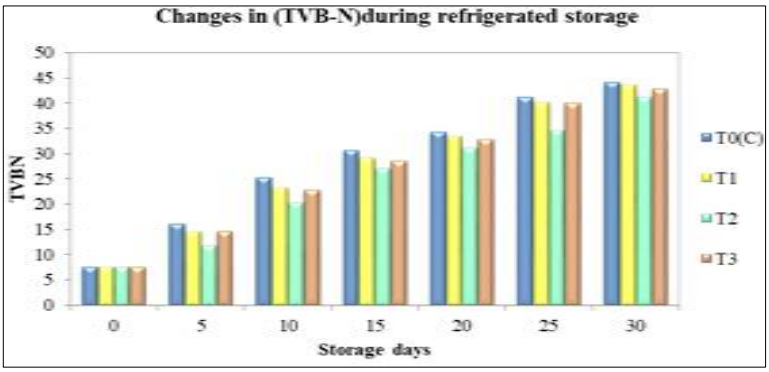

Fig 3: Changes in TVB-N of Streaked Seer fish (*Scomberomorus lineolatus*) during refrigerated storage

4.2.1.4. Changes in Peroxide value PV during refrigerated storage

The interface of treatments was also found significant ($p<0.05$) difference during the complete storage period with CV (%) of 4.29. Changes in PV content for initial PV value were as follows: on day 0, it was 1.89±0.28 for control, 1.9±0.26 for T1, 1.89±0.29 for T2 and 1.89±0.31 PV for T3 and the PV content in the samples increased as the refrigerated storage periods got longer (Table 5 and Figure 4). After 15 days of refrigerated storage at 4±1 °C, the PV value increased to 8.86±0.19 for T0 (control) and 7.72±0.34, 5.81A±0.43 and 6.89A±0.22 for T1, T2 and T3 samples respectively (mean±SD). After 15 days of storage, the control sample showed the highest increase in PV. The highest PV content was recorded for T0 (control) (15.31±0.3) followed by T3 (12.96±0.4) and T1 (14.39±0.38) and the lowest PV content was recorded for T2 (12.41±0.42) on the 30th day of refrigerated storage at 4±1 °C.

Table 5: Changes in PV of Streaked Seer fish (*Scomberomorus lineolatus*) during refrigerated storage

Storage Days	T0(C)	T1	T2	T3	DX
0	1.92±0.28	1.90±0.26	1.89±0.27	1.91±0.31	1.905
5	3.32±0.26	2.28±0.27	2.12±0.13	2.26±0.26	2.495
10	6.40±0.45	5.68±0.31	4.84±0.41	4.95±0.38	5.467
15	7.67±0.23	6.53±0.42	5.44±0.35	6.39±0.21	6.508
20	8.86±0.19	7.72±0.34	5.81±0.43	6.89±0.22	7.321
25	11.25±0.26	10.15±0.23	7.45±0.41	8.76±0.25	9.402
30	14.58±0.38	13.42±0.41	10.38±0.36	12.82±0.26	12.801
TX	7.714	6.811	5.918	6.283	

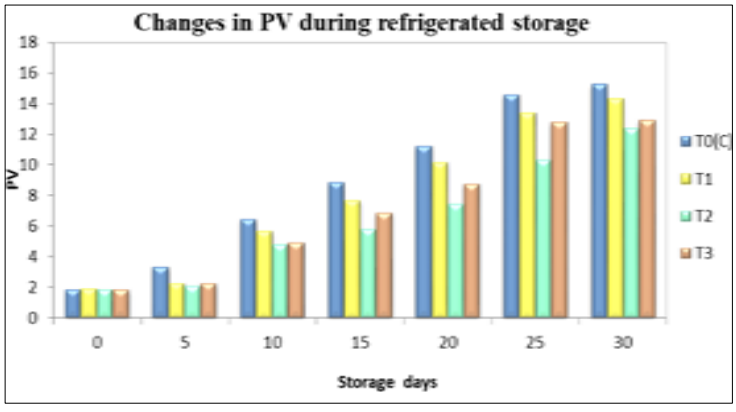

Fig 4: Changes in PV of Streaked Seer fish (*Scomberomorus lineolatus*) during refrigerated storage

Jeon *et al*. (2002) [14] proposed that the permissible peroxide value (PV) limit is 10 m.equ./kg based on their analysis. In the current study, T0 and T1 exceeded the critical limit within 20 days, whereas T2 and T3 remained acceptable for up to 20 days.

4.2.5. Microbiological Characteristics

4.2.5.1. Changes in Total Plate Count (TPC) during refrigerated storage

The interface of treatments was also found a significant ($p<0.05$) difference during the complete storage period with a CV (%) of 3.95. Changes in TPC content for initial TPC value were as follows: on day 0, it was 3.45±0.14 for control, 3.33±0.31for T1, 3.29±0.28for T2 and 3.31±0.3TPC for T3 and the TPC content in the samples increased as the refrigerated storage periods got longer (Table 6 and Figure 5). After 15 days of refrigerated storage at 4±1 °C, the TPC value increased to 6.59A±0.18 for T0 (control) and 6.16A±0.18, 5.32A±0.32and 5.71A±0.31for T1, T2 and T3 samples respectively (mean±SD). After 15 days of storage, the control sample showed the highest increase in TPC. The highest TPC content was recorded for T0 (control) (8.78±0.29) followed by T1 (8.69±0.3) and T3 (8.15±0.16) and the lowest TPC content was recorded for T2 (7.86±0.23) on the 30th day of refrigerated storage at 4±1 °C. The established acceptable threshold for the Total Plate Count (TPC) in fishery products for safe consumption has been set at 5 log CFU/g, as per the guidelines of FSSAI in 2017. In the current study, T0 and T1 exceeded the critical limit within 15 days, whereas T2 and T3 remained acceptable for up to 15 days.

Table 6: Changes in TPC of Streaked Seer fish (*Scomberomorus lineolatus*) during refrigerated storage

Storage Days	T0(C)	T1	T2	T3	DX
0	3.21±0.48	3.16±0.23	3.07±0.28	3.01±0.32	3.112
5	3.45±0.14	3.33±0.31	3.31±0.31	3.62±0.12	3.428
10	4.18±0.21	4.06±0.12	3.93±0.28	4.13±0.24	4.075
15	5.15±0.21	4.45±0.15	4.38±0.22	4.28±0.19	4.565
20	5.62±0.17	5.26±0.26	5.32±0.32	4.68±0.16	5.220
25	6.29±0.18	6.16±0.18	5.71±0.31	5.12±0.14	5.685
30	6.85±0.31	6.59±0.19	6.15±0.35	5.53±0.25	6.280
TX	4.964	4.716	4.553	4.339	

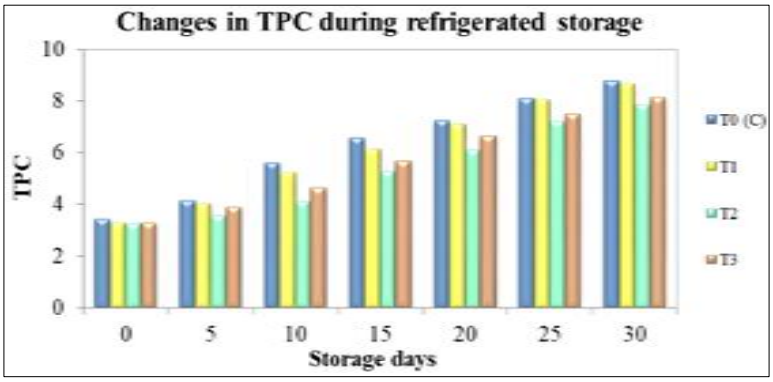

Fig 5: Changes in TPC of Streaked Seer fish (*Scomberomorus lineolatus*) during refrigerated storage

4.2.5.2. Changes in Total Psychrotrophic Counts during refrigerated storage

The interface of treatments was also found a significant ($p<0.05$) difference during the complete storage period with CV (%) of 3.91. Changes in psychrotrophic content for initial psychrotrophic value were as follows: on day 0, it was 2.66±0.32 for control, 2.59±0.38 for T1, 2.44±0.2 for T2 and 2.56±0.27 psychrotrophic for T3 and the psychrotrophic content in the samples increased as the refrigerated storage periods got longer (Table 7 and Figure 6). After 15 days of refrigerated storage at 4±1 °C, the psychrotrophic value increased to 6.38±0.16 for T0 (control) and 6.14±0.24, 5.31±0.33 and 5.96±0.17 for T1, T2 and T3 samples respectively (mean±SD). After 15 days of storage, the control sample showed the highest increase in psychrotrophic. The highest psychrotrophic content was recorded for T0 (control) (9.21±0.27) followed by T1 (9.13±0.14) and T3 (8.78±0.32) and the lowest psychrotrophic content was recorded for T2 (8.13±0.21) on the 30th day of refrigerated storage at 4±1 °C. The permissible threshold for Total psychrotrophic Count in fishery products, for safe consumption, has been determined at 7 log CFU/g, according to the ICMSF in 1986. In the current study, T0, T1, T3 exceeded the critical limit within 20 days, whereas T2 remained acceptable for up to 20 days.

Table 7: Changes in Psychrotrophic of Streaked Seer fish (*Scomberomorus lineolatus*) during refrigerated storage

Storage Days	T0(C)	T1	T2	T3	DX
0	2.66±0.32	2.59±0.38	2.44±0.22	2.56±0.27	2.563
5	4.25±0.31	4.13±0.18	3.43±0.12	3.94±0.18	3.936
10	5.68±0.24	5.25±0.26	4.13±0.27	5.15±0.24	5.053
15	6.38±0.16	6.11±0.24	5.31±0.33	5.96±0.17	5.948
20	7.59±0.21	7.31±0.21	6.46±0.12	7.12±0.18	7.117
25	8.17±0.21	8.12±0.18	7.42±0.19	7.89±0.22	7.900
30	9.21±0.27	9.13±0.14	8.13±0.21	8.78±0.32	8.811
TX	6.276	6.095	5.331	5.912	

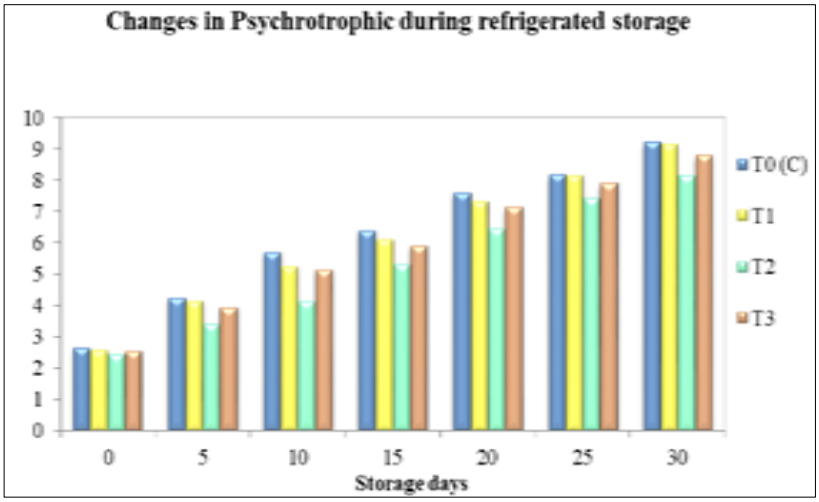

Fig 6: Changes in Psychrotrophic of Streaked Seer fish (*Scomberomorus lineolatus*) during refrigerated storage

Discussion

4.2.1.1. Changes in pH during refrigerated storage

Similar increasing trends were also observed by Pal *et al*. (2017) [23], and Sarah *et al*. (2010) [33], and it was observed that the notable increase in pH was consistently observed in all treated samples throughout the storage period, the reason behind this is attributed to the ability of tea polyphenols to reduce the pH of treated samples by increasing microbial inhibition. Majumder *et al*. (2015) [21] reported similar findings, as they observed a significant ($p<0.05$) impact of storage on the pH values in all formulations containing GTE. These pH values showed a tendency to rise with increased refrigerated storage time. The present study shows the agreement with both reported similar pH values on the first day of refrigerated storage (Hernandez *et al*., 2009; Shinde *et al*., 2015; Sallam *et al*., 2007; and Zambuchini *et al*., 2008) [11, 34, 31, 37] all received the same initial pH value and similar results as the presence study and storage time passing treated samples are found to be better compared to the untreated samples and the reason behind this is that the antibacterial attributes of tea polyphenols because phenolic compounds from tea extract can enhance microbial inhibition, protect fillets from internal protease activity, and ultimately prevent protein degradation and amine production. The initial pH measurement associated with Azizi-Lalabadi *et al*. (2020) [2], and then exhibited a rise over the storage period in the control sample as well as in treatments 1, 2, and 3 respectively. A similar increasing trend was also observed by Bilgin Fıçıcılar *et al*. (2018) [3]. The pH values of brine prepared with green tea extract and bay leaf extract showed no significant differences and the reason behind the increase in pH can be attributed to the formation of nitrogenous compounds, including ammonium and biogenic amines, which arise due to enzymatic and proteolytic activities of psychrophilic bacteria as noted by Lee *et al*. (2009) [16].

4.2.1.2. Changes in Trimethylamine Nitrogen TMA-N during refrigerated storage

Present study related results reported by Hassan and Geethalakshmi (2020) [10] were observed as for GTE stored samples, the TMA-N production remained within acceptable limits until the 16th day of storage and this phenomenon could be linked to either the fast reduction in bacterial population or a decrease in the bacteria's ability to oxidatively deaminate non-protein nitrogen compounds, likely influenced by the presence of phenolic compounds in GTE. Erkan *et al*., also got the same result as present study in TMA-N values exhibited a notable increase in all groups and displayed statistically significant ($P<0.05$) distinctions among all groups from storage period. Findings of Rajesh *et al*. (2002) are equal to the current study the TMA-N levels in the treated samples were less than compared to the control and the reason behind it vacuum-sealed packaging gave good a quality effect compared to simple packaging. As referenced in Pal *et al*. (2017) [23], it is indicated that green tea extracts are notably effective in managing the elevation of TMA-N values when products are stored in ice.

4.2.1.3. Changes in Total volatile base Nitrogen TVB-N during refrigerated storage

Similar increasing trends were also observed by Pal *et al*. (2017) [23], analysis of the mean values indicating a significant ($p<0.05$) reduction in TVBN values for the treated samples when compared to the control samples and because of the significant impact of tea extract as an antimicrobial agent. Shinde *et al*. (2015) [34] reported similar findings, with an initial TVB-N of 7.12±0.58 mg% at the start of storage in both PPE-treated and GTE-treated samples. These results align with the research of Nugraha *et al*., who observed that untreated fish exhibited higher TVB-N levels compared to fish samples treated with green tea and black tea dips. Matching results were obtained as Lin and Lin (2005) revealed that the application of glazing treatment with green and black tea extracts proved highly efficient in sustaining a reduced TVBN value in bonito fillets compared to their untreated counterparts.

4.2.1.4. Changes in Peroxide value PV during refrigerated storage

The present study shows the agreement with Alghazeer *et al*. (2008) confirmed that the addition of instant green tea in Atlantic mackerel (*Scomber scombrus*) fillets resulted in a reduced rate of peroxides and hydroperoxides formation over a storage period in contrast to samples without green tea Mohan *et al*. (2019) [38] reported similar findings, as they observed that the treated samples exhibited very low PV value reaching in fish compared to untreated samples and the reason behind it Green tea extract (GTE) exerts its antioxidant effects by inhibiting chain reactions and breaking down hydroperoxides and free radicals, as noted by Raeisi *et al*. (2014).

4.2.5. Microbiological Characteristics

4.2.5.1. Changes in Total Plate Count (TPC) during refrigerated storage

Fan *et al*. (2008) also attributed to related findings, as they observed lower TPC values in the treated sample compared to the untreated sample because due to green tea extract antibacterial and antifungal impacts occur in sample. The present observation was in line with the result obtained by Nugraha *et al*., as they found that outcomes of dipping fish samples in instant green tea extract indicated a decrease in bacterial growth on the total plate count agar and this suggests that some preservation action had already occurred the reason behind it the antibacterial compounds found in green tea belong to the catechins group, and these compounds can inhibit the growth of bacteria in fish. Hassan & Geethalakshmi (2020) [10] similar result was reported by Chan *et al*., as they observed the TPC of the treated sample with GTE sample is acceptable higher time compared to the control sample and the reason behind it is the that antioxidant activity of green tea can be attributed to the presence of phenolic hydroxyl groups, enabling it to interact with free radicals, thereby inhibiting lipid oxidation and its ability to chelate metal ions contributes to its antioxidant properties.

4.2.5.2. Changes in Total Psychrotrophic Counts during refrigerated storage

A similar increasing trend was also observed by Mohan *et al*. (2019) [38] as they found that for seer fish, the psychrotrophic counts showed an increase over the storage period in all the samples. Mohan *et al*. (2019) [38] reported similar findings, as they observed a significantly ($p<0.05$) greater increase in the control samples compared to the treated samples. Mohan *et al*. (2019) [38] documented comparable findings in psychrotrophic bacterial counts showed a similar increasing trend of the Vacuum packaging however their counts were significantly compared to those higher in air packaging. Azizi-Lalabadi *et al*. (2020) [2] reported similar findings, as they observed the psychrophilic bacteria count was approximately lower on the initial day of storage for all samples after a few days passed the bacterial population of the control sample had significantly increased compared to the untreated sample.

Quality changes during refrigerated storage of fish under different time exposure of green tea extract (GTE) and vacuum packaging treatment

In the experiment fish (*Scomberomorus lineolatus*) was treated with Green tea extract and Vacuum packaging and stored at (4±1 °C) under refrigerated condition for 30 days. Based on Biochemical, Physical, Colour, Microbiological and sensory characteristics it was observed fish treated with GTE & VP has the best performance in terms of the quality as compared to untreated sample. The Biochemical storage study of all samples indicated considerable changes with trends of increase in pH, TMA-N and TVB-N and PV level as storage period increase. While least change in pH, TMA-N and TVB-N and PV were noted in GTE & VP treated sample. The best treatment (T2) according to the biochemical analysis. The Microbiological storage study of fish indicated considerable changes in the form of value of Total plate count and Total psychotropic counts as storage period increased. *Escherichia coli* (cfu/g) and *Staphylococcus aureus* (cfu/g) are not detectable. The level of Total plate count and Total psychrotropic counts were less in GTE & VP treated sample.Study also shown that GTE & VP improve the quality and increase the shelf-life of fish steak. The improvement in the quality of the seafood products is achieved by inhibition of growth of microbial, reduction of oxidation of lipid and enhancement of sensorial attributes.

Conclusion

Finally, the result was that according to Biochemical characteristics the Green tea extract (2%) with Vacuum packaging extent shelf life up to 10 days than control treatment. According to Microbiological character and Sensory characteristics it extend shelf life up to 15 days in T2 than control treatment due to the antimicrobial and antioxidant property of the Green tea extract and Vacuum packaging. Green tea extract act as barrier for lipid oxidation, inhibit the growth of microorganisms, helps to maintain good quality of fish meat during storage. The combined treatment of Green tea extract and Vacuum packaging proved to be a effective strategy for preserving the quality and extending the shelf-life of streaked seer fish steak under refrigerated conditions.

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