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Research Article

Effect of different drying methods on physicochemical characteristics of sardines (*Sardinella longiceps*) incorporated with turmeric extract (*Curcuma longa* L.) for shelf-life extension

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ABSTRACT- In this study, effects of different drying methods, temperatures, and addition of turmeric extract on the quality of sardine (*Sardinella longiceps*) were investigated. Drying methods included sun drying, cabinet thin layer drying (CTL) at 60, 70, and 80 °C, and CTL combined with turmeric extract (CTE) at 70 °C. The study monitored parameters, such as peroxide value (PV), thiobarbituric acid (TBA), total volatile basic nitrogen (TVB-N), and color over a three-month period. Increased drying temperature led to the unfavorable changes in fish quality and lipid oxidation. PV, TBA, and TVB-N values increased with sun drying and CTL at 80 °C. Turmeric extract, known for its antioxidant and antimicrobial properties, was used to enhance the quality and shelf-life of dried fish. Results showed significant improvement in all parameters except TVB-N. The extract also inhibited fatty acid oxidation, and CTE dried fish had better microbial quality at the end of the storage period ($P < 0.05$). Overall, the combination of turmeric extract and drying methods successfully improved the quality of dried sardines.

INTRODUCTION

Seafood is a rich source of nutrients, including proteins, fatty acids, vitamins, minerals, and more, which are essential for human health (Ravichandran *et al*., 2012). However, seafood is highly perishable, and its meat is susceptible to chemical deterioration (oxidation), enzymatic autolysis, and microbial spoilage. Therefore, preserving fish as quickly as possible is crucial (Al-Rubai *et al*., 2020). Drying is one effective method for preserving the nutritional value of fish and extending its shelf-life. However, conventional drying methods, such as sun drying (SD) or open-air drying, can negatively impact the quality and safety of the fish (Abraha *et al*., 2017). Dried fish is a valuable export product for many countries, including Iran, China, Thailand, and the United States. Traditional fish drying is often done in the open air, a method referred to as SD (Darvishi, *et al*., 2013). Although SD is an inexpensive and widely practiced traditional method, it has several drawbacks, including inadequate hygiene and the need for large drying areas (Immaculate *et al*., 2013; Afolabi & Agarry, 2014). These disadvantages have led to the development of newer drying methods, such as microwave drying (Darvishi *et al*., 2013) and electric oven drying (Chukwu & Shaba, 2009). In addition, combination techniques, such as microwave-assisted hot air drying (Duan *et al*., 2011) and salting in combination with SD (Immaculate *et al*., 2013) have been successfully applied to various types of fish. To improve the quality and nutritional value of seafood, the use of advanced processing technologies or methods is crucial. Fish is rich in fatty acids that are highly susceptible to oxidation, which negatively impacts the sensory properties and overall quality of the fish (Cyprian *et al*., 2017). To address this issue, the use of natural plant-based extracts has shown promise in extending the shelf-life and quality of food products due to their beneficial biological properties (Abdou *et al*., 2018; Indira Priyadarsini, 2013). Turmeric is one such plant, with its active ingredients found in the underground stems (rhizomes). The main components of turmeric include curcumin, demethoxycurcumin, and bisdemethoxycurcumin. Among these, curcumin, a diphenolic compound, is considered the most important constituent of turmeric (Naksuriya *et al*., 2014; Yallapu *et al*., 2012). Curcumin, also known as frolyl methane, is a type of polyphenol isolated from the rhizome of *Curcuma longa* (Asouri *et al*., 2013). It is widely used as a spice and coloring agent in the food industry (Meral *et al*., 2019). Furthermore, the consumption of curcumin has been shown to positively affect human health through its various

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biological activities, such as antiviral, anti-inflammatory, antimicrobial, antioxidant, anti-HIV, anti-Parkinson's, anti-Alzheimer's, anti-angiogenesis, and anticancer properties.

Therefore, curcumin exhibits many bioactive properties commonly associated with nutraceuticals and functional foods. Additionally, it can be incorporated into natural and synthetic polymer films, providing them with antioxidant and antimicrobial properties (Musso *et al*., 2017; Wu *et al*., 2018). Several studies have investigated antioxidant films based on curcumin as functional films for use in active packaging or biomedical applications (Liu *et al*., 2016; Manna *et al*., 2015; Wu *et al*., 2018). However, product development with fatty fish, such as sardines, can be challenging due to their susceptibility to spoilage (Senapati *et al*., 2017). Combining drying techniques with natural bioactive compounds, like curcumin, may extend the shelf-life of these products. To date, thin-layer drying in combination with turmeric extract has not been reported. Therefore, the objective of this study was to investigate the effects of different drying methods, including SD and cabinet thin-layer drying (CTL) at 60, 70, and 80 °C, on the physicochemical and microbial properties of sardine (*Sardinella longiceps*). The results were then used to evaluate the impact of turmeric extract combined with the optimum temperature for CTL drying (CTE) treatment.

MATERIALS AND METHODS

Fresh *S. longiceps*, each weighing 16-18 g with an average body length of 5-7 cm, were purchased from a local fish market in Bushehr, Iran. Turmeric rhizomes (*Curcuma longa* L.) were sourced from a local market in Shiraz, Iran. All other chemical reagents were obtained from Sigma-Aldrich.

Fish preparation

Fresh *S. longiceps* samples were transferred to the Department of Food Science and Technology, Shiraz (Iran), within 6 hours while kept in a box filled with ice. Then, they were washed and kept at -18 °C for further analyses.

Turmeric extraction

The aqueous extraction of turmeric rhizomes (*C. longa* L.) was performed following the method described by Mohankumar and McFarlane (2011), with slight modifications. To prepare the turmeric extract, turmeric powder was mixed with distilled water at a concentration of 10% (w/v) and stirred at 45 °C for 10 min. The mixture was then filtered using Whatman No. 1 filter paper and used for fish treatment. For this purpose, the fish were immersed in the turmeric extract for 2 hours and then stored at a standard refrigerated temperature.

Drying methods

The fish were dried using three different methods. The SD process took place over two consecutive days, with an average daylight duration of 10 hours and 57 min, and temperatures ranging from 8.5 to 30.75 °C. For the CTL drying, the fish was dried at temperatures of 60, 70, and 80 °C, with an air velocity of 2.0 ± 0.1 m/s and an inlet relative humidity of 72.0 ± 4.0 %. To compare the different methods, 70 °C (the middle temperature for the CTL method) was selected for drying fish that had been dipped in turmeric extract. The drying process was performed using whole sardines, with each batch precisely measured to reach a mass of 100.0 ± 3 g. After drying, all fish were stored at 20 ± 2 °C for three consecutive months for further analysis.

Proximate analysis

The proximate composition of fresh *S. longiceps*, including moisture, lipid, protein, and ash was analyzed using the AOAC method (1995). Moisture content was determined using an electric oven (SL-908, Iran). A Micro Kjeldahl Apparatus (PECO, Iran) was used for the determination of total protein (crude protein, N = 6.25), while lipid content was measured using a Soxhlet-Henkel Apparatus (KTG, Iran). Ash content was analyzed by incinerating the samples in an electric furnace (Lenton, England) at 550 °C.

Antioxidant activity of turmeric extract by DPPH assay

The antioxidant activity was measured using the 2,2-diphenyl-1-picrylhydrazyl (DPPH) free radical decolorization assay, following the method of Maizura *et al*. (2011), with slight modifications. Methanolic extracts (0.2 mL) of turmeric powder at three different concentrations (0.1, 0.5, and 1.0 mg/mL) were mixed with 1.8 mL of DPPH methanolic solution (0.25 mM). After mixing, the samples were allowed to stand in the dark for 60 min. The absorbance was then measured at 517 nm using a UV-Visible spectrophotometer (Unico, China). DPPH scavenging activity was expressed as the IC_{50} value, which represents the concentration of antioxidant required to inhibit 50% of the DPPH radical activity.

Total phenolic content (TPC)

The total phenolic content (TPC) was measured using the Folin-Ciocalteu method (Lu et al., 2011). To begin, 0.1 mL of the turmeric extract was mixed with 0.75 mL of Folin-Ciocalteu reagent and 0.75 mL of sodium carbonate solution (2% w/v). The mixture was kept in the dark at 20 °C for 45 min, after which the absorbance was measured at 765 nm using a UV-Visible spectrophotometer (Unico, China). TPC values were determined using a calibration curve prepared with gallic acid standards at concentrations of 25, 50, and 100 mg/mL. The results were expressed as mg of gallic acid equivalents per gram of fresh turmeric weight.

Total flavonoid content (TFC)

TFC was analyzed using the method described by Chen *et al*., (2011). As mentioned, the turmeric extract was prepared by mixing turmeric powder with distilled water (10%, w/v) and stirring at 45°C for 10 min, followed by filtration through Whatman No. 1 filter paper. For the TFC assay, 0.5 mL of turmeric extract (20 mg/mL) was mixed with 0.1 mL of 10% $AlCl_2$, 0.1 mL of 1 M potassium acetate (CH_3CO_2K), and 2.8 mL of distilled water. A control sample, in which 0.5 mL of methanol replaced the turmeric extract, was also prepared. Both mixtures were

kept at room temperature for 30 min, and absorbance was measured at 415 nm using a UV-Visible spectrophotometer (Unico, China). A calibration curve was generated using quercetin solutions at concentrations of 6.25, 12.00, and 25.00 mg/mL in methanol, with the control sample containing all reagents except quercetin. TFC was expressed as mg quercetin equivalents per gram of fresh turmeric weight.

Total volatile basic nitrogen (TVB-N)

The TVB-N value of all fish samples was analyzed following the method described by Arulkumar *et al*., (2017). In this procedure, homogenized fish samples (10 g) were mixed with 20 mL of trichloroacetic acid (TCA) solution (6%, w/v). The TCA extract was then absorbed with boric acid, and the resulting boric acid solution was titrated with a 0.05 M sulfuric acid solution. The TVB-N value, expressed as mg of nitrogen per 100 g of fish meat, was calculated based on the amount of sulfuric acid consumed during titration.

Thiobarbituric acid (TBA) value

The TBA assay was conducted following the method described by Moosavi-Nasab *et al*., (2019) with slight modifications. Fish meat (10 g) was mixed with 30 mL of a stock solution containing 0.02 M TBA and 7.5% (TCA). The mixture was heated in a water bath at 100 °C for 35 min to develop a pink color, then cooled with tap water and centrifuged at 600 rpm for 10 min. The absorbance of the final solution was measured at 532 nm using a UV-Visible spectrophotometer (Unico, China), with a control sample containing all reagents except the fish meat. The results were expressed as mg of malondialdehyde (MDA)/kg of fish meat.

Peroxide value (PV)

The PV was determined using the AOCS official method (AOCS, 1997). Five grams of oil samples were mixed with 30 mL of an acetic acid-chloroform solution (3:2 v/v) and shaken thoroughly. Next, 0.5 mL of potassium iodide solution was added to the mixture. After filtration, the mixture was titrated with 0.1 N sodium thiosulfate until the yellow color disappeared. Subsequently, 0.5 mL of starch indicator solution was added, and titration was continued with constant agitation until the blue color disappeared. A blank sample was prepared without the addition of the oil sample. The PV, expressed as mEq of peroxide 1000 g^{-1} of sample, was calculated using the following equation:

$$PV = (S-B) \times N \times 1000/W \quad \text{Eq. (1)}$$

Where B is standard potassium thiosulfate used for titration of blank (mL), S is standard potassium thiosulfate used for titration of the sample (mL), N is the normality of sodium thiosulfate solution, and W is weight of the sample (g).

Fatty acids analysis

Fatty acids were analyzed using gas chromatography (GC) with an SP-3420 A gas chromatograph apparatus (Beijing, China) equipped with a flame ionization detector and a BPX-70 fused silica capillary column (30 m × 0.25 mm, 0.25 μm film thickness). Nitrogen (N_2) was used as the carrier gas, with an injection volume of 1 μL and a split ratio of 1:10. The injector and detector temperatures were set at 250 and 300 °C, respectively. The oven temperature program began at 140 °C, maintained for 5 min. It was then increased to 180 °C at a rate of 20 °C/min and held for 9 min, followed by a further increase to 200 °C at the same rate, where it was held for 3 min. Fatty acids were identified by comparing their retention times with those of corresponding standards. The results were expressed as percentages of the relative peak area. Data analysis was conducted using the PEAK-ABC chromatography workstation, version 2/24.

Microbial assay

To determine the total plate count for each treatment, samples were collected from multiple parts of the fish and combined. Ten grams of each sample were aseptically weighed and transferred to a sterile stomacher bag, followed by the addition of 90 mL of sterile saline. The mixture was then homogenized. For enumeration, 0.1 mL of the serial dilution of the homogenate was plated on plate count agar and incubated at 37 °C for 2 days. The average number of colonies for each treatment was expressed as $\log_{10}$ CFU/g of the sample (AOAC, 1996).

Statistical analysis

The results were expressed as mean values ± standard deviation (SD) and analyzed using one-way analysis of variance (ANOVA), followed by Duncan's test at a significance level of $P < 0.05$. The analysis was performed using SPSS version 19.0 software.

RESULTS AND DISCUSSION

Proximate composition of sardines

The proximate composition of fish is crucial due to its role in determining the most effective processing and storage pathways for final products (Mohan *et al*., 2012). The proximate composition of *S. longiceps* is presented in Table 1. As shown, *S. longiceps* contained a considerable lipid content (8.26 ± 0.32%), along with moisture and protein contents of 72.26 ± 0.28% and 16.8 ± 0.3%, respectively. Similar findings were reported by Mohan *et al*., (2012), who evaluated the proximate composition of Indian oil sardine (*S. longiceps*). However, the chemical composition of fish varies based on multiple factors, including environmental and nutritional conditions, sex, age, and season, leading to diverse reports in the literature (Mohan *et al*., 2012).

Table 1. Proximate composition of Sardine (*S. longiceps*)

Moisture (%)	Lipid (%)	Protein (%)	Ash (%)
72.26 ±0.28	8.26±0.32	16.8±0.3	2.33±0.12

Data are expressed as mean ± SD (n = 3).

TPC, TFC, and IC_{50}

TPC, TFC, and IC_{50} of turmeric powder extract are presented in Table 2. The TPC was 15.03 ± 1.8 mg of gallic acid equivalents per gram of sample, the TFC was

3.83 ± 0.29 mg of quercetin equivalents per gram of sample, and the IC_{50} value was 0.047 ± 0.0 mg/mL. These findings highlight the relationship between turmeric extract and its antioxidant activity. Reactive oxygen species (ROS) can react with macromolecules such as lipids and proteins, leading to cellular damage. Antioxidants mitigate this damage by reducing ROS and preventing the oxidation of macromolecules (Klein *et al*., 2012). Phenolic compounds in natural substances play a significant role in antioxidant activity, with a linear correlation observed between TPC and antioxidant efficacy. This is attributed to their ability to donate hydrogen atoms or electrons, neutralizing free radicals (Maizura *et al*., 2011). Curcumin, the main antioxidant component in turmeric, contributes to its antioxidant activity. However, the content of curcumin can vary depending on species and acidity. Differences in TPC and TFC among turmeric samples may also result from variations in species and varieties (Akter *et al*., 2019). The antioxidant activity of turmeric was evaluated using the DPPH assay, which is widely regarded as a reliable and straightforward method for assessing antioxidant activity (Maizura *et al*., 2011). In this assay, plant extracts reduce DPPH free radicals, transforming them into stable DPPH molecules and causing a color change from deep violet to pale yellow (Nisar *et al*., 2015). The IC_{50} results demonstrated the antioxidant potential of turmeric extract, suggesting its suitability as a natural substance for enhancing food quality. However, the higher IC_{50} value of turmeric extract could be attributed to the lower solubility of curcumin in water and the reduced yield of curcumin when isolated using distilled water (Sukati & Khobjai, 2019). The antioxidant activity of turmeric extract is linked to its ability to donate hydrogen and scavenge hydrogen peroxide, superoxide, and free radicals (Xin *et al*., 2020). The findings of this study align with those of Maizura *et al*. (2011), who reported high antioxidant activity in turmeric, with a TPC of approximately 67.89 mg of gallic acid equivalents per gram and 64% free radical scavenging activity. Asouri *et al*. (2013) also confirmed curcumin's antioxidant properties and its protective effects against oxidative damage, reporting a free radical scavenging activity of approximately 69%. Additionally, Shang et al. (2010) observed a correlation between curcumin's antioxidant activity and its chemical structure, noting that specific structural features enhance its efficacy. The type of solvent used also influences antioxidant activity. Nisar *et al*. (2015) reported that ethanolic extracts of turmeric exhibited higher antioxidant potential than aqueous extracts under similar conditions. Similarly, Sukati and Khobjai (2019) found that aqueous extracts of fresh and ripened rhizomes had TPC values of 18.38 ± 0.41 and 19.79 ± 0.64 mg of gallic acid equivalents per gram, respectively.

Table 2. TPC, TFC, and IC_{50} value of turmeric extract

TPC (mg of gallic acid equivalents/g of sample)	**TFC (mg of quercetin equivalents/g of sample)**	**IC_{50} (mg/mL)**
15.03 ± 1.8	3.83±0.29	0.047 ±0.0

Data are expressed as mean ± SD (n = 3).
Total phenolic content (TPC); total flavonoid content (TFC); half-maximal inhibitory concentration (IC_{50}).

PV and TBA value

Lipid oxidation is a major factor limiting the shelf-life of fatty fish due to the deteriorative reactions that occur in unsaturated fatty acids. PV and TBA value are widely used as indicators to evaluate primary (peroxides) and secondary (aldehydes, ketones, and short-chain fatty acids) lipid oxidation products in fat-enriched foods (Al-Rubai *et al*., 2020). The PV and TBA values of dried fish are presented in Fig. 1. The initial PV was 5.43 ± 0.58 mM of peroxide per kg. After the drying process, the PV increased significantly across all treatments (P < 0.05). Furthermore, an increase in temperature during the controlled temperature and light C.T.L. method led to a dramatic rise in the PV, likely due to the enhanced rate of oxidation induced by higher temperatures.

Additionally, turmeric-treated fish showed a reduction in PV value from 16.8 to 12.26 mM of O_2/kg, attributed to the antioxidant activity of turmeric. Turmeric extract effectively mitigated primary oxidation during drying and storage, likely due to its curcuminoids, which act as natural antioxidants and effectively control PV development (Sathishkumar *et al*., 2015). A similar study by Immaculate *et al*. (2013) reported that fish dried using traditional methods did not achieve the desired physicochemical quality. Another study observed an increase in PV values during storage, with the highest PV reported in the SD. treatment (Arulkumar *et al*., 2017). Multiple studies have also documented increased lipid oxidation as a result of the drying process (Ali *et al*., 2011; Kilic, 2009; Ortiz *et al*., 2013). These findings suggest that traditional drying methods, such as SD., are insufficient for preserving the quality of fatty fish.

The initial TBA value was 0.33 ± 0.01 mg of MDA/kg of fish meat, consistent with the findings of Mohan *et al*. (2012), who reported an initial TBA value of approximately 0.32 mg MDA/kg for sardines. However, the TBA content increased after drying, with the highest value observed in fish dried at 80 °C (CTL 80 °C). This suggests that higher temperatures result in greater TBA content, indicating a direct correlation between temperature and secondary lipid oxidation products. These findings align with Fu, *et al*., (2015), who reported that drying at higher temperatures accelerates the breakdown of peroxides into carbonyl compounds, leading to higher TBA values in fish dried at 90 °C in comparison with 60 °C. During three months of storage, both TBA and PV values increased across all batches, consistent with the observations of Can (2011). However, the increase was not statistically significant in CTE at 70 °C. Turmeric extract demonstrated its antioxidant properties by mitigating lipid oxidation during storage. Overall, the TBA value was the highest in the SD treatment and lowest in CTE at 70 °C by the end of the storage. The TBA value in the S.D treatment exceeded the maximum permissible limit for fish at the end of the storage. According to Schormüller (1965), the acceptable limit for TBA in fish is 5 mg of MDA/kg.

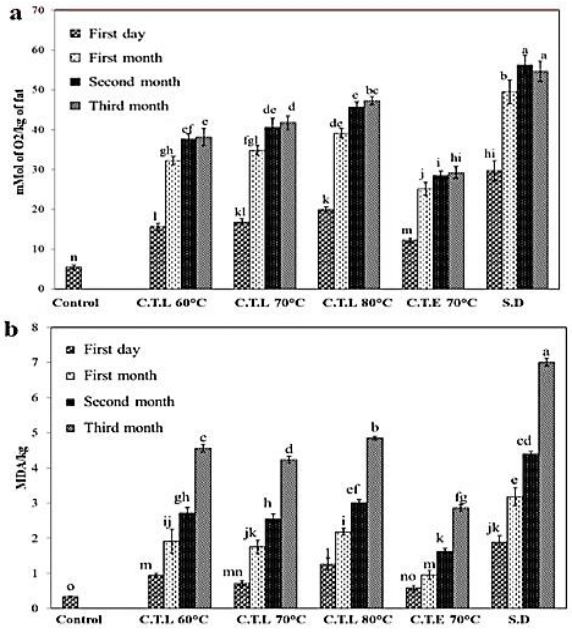

Fig. 1. Effect of different drying methods on PV (a) and TBA index (b) of Sardine. Data are expressed as mean ± SD *(n=3)*. Different lowercase letters within each column are significantly different (P < 0.05). Peroxide value (PV); Thiobarbituric acid index (TBA); malondialdehyde (MDA).

TVB-N value

TVB-N is widely recognized as a critical indicator for assessing fish quality and spoilage level. It reflects protein decomposition caused by bacterial and enzymatic activity, which produces dimethylamine and trimethylamine (Li *et al*., 2011). Proteins and nitrogenous compounds in fish degrade into ammonia, amines, and other volatile substances through enzymatic reactions and microbial activity. TVB-N values also indicate the accumulation of volatile alkaline nitrogen compounds during storage and serve as an essential measure of product freshness (Xin *et al*., 2020). According to Fig. 2, comparative results of different drying methods on TVB-N values are presented. The initial TVB-N value was 12.8 ± 0.58 mg/100 g, consistent with Mohan *et al*. (2012), who reported a value of 14.81 mg/100 g for Indian sardine. After drying, all samples exhibited significantly higher TVB-N values compared to the fresh fish (P < 0.05). Among the drying methods, the best results were achieved using the CTE method at 70 °C, while the highest and lowest TVB-N values were observed in the SD method (23.26 ± 0.56 mg/100 g) and CTE at 70 °C (16.7 ± 0.45 mg/100 g), respectively. These findings align with the acceptable limit for TVB-N (30-35 mg/100 g) in ice-stored marine fish (Behnam *et al*., 2015). Similarly, Immaculate *et al*. (2013) demonstrated that TVB-N values in SD fish ranged from 16.41 to 22.72 mg/100 g. Over three months of storage, TVB-N levels increased significantly across all batches (P < 0.05), with sun-dried fish showing the highest TVB-N values at the end of the storage. Among the three drying temperatures in the cabinet dryer, fish dried at 60 °C exhibited the lowest TVB-N values, while those dried at 80 °C had the highest. The deteriorating effect of temperature on TVB-N values has been corroborated by other studies. For instance, Arulkumar *et al*. (2017) observed an increase in TVB-N content in *S. brevimana* treated with turmeric extract, with final values reaching 33.6 mg/100 g. TVB-N levels depend on factors such as fish species, specific treatments, and processing conditions (Arulkumar *et al*., 2017). Interestingly, the rate of TVB-N increase was significantly lower in the presence of turmeric extract (CTE at 70 °C) compared to other treatments (P < 0.05). This improved preservation was attributed to the antioxidant properties of turmeric extract, which reduced microbial and enzymatic activities, thereby limiting the formation of volatile bases during spoilage (Arulkumar *et al*., 2017). Similar observations were reported by Xin *et al*. (2020), who found that the gradual release of curcumin from a zein-potato starch film reduced lipid oxidation in fish fillets. Curcumin's effectiveness is linked to its ability to scavenge free radicals, as its structure contains active phenolic hydroxyl groups.

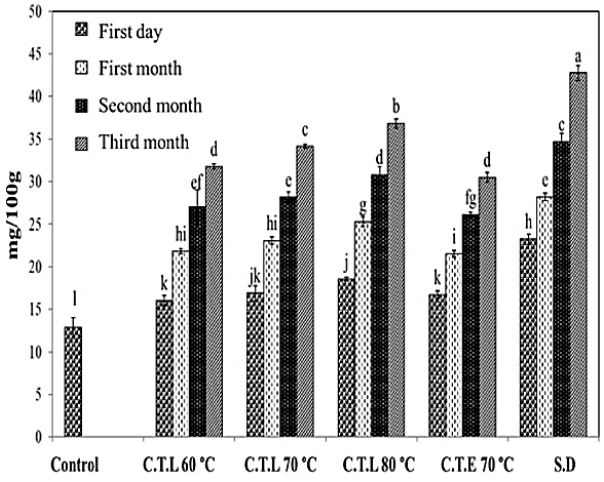

Fig. 2. Effect of different drying methods on Total volatile basic nitrogen value of sardines. Data are expressed as mean ± SD (n = 3). Different lowercase letters are significantly different (P < 0.05). Total volatile basic nitrogen (TVB-N) value.

Fatty acid profile

The fatty acid profile of *S. longiceps* is summarized in Table 3. Saturated fatty acids (SFAs) were the predominant fatty acids, comprising 42.68 ± 1.52% of the total, followed by polyunsaturated fatty acids (PUFAs) at 29.75 ± 0.33%, and monounsaturated fatty acids (MUFAs) at 27.55 ± 1.19%. Among the SFAs, myristic acid (C14:0), palmitic acid (C16:0), and stearic acid (C18:0) were the most abundant, with relative contents of 7.96 ± 0.71%, 29.38 ± 0.45%, and 11.88 ± 0.75%, respectively. Oleic acid (C18:1) dominated the MUFAs, with a relative content of 11.83 ± 0.37%. PUFAs were primarily represented by docosahexaenoic acid (DHA, C22:6n-3) and eicosapentaenoic acid (EPA, C20:5n-3), which are well-known for their therapeutic effects on human health. However, DHA and EPA are highly susceptible to degradation and serve as benchmarks for evaluating fish quality. Thus, the effects of different drying methods on DHA and EPA were

assessed. The relative percentage of EPA in fresh *S. longiceps* was 6.57%, but it decreased to 5.36% in fish dried using the CTL method at 80 °C (Table 4). This finding highlights the adverse effects of high processing temperatures on fatty acids. Additionally, fatty acid degradation occurred during storage, with turmeric extract showing the highest protective effect over a three-month period ($P < 0.05$). The reduction in PUFA content following SD and CTL methods, compared to CTE, was attributed to the structural and chemical damage to cells during drying. High temperatures likely caused the breakdown of double bonds in EPA and DHA, resulting in lipid oxidation (Tenyang *et al*., 2020). Fish dried using the CTE method at 70 °C retained the highest EPA content (5.62 ± 0.16%) at the end of storage ($P < 0.05$), demonstrating curcumin's role in reducing fatty acid decomposition due to its antioxidant properties. Similar trends were observed with DHA content. The initial DHA content of 17.14% decreased significantly by increasing drying temperatures, with the greatest reduction seen in the CTL method at 80 °C. Conversely, the highest DHA retention was observed in fish treated with CTE at 70 °C ($P < 0.05$), underscoring turmeric extract's ability to prevent fatty acid losses as a natural antioxidant. Ortiz *et al*. (2013) reported significant reductions in EPA and DHA after drying. They also noted that increasing the drying temperature from 40 °C to 60 °C caused a noticeable decline in DHA, while EPA remained relatively stable. Similarly, Fu *et al*. (2015) found low EPA and DHA levels in dried silver carp slices, though turmeric extract provided some protection against lipid oxidation. These findings align with the current study, which demonstrated higher EPA and DHA retention in the CTE treatment.

Table 3. Fatty acid distribution in Sardine

Fatty acid	Relative peak area (%)
C11	0.22 ± 0.02
C12	0.12 ± 0.01
C13	1.39 ± 0.03
C14	7.96 ± 0.71
C15	0.15 ± 0.21
C16	29.38 ± 0.45
C16:1	3.31 ± 0.17
C17	2.51 ± 0.13
C18:0	11.88 ± 0.75
C18:1	11.83 ± 0.37
C19:0	1.71 ± 0.06
C18:2	0.11 ± 0.01
C20:0	2.17 ± 0.08
C20:1	2.09 ± 0.18
C21:0	0.28 ± 0.01
C20:4	3.73 ± 0.95
C22:1	0.46 ± 0.01
C20:5	4.79 ± 0.87
C22:5	0.25 ± 0.00
C22:6	15.85 ± 1.02
SFA	42.68 ± 1.52
MUFA	27.55 ± 1.19
PUFA	29.75 ± 0.33

Data are expressed as means ± SD ($n = 3$).
Monounsaturated fatty acid (MUFA); polyunsaturated fatty acid (PUFA)

Microbial assay

Fig. 3 illustrates the total microbial count in sun-dried and thin-layer-dried fish. According to the data, the initial microbial count for sardines was 4.69 log CFU/g, which aligns closely with findings by Can (2011) and Mohan *et al*. (2012), who reported initial counts of 4.84 log CFU/g and 4.9 log CFU/g, respectively. CTL drying significantly improved the microbial quality of dried fish, maintaining total plate counts below 7 log CFU/g, which is the acceptable limit for fresh fish (Ojagh *et al*., 2010). Among the drying methods, SD was the least effective, resulting in higher microbial counts, particularly by the end of the storage period. These findings are consistent with Immaculate *et al*. (2013), who observed that naturally sun-dried fish had higher microbial counts compared to solar-dried fish. Similar observations were made by Tarle *et al*., (2015) in dried fish species such as *Oreochromis niloticus*, *Pylodictis olivaris*, and *Cyprinus carpio*. Drying generally enhances microbial quality and extends fish shelf-life by reducing moisture content and water activity (Xie *et al*., 2019). In addition, the use of turmeric extract further inhibited bacterial growth. In turmeric-treated fish, CTE drying with turmeric extract reduced the total microbial count to 4.54 log CFU/g. The results indicate that turmeric extract has a significant antimicrobial effect, further enhancing the microbial quality compared to the other drying methods. This effect is attributed to the compounds in turmeric that inhibit decarboxylase enzyme activity in fish muscle during storage (Sathishkumar *et al*., 2015). The antimicrobial activity of turmeric is linked to its amphipathic and lipophilic structure, which disrupts bacterial membranes and causes cell damage (Karimi *et al*., 2018). Curcumin, a key component of turmeric, has been shown to lyse bacterial cells through cell wall or membrane damage, potentially involving specific mechanisms of action (Bound *et al*., 2015). Several studies support the antimicrobial effects of turmeric extract. For example, Mohamed *et al*., (2016) found that fermented turmeric extract exhibited antibacterial effects against *Trichoderma* spp., with minimum inhibitory concentrations ranging from 7.5 to 125 mg/mL. Arulkumar *et al*. (2017) demonstrated that turmeric-treated cuttlefish exhibited antimicrobial activity and extended shelf-life by up to 10 days. Similar findings were reported in other studies (Kalaycıoğlu *et al*., 2017; Wang *et al*., 2017). Minh *et al*. (2019) used a combination of honey, curcumin, drying, and salt to produce high-protein snakehead fish (*Channa striata*), leveraging curcumin's antioxidant and antimicrobial properties alongside drying as an effective seafood processing method. Additionally, Ceylan *et al*. (2020) highlighted the antibacterial effects of curcumin-containing emulsions in preserving rainbow trout fillets, attributing its efficacy to the flavonoids and terpenes such as borneol, cuparene, carene, and cymene.

Table 4. Effect of different drying methods on DHA and EPA of sardine

Drying method	Docosahexaenoic acid (DHA)			Eicosapentaenoic acid (EPA)		
	Fresh	Day 1	Month 3	Fresh	Day 1	Month 3
CTL at 60 °C	17.14 ± 0.21^{Aa}	16.59 ± 0.36^{Ab}	13.52 ± 0.21^{Bc}	6.57 ± 0.47^{Aa}	5.51 ± 0.21^{DCb}	4.96 ± 0.03^{ABc}
CTL at 70 °C	17.14 ± 0.22^{Aa}	16.83 ± 0.13^{Ab}	12.92 ± 0.28^{Cc}	6.57 ± 0.48^{Aa}	5.84 ± 0.33^{BCb}	4.97 ± 0.12^{ABc}
CTL at 80 °C	17.14 ± 0.23^{Aa}	15.07 ± 0.56^{Bb}	11.27 ± 0.17^{Cc}	6.57 ± 0.49^{Aa}	5.36 ± 0.12^{Db}	4.85 ± 0.40^{Bb}
CTE at 70 °C	17.14 ± 0.24^{Aa}	16.59 ± 0.42^{Aa}	15.05 ± 0.42^{Ac}	6.57 ± 0.50^{Aa}	6.28 ± 0.14^{ABa}	5.62 ± 0.16^{Ab}
SD	17.14 ± 0.25^{Aa}	16.89 ± 0.63^{Aa}	13.96 ± 0.63^{Bc}	6.57 ± 0.51^{Aa}	6.41 ± 0.17^{Aa}	4.91 ± 0.80^{ABb}

Data are expressed as mean ± SD (n = 3). Different capital letters in each column and small letters in each row indicate significant differences (P < 0.05). Cabinet thin layer method (CTL); Cabinet thin layer + turmeric extract (CTE); Sun drying (SD).

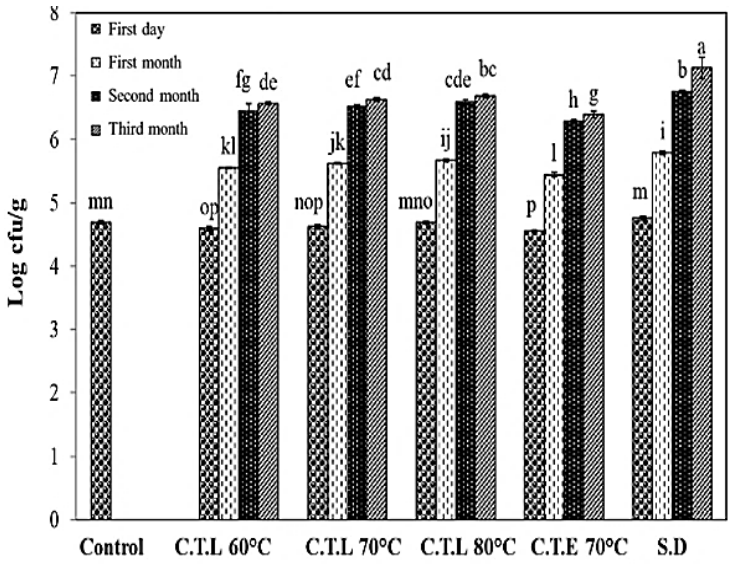

Fig. 3. Microbial total count of dried Sardine with different methods. Data are expressed as mean ± SD (n = 3). Different lowercase letters are significantly different (P < 0.05). Cabinet thin layer method (CTL); Cabinet thin layer + turmeric extract (CTE); Sun drying method (SD)

CONCLUSION

This study aimed to evaluate the effects of various drying techniques, including controlled thin-layer drying at 60, 70, and 80 °C, controlled thin-layer drying at 70 °C combined with turmeric extract, and sun drying, on the physicochemical properties and shelf-life extension of sardines. The findings demonstrated that controlled thin-layer drying outperformed SD in preserving the physicochemical quality of sardines. However, higher drying temperatures had adverse effects, particularly on lipid oxidation, total volatile basic nitrogen, and fatty acid degradation. Specifically, a drying temperature of 80 °C caused significantly more deterioration compared to lower temperatures. The study highlighted that controlled thin-layer drying at 70 °C, combined with turmeric treatment, was the most efficient and reliable method for fish preservation and processing. Sardines treated with turmeric extract during controlled thin-layer drying exhibited superior quality in terms of PV, TBA reactive substances, and TVB-N compared to those subjected to CTL drying alone. Seafood preservation through drying is based on reducing water content and water activity, thereby inhibiting microbial growth. However, the rates of other deteriorative processes in fish tissues are influenced by water activity levels, which reach a minimum at specific thresholds. Incorporating natural antioxidants, such as turmeric extract, into the drying process is therefore beneficial. This study demonstrated that turmeric-treated fish had better microbial quality, with reduced microbial and enzymatic activity. The combination of CTL drying at a lower temperature (70 °C) and turmeric extract effectively extended the shelf-life of fish. Furthermore, this research introduced a novel preservation method as a potential alternative for extending the shelf-life of marine products.

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DECLARATION OF COMPETING INTEREST

The authors declare no conflicts of interest.

DATA AVAILABILITY

The data that support the findings of this study are available on request from the corresponding author.

ETHICAL STATEMENT

This work is not related to experimental animals or specific human diseases that requires publication and approval of publication ethics.

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