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Effect of active and vacuum packaging on the quality of dried sardine (*Sardinella longiceps*) during storage

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Abstract
Changes in biochemical, microbiological and sensory attributes of dried sardine (*Sardinella longiceps)* during storage in air pack (AP), vacuum pack (VP) and active pack (oxygen scavenger) (OSP) were investigated. For this purpose, total volatile basic nitrogen (TVB-N), trimethylamine nitrogen (TMA-N), TBA values (TBA), free fatty acids value (FFA) and peroxide value (PV), sensory attributes and micro-biological analyses were carried monthly during storage. Dried fish packed with oxygen scavenger (active packaging) was found to be the best in terms of biochemical, microbiological, sensory score and long shelf life compared to others. Instrumental analysis by colour reader (L*-value, a*-value, b*-value), textural analysis (shear force) also complimented the findings based on microbiological, biochemical and sensory analyses. Therefore, oxygen-absorbing packaging may be used to pack dried sardine. Sardine as long as 180 days without indicating sign of spoilage observation in quality.

Keywords: Drying, sardine, *Sardinella longiceps,* active packaging, vacuum packaging

Introduction

Fresh fish is highly perishable and various preservation techniques such as chilling, freezing, drying, salting and smoking are commonly used to extend its shelf life. Drying is the most affordable traditional fish preservation methods that are commonly used in many developing countries [1]. The preservative effect of salting and drying is mainly due to the decrease in water activity, which turn prevents the growth of many spoilage organisms is prevented.

Dried fish has been relished as food all along the coastal states of India and there is a tradition to consume it during special functions, especially among the lower strata of the society. It is widely consumed in the interior parts of India since availability and transport of fresh fish is poor. Dry fish processing is a common practice of the Gujarat coastal region and about 20% of the catch is being processed regularly for domestic and overseas consumption. Totally, 11 export units and 350 domestic dry fish units are involved in processing of dry fish in Gujarat state. About 5000 MT of dry fish per season is regularly exported from India, which accounts for about 80% of total exports from Gujarat [2]. Dried fish products have low water content and a low water activity, so the main problems during handling and storage are water adsorption by the product surface in contact with the surroundings, and insect infestation. However the maintanence of quality of dried fish is a serious problem during transportation, storage and distribution in the market. It calls for improvement in packaging of dried fish for long term without compromising the quality. Several packaging methods to improve the quality of dried fish has been tried for maintenance of quality during storage. So, that its value does not decrease during marketing. Active packaging and vacuum packaging are a new concept of food packaging, developed in response to changes in current consumption and market trends and is designed to improve processed sea food product quality safety and shelf life. Vacuum packaging represents a static form of hypobaric storage which is widely used in the food industry due to its effectiveness in reducing oxidative reactions in the product at relatively low costs [3]. Oxygen absorbing systems provide an alternative to vacuum and gas flushing technologies as a means of improving product quality and shelf life. The oil sardine, *Sardinella longiceps* forms one of the major single species fisheries contributing along the Indian coasts. The oil sardine became the second highest contributor in the marine fish landing rankings during 2016 with 2.45 lakh tons [4]. Salt-dried sardine is largely consumed as well. Dried sardine contains a good source of protein and other nutritive value.

Materials and Methods

Fresh sardine (*Sardinella longiceps)* measuring 18.53 ± 1.02 cm in length & weighing 15.39 ± 0.89 g caught by trawl net along the coast off Veraval, India (Lat- 20°55’12”N and long- 70°20’24”E) were used for the study. Immediately after harvest, fishes were washed in fresh water and iced in the ratio of 1:1 (fish: ice) and transported to the fish curing yard of Veraval for processing. The whole fish was washed in chilled water prior to processing. Descaling process was carried out manually and fishes were further washed with chilled water before drying. Plastic bags (O_2 transmission: 46 cc /m^2 / atmosphere / 24hrs /room temperature 28-32°C; vapour permeability: 2.09 g/m^2/24 hrs) with poly propylene (PP) and polyester layered (300 gauge) were used for packing the dried fishes. Dried fish packed in different packaging viz. air packaging (AP), Vacuum pouch packaging (VP) and pouch packaging with iron based O_2 scavenger (50 CC - Pack Fresh, USA) (O_2 scavenger packaging) (OSP). All dried products sample with different treatments were stored at room temperature. Samples from each were drawn randomly at regular intervals of 30 days for biochemical, microbiological and sensory quality analysis for 180 days.

Peroxide value (PV) was also determined as per standard methods [5]. Total volatile based nitrogen (TVBN) was determined by the Conway micro diffusion method [6]. Free fatty acid (FFA) was determined by improved titrimetric method [7]. pH was estimated as per standard methods [8] and TBA (Thiobarbituric Acid) [9]. The microbiological characteristics of sardine sample was carried out according to standard method of TPC (Total plate count) and TFC (Total fungal count) [10]. Organoleptic evaluation was carried out by 9 point Hedonic scale [11].

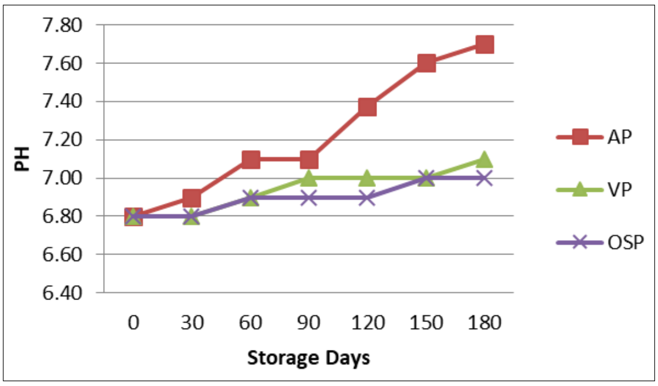

Fig 1: Changes in pH of the dried sardine (*Sardinella longiceps*) during storage.

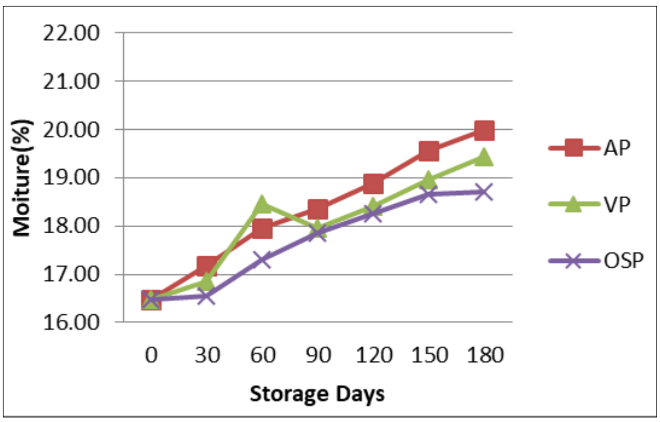

Fig 2: Changes in moisture (%) content of the dried sardine (*Sardinella longiceps*) during storage.

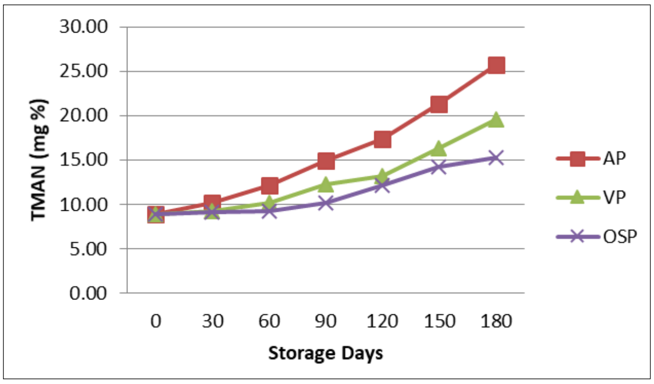

Fig 3: Changes in TMA-N (mg/100g) of the dried sardine (*Sardinella longiceps*) during storage.

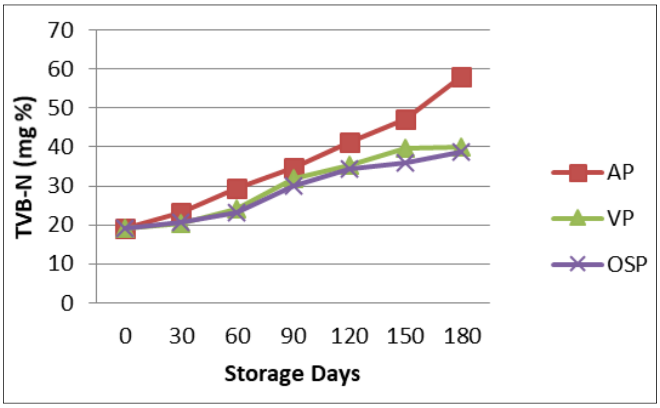

Fig 4: Changes in TVB-N (mg/100g) of the dried sardine (*Sardinella longiceps*) during storage.

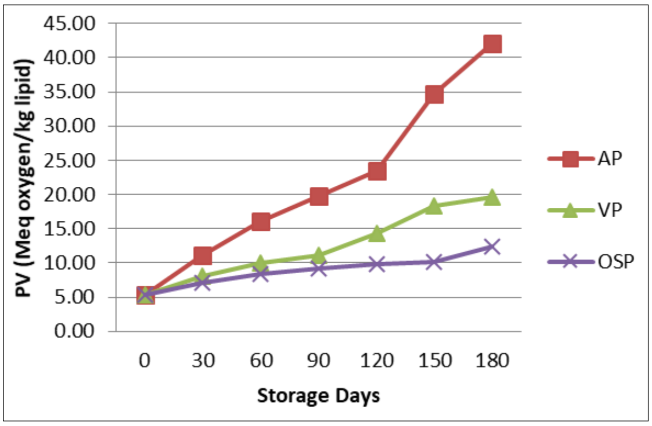

Fig 5: Changes in PV (meq/kg) of the dried sardine (*Sardinella longiceps*) during storage.

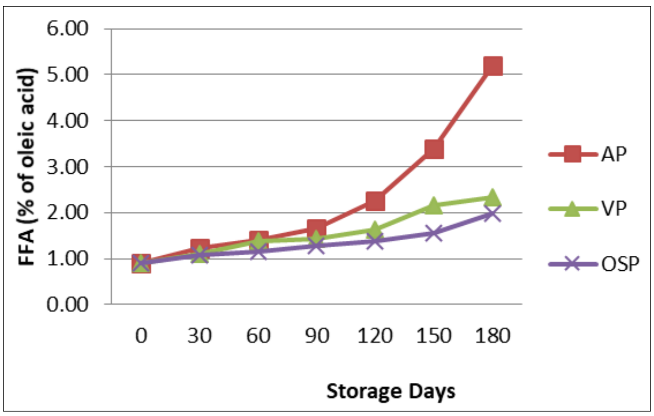

Fig 6: Changes in FFA (% of oleic acid) of the dried sardine (*Sardinella longiceps*) during storage.

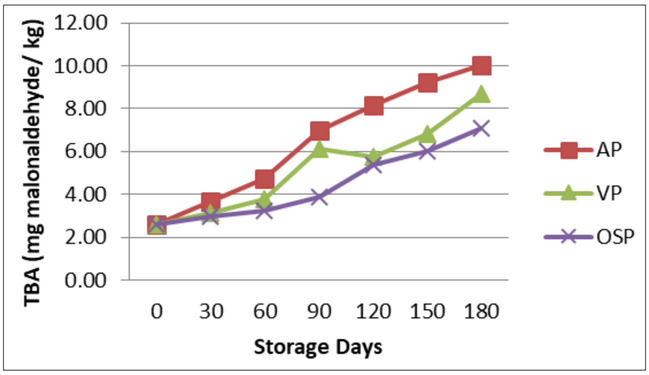

Fig 7: Changes in TBA (mg malonaldehyde/kg) of the dried sardine (*Sardinella longiceps*) during storage.

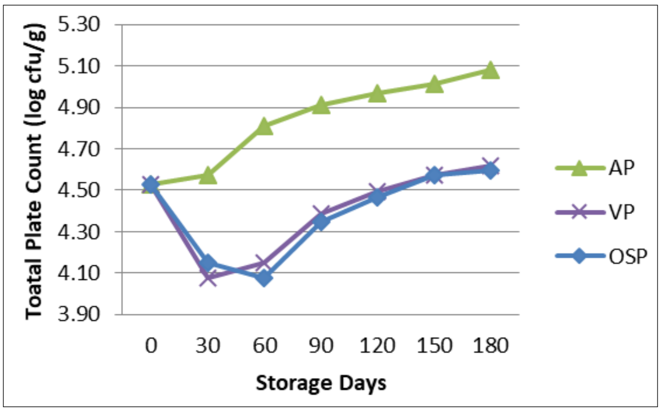

Fig 8: Changes in Total plate count (log cfu/g) of the dried sardine (*Sardinella longiceps*) during storage.

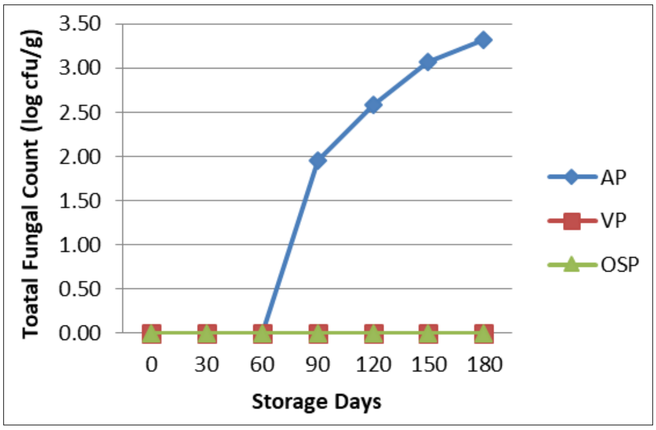

Fig 9: Changes in Total fungal count (log cfu/g) of the dried sardine (*Sardinella longiceps*) during storage.

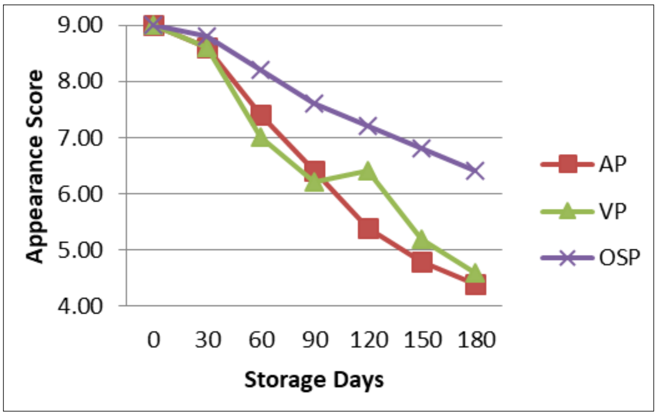

Fig 10: Changes in Appearance score of the dried sardine (*Sardinella longiceps*) during storage.

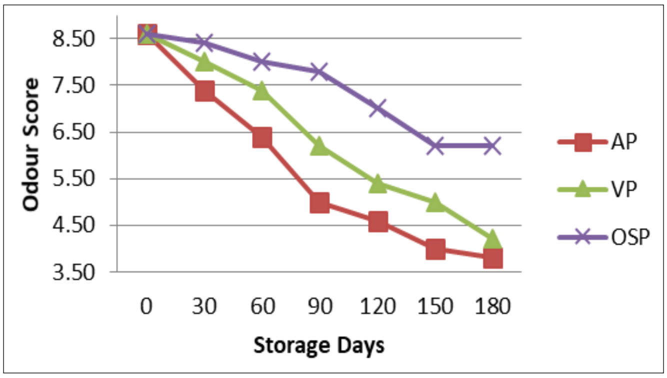

Fig 11: Changes in Odour score of the dried sardine (*Sardinella longiceps*) during storage.

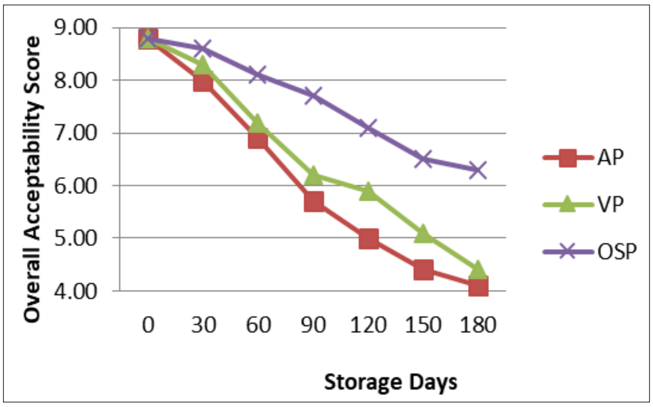

Fig 12: Changes in Overall acceptability score of the dried sardine (*Sardinella longiceps*) during storage.

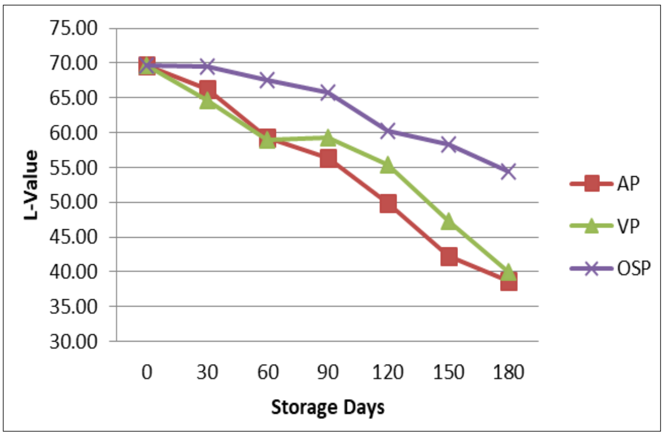

Fig 13: Changes in L*-value of the dried sardine (*Sardinella longiceps*) during storage.

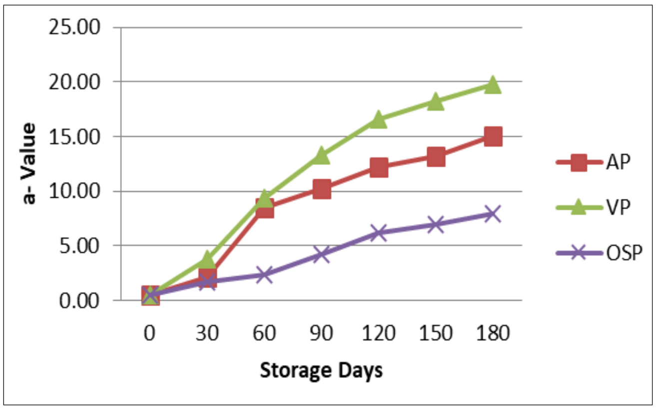

Fig 14: Changes in a*-value of the dried sardine (*Sardinella longiceps*) during storage.

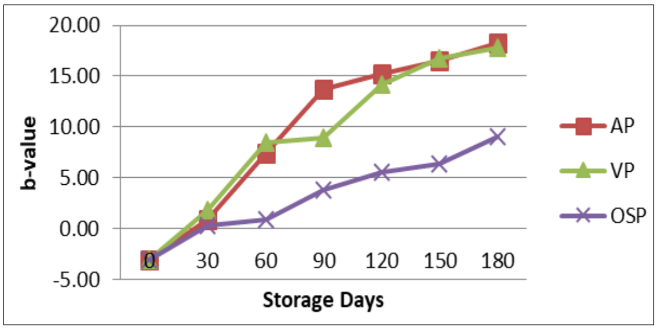

Fig 15: Changes in b*-value of the dried sardine (*Sardinella longiceps*) during storage.

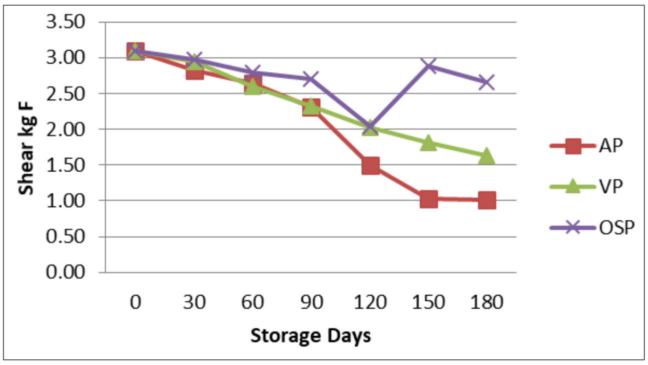

Fig 16: Changes in shear force (Kgf) of the dried sardine (*Sardinella longiceps*) during storage.

Results and Discussion

Dried fish having pH in the range of 6.0-6.9 are considered to be of very good quality [12]. Highest values of pH at the end of storage days were observed in air pouch packed sun dried sample revealing 7.70 for sardine in Figure 1. pH value of dried fishes increased in the time interval due to increase of basic compounds, indicating the loss of quality in fishes. This result is in agree [13], who revealed that there was increase in pH values of marketed sardine during storage. Moisture content is an important factor affecting the ultimate quality and storage life of salt dried fish [14]. It is the major of the total amount of chemically unbound water in sample [15]. Mean value of moisture content increased steadily with increasing storage period in dried sardine storage samples in Figure 2. Increase in moisture content of salt cured fish during storage was observed by researchers [16] and attributed this to absorption of water by complex compounds formed by salt and proteins in the fish tissue. Food Standard Safety Authority of India [17] had specified not less than 16% as the moisture level for dry salted fish. Salting and drying are the interrelated to reduce the moisture sufficiently the decreasing the moisture due to osmotic migration of salt into and water outer the fish [18].

In marine fish, TMA which is formed from trim ethylamine oxide (TMAO) as a result of bacterial enzyme activity, is the main component responsible for an unpleasant ''fishy'' odour [19]. The acceptable of TMA value is 50 mg% for dried fish for human consumption [20]. In present study a relatively high TMA-N value was found in the air packed sardine sample (AP) as compare to vacuum packed (VP) and oxygen scavenger (OSP) in Figure 3. The acceptable of TMA value is 50 mg% for dried fish for human consumption [20]. In case of dried fish, spoilage is considered when TVBN content is 100-200mg% on dry weight basis [21]. Similarly, the value for fresh fish remains 35-40 mgN/100 g [22]. If 100-200 mg% is taken as acceptable in dried fishes, the experimental dried fish samples found to be fit for human consumption. Initial TVB-N value of all dried sardine (*Sardinella longiceps*) was 48.13 ± 0.70 (mg/100g). At the end of storage period TVB-N value were found to be 145.25 ± 0.81, 105.75 ± 0.81 and 97.13 ± 2.39 (mg/100g) in AP, VP and OSP respectively (mean ± SD) (Figure 4). In case of dried fish, spoilage is considered when TVBN content is 100-200 mg% on dry weight basis [21]. Oxygen scavenger packaging was found more effective followed by vacuum packaging and air packaging.

Peroxide value (PV) is used to express the oxidative state of lipid-containing foods. It measures the first stage of oxidative rancidity [23]. Author has suggested that the acceptable level of PV is 20 to 40 millimoles of oxygen/kg of fish fat [24]. Similar higher PV have been reported in dried fishes by several studies [25]. In this study the changes in PV value in all samples of dried sardine showed significant ($p<0.05$) but limited increasing trends during storage period (Figure 5). Increase in peroxide value may be attributed to the oxidation of highly unsaturated fatty acids in fish lipids by the catalytic activity of common salt [26], iron impurities that are probably present in the crude salt, proxidant action of moisture and auto oxidation by atmospheric oxygen [27]. Lowest PV value in oxygen scavenger packed fish sample as compared to other shows the antioxidant effect, thus indicating strong antioxidant effect with vacuum and removal of oxygen by active packaging.

FFA acid progressively increased throughout storage period in all samples (Figure 6) but relatively low in OSP. Author reported that FFA is a measure of hydrolytic rancidity, the extent of lipid hydrolysis by lipase action and that fish oil containing high levels of polyunsaturated fatty acids, is very susceptible to oxidative deterioration at varying velocities, strongly depending on the storage conditions and fatty acid profile [28]. Author suggested that high level of FFA is an indication of microbial spoilage activity [29]. Most fat acidity begins to be visible to the palate when the FFA value calculates as oleic acid is about 0.5 to 1.5% [30]. Increase in FFA values of dried fishes with storage period may be due to hydrolysis of fats along with increase in microbial load as also reflected in PV and TBA values. The TBA was employed for measuring of advanced oxidative rancidity of the product. TBA index is a widely used indicator for the assessment of degree of lipid oxidation, which measures malonaldehyde content [31]. TBA values increased throughout the storage periods in Figure 7. The acceptable limit is from 7-8 mg of malonaldehyde/kg [32]. Fish containing high content of high unsaturated fatty acids such as sardine and mackerel is rapidly oxidized in the presence of oxygen and natural light [33].

Microorganisms play an important role in the spoilage of fish [34]. However, samples packed with oxygen scavenger showed the lowest rate of bacterial growth compare with other packed samples (Figure 8). The result thus indicates that microbiological growths were restricted by oxygen removal and extended the shelf life of sample. Active packed and vacuum samples showed the longest shelf life (180 days) as compared to other samples. The limit of TPC in salted and dried products is $1x10^5$ cfu/g [17]. The absence of total fungal count was observed in treatment OSP (oxygen scavenger packed) & VP (vacuum packed) and 3.31 ± 0.01 log $cfug^{-1}$ found in AP (air packed) at the end of 180 days storage period (Figure 9). Other workers have also reported the presence of fungi in dried fish at 2-4 log $cfug^{-1}$ concentration [35].

Variation in appearance of dried sardine (*Sardinella* *longiceps*) exhibited a decreasing trend during the storage in Figure 10. The reactions for appearance was not known, it could be assumed that excessive microbial and enzymatic proteolysis of the tissue causing tissue disintegration [36]. Development of odour in stored dried sardine (*Sardinella longiceps*) are shown in Figure 11. The result showed characteristic natural odour in initial period of storage but it was lost during the storage (Figure 11). The oxidation of lipids in fish muscle usually develops off-flavor and off-odor that ultimately affect the storage time [37]. The sensory characteristics of fish are clearly visible to the consumer and are essential for consumer satisfaction [38]. Overall quality is a critical factor in determining the acceptability of dried fish products. The reduction in the sensory qualities with increase in the storage period of processed fish could be attributed to higher activities of the spoilage agents. Similar trend was observed in dried fish under storage [39]. Oxygen scavenger packed dried sardine was comparatively better than air packed and vacuum packed samples (Figure 12). Active packaging of sardine maintaining certain quality is obvious.

Colour is the important physical characteristics of fish products and directly related to the acceptability of food products. The color of any product can be represented in terms of tristimulus L *, a *, b *, or by a combination of them, depending on the nature of the pigment present in the foodstuff [40]. Therefore decrease in *L* value indicates a decrease in whiteness. 'L'-value of dried sardine showed decreasing trend during the storage (Figure 13). Authors reported decrease in brightness (L*) ($p < 0.05$) during storage in the samples of dehydrated chub mackerel (*Scomber japonicus*) stored at 25 and 35°C [41]. Authors observed that the deterioration of colour of herring fillets could be related to the changes in proximate composition of the fish muscle [42]. The maximum value for *a** is +127 indicating redness. The changes in 'a'-value in dried sardine showed decreasing trends toward redness with the increasing storage periods (Figure 14). This may be attributed to oxidation and denaturation of myoglobin [43]. The maximum value for *b** is +127 indicating yellowness. Yellow color is associated with lipid oxidation in muscle foods such as fish [40]. 'b'-value of dried sardine showed increasing trend during the storage (Figure 15). The interaction effect of different packaging style and storage period (days) were found to be significant. The Warner-Bratzler apparatus is widely known as a device for tenderness measurement of livestock meat [44]. Dried fish meat has a heterogeneous structure. That is, the surface became harder than its inside as the moisture evaporation proceeded [45]. Decrease in shear force during the storage period (Figure 16) may be due to proteolysis or weakening of connective tissue and also increasing moisture content in dried fish during storage period.

Conclusion

The results from the present study suggests that traditi onally dried sardine (*Sardinella longiceps*) performed well in terms of quality and safety as stored products when packed with oxygen scavenger (active packaging). It was observed that the active packaging using oxygen scavenger in the packaging of dried fish, may assure an effective packaging for dried fish, quality for long term storage. Quality losses due to fatty oxidation in fatty fish are important for a traditional salted dried fish product. While the preferred vacuum packaging for fatty dried fish requires equipment, active packaging by oxygen scavenger, appears to be the most effective and convenient to packaging of dried fish products sensitive to fatty oxidation, as well as other deteriorative changes detrimental to quality during long term storage and transportation. When comparing types of packaging, it was concluded that the active package, which is a new approach, is the best option to vacuum packaging and air packaging in terms of quality.

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