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Quality changes in fish balls prepared using spice mix cooked both in microwave and conventional oven stored under refrigerated condition

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Abstract
In the present study an attempt has been made to prepare ready to eat fish balls which were packed in stand up pouches and its shelf life was studied at 4°C after an interval of 4 days each. The organoleptic scores for overall acceptability of the fish ball were slightly decreased within the storage period for spiced and non-spiced ones (12 days) cooked both in microwave and conventional oven. With the increase in storage period the pH value increased non significantly in all the treatments. Products cooked in microwave oven contained more TBA value than conventionally cooked ones irrespective of addition of spices and storage duration. Methods of cooking showed non-significant effects on bacterial load. Proliferation of microbes was noted during storage and TPC increased significantly on 12th day of storage which is within acceptable range for spiced products but non-spiced products deteriorated completely. The increase in coliform count up to 4th day of storage was non-significant. It was observed that the products stored at 4°C were acceptable organoleptically and stable microbiologically up to 9 days in non-spiced ones and up to 12 days in spiced fish ball.

Keywords: catla catla, fish ball, organoleptic, microbiological evaluation, refrigerated storage

Introduction
Fish products are perceived by consumers to be healthful and nutritious food because of plethora of scientifically substantiated and documented health benefits. Fish and fishery products play an important role in human nutrition as a source of proteins, fatty acids, fat-soluble vitamins macro and trace elements (Belitz *et al.*, 2009). However, fish is a highly perishable commodity and its post-harvest handling and processing being labor intensive, result in increased microbial contamination. There is an increasing growth in the demand for convenience ready-to-cook/eat minimally processed meat products in both developed and developing countries. Ready-to-cook/eat meat products are susceptible to environmental contamination with spoilage and pathogenic microorganisms during handling of these products after cooking and before or during packaging. There are numerous food processing tools available that provide food protection. Individually frozen fish and fish products are available in the market. However, fish and fishery products can undergo undesirable changes during refrigerated storage and deterioration may limit the storage time. The aim of this study was to investigate the quality changes during refrigerated (4^0 C) storage. The microbial evaluation was done to make the present findings a wholesome one in order to know the microbial load of spiced and non-spiced fillets and balls cooked conventionally and in microwave oven. The present investigation was therefore undertaken to assess the microbial stability of the products, in terms of total plate count (TPC) and coliform counts on different treatments along with sensory attributes of fish fillet and ball.

Material and Methods
Sample preparation
Fresh catla (*Catla catla*) were obtained from local fish market, Hisar, Haryana (India). After purchase, the fish were transferred to the laboratory in polystyrene boxes with crushed ice within 1 h. On arrival at the laboratory, quality control analysis of the fish were performed and balls were prepared. The fish were gutted, filleted and washed. After the handling process, the fish were minced using a 3 mm diameter holes plate. Ingredients were added to the minced fish. The ingredients were homogenized with a kitchen blender. The mixture was kept in a refrigerator at 4º C for 1 h and processed into fish balls (1.5 cm thick and 100 mm diameter) by using a metal shaper. Balls of approximately 25 g in weight were made. Balls were then cooked in conventional and microwave oven.

Microbiological evaluation
Microbiological condition of the product prepared by conventional cooking and microwave cooking using fish meat after addition of additives and spices was evaluated at 0, 4, 8 and 12 days of storage at 4 ± 1°C. Total plate count and coliform count was done according to the method prescribed by APHA (1966) [1].
Method of Trout *et al.* (1992) [9] was followed for determining the pH of sample.

Organoleptic evaluation
All the processed products were subjected to sensory evaluation. The organoleptic evaluation was conducted by a

semi-trained panel for color, flavor, texture, taste, appearance and over all acceptability. The judges scored quality characteristics of each sample on 9 point hedonic rating scale (Annexure-1). Score of 9 indicated "very desirable" trait, which gradually decreases with reduction in acceptability. The score of five shows "neither desirable nor undesirable trait. The characteristics with mean score of 6 and above were considered acceptable. The mean scores obtained after evaluation of different samples for each trait were calculated (Ranganna, 1986) [6].

Results and Discussion

Changes in pH value

With the increase in storage period the pH value increased non-significantly in all the treatments. The mean value pH in fish products for different treatments increased and indicated that there was non-significant difference in pH values of different treatments on various storage days i.e. at 0,4,8 and 12 days throughout the storage.

Variation in Total plate Count

The data pertaining to microbial evaluation of fish ball during storage is presented in Table 7.1 (Fig. 7.1). The highest value of TPC at 0 day of cooking was found in P1S1M1 (2.28 log cfu/g) followed by P1S0M1 (2.23 log cfu/g), P1S0M0 (2.11 log cfu/g) and P1S1M0 (2.04 log cfu/g). TPC of balls in both kinds of cooking did not differ significantly and ranged from 2.04 log cfu/g (P1S1M0) to 2.28 log cfu/g (P1S1M1). Proliferation of microbes was noted at storage intervals and TPC increased significantly on 12th day of storage. At 4th day of storage highest count was found in P1S0M0 (4.44 log cfu/g).

Table 1: Effect of different treatments as affected by spices and method of cooking on total plate count (log cfu/g) of fish ball during storage

Treatment	Total plate count (log cfu/g)				
	Storage period (days)				
	0	4	8	12	Mean
P1S0M0	$_{a}2.11^{c}\pm0.69$	$_{a}4.44^{b}\pm0.65$	$_{a}6.33^{a}\pm0.53$	-	4.29
P1S0M1	$_{a}2.23^{c}\pm0.73$	$_{a}4.26^{b}\pm0.88$	$_{a}6.12^{a}\pm0.57$	-	4.21
P1S1M0	$_{a}2.04^{d}\pm0.68$	$_{a}3.39^{c}\pm0.68$	$_{b}5.06^{b}\pm0.73$	$_{a}6.51^{a}\pm0.69$	4.25
P1S1M1	$_{a}2.28^{d}\pm0.68$	$_{b}3.33^{c}\pm0.89$	$_{b}4.83^{b}\pm0.63$	$_{a}6.26^{a}\pm0.78$	4.18
Mean	2.19	3.84	5.63	6.35	
CD at 5%	p×s×M×D = 0.829, Duration (mean) = 0.030, p×s×M (mean) = 0.414				

P1S0M0 (Balls without Spice cooked in Conventional oven)
P1S0M1 (Balls without Spice cooked in Microwave oven)
P1S1M0 (Balls with Spice cooked in Conventional oven)
P1S1M1 (Balls with Spice cooked in Microwave oven)
Mean values are the average of 6 replications
Similar superscripted letters along the row and subscripted letters along the column depicts non-significant difference

The trend in variation of TPC in balls were almost identical with respect to method of cooking and addition of spice mix. Balls with spice possessed higher TPC than without spice P1S0M1 (4.26 log cfu/g). At 8th day of storage, both the methods for preparation of balls with or without spice exhibited almost similar trend as that of 4th day. Excluding the non spiced products which were completely spoiled at 12th day the highest count (6.51 log cfu/g) was found in P1S1M0. The trend achieved in ball at 12th day was such that conventional oven cooked (P1S1M0) balls had higher TPC (6.51 log cfu/g) than microwave cooking (6.26 log cfu/g). As the storage duration increased TPC also increased significantly in each treatment from 0 to 12 days of storage. The mean value for TPC increased from 2.04 log cfu/g (P1S1M0) on zero day to 6.51 log cfu/g (P1S1M0) on 12 days of storage.

Table 1 illustrated that method of cooking had non-significant effect on TPC throughout the storage duration and it is evident that at 0 day, microwave cooked balls possessed higher TPC than conventionally cooked (non-significant). Sharma (1997) [8] also reported non-significant difference in TPC in chicken patties cooked by conventional and microwave oven. Mendiratta *et al.* (1998) [5] reported higher TPC in microwave cooked than gas oven cooked chicken meat. At 0 day higher TPC was found in spiced products, which might be due to initial microbial load of spices which contributed to higher TPC in spiced balls. Whereas, due to extended period of storage non spiced products were found to have higher TPC than spiced products; particularly at 4th day where non-spiced balls exhibited highest counts, thereby confirming the antimicrobial action of the spice ingredients owing various microbial inhibitory substances like cinnamic aldehyde in cinnamon and eugenol in cloves. Sethi and Anand (1984) [7] also observed various degrees of inhibition against microbial contamination and growth when essential oils and oleoresins from most of the spices were used in product formulation. They also observed that spices either in powder or oils are known to control microbial spoilage. The increase in TPC with the increase in storage duration was also reported by Cunningham and Bowers (1977) [3] who found that initial total count of fresh chicken patties was less than 10^4 /g and after storage of 10 days at 3˚C count increased to 7 x 10^6/g. Proliferation of microbes was noted during storage and TPC increased significantly on 12th day of storage which is still confined for being the acceptable range was also in agreement with the finding of Sharma (1997) [8]. Additives and spice mix used in the study for preparation of balls helped to extend their shelf life at 4±1ºC temperature by their antimicrobial action.

Changes in Coliform during storage

The perusal of data regarding changes in coliform count (log cfu/g) of fish ball as affected by method of cooking, spice mix and duration of storage are presented in Table 7.3. At 0 day of

fish ball preparation highest coliform count was reported in P1S0M1 (0.78) followed by P1S0M0 (0.72), P1S1M0 (0.60) and P1S1M1 (0.56 log cfu/g). At 4th day of storage highest count (1.38 log cfu/g) was found in P1S0M1, this shows spices produced their antimicrobial effects leading to lowest count in spiced conventional oven cooked ball and highest in non-spiced microwave cooked ball. At 12th day, non spiced products get deteriorated completely indicating that spices must have shown their antimicrobial effects in the treatments containing spice mix. With the increase in storage duration coliform count increased non-significant till 4th day of storage and significant difference was observed from 8th day of storage in P1S1M1 (1.38 log cfu/g). At 12th day non spiced products were completely spoiled and spiced products contained a moderate amount of coliform counts. Table 2 suggests that the increase in coliform count up to 4th day of storage was non-significant. Application of heat processing (conventional and microwave oven) stopped the twitter increase in coliform counts. Out results are also in agreement with the findings by Gashti (2002) [4], that the coliforms are killed during heat processing of products leading to lower counts. The reason for the spoilage of non-spiced product at 12th day of storage could be higher proliferation of bacteria where spices were not utilized as ingredients.

Table 2: Effect of different treatments as affected by spices and method of cooking on coliform count (log cfu/g) of fish balls during storage

Treatment	Coliform count (log cfu/g)				
	Storage period (days)				
	0	4	8	12	Mean
P1S0M0	$_{a}0.72^{ab}\pm0.08$	$_{a}1.23^{ab}\pm0.07$	$_{a}1.82^{a}\pm0.05$	-	1.26
P1S0M1	$_{a}0.78^{ab}\pm0.08$	$_{a}1.38^{ab}\pm0.06$	$_{a}1.87^{a}\pm0.05$	-	1.33
P1S1M0	$_{a}0.60^{ab}\pm0.06$	$_{a}1.14^{ab}\pm0.06$	$_{a}1.49^{a}\pm0.05$	$_{a}1.91^{a}\pm0.05$	1.29
P1S1M1	$_{a}0.56^{ab}\pm0.08$	$_{a}1.20^{ab}\pm0.07$	$_{a}1.38^{a}\pm0.09$	$_{a}1.82^{a}\pm0.05$	1.24
Mean	0.69	1.22	1.64	1.89	
CD at 5%	p×s×M×D = 0.743, Duration (mean) = 0.263, p×s×M (mean) = 0.372				

Organoleptic Quality Evaluation

After evaluating organoleptic quality of the products better scores in terms of color was observed in conventional oven cooked spiced balls at the time of preparation and even after 12 days of storage. In conventional oven cooking due to maillard reaction browning takes place which imparts better color to the products. Spices further aggravated the color in the products. Due to proper mixing of fish mince, spice and oil, better color development appears in fish balls. It can be stated that microwave oven products just after cooking achieved better flavor scores than conventionally cooked products which underwent more flavor deterioration due to oxidative rancidity during storage. Our study showed that the spiced microwave cooked products resulted in better texture than without spice products cooked conventionally. On the basis of the results obtained through sensory evaluation it can be concluded that all the organoleptic traits deteriorated with the days of storage. However, at 12th day of storage non spiced products get completely spoiled irrespective of color, flavor, texture, appearance and taste. This shows that addition of spice mix helped in better enhancement of the shelf life along with overall product acceptability.

Conclusion

Spice mix application was found to be most desirable in enhancing the microbiological quality of fish products by reducing the microbial load. This might be due to the antimicrobial action of spice ingredients containing microbial inhibitory substances like cinnamic aldehyde in cinnamon and eugenol in cloves in the spice mix. However, methods of cooking and products showed non significant effects on bacterial load. Proliferation of microbes was noted during storage and TPC increased significantly on 12th day of storage which is within the acceptable range for spiced products but non spiced products deteriorated completely. The increase in coliform count up to 4th day of storage was non significant. This indicates that raw fish procured was of acceptable quality and the hygienic conditions of the laboratory were also adequately maintained during product preparation. Application of heat processing (conventional and microwave oven) restricted the increase in coliform counts. The reason for the spoilage of non-spiced product at 12th day of storage could be higher proliferation of bacteria where spices were not utilized as ingredients. Additives and spice mix used in the study for preparation of fillets and balls helped to extend shelf life by their antimicrobial action.

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