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COMPARATIVE ASSESSMENT OF MICROBIAL LOAD OF RAW FISH MUSCLE (*LABEO ROHITA)* SUBJECTED TO VARIOUS PRESERVATION TECHNIQUES

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ABSTRACT

The present study was aimed to determine the influence of different preservation methods viz. refrigeration at -12±2^{0}C (G-I); freezing at -20±2^{0}C (G-II); sun drying (G-III) and oven drying (G-IV) on the microbial profile of raw *Labeo rohita* muscle for a period of 7 weeks. The results revealed that after 7 days of storage,the microbial load increased gradually in all the preserved samples. The increase was found in the following trend i.e. G-IV<G-III<G-II<G-I. The Total Plate Count (TPC) was found to be within the permissible limits (6 log cfu/g) till the end of experiment in G-IV whereas it crossed the permissible limit on 42nd day in G-III, 28th day in G-II and 14th day in G-I. The value of Coliform Count (CC) was found to be negligible in G-IV; it crossed the permissible limits on 49th day in G-III, on 28th day in G-II and on 14th day in G-I. Hence, on the basis of present data on the microbial quality of fish muscle, the oven dried fish muscle can be suggested to be safest for human consumption as compared to other preservation techniques.

KEY WORDS: -12±2^{0}C, -20±2^{0}C, sun drying, oven drying, microbial analysis, *Labeo rohita*.

1. INTRODUCTION

The rapid escalation in human population combined with the parallel shortage of animal protein has directed the attention to fish as a healthy compensatory source of good quality animal protein. In addition to high percentage of protein, fish muscle is endowed with various essential nutrients in the form of vitamins and minerals required for supplementing both infant and adult diets. Despite its high nutritional worth, fish is extremely perishable commodity. Microbial action has been known to play a large part in the spoilage of fish which results in substantial amount of quality loss before its consumption. Delay in microbial contamination can be achieved by combination of various techniques.

One of the most widespread methods of preserving fish is by means of freezing. Different freezing temperatures are known to have different effects on the shelf life extension of fish. Low temperature aids by making micro-organisms inactive, slowing down the enzymatic activity and thus causing a decline in the biochemical activities thus keeping the fish free from spoilage for longer duration.[1] Although many damaging processes are inhibited by such low temperature storage methods, but the undesirable reactions associated with lipids and proteins are shown to occur, leading to detrimental changes in nutritional and sensory properties of fish.[2]

Salting in combination with smoking and drying is one of the most traditional methods of preservation since prehistoric times. Drying reduces or completely eliminates physiological, microbial and enzymatic degradation of biological materials such as fish.[3] Fish drying checks the growth of microbes as well as prevents chemical degradation in fish muscle. Drying works by reducing the moisture content of a material to considerably low levels. Wide availability of dried fish in market reflects the popularity of this traditional food among masses. Further the quality of fish dried in an electrically-operated oven varies from that of fish dried under the sun in open air.

The present study was therefore conducted with the objective to assess the influence of different preservation methods viz. freezing (at -12±2^{0}C and -20±2^{0}C) and drying (sun drying and oven drying) on the microbial profile of raw *Labeo rohita* fish muscle.

2. MATERIALS AND METHODS

2.1 Collection of fish samples: Fresh *Labeo rohita* samples were procured from local market of Jammu city. They were immediately brought to the laboratory in polythene bags with crushed ice.

2.2 Sample processing: The viscera of fish were removed and the fish was washed with large amount of water to remove blood residues and dirt particles. Then, it was cut into fillets of uniform size. The raw fish fillets were washed properly, wrapped in aluminum foil, kept in air tight plastic container followed by storage at -12±2^{0}C **(G-I)** and -20±2^{0}C **(G-II).** Some of the raw fish fillets were subjected to dry salting in a salt to fish ratio of 1:3 for 24 hrs. Half of the salted samples were subjected to sun drying **(G-III)** under bright sun light in open air by placing them on an elevated rack. The remaining half of the salted samples was subjected to oven drying **(G-IV)** with an initial temperature of 40^{0}C for 1 hr following which the temperature was raised to 55^{0}C. Dried samples were obtained after 24 hours.

After drying, the G-III and G-IV samples were allowed to cool down, packaged tightly in separate plastic bags, labeled and stored at room temperature (27^0-31^{0}C).

The complete experiment was carried out for a period of 7 weeks. Analytical procedures for microbiological changes were done after every 7 days.

2.3 Analysis: The microbiological profile was determined according to APHA method (1984).[4] Data was expressed as mean ± SD and were analyzed by one-way ANOVA test using SPSS statistical programme.

2.4 Statistical Analysis: Mean and standard errors were calculated for different parameters. The data analyses were performed using SPSS software (12.0 for Windows).

3. RESULTS AND DISCUSSIONS

3.1 Microbial Analysis - To assess the microbial load of preserved samples Total Plate Count (TPC), Coliform Count (CC) and Psychrophilic Count (PC) were analyzed for a period of 7 weeks.

The bacteriological changes in all the four groups has been shown in following tables:

Table 1: Bacteriological changes in raw muscle of *Labeo rohita* kept under frozen conditions at -12±2^{0}C (G-I).

Days of storage	0 day	7th day	14th day	21st day	28th day	35th day	42nd day	49th day
TPC*	2.88±0.09	**6.79±0.11**	8.17±0.07	9.95±0.02	12.11±0.30	14.91±0.57	16.15±0.28	20.06±0.01
CC**	1.20±0.15	1.98±0.08	**2.85±0.07**	4.11±0.22	6.19±0.1	7.99±0.02	9.16±0.07	14.12±0.02
PC***	2.12±0.44	3.36±0.04	**4.84±0.1**	5.98±0.14	6.21±0.06	8.16±0.27	9.01±0.08	12.25±0.05

Table 2: Bacteriological changes in raw muscle of *Labeo rohita* kept under frozen conditions at -20±2^{0}C (G-II).

Days of storage	0 day	7th day	14th day	21st day	28th day	35th day	42nd day	49th day
TPC*	2.75±0.2	3.01±0.11	4.30±0.23	5.10±0.06	**6.20 ±0.01**	7.50±0.08	8.08±0.1	9.10±0.03
CC**	1.08±0.15	1.92±0.2	2.04±0.1	2.53±0.09	**3.30±0.4**	4 4.4.27±0.11	5.29±0.19	6.85±0.12
PC***	2.04 ±0.12	3.12±0.04	3.72±0.08	4.00±0.44	**5.12 ±0.05**	6.09 ±0.12	7.15±0.05	8.66±0.44

Table 3: Bacteriological changes in experimentally sun dried muscle of *Labeo rohita* kept at room temperature (G-III).

Days of storage	0 day	7th day	14th day	21st day	28th day	35th day	42nd day	49th day
TPC*	1.91±0.01	2.44±0.12	2.90±0.8	3.28±0.76	4.65 ±0.09	5.71±0.03	**6.35 ±0.07**	7.04 ±0.08
CC**	0.47±0.03	0.89±0.5	1.04±0.01	1.18±0.12	1.65±0.06	1.80±0.02	2.47±0.07	**3.09±0.11**
PC***	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0

Table 4: Bacteriological changes in experimentally oven dried muscle of *Labeo rohita* kept at room temperature (G-IV).

Days of storage	0 day	7th day	14th day	21st day	28th day	35th day	42nd day	49th day
TPC*	1.04±0.04	1.35±0.5	1.84±0.15	2.12±0.09	2.87±0.41	3.32±0.18	3.90 ±0.06	4.8±0.20
CC**	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
PC***	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0

3.1.1 Total plate count (TPC)

Table 1 to 4 depict a gradual increase in TPC of all the groups (G-I, G-II, G-III and G-IV) from 0 day to 49th day.The increase was more rapid in G-I as compared to other groups. On the last day of storage i.e. 49th day, it increased up to 20.06±0.01 log cfu/g in **G-I**; 9.10±0.03 log cfu/g in **G-II**; 7.04±0.08 log cfu/g in **G-III** and 4.8±0.20 log cfu/g in **G-IV**. The TPC of **G-IV** was found to be within the permissible limits till the last day i.e. 49th day of storage (4.8±0.20 log cfu/g) whereas in **G-III** it was within the permissible limits on 35th day of storage (5.71±0.03 log cfu/g). On the other hand, in **G-II,** TPC was within the permissible limit on 21st day (5.10±0.06 log cfu/g) whereas in **G-I**, the maximum permissible limit of **6 log cfu/g** was crossed on 7th day (6.79±0.11log cfu/g).[5-9]

3.1.2. Coliform count (CC)

The CC shows an increasing trend with the increase in storage period in the groups - G-I, G-II and G-III from 0 day to 49th day. Among these three groups, the increase was most rapid in **G-I** i.e. raw muscle stored at -12±2°C.

At the end of storage i.e. 49th day, it escalated up to 14.12±0.02 log cfu/g in **G-I**; 6.85±0.12 log cfu/g in **G-II** and **3.09±0.11** log cfu/g in G-III. In oven dried muscle stored at room temperature **(G-IV),** the coliform count was found to be negligible throughout the study period of 49 days. In **G-III,** CC crossed the permissible limits on 49th day of storage (3.09±0.11 log cfu/g); whereas in **G-II,** CC was within the permissible limit on 21st day (2.53±0.09 log cfu/g) while in **G-I**, the maximum permissible limit of **2.69 log cfu/g** was crossed on 14th day (2.85±0.07 log cfu/g). Similar results in the Coliform Count of different fish samples was reported by many researchers.[10-14]

3.1.3. Psychrophilic count (PC)

The Psychrophilic Count showed an increase with increase in storage time in G-I and G-II samples (Table 1 and 2). The increase was more rapid in raw muscle stored at -12±2°C **(G-I)** as compared to raw muscle stored at -20±2°C **(G-II)**. It reached up to 12.25±0.05 log cfu/g in G-I and 8.66±0.44 log cfu/g in G-II by the end of 49 days. In G-II, the maximum permissible limit of **4.6 log cfu/g** was crossed on 28th day (5.12±0.05) while in G-I it was crossed on 14th day (4.84±0.1 log cfu/g) of storage.[15-19]

4. CONCLUSION

Thus, the following trend emerges in terms of microbial load:

G-IV< G-III< G-II < G-I

This clearly depicts that the method of oven drying is most effective in retarding the growth of microorganisms in the fish muscle and extends its shelf life.

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