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# Preparation of Co-Dried Fish Silage by Using Fish Market Waste and Its Comparative Study

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**ABSTRACT**

*In the present study biological method with 15 % molasses and 10% curd were used for preparation of fish silage. The important findings are summarised as: the chemical analysis of fish market waste was observed on wet basis to be moisture 77.09 ± 0.14 %, crude protein 15.20 ± 0.15 %, lipid 4.03 ± 0.07 % and ash 3.30± 0.11 %. Rice bran was used as co-drying material for drying liquid silage. The proximate composition of rice bran contained moisture, crude protein, fat and ash as 9.45 ± 0.19 %, 16.05 ± 0.08 %, 13.42 ± 0.15 % and 10.44 ± 0.14 % respectively on dry weight basis. Powder fish silage was prepared by mixed liquid silage with different quantity of rice bran. For neutralizing the liquid fish silage 1.5% sodium carbonate was added and measured pH value was 6.45. Mixtures were dried within 2-3 days in solar tunnel drier. The proximate composition of powder fish silage made with different quantity of rice bran 10, 20, 30, 40 and 50% contained moisture content as 13.74 ± 0.12, 12.54 ± 0.30, 10.91 ± 0.13, 10.18 ± 0.05, 9.18 ± 0.02 % respectively; crude protein content as 29.50 ± 0.22, 28.56 ± 0.12, 27.66 ± 0.10, 26.53 ± 0.18, 25.73 ± 0.08 % respectively; fat content as 16.28 ± 0.11, 15.71 ± 0.14, 14.45 ± 0.11, 13.73 ± 0.17, 12.60 ± 0.10% respectively; ash content 14.21 ± 0.12, 14.55 ± 0.11, 15.27 ± 0.13, 15.65 ± 0.15, 15.99 ± 0.12 % respectively. Considering all the limitation, powder fish silage with 30 % rice bran was found to be better and carried for further storage study. During storage, moisture content of powder silage was increased in the range from 10.91 ± 0.14 to 11.15 ± 0.10 ,whereas the protein content was decreased in the range from 27.66 ± 0.10 to 27.04 ± 0.06 %, fat content decreased in the range from 14.45 ± 0.11 to 13.42 ± 0.10 %, ash content decreased in the range from 15.27 ± 0.11 to 14.60 ± 0.09 %, Fiber content decreased in the range from 9.89 ± 0.06 to 9.64 ± 0.07 %, Carbohydrate content increased in the range from 21.82 ± 0.07 to 23.14 ± 0.11 %, pH increased in the range from 6.45 ± 0.05 to 6.59 ± 0.14 and TPC increased in the range from 2.10×10$^4$ to 2.36 ×10$^4$ cfu/g during the storage period from 0 to 90 days.*

*From above study it can be concluded that fish market waste is suitable for preparation of fish silage powder. At room temperature powder fish silage could be stored up to more than 3 months without loss of major nutrient components.*

***Key words:*** *Fish silage, Co-drying, Rice bran and proximate composition.*

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## INTRODUCTION

During processing of sea foods large amount of fish wastes and deteriorated whole fishes are daily discarded in the fish markets. Fish waste, includes frames and rests from trimming, guts, skins, fats, viscera, roes/eggs, heads, breasts, scales and deteriorated filets. A fish contain 45 % flesh, 24-27 % head, 12 % skeleton, 3 % skin, 4 % cut off and 12 % viscera including eggs, milts and liver of its total body weight[10]. These wastes are a potential source of pollution and contamination of the environment, as they degrade rapidly in warm temperatures. If it is not appropriately stored or managed, it creates aesthetic problems and strong odours due to bacterial decomposition. On the other hand, they contain high amount of nutrients such as protein, fat and minerals which is available in low cost. So that, there is need for developing new methods for biotransformation of this fishery waste into animal feed to reduce aqua production cost.

The best alternative way of utilizing fish waste material is the production of fish silage which does not release any off odour during preparation[19]. The product has a good nutritive quality and can be sufficient for animal feeding. This procedure is safe, cost effective, eco-friendly and has a good nutritive quality which can be adequate for animal feeding[15]. Fish silage is defined as a liquid product produced from the whole fish or parts of it, to which acids, enzymes or lactic acid-producing bacteria are added, with the liquefaction of the mass provoked by the action of enzymes from the fish[12]. The disadvantage of fish silage is that it has high water content make it bulky and difficult to transport and store. The most effective method is to co-dried fish silage with other dry ingredients or other filler materials for convenient use. Co-drying is a process where dry products are added to the wet silage to absorb the solublized protein and some of the moisture[11]. Co-dried fish silage used as an aqua feed ingredient that is easy to package, stored, and transport. Due to the similarity of the protein source with the raw material and low cost, especially when compared to fish meal, silage has a potential use in aquaculture[27].

Silage can be effectively used for animal feeding, like powder fish silage is used to feed beef cattle, milk cow, swine, sheep, mink and many other terrestrial animals[22,1,2]. Fish silage can be used as a protein source for broiler chicks. Replacing fish meal protein by fish silage protein resulted in similar or increased weight gain and feed conversion ratio in broiler chicks, slaughtered at 4 or 5 weeks of age[17]. In many countries, it is used as bird feed[6]. It can be very vitally used as a feed supplement in aquaculture to convert nutrients into flesh. Powder fish silage was found to give better growth than fish meal when fed to carp[9]. In case of pink abalone, *Haliotis fulgens*, powder fish silage has been used as an alternative to natural food (microalgae)[26]. Fish silage could also find use as a fertilizer, and trials with various vegetables on different types of soil have been underway for several years at research station of many countries[8].

## MATERIAL AND METHOD

Fish market wastes were procured from fish market of Ratnagiri. The fish mainly consisted of heads, tails, gills, fins and visceras of Sardine (*Sardinella Fimbriata, Sardinella longiceps*), Mackerel (*Rastrelliger kanagurta*), Tuna (*Euthynnus affinis, Auxis thazard*), Pink perch (*Nemipterus japonicus*), Ribbon fish (*Trichiurus lepturus, Lepturocanthus savala*), Bombayduck (*Harpadon nehereus*), Seer fish (*Scomberomorus guttatus, Scomberomorus commerson*), Mullet (*Mugil cephalus*). The collected fish wastes were washed with

potable water and were grinded into paste by using mixer. The resulting compound fish waste was used as raw material for preparation of silage

Curd was used as lactic acid bacteria source for preparation of fermented silage**.** Curd was prepared fresh from boiled and cooled milk and kept for overnight. Lactic acid bacterial (LAB) count of curd was estimated by pour plating technique on MRS agar and Lactic Acid Bacteria (LAB) count of 4.20 x $10^6$ cfu /gram used for preparation of biological silage. Rice bran was used as co-dried material for preparation of dried fish silage powder. It was collected from Joshi rice meal factory, Basani, Tal. Dist. Ratnagiri.

**Method for preparation of dried fish silage powder[16]:**

500 g of minced fish waste was poured in plastic container. Then, sugar cane molasses, Butylated Hydroxyl Toluene (BHT) and water were added in container with weight percentages of 15% (v/w), 65 mg and 30% (v/w) respectively. Buckets were stirred properly. After this sample was heated in water bath for 15 minutes at 90 °C and then cooled at a room temperature. Then curd (starter culture) with a weight percentage of 10 % (w/w) was added to them. Mix the samples thoroughly and stored in airtight plastic container. Silage was kept for 10-12 days at ambient temperature. After 10-12 days, 1.5 % of sodium carbonate $Na_2CO_3$ was used for neutralised pH of silage. After neutralising the pH of the 15% of biological silage, rice bran were added 10%, 20%, 30%, 40%, 50% respectively to prepare a semidry feed. The semidry fish silage was spread on the plastic film and dried in a solar tunnel drier for further used. Traditional mortar and pestle or mixer was used for grinded the dried fish silage powder. Proximate composition of the dried silage samples were analysed (10%, 20%, 30%, 40% and 50% respectively.).Cooled powder mixture was packed in air-tight polythene (LDPE) packets and packets were sealed by using sealing machine. Packets were kept at room temperature to study the shelf life of the product up to 3 months. During storage, changes in proximate composition, pH and TPC were observed.

**Proximate compositions:**

Proximate composition i.e. Moisture, crude protein, crude fat, ash, fiber content and pH of prepared dry silage powder were determine as per standard methods of[4]

The carbohydrate content was determined by subtracting the summed up percentage compositions of moisture, protein, lipid, fiber, and ash contents from 100[18].

**Microbiological quality**

**Enumerations of total plate count (TPC)**

Samples were analysed for total plate count (TPC) by the method recommended by APHA[5]. The TPC was founded as under

Total plate count cfu /g =Avg. of Count in duplicate plates x dilution factor.

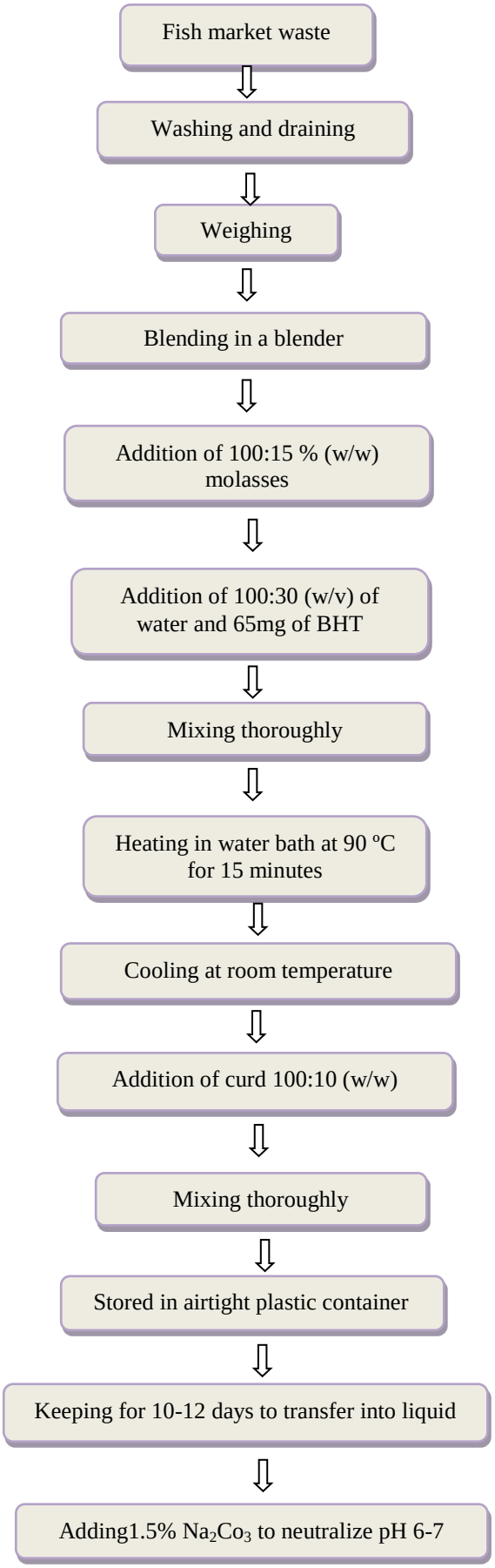
**Lactic acid bacteria count:**

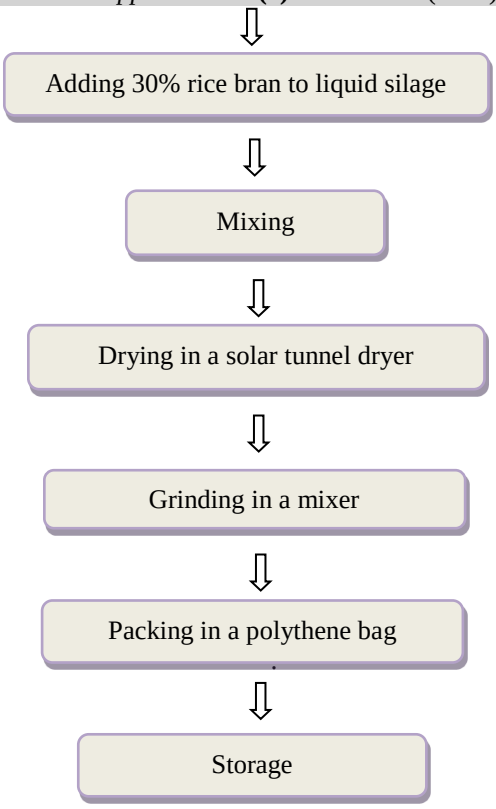
Appropriate dilutions were plated by pour-plate technique in duplicate using MRS medium[3] and were incubated at room temperature for 48 hours.

**Statistical analysis:**

The data were analysed to test significance difference by applying analysis of variances (ANOVA) tool available in MS-EXEL 2010. The significant differences were tested by 5% level of significances and are mentioned as $p < 0.05$ for significance difference[24].

**Method for preparation of powder fish silage[16]**






## RESULTS AND DISCUSSION

In present study fish market waste can successfully converted into powder fish silage by using locally available cheap sources of molasses and curd for lactic acid fermentation. Biological silage with 15% molasses was used for preparation of fish silage powder. Fermented silage would be a good potential source for animals in arid regions as it contained high protein[1,20,21]. Rice bran was used as co-drying material for drying liquid fish silage. This process is safe, economical and eco-friendly as compared to fish meal. Various Biochemical and microbiological quality changes in different percents of powder fish silage during storage have been discussed in this section.

The biochemical and microbiological characteristics of fish waste were analysed during the initial stage of present work. The fish waste used for the present work was having acceptable fishy smell and appearance.

**Proximate composition**

Proximate composition of fish waste on wet basis is shown in Table no. 1.1.and Fig.1.1. Fish waste contained moisture 77.09 ± 0.14 %, crude protein 15.20 ± 0.15 %, fat 4.03 ± 0.07 % and ash 3.30 ± 0.11 % and NFE (Nitrogen Free Extract) 0.38 % ±.0.06. Similar results were found by Palekar[21] depicted moisture 79.47 %, protein 15.78 %, lipid 2.13% and 2.32 % ash in fish waste. Rahmi *et al*.[22] reported similar result that Fish silage made from fish wastes contains 13.62% protein, 8.8% lipid and 17.35% ash.

**Biochemical and microbiological analysis of fish waste**

The pH, α- amino nitrogen, TVB-N and TPC of fish waste were 6.8 ± 0.49, 10.64 ± 0.13 mg-N100 g $^{-1}$ 18.64 ± 0.09 mg-N100g $^{-1}$, and 5.1× $10^{6}$ cfu/g respectively as shown in Table 1.2 .Similar results were depicted by Palekar[21]

**Proximate composition of rice bran**

Rice bran was used as dry ingredient for convenient use of liquid fish silage. The proximate composition of rice bran was shown in Table no.1.3 and Fig.1.2. The moisture, crude protein, fat, ash content and NFE of rice bran were 9.45 ± 0.19, 16.05 ± 0.08, 13.42 ± 0.15, 10.44 ± 0.14, 50.64 ± 0.19 (%) respectively. Similarly Tao *et al*.[25] analysed the proximate composition of rice bran from the long grain and medium grain showed similar results. They reported protein, lipid,

ash, moisture, crude fibre and NFE content were 16.07 %, 19.20 %, 9.23 %, 11.20 %, 8.49 %, and 47.01 % in rice bran from long grain and 16.20 %, 21.97 %, 9.46 %, 10.83 %, 8.41 % and 44.07 % in rice bran from medium grain on dry weight basis. Satter *et al*.[23] analysed rice bran variety BRRI-28 that contained 6.54 to 9.48 % moisture, 7.24 to 10.63 % ash, 12.26 to 14.01 % proteins, 23.53 to 27.86 % fats, 2.5 to 10.10 % fibre, 42.19 to 45.74 % carbohydrate.

Rice bran contained moisture, protein, fat, ash and carbohydrate were 4.30 and 5.40, 17.50 and 19.25, 13.10 and 17.20, 4.92 and 4.64, 52.33 and 48.55 g/100g respectively. Similar results were given by the Bhosale and Vijayalakshmi[7]. Hossain and Alam[16] depicted slightly different results of rice bran. Rice bran contained an average 9.32 % protein, 17.94% lipid, 18.67% ash and 9.65 % moisture and 44.42 % NFE.

**Table 1.1 Proximate composition of Fish waste**

| Fish waste | Proximate composition (%) |
|---|---|
| Moisture | 77.09 ± 0.14 |
| Crude protein | 15.20 ± 0.15 |
| Fat | 4.03 ± 0.07 |
| Ash | 3.30 ± 0.11 |
| NFE | 0.38 ± 0.06 |

**(Mean ± standard error)**

**Table 1.2 Biochemical and microbiological analysis of fish waste**

| Characteristic | Fish waste |
|---|---|
| pH | 6.8 ± 0.49 |
| α-Amino nitrogen(mg-N100g $^{-1}$) | 10.64 ± 0.13 |
| TVB-N (mg-N100g $^{-1}$) | 18.64 ± 0.09 |
| TPC (cfu/g) | $5.1 \times 10^6$ |

**(Mean ± standard error)**

**Table 1.3 Proximate composition of Rice bran**

| Rice bran | Proximate composition (%) |
|---|---|
| Moisture | 9.45 ± 0.19 |
| Crude protein | 16.05 ± 0.08 |
| Fat | 13.42 ± 0.15 |
| Ash | 10.44 ± 0.14 |
| NFE | 50.64 ± 0.19 |

**(Mean ± standard error)**

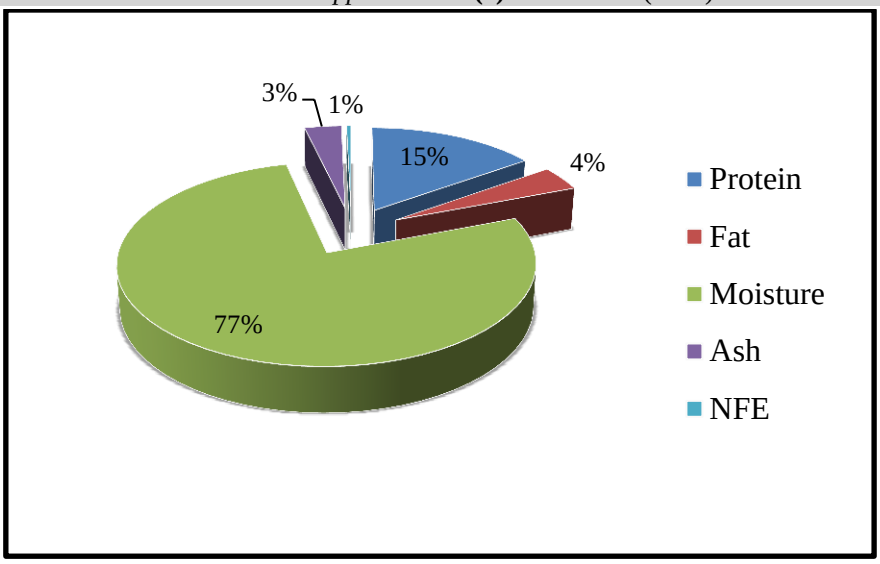



**Fig. 1.1 Proximate composition of Fish waste**

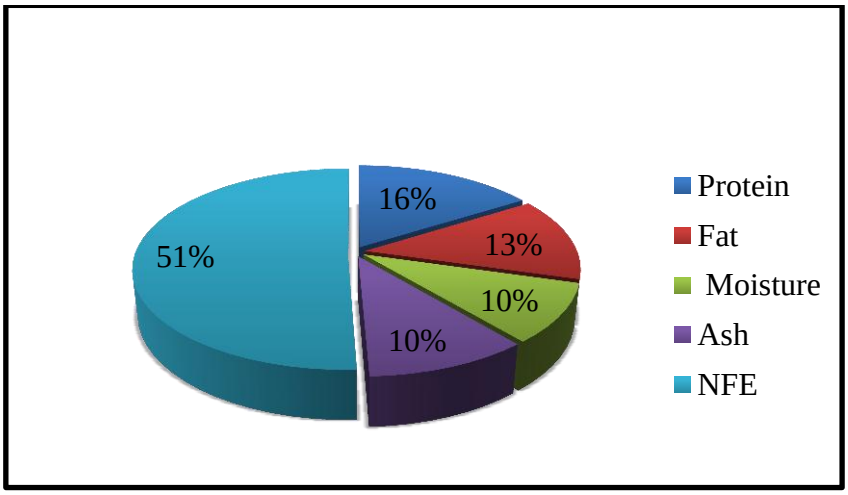



**Fig. 1.2 Proximate composition of Rice bran**

**Drying rate of powder fish silage containing different concentration of rice bran**

After 10 days of storage recorded pH of biological fish silage was 3.9 and TPC was 2.69 x $10^4$ cfu/g. Rice bran was used as co-drying material for drying liquid fish silage. The current study demonstrated that fermented silage was successfully dried with rice bran in solar tunnel drier. The moisture levels of co-dried mixtures were reduced to less than 10% within 2-3 days. Solar tunnel dryer had temperature range between 35°C to 50°C. In present study the initial moisture content of 10 %, 20%, 30%, 40%, 50% rice bran were 67.63, 56.36, 53.83, 50.91% respectively. Although the three mixture i.e 30%, 40%, 50% rice bran appeared to reach equilibrium moisture content below 15% after 48 hours. The last recorded moisture content in 30%, 40%, 50% rice bran were 13.23, 11.88 and 9.84 % respectively. Two mixtures 10 and 20 % rice bran took 72 hours to lower down the moisture content below 15 %. Similar trend was observed by Al-Abri *et al.*[1], Goddard and Al-Yahyai and Goddard and Perret[14]. Two-way ANOVA and SNK test also revealed that all percentages (10 %, 20%, 30%, 40%, 50% rice bran) were significantly different from each other.

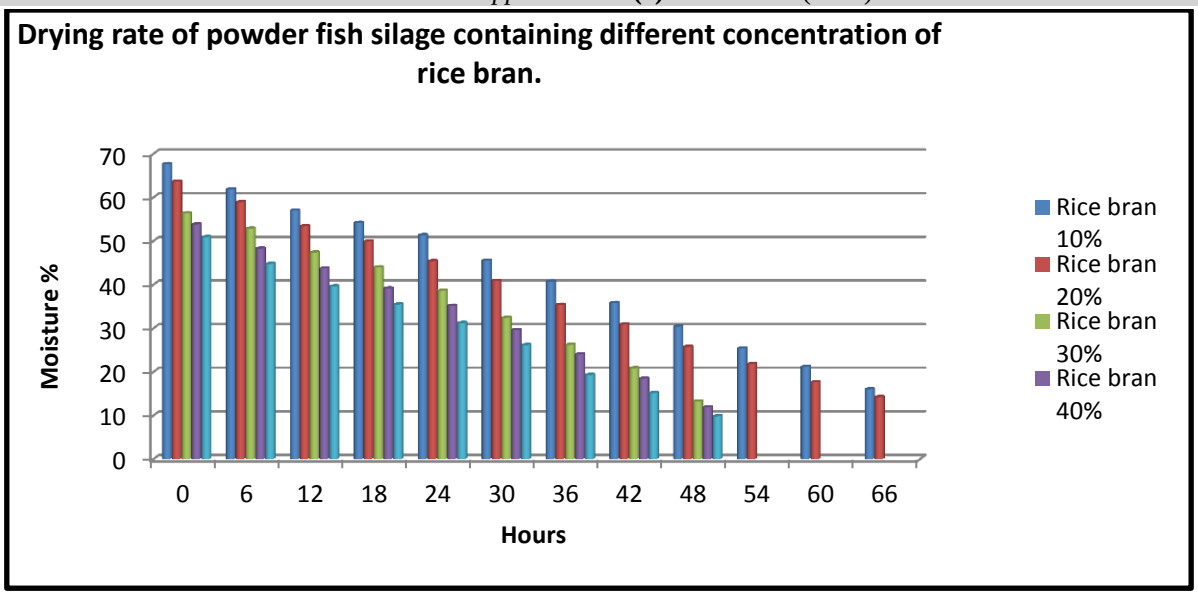



**Fig no.1.3: Drying rate of powder fish silage containing different concentration of rice bran**

**Table 1.4 Proximate composition of powder fish silage made with different quantity of rice bran**

| Rice bran (%) | Moisture (%) | Protein (%) | Fat (%) | Ash (%) |
|---|---|---|---|---|
| **10** | 13.74 ± 0.12 | 29.50 ± 0.22 | 16.28 ± 0.11 | 14.21 ± 0.12 |
| **20** | 12.54 ± 0.30 | 28.56 ± 0.12 | 15.71 ± 0.14 | 14.55 ± 0.11 |
| **30** | 10.91 ± 0.13 | 27.66 ± 0.10 | 14.45 ± 0.11 | 15.27 ± 0.13 |
| **40** | 10.18 ± 0.05 | 26.53 ± 0.18 | 13.73 ± 0.17 | 15.65 ± 0.15 |
| **50** | 9.18 ± 0.02 | 25.73 ± 0.08 | 12.60 ± 0.10 | 15.99 ± 0.12 |

**(Mean ± standard error)**

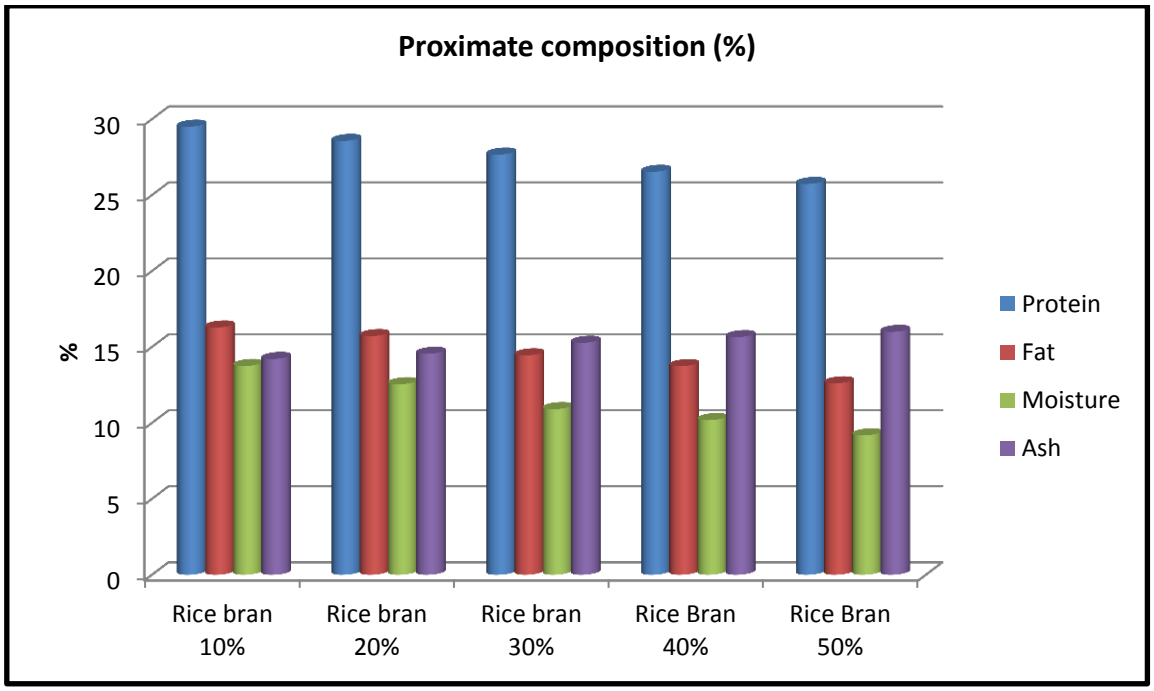



**Fig no.1.4: Proximate composition of powder fish silage made with different quantity of rice bran**

**Proximate composition of powder fish silage made with different quantity of rice bran**

In the present study the protein content of 10% , 20%, 30 %, 40 % and 50% rice bran were 29.50%, 28.56%, 27.66%, 26.53% and 25.73% respectively. Fat content of 10%, 20%, 30 %, 40 % and 50% rice bran were 16.28%, 15.71%, 14.45%, 13.73% and 12.60% respectively. Moisture and ash content of 10%, 20%, 30 %, 40 % and 50% rice bran were 13.74%, 12.54%, 10.91%, 10.18%, 9.18 % and 14.21%, 14.55%, 15.27%, 15.65%, 15.99% respectively. Similar trends were reported by Hossain and Alam[16], protein content of 20 %, 30 %, 40 % and 50% rice bran were 21.75, 20.84, 19.87 and 18.73 % respectively. Fat content of 20%, 30 %, 40 % and 50% rice bran were 34.71, 33.73, 32.88 and 30.74 respectively. Moisture and ash content of 20%, 30 %, 40 % and 50% rice bran were 11.68, 10.83, 10.17, 9.66 % and 13.36, 14.05, 14.28, 14.55 % respectively.

With increasing the quantity of rice bran, the levels of protein, lipid, ash and moisture were decreased. Nutrient content of 40 and 50% rice bran were low compared to 10, 20 and 30 % rice bran similar observation were reported by Hossain and Alam[16]. At 30% rice bran it was found that the protein content was enhanced to about 10 % and 20 % and other nutrient contents were also comparatively acceptable for animal feeding.

It was possible to increase protein content more by decreasing the quantity of rice bran. The mixture of 10 % and 20 % rice bran might be difficult to dry due to the lesser content of dry matter and there would be a probability of mould attack. Therefore, considering all the limitation, powder fish silage with 30 % rice bran was found to be better and carried for further storage study. Similar observation was discussed by Hossain and Alam[16].

**Storage study of powder silage at ambient temperature**

**Biochemical and microbial changes in 30 % Rice bran packaged fish silage powder during storage**

In the present study the moisture content of powder fish silage sample was found to be increasing in trends during through the storage study (0 to 90 days) in the range of 10.91 to 11.15 %. Similar trend were observed by Hossain and Alam[16] reported moisture content of 30% rice bran was increased from 10.83 to 10.98 % at the end of 4 months. Protein content of powder fish silage sample was found to be decreasing in trends during through the storage study (0 to 90 days) in the range of 27.66 to 27.04 %. Hossain and Alam[16] reported protein content of 30% rice bran was decreasing from 20.84 to 20.30 % at the end of 4 months. Fat content of powder fish silage sample was found to be decreasing in trends during through the storage study (0 to 90 days) in the range of 14.45 to 13.42 %. Hossain and Alam[16] reported lipid content of 30% rice bran was decreasing from 33.73 to 32.41 % at the end of 4 months. Ash content of powder fish silage sample was found to be decreasing in trends during through the storage study (0 to 90 days) in the range of 15.27 to 14.60 %. Hossain and Alam[16] reported ash content of 30% rice bran was decreasing from 14.05 to13.49 % at the end of 4 months. Fiber content of powder fish silage sample was found to be decreasing in trends during through the storage study (0 to 90 days) in the range of 9.89 to 9.64 %. Hossain and Alam[16] reported fiber content of 30% rice bran was decreasing from 6.61 to 6.32 % at the end of 4 months. Carbohydrate content of powder fish silage sample was found to be increasing in trends during through the storage study (0 to 90 days) in the range of 21.82 to 23.14 %. Hossain and Alam[16] reported carbohydrate content of 30% rice bran was increasing from 13.94 to 16.50 % at the end of 4 months. pH of powder fish silage sample was found to be increasing in trends during through the storage study (0 to 90 days) in the range of 6.45 to 6.59. Hossain and Alam[16] reported pH of 30% rice bran was increasing from 6.54 to 6.76 at the end of 4 months. TPC content of powder fish silage sample was found to be increasing in trends during through the storage study (0 to 90 days) in the range of $2.10\times10^4$ to $2.36\times10^4$ cfu/g.

**Table 1.5 Biochemical and microbial changes in 30 % Rice bran packed powder silage during storage**

| Days | Crude Protein (%) | Fat (%) | Moisture (%) | Ash (%) | Fiber (%) | Carbohydrate (%) | pH | TPC (cfu/g) (Log value) |
|---|---|---|---|---|---|---|---|---|
| 0 | 27.66 ± 0.10 | 14.45±0.11 | 10.91± 0.14 | 15.27±0.11 | 9.89±0.06 | 21.82 ±0.07 | 6.45±0.05 | $2.10\times10^4$ (4.32) |
| 30 | 27.31 ± 0.11 | 14.16±0.07 | 10.99± 0.10 | 15.07±0.08 | 9.84±0.06 | 22.21 ±0.08 | 6.48±0.09 | $2.15\times10^4$ (4.33) |
| 60 | 27.16 ± 0.33 | 13.83±0.08 | 11.06 ±0.18 | 14.87±0.06 | 9.75±0.13 | 22.64 ±0.13 | 6.53±0.10 | $2.25\times10^4$ (4.35) |
| 90 | 27.04 ± 0.06 | 13.42± 0.10 | 11.15± 0.10 | 14.6 ± 0.09 | 9.64±0.07 | 23.14 ±0.11 | 6.59±0.14 | $2.36\times10^4$ (4.37) |

**(Mean ± standard error)**

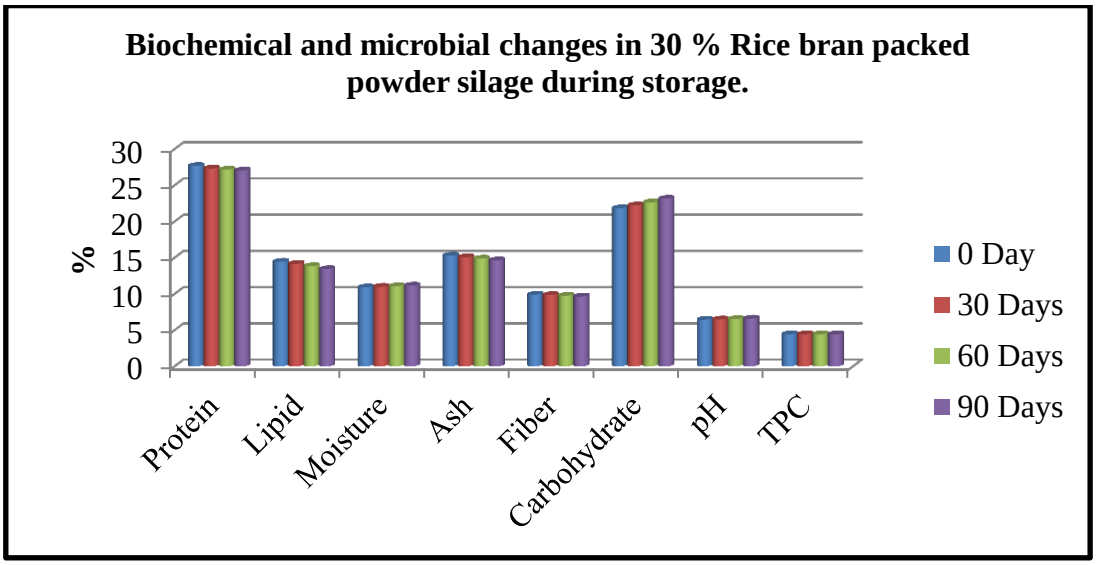



**Fig. no.1.5: Biochemical and microbial changes in 30 % Rice bran packed powder silage during storage**

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