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Research Article

# Effect of Incorporation of Cabbage Powder on Physicochemical and Microbiological Properties and Stability of Emulsion Sausage During Refrigerated Storage

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#### **Abstract**

The objective of research was to evaluate the effect of different levels of cabbage powder on Physicochemical Properties of emulsion sausages of buffalo meat with two level of fat (15% and 20%) during refrigerated storage. Treatment consisted of control sample (C,) and other four samples consisted S1, S2, S3, S4 which had cabbage powder content 1.5%, 3.0%, 4.5% and 6.0% respectively. Analysis of variance (ANOVA) was used for the analysis of statistical analysis. Quality criteria of the product sausage was determined on the basis of physicochemical properties rich moisture, ash, fat, protein content. pH and TBA number was also the part of physicochemical properties. To determine the shelf-life it was decided to packed the product in a suitable packaging. Shelf life was evaluated at 0°C. All quality parameters were determined periodically every after 7 days. The profile of quality during storage was noted as shelf-life was reported accordingly. The result showed that moisture content was found between 57.42 to 62.35% in fresh condition. Effect of incorporation of cabbage powder in the targeted product sausage was noted as the amount of cabbage powder incorporation increase, it was found that moisture content decrease in fresh condition. During storage extent of decrement of moisture was found less in control sample as compared to treated sample. The ash content of the sample was found in between 1.9 to 2.3% in fresh product and had higher moisture content as compared to moisture content of stored sample. The effect of moisture content on ash content was noted that the decrease in moisture content during storage caused slight increase in ash content of the product. Fat content of the product was found in between 14.12 - 16.87% in fresh product, fat content of the product was found 14.32 to 16.85% in between, during refrigerated storage increment in fat content was noted. During storage decrease in moisture content was noted. It caused increase in the protein content of the sausage sample. In fresh condition, protein content of the sample was found in between 16.18 to 17.38%, was caused due to lower down of moisture content. In fresh condition pH values of the sausage samples were found in between 5.75 - 5.85. It was found that decrease in moisture content was affected by incorporation of cabbage powder and sausage sample having 25% fat content and 6.0% cabbage powder was found to have least decrement in pH as compared to their other level of treated counterpart. It was reported that the TBA value for which group of panelist first detected a difference in intensity of oxidized flavour (rancidity, wof was found 0.6 to 2.0) (Tarladgis., et al. 1960). The shelf life of emulsion sausage indicated by microbiological condition was found to be 14 days during the refrigerated storage.

Keywords: Buffalo Meat; Cabbage Powder; Fat; Shelf Life

#### **Abbreviations**

ANOVA: Analysis of Variance; TBA No: Thiobarbituric Acid; TPC: Total Plate Count

#### Introduction

Meat is define by the Codex Alimentarius as "All parts of an animal that are intended for or have been judged as safe and suitable for human consumption. Meat is comprised of high quality protein with essential amino acids, proper moisture content and fat. Meat is rich in Vitamin B complex and minerals like phosphorus, calcium, zinc, magnesium iron etc. There is no dietary- fiber in meat and meat product. The absence of dietary fibre leads to constipation, several stomach and intestinal disorder. Meat sausage is incorporated with cabbage powder to fulfill dietary requirement of buffalo meat. It have been observed that the metabolic rate is affected by the food rich in dietary fiber and it also helps in the prevention of the diabetes [1]. Except carbohydrate and dietary fibre, meat has all the nutrient which are required for the growth and health of human being. Meat and their product has enough amount of protein, vitamin, minerals and micronutrient which are needed for the growth and health of human being. Meat production and processing offers a golden chance to improve the values of this product and by adopting HACCP plan we can produced safe meat with improved shelf-life. According to WHO, around 2 billion people in the world are deficient in key vitamins and minerals, particularly vitamin A, zinc, iron and iodine. People becomes deficient in nutrition, when they do not take the nutrient from different sources, such as cereals, fruits, vegetables and fresh fruits.

According to FAO (outlook 2014) meat is highly nutritious food which is particularly require for HIV AIDS infected communities and also for women and children. 20g of animal protein per person per day or 7.3 kg per year should be provided to overcome the malnutrition and under-nourishment problem. India has 50.34% share in buffalo meat production in the world. Buffalo meat is abundance in our country. It has high quality protein and it gives a source of several commercial products which can be value added and marketed, good income and employment can be generated. Due to the fact that meat and meat product do not have fiber therefore continuous consumption of meat product lead to stomach and intestinal disease like constipation and even colon cancer. All food having high nutrient consume by human being but does not conta-

in dietary fiber may cause some stomach and intestinal disease. In this research, cabbage powder incorporation was plan to make the meat product healthy for human beings. Cabbage (Brassica oleracea L. var. capitata), it is the most grown vegetables in the world. Cabbage belongs to the Cruciferous family, which includes broccoli, cauliflower, and kale. There are many varieties of cabbage differing in cultivation, size, shape, color and texture off the head [2]. Medicinal properties which are exploited, to combat several disease and provide health effect in human being, when cabbage is incorporated in traditional processed food product. Research on medicinal effect of cabbage have shown to control complex stomach disease like peptic ulcer [3], and it also assist the lowering of serum LDL levels [4]. Analytical determination had shown that carbohydrates is being principal component of cabbage is consisting of nearly 90% dry weight, in which one-third is dietary fiber and two-third represent low molecular weight carbohydrate. While glucosinolates are other typical constitutes presents [5]. There is significant role of dietary fiber in human diet. The scarcity of dietary fiber in human diet leads to several complex diseases including constipation, type 2 diabetes, cardiovascular disease, colon cancer and other gastrointestinal disease [6]. The principal objectives of the study were taken to determine several physicochemical properties viz. the effect of refrigerated storage on pH value, TBA number, protein, fat, ash and moisture content periodically every after seven days till the end of shelf- stability of the product.

#### **Objective of the Study**

The objective of the research was to incorporate the cabbage powder for development of sausage.

### **Materials and Methods**

Samples of meat were arranged from the meat shop located in the vicinity of the University, slaughter house of Aligarh municipal corporation had traditional techniques of slaughtering buffalos as per halal method. To bring consistent post-mortem glycolysis in the animal after slaughter, they were kept in a lairage for a time of 20 hours. To get enough meat for samples preparation, the part biceps femoris muscle was choosen of the female buffalo, between the age of 2.5 to 3.5 years. Sample of meat were brought to laboratory within four hours after slaughtering of the buffalo. Combination of film was used for packaging the meat chunk which was obtained from the market and brought to the laboratory within 20

minutes. Cabbage, Spices, salts, condiments and packaging material (combination film) were arranged from the market located in the vicinity of the university. The samples of meat and fat are stored in low temperature cabinet (Yarco India) at 2°C. The casing material for sausages were arranged from PRS technology Delhi. Deboning of meat was done immediately after washing. Fat and minced meat was chopped in a bowl chopper and added with cabbage powder, spices, condiments and preservative. At the end of mincing the mix was stuff into polymer casing and put under refrigerated storage. As usual in the sausage 20 - 25% white fat was taken in the formulation while level of cabbage was kept between 1.5 to 6.0%. In the beginning levels of cabbage were taken as 1.5, 3.0, 4.5 and 6.0% and finally ANOVA provided optimal solution. Further storage study was carried out under refrigerated storage periodically after seven days to study self-stability.

#### **Evaluation of physicochemical properties**

Ash and moisture content was determined by standard hot air oven [7]. Fat content was determined by Soxhlet apparatus. Measurement of pH was done using pH meter (Khera model, India). Measurement of protein was done by Kjeldahl method. Pearson 1973, method was used to measure TBA number taking the help of digital spectrophotometer (model 310E, India).

### Statistical analysis

Analysis of variance (ANOVA) was used for analysis of statistical analysis (Cochran and Cox 1992).

#### **Results and Discussion**

#### **Effect on moisture content**

Moisture content of sausage is an important property to determine the quality and shelf life of the product. Higher the moisture content, the sample would be more susceptible for growth of micro-organism but it has good sensory characteristics such as texture, juiciness, mouth feel etc.

### At 15% fat

The moisture content of emulsion sausage incorporated with different concentration of cabbage powder has been shown in table 1. Moisture contents of the samples were found in between 58.25 - 62.66%. During refrigerated storage at  $0^{\circ}$ C, moisture contents of these samples significantly significant (p < 0.05) decreased. The decrease were due to evaporation of moisture through the perme-

able packaging film. It was noted that in case of sausage sample treated with different levels of cabbage powder, decrease in moisture content during storage was less as compared to their controlled counterpart and sample treated with 6.0% cabbage powder sample was noted minimum decrease in moisture content. This was due to increased water binding capacity (WBC) of the samplesincorporated with cabbage powder because it has an antioxidant property N Cumby., et al. (2008) [8], reported that improved water holding capacity of meat by incorporation of antioxidant. The final moisture contents of these samples were found in the range of 53.86 - 58.25% on 14 day of storage (Figure 1).

#### At 20% fat

It was noticed that by increasing levels of fat constantly decreased moisture content of sausages samples. Ahmad and Srivastava (2007) [9] reported that increasing levels of fat decrease significant (p < 0.05) the moisture content of the sausages because the required medium for making the emulsion is fat and water and once the proportion fat increases, obviously moisture content decreases. These results are in similarity of Olivares.,  $et\ al.\ (2010)\ [10]$  found higher water content in low fat sausages.

Table 2 presents the results of moisture contents of buffalo meat emulsion sausage prepared with 20% fat. The moisture contents of all samples of sausage were found in the range of 57.42 - 61.55% in emulsion product. During refrigerated storage of emulsion sausage at  $0 \circ C$  significant (p < 0.05) decrease in moisture content was noted. The decrease were due to evaporation of moisture through the permeable packaging film. Figure shows the moisture loss behavior of emulsion sausages samples during refrigerated storage ( $^{\circ}C$ ). In both levels of fat, significant (p < 0.05) effect of different antioxidants on moisture content had been observed.

**Figure 1:** Evaluation of moisture content of sausages incorporated with 15% fat during refrigerated storage at 00C.

**Figure 2:** Evaluation of moisture content of sausages incorporated with 20% fat during refrigerated storage at 00C.

Sample	Storage Periods (no. of days) 15% fat			
	0	7	14	
C1	62.66 ± .02887	61.57 ± .0200	60.46 ± .01528	
S1	60.78 ± .02517	58.87 ± .03215	57.86 ± .03606	
S2	59.14 ± .02646	57.6 ± .0400	56.41 ± .60754	
S3	58.25 ± .0200	56.77 ± .02517	55.97 ± .00577	
S4	56.2 ± .02010	54.6 ± .02310	53.58 ± .00358	
Sample	Storage period (No. of days) 20% fat			
	0	7	14	
C1	61.55 ± .01155	60.47 ± .02000	59.42 ± .05508	
S1	59.48 ± .01000	58.03 ± .22368	56.81 ± .06557	
S2	58.76 ± .03055	56.55 ± .2610	56.09 ± .25482	
S3	57.42 ± .27154	55.8 ± .21962	54.6 ± .22811	
S4	56.3 ± .02314	53.53 ± .2014	52.15 ± .0258	

**Table 1:** Values are means of three replicates ± SD; Means with different letters differ significantly (p < 0.05).C = control, s1= sample with 1.5% cabbage powder, s2= sample with 3.0% cabbage powder, s3= sample with 4.5% cabbage powder, s4= sample with 6.0% cabbage powder.

### Effect on thiobarbituric acid number (TBA)

Thiobarbituric acid (TBA) number is a property which determined the extent of rancidity in the meat product. Air packaging system and combination film as a packaging material was used as packing of emulsion sausages. The meat sausage sample have enough fat and there is probable risk of oxidation of the fat by the oxygen present in the atmosphere ultimately it may caused development of rancidity but in meat product it is known as wof (warm

off flavour). The extent of rancidity is co-related with a sensory score of aroma attributes there is a stage of rancidity/wof when the relevant aroma is detected. The TBA values when exceeded in the range of 1-2 mg of malonaldehyde/kg, the WOF was detected as per sensory evaluation [11]. Malonaldehyde as the ultimate product of TBA reagent with the oxidize fat, it gives a reddish complex compound. Expressed as mg of malonaldehyde of sample. The spectrophotometric result provides the values in terms of absorption of the ultimate red color. The range is taken 530 - 538 nm. Two moles of TBA reagent with 1 mole of malonaldehyde gives a red pigment product [12].

#### At 15% fat

In emulsion condition, TBA number values were between 0.107 - 0.1580 mg of malonaldehyde/kg of meat (Table 2 and figure 2). When the samples was kept under stored condition at 0°C, TBA value of all samples was increased. It happened because during preparation of sausage some oxygen molecules entrapped inside casing and exposure of light which causes the oxidation of fat. The antioxidative property of cabbage powder prevents oxidation of fat. Samples incorporated with 6.0% powder concentration showed minimum TBA number. The TBA profile during storage was found to be increasing (Table 2 and figure 2). after 14 days of storage, TBA number values were between 0.202 - 00.317 mg of malonal-dehyde/kg of meat. Greene and Cumuze (1981) [13] reported that oxidised flavour developed in range of 0.6 - 2.0. Ali (2011) [14] reported that TBA values of ground beef increased gradually and significantly (p < 0.05) during storage period.

### At 20% fat

Table 2 and figure 3, presents the results of TBA number analysis of emulsion sausages incorporated with 20% fat during refrigerated storage (0°C). TBA number values were between 0.127 - 0.167 mg of malonaldehyde/kg of meat in emulsion condition. Samples incorporated with 6.0% cabbage powder concentration showed minimum TBA number in emulsion condition. Refrigerated storage significantly (p < 0.05) increased the TBA number of all samples. Control sample (without cabbage powder concentration) showed highest TBA number. Increasing fat level consistently increased TBA number in all samples of emulsion sausages. After 14 days of storage, TBA number was between 0.205 - 0.312 mg of malonaldehyde/kg of meat. That results were similar to the results reported by Ahmad., *et al.* (2005) [15]; Ahmad and Srivastava (2007) [10]; Liaquati and Srivastava, (2010)

[17]. However iin both levels of fat, the values of TBA number were under safe limit on the 14 day of storage. Previous reports indicated that the meat samples containing TBA numbers from 0.5 to 1 possess no off odour [18], additionally Watts (1962) [19] reported that values of TBA number of 1-2 mg/kg of malonaldehyde was the minimum detectable level for oxidized flavour in beef and its products for an inexperienced panel [20] Figures indicates the TBA number of emulsion sausages with the help of regression analysis, when they are kept under refrigerated storage at (00C) incorporated with 15% and 20% fat respectively.

Sample	Storage periods (no. of days) 15% fat			
	0	7	14	
C1	0.158 ± .001	0.195 ± .00058	0.232 ± .001	
S1	0.115 ± .00058	0.145 ± .00050	0.185 ± .0063	
S2	0.115 ± .00624	0.136 ± .001	0.168 ± .00071	
S3	0.107 ± .0026	0.127 ± .002	0.158 ± .00121	
S4	0.101 ± .0050	0.118 ± .0001	0.147 ± .0010	
Sample	Storage Periods (no. of days) 20% fat			
	0	7	14	
C1	0.167 ± .00000	0.205 ± .02994	0.229 ± .01646	
S1	0.145 ± .03522	0.16 ± .00100	0.22 ± .02914	
S2	0.143 ± .04071	0.139 ± .01819	0.182 ± .01242	
S3	0.127 ± .02858	0.146 ± .01159	0.174 ± .02623	
S4	0.115 ± .02061	0.129 ± .01248	0.158 ± .02030	

**Table 2:** Values are means of three replicates  $\pm$  SD; Means with different letters differ significantly (p < 0.05).C = control,

s1= sample with 1.5% cabbage powder, s2= sample with 3.0% cabbage powder, s3= sample with 4.5% cabbage powder, s4= sample with 6.0% cabbage powder.

**Figure 3:** Evaluation of TBA number of sausages incorporated with 15% fat during refrigerated storage at 0oC.

**Figure 4:** Evaluation of TBA number of sausages incorporated with 20% fat during refrigerated storage at 00C.

### Effect on pH value

The pH value is an important physicochemical characteristic to determine the shelf life and quality of the food products. The food products with low pH are least susceptible for microbial growth. The pH of meat products also affects the sensory characteristics. The acceptable limit of pH for meat consumption lies between slightly acidic to neutral pH value.

### At 15% fat

The sausage sample (15% fat) was found to have pH values in the range of 5.71 - 5.85 at  $0^{th}$  day (Table 3 and figure). Effect of refrigerated storage was determined at  $0^{\circ}$ C, consistently decrease in pH was noted when the observation was made periodically after  $7^{th}$  day. The reduction in pH was due to formation of free fatty acids and carbonyl compounds as a result of oxidation of fat and protein and also micro-organism metabolise carbon compounds in acids. Refrigerated storage significantly (p < 0.05) reduced the pH of emulsion sausages. It was noted that in case of sausage sample treated with different concentration of cabbage powder decrease in pH during storage was less as compared to their controlled sample due to antioxidative and antimicrobial property of cabbage powder. At the 14 days of storage pH values were found to be between 5.75 - 5.95.

### At 20% fat

pH values of all samples were found between 5.75 - 5.98 just after product preparation of samples (Table 3, figure). Incorporation with different concentration of cabbage powder significantly (p < 0.05) affected the pH of sausage sample. During refrigerated storage at  $0^{\circ}$ C, pH values were found to significantly (p < 0.05)

decrease. At the 14 days of storage pH values were found to be between 5.84 - 5.94 Increasing the fat level from 15 to 20% increased the pH due to formation of more fatty acids of all sample and that are in agreement with the results of studies conducted by Ahmad, 2005; Liaquati and Srivastava, 2010; Olivares, *et al.* 2010.

Sample	Storage Periods (no. of days) 15% fat			
	0	7	14	
c1	5.71 ± .02261	5.73 ± .11408	5.78 ± .00551	
s1	5.73 ± .00892	5.76 ± .00100	5.79 ± .00100	
s2	5.79 ± .00777	5.83 ± .04965	5.85 ± .00252	
s3	5.81 ± .00503	5.86 ± .00902	5.88 ± .02126	
s4	5.85 ± .00302	5.88 ± .00709	5.95 ± .01021	
Commis	Storage Periods (no. of days) 20%fat			
Sample	0	7	14	
c1	5.75 ± .01000	5.78 ± .12082	5.84 ± .02000	
s1	5.78 ± .00577	5.81 ± .02309	5.87 ± .15617	
s2	5.85 ± .00577	5.86 ± .01155	5.88 ± .04041	
s3	5.89 ± .01000	5.91 ± .08145	5.94. ± .03606	
s4	5.91 ± .00302	5.93 ± .00709	5.98 ± .01021	

**Table 3:** Values are means of three replicates  $\pm$  SD; Means with different letters differ significantly (p < 0.05).C = control, s1= sample with 1.5% cabbage powder, s2= sample with 3.0% cabbage

powder, s3= sample with 4.5% cabbage powder, s4= sample with 6.0% cabbage powder.

**Figure 5:** Evaluation of pH of sausages incorporated with 15% fat during refrigerated storage at 0oC.

**Figure 6:** Evaluation of pH of sausages incorporated with 20% fat during refrigerated storage at 0oC.

### **Microbiological characteristics**

### **Total plate counts**

TPC of all samples of emulsion sausages was determined periodically after every 7 days during refrigerated storage. The table 4 present the result of TPC of sausage sample. In fresh condition the TPC of the samples expressed in log cfu/g was found in the range of 2.367 - 3.276 cfu/g. Effect of storage was noted on the values of TPC (log cfu/g), after 14 days the result showed that the TPC values in between 3.321 - 4.63 in log cfu/g. The sausage sample were in still in edible condition. The marginal limit for TPC of sample had reached at TPC of  $10^7$ /g, it would have spoiled but due to lockdown period study was stopped after 14 days and the sample were found in better condition.

Sample	Storage Periods (no. of days) 15% fat			
	0	7	14	
c1	2.367 ± .00577	2.372 ± .01155	3.321 ± .02517	
s1	2.360 ± .57303	2.492 ± .00577	3.350 ± .00153	
s2	2.357 ± .00076	2.620 ± .00153	3.379 ± .00058	
s3	2.355 ± .00126	3.320 ± .00071	3.390 ± .00098	
s4	2.352 ± .00210	3.569 ± .00105	4.630 ± .00059	
Commis	Storage Periods (no. of days) 20% fat			
Sample	0	7	14	
c1	2.369 ± .00577	2.375 ± .01155	3.223 ± .02517	
s1	2.495 ± .57303	2.575 ± .00577	3.360 ± .00153	
s2	2.565 ± .00076	2.641 ± .00153	3.705 ± .00058	
s3	2.658 ± .00126	2.695 ± .00071	3.863 ± .00098	
s4	3.190 ± .00210	3.769 ± .00105	4.889 ± .00059	

**Table 4:** Values are means of three replicates ± SD; Means with different letters differ significantly (p < 0.05). C = control, s1= sample with 1.5% cabbage powder, s2= sample with 3.0% cabbage powder, s3= sample with 4.5% cabbage powder, s4= sample with 6.0% cabbage powder.

#### Conclusion

Incorporation of cabbage powder brought improved in the nutrition value of the emulsion sausage. Nutritional value was supported by dietary fibers and flavonoids (antioxidants) in the cabbage. Texture of the product remains unaffected by incorporation of cabbage powder. Slight effect of incorporation of cabbage was noted on moisture content of the sausages but however did not make any hurdle in emulsion making. People who make choice of sausage consumption these products will minimize the constipation and hence several coronary disease. Expected shelf life of sausage may be more what it has been reported.

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