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INVESTIGATION OF SOME QUALITY CHARACTERISTICS OF SMOKED COMMON CARP (*Cyprinus carpio*) SAUSAGES SUPPLEMENTED WITH PROPOLIS EXTRACT

ABSTRACT

In this study, some quality attributes of smoked *Cyprinus carpio* sausages supplemented with %1 Propolis Extract, stored under cold condition (2°C) for 9 weeks were investigated. Quality of sausages were determined using physicochemical (Thiobarbituric acid (TBA), microbiological (Coagulase-positive Staphylococci, Sulphite-reducing Clostridium, *E. coli* and *Salmonella*) and sensory analyzes. During the storage period, propolis added group was measured lower TBA than control (Without propolis) group. *E. coli* and *Salmonella* were not detected in the all sausage samples. In addition, significant differences were found each two groups as sensorial quality ($p<0.05$). Consequently, %1 propolis treatment increased the shelf life of sausages by 3 weeks as compared to the control samples. Therefore, propolis extract as natural preservative can be used on other studies to enhance seafood product quality.

Keywords: *Cyprinus carpio*, Fish sausage, Propolis, Natural preservative

1. INTRODUCTION

The demand for ready-to-eat food has increased due to the increment of world population and socioeconomic changes. Fish sausages are one of these ready-to eat foods [1]. Fish sausages is produced from flesh or frozen fish minced like meat sausage. However, fish sausages that do not contain any preservatives have a short shelf life in a cold condition. Deterioration in fish and fish products may be prevented by storage at low temperatures or by the addition of suitable antioxidants [2 and 3]. Nowadays, there has been a lot of worrying about the safety of synthetic food preservatives because of their potential toxicity. Therefore, health-conscious nutritionist and food experts have focused on the use of natural protectives to stabilize fatty food [4]. Propolis is a natural preservative. It is a natural resinous mix breed by honeybees from substances gathered from various parts of plants and sprout. Honey and propolis ensure useful influence on human health. Since the earliest times propolis has been widely used by human, especially in public medicine to cure several illnesses [5 and 6]. In recent years, propolis is a natural drug found in many health food stores in various forms for local use. Propolis is used in mouthwashes to forestall caries and to treat gum-boil and stomatitis. It is commercially existent in the shape of

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capsules and powder. Owing to its antimicrobial, antiviral, and antioxidant properties, it is widely usaged in human and veterinary medicine, food, pharmacology and cosmetics [6 and 7]. Propolis contains; polyphenols, benzoic acids, cinnamic alcohol and cinnamic acid, sesquiterpene and triterpene hydrocarbons, alcohols, ketones, and heteroaromatic compounds, terpene, aliphatic hydrocarbons, minerals, sterols [8]. The aim of this study was to investigate the lipid oxidation, sensory and microbiological characteristics of smoked Cyprinus carpio sausages supplemented with propolis extract which were stored at 2°C for 9 weeks. According to our knowledge, this study will be the first study on propolis added smoked sausage.

2. RESEARCH SIGNIFICANCE

According to our knowledge, this study will be the first study on the use of propolis extract in fish sausages. Propolis is a powerful and natural antioxidant and antibacterial.

3. EXPERIMENTAL METHOD-PROCESS

3.1. Reagents

Propolis extract was obtained from a commercial company (Xi'a Xin Sheng Bio-chem Co., Ltd, China). This extract was used for food purposes and had ISO 22000, GMP, FDA and Hallal certificates Cellulose casings were bought from the Wienie-Pak (21mm×15m, Lommel, Belgium). Other ingredients for sausage were purchased from local markets.

3.2. Fish

The fresh common carp (*Cyprinus carpio*) were taken from a local market, in Elazığ (Turkey). A total of two fish were used in sausage production. The weight of each fish was approximately 7-8kg. Fish were transported to the laboratory on ice in polystyrene and then they were cleaned, skinned and filleted under aseptically conditions.

3.3. Formulation and Processing of Pausages

Sausages were made according to the method of Dincer and Caklı [3] with minor change. The list of the components usaged for sausages making and their percentages were demonstrated in Table 1. Production of fish sausages were made from *Cyprinus carpio* fillets and beef fat in a ratio of 70:10. Carp and beef fat were minced with a kitchen meat mincer with a pore size of 3.0mm. Afterwards, spices were added. The crushed batter was filled into a cellulose casing (21mm diameter×15m long) by using a manual filler. The filled casing was linked in 10cm length and smoked at 75±3°C and RH=75-85% for 30min. After smoking, sausages were immediately cooled in ice-water (1:1, 6-7°C). Sausages samples were packed in a plastic bag and stored for 9 week at 2°C. Analyses of lipid oxidation as well as sensory and microbiological analysis were conducted on 9 weeks of storage. All analysis was carried out two samples from each batch (without propolis and propolis extract added sausages) in duplicate.

Table 1. Rates and ingredients used in formulations of smoked carp sausages

Ingredients and Additives	Control (%)	Supplemented with Propolis (%)
Minced Fish	70	70
Beef Fat	10	10
Bread Crumbs	3	3
Salt	2	2
Sugar	0.8	0.8
Ascorbic acid	0.02	0.02
Hot paprika powder	0.2	0.2
Ginger	0.18	0.18
Caraway	0.2	0.2
Garlic	0.2	0.2
Sodiumnitrit	0.2	0.2
Sodiumphosphat	0.2	0.2
Propolis	-	1
Ice/water	13	12

3.4. Proximate Analysis

The proximate analysis of carp fillets and the sausages were made according to AOAC methodology [9]. Proximate compositions of produced sausages were determined on the first day.

3.5. Lipid Oxidation Analysis

Thiobarbutiric acid (TBA, mg malonaldehyde/kg) was detected using the spectrophotometric method with change [10]. TBA value was measured at 535nm in a spectrophotometer and it was expressed as mg of malonaldehyde/kg of samples.

3.6. Sensory Analysis

Sensory evaluation of flavour and odour was done. The cellulose casing of sausages was removed and cut into strips of approximately 4 mm thickness and dish out at room temperature on white plastic dishes. Three strips were served from each group. Water was used to cleanse the palate between samples. Flavour and odour were performed by a panel beginning 8 trained members of different ages [11 and 12]. Assesment was made using five point scale by quantitative descriptive analysis (QDA). According to this scale; 5:Extremely Good Quality, 4:Good Quality, 3:Drawbacks And Defects of Quality, 2:Defects of Quality, 1:Product Unacceptable [13 and 15].

3.7. Microbiological Analysis

The assessment of the microbiological feature of the smoked sausages (control and supplemented with propolis extract) was performed during storage at 2°C through the description of the Most Probable Number (MPN) of total and fecal coliforms per gram of food, the number of coagulase-positive Staphylococci and sulfite-reducing Clostridium and of the existence of Salmonella, since the existence of such groups or species in great amounts may show the exposure of food to conditions that may let the multiplication of infectious or toxigenic species. This analyzes were made according to methodology recommend by Harrigan [16] and ICMSF [17].

3.8. Statistical Analysis

Statistical analysis was applied using the SAS software [18]. All data was given as mean values with their standard deviations (mean±SD). Variance analysis (ANOVA) was performed to evaluate the statistical significance ($p<0.05$). Duncan's multiple range test was used to evaluate the comparison of means.

4. RESULTS AND DISCUSSION

The moisture, protein, fat, ash of raw carp fillets and groups are indicating in Table 2. Carp fillets were 75.13% moisture, 17.56% protein, 2.20% fat and 0.92% ash content. In accordance with the Institute of Turkish Standards 1070 sausage standard are the maximum moisture, protein and fat content in sausage is 40%, 20% and 30%, respectively [19]. These data are not compatible with our findings. This can be explained with difference of meat and used fat ratio.

Table 2. Proximate composition of sausages and raw carp fillets (%)

Proximate Analysis	Carp Fillet	Control	Added Propolis Extract 1%
Moisture	78.13±0.12 ^a	68.39±0.18 ^b	68.38±0.06 ^b
Protein	17.56±0.02 ^a	18.84±0.02 ^a	18.86±0.03 ^a
Fat	2.20±0.08 ^a	10.13±0.2 ^b	10.17±0.09 ^b
Ash	0.92±0.01 ^a	2.04±0.03 ^b	2.25±0.07 ^b

TBA value is a widely operated sign for the assessment of degree of lipid oxidation. Fatty fishes are very sensitive to lipid oxidation. As the number of double bonds increases, oxidation reactions accelerate [20 and 22]. Alcohols, aldehydes and ketones which are necessary to enhance the flavour are produced by these reactions [22 and 24], however more of these components cause undesirable rancidity. Acceptable limit value of TBA content is between 7-8mg MDAkg⁻¹ [25]. TBA values were expressed as milligrams of malonylaldehyde content per kilogram, was shown in Figure 1.

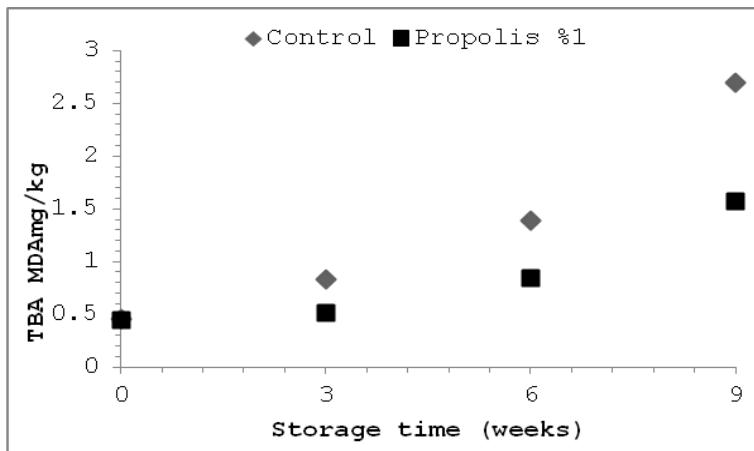


Figure 1. The TBA results of smoked Cyprinus carpio sausage supplemented with propolis extract

Each two groups demonstrated an increase in TBA values during storage, they did not exceed the limit value at a dose of 8 mg MDA/kg. Schormüller [26] proposed that the maximum level of TBA value indicating good quality of fish is 5 mg MDA/kg and the consumption level is up to 8mg MDAkg⁻¹. At the initial day, TBA value was 0.46 and 0.45mg MDA/kg for control group and supplemented with propolis extract group, respectively. Control group showed highest TBA (2.70mg MDA/kg) at 9th weeks. During storage, TBA increased gradually for two groups ($p<0.05$). This could be due to antioxidant activity of propolis extract [6]. This is a valuable result, which can be used in further studies on seafood product shelf life prolongation using propolis extract. TBA values from this research were in accordance with results acquired by some researchers [2, 15 and 27]. TBA values in both groups were lower than the limit value at the end of the storage. Some

researchers found lipid oxidation in sausage made from six different marine fish as TBA values from 10 to 40mg/kg. Fermented sausages made from six different marine fish species were showed high oxidation when compared to commercial fermented sausage made from freshwater fish ranging in TBA values from 5 to 14mg kg⁻¹ [22]. Compared to day 0, both without propolis extract and propolis extract added sausages were scored with lower points for flavour and odour after 9 weeks of storage (Figure 2).

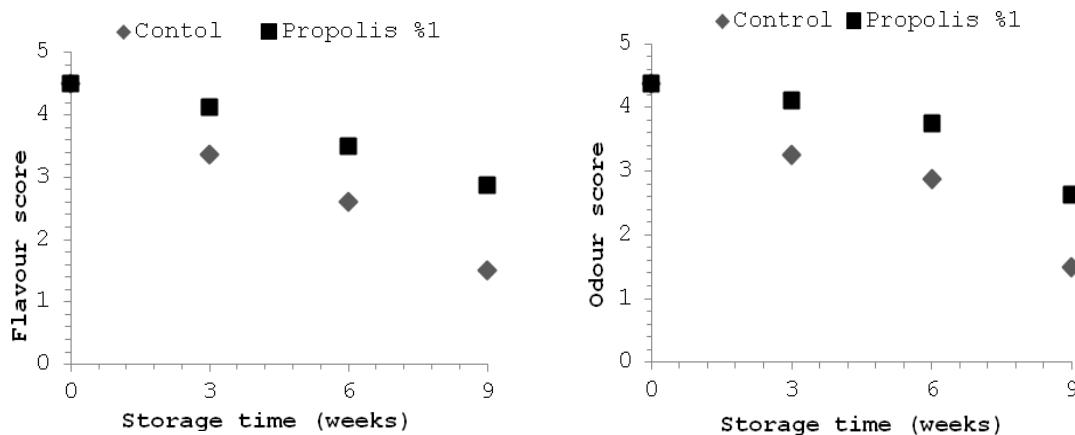


Figure 2. The results of the sensory evaluation of smoked *Cyprinus carpio* sausage suplemented with propolis extract

With without propolis extract sausages, panelists noticed drawbacks and flaws of flavour and odour in the form of slight rancidity. For sausages suplement with propolis extract, different a aroma resulting from propolis was not detected. No other changes in flavour or odour were detected but, because of an atypical aroma, sausages suplement with propolis extract was scored lower after storage than the starting sausage. These results correspond to TBA analysis results. Rancidity was avoided in propolis extract added sausages. The microbiological assessment realizes during storage indicated that the bacterial counting of the smoked carp sausages control group and supplemented with propolis extract 1% group, was within the limits given by the TS 1070 [19]. Total coliforms (MPN) ranged from 2.3×10 cfu/g for fish sausage Control group to 1×10 for sausage added propolis extract 1%, while *E. coli* was not observed for both groups. Sulphite-reducing Clostridium count was determined as <10 log cfu/g, for both sausages (control group and suplemented with propolis extract). Coagulase-positive Staphylococci was found as 1.5×10^2 cfu/g for sausage control group and <10 log cfu/g for sausage suplemented with propolis extract ($p<0.05$). This result can be explained by effect antimicrobial activity of propolis [6 and 8]. The presence of *Salmonella* was not determined in the sausage samples. Similar results can be seen in different studies [28 and 29].

5. CONCLUSION AND RECOMMENDATIONS

Propolis extract had positive effect in slowing down the degradation of the sausage lipid oxidation and microbial growth were slowed down, probably due to propolis extract antioxidant and antimicrobiyal activities. Supplemented with propolis extract sausage was obtained better results from sensory evaluation of flavour and odour. These results are encouraging for further work on optimization of propolis extract based (propolis as a natural additive) for smoked

sausage shelf life prolongation, without using vacuum packaging and special packaging conditions.

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