O EFEITO DA EMBALAGEM DE FOLHA DE BANANEIRA NA VIDA DE PRATELEIRA DO FILÉ DE TRUTA ARCO-ÍRIS EM COMPARAÇÃO COM SACOS DE PLÁSTICO

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ABSTRACT: The aim of this study was to evaluate the effect of banana leaf extract on the quality and shelf life of rainbow trout compared to plastic bags at freezing temperature for 40 days. For evaluating this propose, the antioxidant activity of banana leaf extract was assessed. In addition, the shelf life of fish filets was determined by measuring thiobarbituric acid (TBA) and pHof fish. The banana leaves extract showed the highest content of vitamin E ($5.8 \pm 0.61 \text{ mg}/g$) and carotenoids ($12.8 \pm 0.1 \text{ mg}/g$). The potential of Cu (II) reduction the extract was 1.76 ± 0.09 . The magnitude of modification in TBA and pH of the packed fish with banana leaves were less than the control samples. The present study demonstrated that the use of banana leaf extract will retard lipid oxidation in fish. fillet during freezing storage that may due to its strong antioxidant properties.

KEYWORDS: Banana leaf extract. Antioxidant. Shelf life. Fish.

INTRODUCTION

The world's waters have a total of 40,000 fish species, many of which are nutritious and can be consumed by humans (SHAHIDI; KAMIL, 2001). Global production of fish increased significantly in recent years. Consequently, fish consumption has also gained great popularity, both in developed and developing countries. It has been an important source of proteins, vitamins, useful fatty acids and other nutrients for humans throughout recorded history (CALDER; YAQOOB, 2009).

Fresh fish and marine products are extremely perishable as compared to other fresh meat commodities (CAN, 2011). Rainbow trout undergoing oxidative damages are highly perishable products due to its unsaturated fat composition. Lipid oxidation may produce modulations in seafood quality parameters such as color, off-flavor, rancidity, odor, texture, and even nutritional value (NIELSEN; JACOBSEN, 2013).

The increase in salmon farming in the last decade has led to a saturation of the world market. Because of the high moisture content and neutral pH in rainbow trout and consequently, the low shelf life has led to the attention to maintaining the quality and health of this product, especially in distant markets of production areas (GONZALEZ-FANDOS et al., 2004).

Temperature control, cooling and freezing, controlling during processing, vacuum packaging, packaging in the modified atmosphere, as well as the use of antioxidants are the techniques to postpone the corruption of fish (ABEDI et al., 2016).

Musa sapientum var. sylvesteris (M. sapientum var. sylvesteris) is a valuable medicinal plant which belongs to the family Musaceae (SAHAA et al., 2013). Banana is native to the tropics of Africa and Asia that is implanted at latitude 30 degrees north and south.

The fruit, peel, root, and leaves of banana have been reported to possess considerable antioxidant and antimicrobial activities due to Alkaloid, flavonoid, steroid, glycoside, saponin and proanthocyanidin compounds (AGARWAL et al., 2009). The leaf of banana is suitable for packaging large volumes of food due to its large surface area and its waxy nature.

The aim of this study was to evaluate the effect of banana leaves on the quality and shelf life of frozen trout fillet compared to plastic bags.

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MATERIAL AND METHODS

Sample collection and preparation of banana leaf extract

Samples of the fresh leaf of banana were collected. The ethanolic extract of leaf was prepared. Briefly, 2 g of the powdered of banana leaf was mixed with 20 mL of 70% ethanol in a shaking incubator at 40°C for 24 h. The solutions were filtered through filter paper (110 mm) and evaporated under vacuum by using a rotary evaporator (IKA RV 10 digital) below 50°C. After the evaporation of ethanol, the ethanolic extract was stored at 4°C until use.

Chemical evaluations of leaf extract of banana Determination of total carotenoids

The total carotenoid content was measured by β carotene standard curve as well as the spectrophotometric method at 470 nm. The total carotenoid content of the samples was calculated on basis of the \Box carotene standard curve (THAIPONG et al., 2005). The concentration of carotenoid pigment in the extracts was calculated using the standard curve obtained by commercial carotene. y = 0.2495x - 0.3047, R2=0.817. The antioxidant activity level of banana leaf extract was expressed as 1% vitamin C equivalent by CUPRAC assay.

Antioxidant activity screening assays

For measuringantioxidant activity, we used the cupric ion reducing antioxidant capacity assay. This assay is fast, selective, applicable and accessible than other chromogenic assays (APAK et al., 2008). The method based on the reduction of Cu (II) to Cu (I). Antioxidant activity measured by copper (II) reduction assay at 450 nm using DR 5 000TM UV-Vis Spectrophotometer.

Chlorophyll content

Chlorophyll content was determined by the spectrophotometric method at 663 and 645 nm (Şükran et al., 1998). The chlorophyll a, b contents were calculated as follows:

Chlorophyll a = $(19.3 \times A663 - 0.86 \times A645)$ V/100W

Chlorophyll b = (19.3 ×A645 - 3.6 ×A663) V/100W Measurement of vitamin E content

Vitamin E content was measured according to the published method (WONG et al., 2006).

Samples were exposed to the Fe₃ solution, TPTZ and acetate buffer (pH=4). The standard curve was prepared with appropriate vitamin E concentration. The absorbance of samples was read at 595 nm wavelength.

Measurement of total flavonoids content

The total flavonoids content was determined by the previous method. The leaf extract was mixed with 2% AlCl3, 6H2O in methanol. Samples were incubated at 37 ° C for 15 minutes. The absorbance of these samples was measured at 430 nm by a spectrophotometer (TIAN et al., 2006).

Sample collection and preparation of trout fish fillet

The fresh rainbow trout fish samples were obtained from the fishmonger shop and the fillet was removed. The fillets were transferred to the chemistry laboratory and stored at 2- 4°C until use. The trout fillet samples were divided into between two groups (100 g each sample): one group was packed with a plastic bag and another packed and with the banana leaves. Both of them were analyzed at 20th and 40th days.

Measurement of lipid peroxidation level

In order to determine the inhibition of lipid peroxidation, we wereused thiobarbituric acid reactive substances (TBARS). The formation of thiobarbituric acid in trout fillet samples was assessed for measuring of lipid peroxidation in trout fillet according to an original method. Briefly, the supernatant of the fillet homogenate was a mixture, then centrifuged. In the next step, thiobarbituric acid was added to the supernatant and heated in 90°C for 90 minutes. The absorbance of the supernatant was measured at 532 nm. The values expressed in mg malondialdehyde (MDA)/L. All tests were performed in triplicate (SHAHIDI; ZHONG, 2005).

pH evaluation

The pH value of samples were determined using a digital pH meter (Kent, EIL7020, Kent Industrial Measurement Limited, Surrey, England) using 10 g of the blended sample with 50 mL distilled water. Means of three replicates were reported for each treatment.

Statistical analysis

All the experiments were repeated in triplicate. Statically analysis was performed by one-way Analysis of Variance (ANOVA). The data obtained were assessed using the SPSS 21.0 software package. Differences of p<0.05 were expressed significant.

RESULTS AND DISCUSSION

Results are given in table 1 and 2. Table 1 shows the properties of banana leaf extract including total carotenoids, total flavonoids, Chlorophyll a and b, Vitamin E and antioxidant activity.

Chemical composition	Amount	
Total carotenoids (mg/g	12.8±0.1	—
Total flavonoids (mg/g)	4.88±0. 21	
Chlorophyll a (mg/g)	114.3±3.08	
Chlorophyll b (mg/g)	102.8±2.55	
Vitamin E (mg/g)	5.8±0.61	
Antioxidant activity	1.76±0.09	

Table 2. Effect of banana leaf package on the shelf life of fish filet

	The days of storage	Rainbow trout fillet packaged with banana leaf	Rainbow trout fillet packaged with a plastic bag
Lipid oxidation mM/10g	Samples stored in 20 days	110± 6	140± 3
	Samples stored in 40 days	150±6 +	190 ± 7
рН	Samples stored in 20 days	6.1±0.02	6.3±0.02
	Samples stored in 40 days	6.2±0.05 *, +++	6.5±0.03

*: Significant differences between banana leaf package group and plastic bag group: *p*<0.05; +: Significant differences between samples stored in 20 days and 40 days in each group: *p*<0.01, *p*<0.001

The result of Analysis of Variance showed there was a significant difference between pH value in the treatment group and control group (plastic packaging) after 20 (P=0.01) and 40 (P=0.006) days. According to findings, the banana leaf extract reduces the pH of fish meat during storage.

In the present study, the TBA values of controls and treated samples significantly increased (p<0.05) with increasing the storage time. At the end of the first 20 days, there was a significant difference (p=0.02) in the TBA level in fish fillet treated with the banana leaf extract and control sample. However, at the end of the 40th day, there was no significant difference (P=0.09) in the TBA content between fish fillet treated with the banana leaf extract and control sample extract and control sample. After the end of the storage period, the highest TBA value was observed in the control sample (190 mM MDA/10g) as well as, the lowest of the TBA value was significantly reported in trout fillet treated with the banana leaf extract (110 mM MDA/10g).

In our study, we measured the total carotenoids, flavonoids, chlorophylls, vitamin E contents as well as, the antioxidant capacity of the banana leaf extract and also level of pH and lipid peroxidation of trout fillet treated with the banana leaves.

Extracts and natural compounds of plants have antioxidant effects due to the antioxidant compounds such as polyphenols, flavonoids and etc. (PEZESHK et al., 2011). The banana fruits, as well as various other parts of the banana tree, find various uses in medicine, fiber making, religious rituals. The leaves are used by the tribals of the Western Ghats in India for bandaging cuts, blisters, and ulcers (SAHAA et al., 2013). Therefore, this plant can be used to increase the shelf-life of foods as well as prevent the oxidation process in food formulations.

In our study, the antioxidant activity of banana leaves was 1.76 ± 0.09 . The results of

Vitamin E reacts rapidly with organic free radicals. It is widely accepted that the antioxidant properties of this compound are partly responsible for its biological activity. Vitamin E has many biological functions, including fat-soluble antioxidant. The vitamin E content in our study was 5.8±0.61 mg/g, which was higher than other studies (CHANWITHEESUK et al., 2005).

The total carotenoids content of banana leaf was 12.8 ± 0.1 mg/g. It has been obtained in the otherstudies; (FEYZI et al., 2015). However, the total carotenoids content of Scutellaria pinnatifida as 3.52 ± 0.56 which was much lower than the present study. Carotenoids, as natural antioxidant pigments in the banana leaf extract played a protective role in trout fillet. The extract inhibited the disruptions of trout fillet due to oxidative processes or lipid oxidation. On the other hand, the presence of carotenoids increases the oxidation stability of salmon fillet.

Phenolic compounds are important fruit, which mainly include components of (flavanones. flavonoids flavonols. flavones. isoflavones, flavanols, anthocyanidins), phenolic acids, coumarins, and tannins. The antioxidant properties of the phenolic compounds are related to their structures, including the substitutions on the aromatic ring and the structure of the side chain (MATHEW et al., 2015). The total phenol and total flavonoid contents of the banana leaf extract are reported as pharmaceutical and nutritional substances (ELEAZU et al., 2010). Our study showed the total flavonoid content of banana leaf extract was 4.88 ± 0.21 mg/g. The anti-radical activity of extract could be associated with the level of total phenolic compounds (SAEED et al., 2012).

Table 2 shows the variation of pH and TBA in rainbow trout fillet in 40 days of storage. The highest level of pH (6.5) was in control group on day 40 and the lowest of level pH (6.1) was in the trout fillet treatment with banana leaf extract on day 20.

The pH in fresh fish is almost neutral. Fish have a small content of carbohydrates (less than 5%) in their muscle tissue. Therefore, a small lactic acid content is produced after death. The pH values of trout fillet treated with banana leaves were Less than the control sample during freezing storage (p<0.05).

There was the significant difference (p<0.001) in pH between the control sample at the end of 20 and 40 days. therefore, it suggests that fish in this type of packaging (packaging with plastic without any extract) will change gradually over time. In the postmortem period, nitrogenous compounds will be decomposed by proteolytic enzymes activity and increased pH in fish meat during freezing storage (GÖKOĞLU et al., 2004).

Fish are more vulnerable than red meat and chicken as it contains relatively large contents of unsaturated fatty acids, and free amino acids with a higher pH which limiting the product of shelf life (MEXIS et al., 2009). Lipid oxidation occurs in various food particularly enriching with polyunsaturated fatty acids (SADIGHARA, 2012). Thiobarbituric acid reactive substances are used for measuring secondary oxidation products (SHAHIDI; ZHONG, 2005). The increased TBA indicates the progression of lipid oxidation, decomposition of hydroperoxides, and production of volatile substances, especially aldehyde. It has been proposed that the maximum level of TBA indicating good quality of the fish.Our data showed 2 mg of MDA/kg of fish flesh. According to the results obtained which shown in Table 2, although in both samples (control and treated with banana leaf), TBA and pH increased during freezing storage, however, the TBA and pH level was lower in trout fillet treated with the banana leaf compared to the control sample. Increasing in TBARS levels in the banana leaf extract treated samples was very slow compared to the control sample. The reduction in TBARS level was significantly associated with the total phenolic, total carotenoid and flavonoid contents. In conclusion, the present study demonstrated that the banana leaf extract acts as natural antioxidants that retard lipid oxidation in fish fillet during freezing storage. The present study suggests that the banana leaf extract has beneficial effects in delaying lipid oxidation in fish fillet tissue due to its natural antioxidant compounds such as vitamin E, total phenol, and carotenoid compounds.

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RESUMO: O objetivo deste estudo foi avaliar o efeito do extrato de folhas de bananeira sobre a qualidade e vida de prateleira da truta arco-íris comparada a sacolas plásticas na temperatura de congelamento por 40 dias. Para avaliar essa proposta, foi determinada a atividade antioxidante do extrato de folhas de

bananeira. Além disso, a vida de prateleira dos filés de peixe foi determinada medindo o ácido tiobarbitúrico (TBA) e o pH do peixe. O extrato de folhas de bananeira apresentou o maior teor de vitamina E $(5,8 \pm 0,61 \text{ mg/g})$ e carotenóides $(12,8 \pm 0,1 \text{ mg/g})$. O potencial de redução de Cu (II) no extrato foi de $1,76 \pm 0,09$. A magnitude da modificação no TBA e pH do peixe embalado com folhas de bananeira foi menor que as amostras controle. O presente estudo demonstrou que o uso de extrato de folhas de bananeira é capaz de retardar a oxidação lipídica no filé de peixe durante o armazenamento de congelamento, devido às suas fortes propriedades antioxidantes.

PALAVRAS-CHAVE: Extrato de folhas de bananeira. Antioxidante. Vida de prateleira. Peixe.

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