

The Restraining Effects Of Pre-Treatment With Turmeric Extract And Green-Tea Extract In Lipid Peroxidation Of Refrigerated Eels

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ABSTRACT

Turmeric and green-tea was extracted using water to investigate their effects in restraining the lipid oxidation. 80 g of eel was dipped into 150 mL of extracted solution for one hour, cleaned up with fresh water, pre-treated with applicable solution, and then vacuumed. Eel was refrigerated for 16 weeks for the measurement of its oxidation degree. Acid value, peroxide value, COV, and TBA value were measured using extracted eel oil from refrigerated eels. While untreated eels were in the control, eels pre-treated with Vitamin C 10 mM solution were in the comparison group. During 16 weeks of refrigeration, the group pre-treated with green-tea extract or turmeric extract, and Vitamin C 10 mM solution restrained significantly the lipid peroxidation ($p < 0.05$). For the acid value, the pre-treatment with green-tea extract was the lowest; the pre-treatment with turmeric extract recorded the lowest peroxide value. Both cases showed similar antioxidant effects as in Vitamin C 10 mM solution; however, the pre-treatment with green-tea extract or turmeric extract reported significantly higher antioxidant effects in carbonyl and TBA value than Vitamin C 10 mM solution did ($p < 0.05$). In conclusion, eels pre-treated with green-tea extract or turmeric extract and refrigerated showed similar or better antioxidant effects than those of Vitamin C 10 mM solution; therefore, it was reasonably concluded that eels' quality could be maintained during the distribution by preventing the lipid oxidation with pre-treatment of green-tea extract or turmeric extract.

Keyword: eel, green-tea, turmeric, lipid peroxidation, peroxide value

INTRODUCTION

Eel was traditionally known not only as stamina food, but as one of the most favorite food in summer. However, as the skills of eel farming have advanced, eel's supplies and their consumptions increased, which also promoted the demands for researches on how to control their quality and improve the storage and distribution through the peroxide prevention (Song & Kim, 2018). In order to resolve the problem of easy rancidity in fishes with high unsaturated fat like eels, several researches have been undertaken in relation to various fishes, such as eels with the antioxidant solution pre-treatment (Song 2019), half-dried eels (Song & Kim, 2018), salted mackerel (Nam *et al.*, 2011), anchovy oil (Kang *et al.*, 2007), saury (Cook, 1995), seasoned squid (Yang *et al.*, 1999), white fish meat (Lee *et al.*, 1997), and shellfishes (Choi *et.al*, 1998), focusing on peroxide prevention. The aim of this research was to find out the antioxidant effects of extracts by measuring the degree of eel's peroxidation when green-tea extract and turmeric extract were pre-applied and stored in refrigeration as reported in many literatures (Oh *et al.*, 2010; Nam *et al.*, 2011; Choi *et al.*, 2012; Jung *et al.*, 2012; Kim & Jeong, 2017; Ryu *et al.*, 2017; Song & Kim, 2018; and Song, 2018).

MATERIALS AND METHODS

Samples

Eels (*Anguilla bicolor pacifica*) used in the experiment were raised in the city of Naju, weighed 250-300 g, eviscerated, and cleaned before the use. Green-tea used for extracting solutions to be pre-applied to eels for the purpose of preventing eel's peroxide was dried green-tea (Green tea, Damian, Korea) and turmeric (Turmeric, Woldeung Food, Korea) available in the market. 50 g of green-tea or turmeric was added to 2L water, and then, it was extracted for 2 h at 112°C using a pressured double boiler function in an electronic boiling pot (OC-2300R, OCOO, co., Ltd., Korea). After the first try, the extracted solution was poured out into a different cup. Then, the second solution was obtained through the same steps as in the first try. After the first solution and the second solution were made, two solutions were put together and were filtered using gauze and paper filter (No.1, Whatman, UK) in a sequential manner.

Pre-treatment of eels and eel oil extraction

80 g of eel and 150 mL of extracted solution were put together in a clean vinyl bag, and laid for one hour in a tied-up bag. After 1 h, eel was taken out to get a clean-up shower. Once cleaned and dried up,

eel was vacuumed and refrigerated for 16 weeks (Song, 2018). On the 1st, 3rd, 8th, 12th, and 16th week of refrigeration, eel oil was extracted for measuring the acid value, peroxide value, carbonyl value, and TBA value. Eel oil was extracted using the method specified by Folch *et al.*(1957). 80 g of chopped eel was mixed with 300 mL of blended solution of chloroform and methanol (2:1, v/v), then they were extracted using a homogenizer (SMG-G, Shinsang, Co., Ltd., Korea), and filtered using a paper filter (No.1). Another 250 mL of blended solution of chloroform and methanol was added to the residue, then they were extracted using a homogenizer, which constitute the second time of filtration. All obtained solutions were poured into a separatory funnel and mixed with distilled water, and laid for 15 to 20 h. After that, the layer of chloroform was separated, dehydrated by Na₂SO₄ and filtered (No.1). Eel oil was obtained by concentrating all filtered solutions under reduced pressure using a rotary evaporator (Rotavaor R-215, Büchi, Germany) at 40°C.

The measurement of acid value

After 1 to 2 gram of eel oil using a method specified in Korean food code was taken into a flask, 100 mL mixed solution of methanol and ether (2:1) was added. With 1-2 drop of phenolphthalein indicator injected, the point with pink color was chosen as the end point through titration by 0.1 N KOH (Kim *et al.*, 2015; Ministry of Food and Drug Safety, 2017; and Song, 2018).

The measurement of peroxide value

After 1 to 2 gram of lipid recovered using a method specified in Korean food code was taken into a flask, 25 mL mixed solution of acetic acid and chloroform (3:2, v/v) was added. On top of it, 1 mL of KI saturated solution was also added. Then, it was laid in the dark room for 10 minutes after being shaken for a minute. With 30 mL of distilled water and starch indicator solution injected, the point without color was chosen as the end point through titration by 0.01 N Na₂S₂O₃ (Kim *et al.*, 2015; Ministry of Food and Drug Safety, 2017; and Song, 2018).

The measurement of carbonyl value

After placing 0.05g of eel oil into 100 mL glass bottle with a cap, benzen 5 mL, 0.05% 2,4-DNPH(dinitrophenyl hydrazine) benzen 5 mL, and 4.3% trichloroacetic acid 3 mL were arranged to be injected into the bottle. Then, the mixture was warmed up in a double boiler of 60°C water bath for 30 minutes. After cooling it off at room temperature and popping the color with 10 mL of 4% KOH-ethanol, the absorbance was measured at 440 nm (Choi *et al.*, 2006; and Song, 2018).

The measurement of TBA value

After mixing and sonicating 200 mg of TBA with 100 mL of 95% butanol at 60°C sonicator (Ultrasonic, JAC 4020, KODO, Korea) for 30 minutes using a method specified in Korean food code (Ministry of Food and Drug Safety, 2017), TBA (thiobarbituric acid) reagent was taken into a form with a direct mix of glacial acetic acid at the rate of 1:1. TBA value was obtained through the absorbance measurement at 530 nm after cooling off the solution, a blend of 0.05 gram eel oil, 10 mL benzen, and 10 mL of TBA reagent, which was kept for 2 h in 95°C water bath, in the flowing water (Ministry of Food and Drug Safety, 2017; and Song, 2018).

Statistical analysis

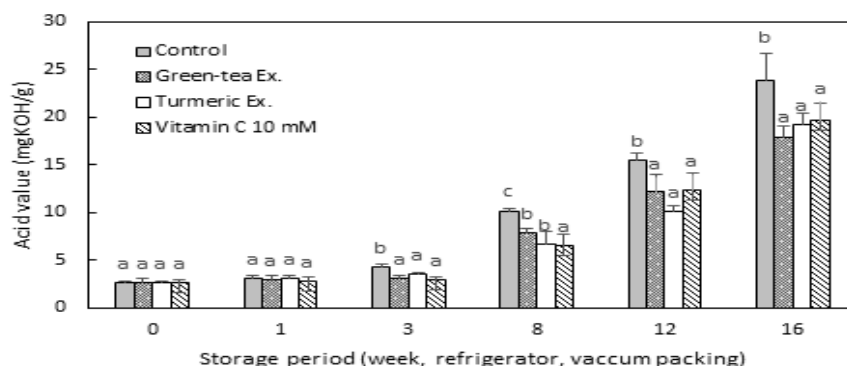
Comparative analysis among experiment groups was performed using IBM SPSS Statistics 20. After ANOVA was undertaken, Duncan's multiple range test was also performed with 5% confidence level ($\alpha=0.05$).

RESULTS AND DISCUSSION

Acid value

The acid value of the control increased 7.9 times during 16 weeks of refrigeration. On the contrary, the acid value increased only 5.8 times, 6.3 times, and 6.4 times when green-tea extract, turmeric extract, and Vitamin C 10mM solution were respectively applied, which was deemed effectively preventing the peroxidation ($p<0.05$). Up to 8 weeks of refrigeration, Vitamin C 10mM solution was more effective in lowering the acid value than the others, but there was no meaningful difference among green-tea extract, turmeric extract, and Vitamin C 10mM solution after 8 weeks of refrigeration ($p<0.05$).

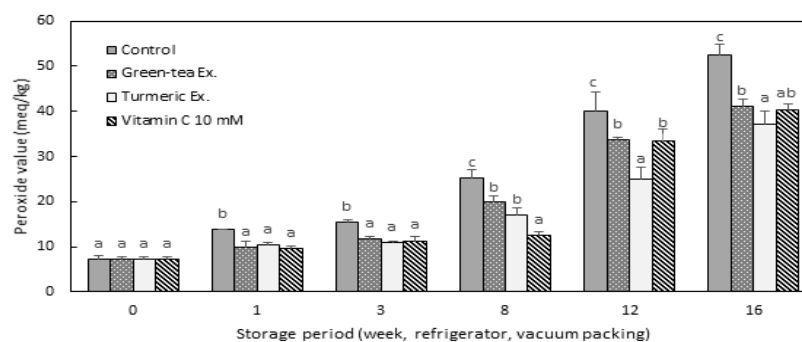
Fig. 1 Acid value of eels refrigerated for 16 weeks



Peroxide value

While the peroxide value of the control increased 2 times on the 3rd week compared to the beginning, it was less than 2 times when green-tea extract, turmeric extract, and Vitamin C 10mM solution was applied ($p<0.05$). On the 12th week, the peroxide value of the control went up 5.6 times, but those of the groups treated with green-tea extract and Vitamin C 10mM solution went up 4.7 times, and that of group treated with turmeric extract increased 3.5 times. After the 16th weeks of refrigeration, the groups treated with green-tea extract, turmeric extract, and Vitamin C 10mM solution revealed that the peroxide values were low enough to be deemed statistically meaningful. When treated with turmeric extract, the value decreased meaningfully after 3 weeks, compared to that of the other antioxidant solutions ($p<0.05$).

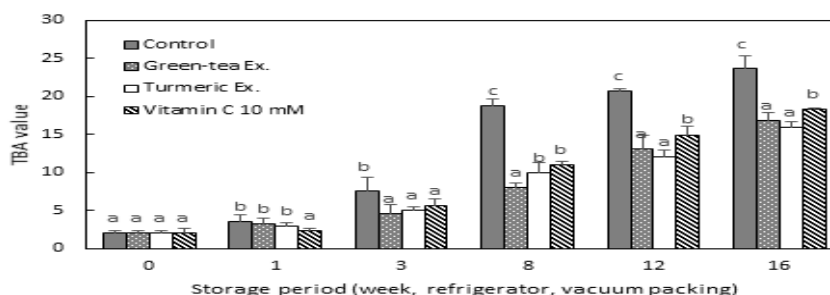
Fig. 2 Peroxide value of eels refrigerated for 16 weeks



TBA value

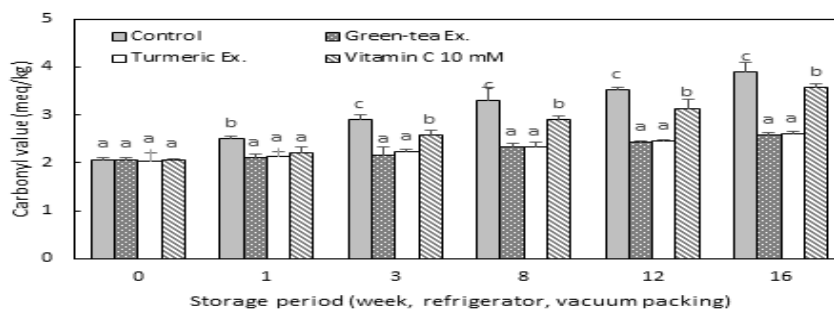
Throughout 16 weeks of refrigeration, TBA value of the control increased 11.2 times, and that of the groups treated with green-tea extract, turmeric extract, and Vitamin C 10mM solution went up 7.9 times, 7.5 times, and 8.7 times, respectively. Up to 8 weeks of refrigeration, green-tea extract treatment meaningfully prevented the creation of malonaldehyde more than the other treatments; however, from the 12th week, green-tea extract treatment and turmeric extract treatment yielded stronger antioxidant effects than Vitamin C 10 mM solution treatment. Although there was no meaningful difference, turmeric treatment prevented malonaldehyde more than green-tea extract treatment.

Fig. 3 TBA value of eels refrigerated for 16 weeks



Carbonyl value

Throughout 16 weeks of refrigeration, carbonyl compound of the control increased 1.9 times, and that of the groups treated with green-tea or turmeric extract, and Vitamin C 10mM solution went up 1.3 times, and 1.8 times, respectively. From the 3rd week of refrigeration, green-tea or turmeric extract treatment meaningfully prevented the creation of carbonyl compound more than Vitamin C 10mM solution treatment; in other words, from the 3rd week of refrigeration, green-tea or turmeric extract treatment was more effective in preventing carbonyl compound than Vitamin C 10mM solution treatment. Many researches have asserted that the pre-treatment of green-tea water and ethanol extract was as effective as Vitamin C 10mM solution in preventing the peroxide of eels (Song, 2018), and that green-tea water extract was effective in preventing the peroxide of anchovy oil (Kang *et al.*, 2011). However, there was few research investigating turmeric's antioxidant effects preventing eel's peroxidation among many researches reporting turmeric's antioxidant activities (Oh *et al.*, 2010; Choi *et al.*, 2012; Jung *et al.*, 2012; and Kim & Jeong, 2017). Ginger extract, which was said to have radical scavenging effects similar to turmeric (Jung *et al.*, 2012), was reported to be effective in controlling not only half-dried eels' peroxidation (Song, 2019) but also that of sardine oil, mackerel, and saury (Byun *et al.*, 1986; Lee & Lee, 1990; and Cook, 1995). Upon reviewing the results on the acid value, the peroxide value, the TBA value, the carbonyl value, the findings were as follows. Green-tea water extract restrained the lipid peroxidation of eels more than Vitamin C 10 mM did even in the long-term refrigeration between 3 weeks and 16 weeks. And turmeric water extract was found to have higher antioxidant activities than green-tea extract and Vitamin C 10 mM did in the long-term refrigeration over 12 weeks. Green-tea extract or turmeric extract was also proved to more effectively restrain the second oxidative compound, such as malonaldehyde or carbonyl compound than Vitamin C 10 mM did. Therefore, it was reasonably concluded that eels' quality could be maintained during the distribution by preventing the lipid oxidation with pre-treatment of green-tea extract or turmeric extract.

Fig. 4 Carbonyl value of eels refrigerated for 16 weeks

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