

Monitoring Color Values of The Fillet and Skin of Seabass and Seabream Pre-treated with Bleaching Agent Solutions

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Abstract

This study monitored the color changes of seabream and seabass fillet and skin parts immersed in hydrogen peroxide (H_2O_2), and colloidal silver added H_2O_2 solutions (800 ppm). The images were taken on three-point of the process: fresh, after immersion, and one-day storage at 4°C. Computer-based image analysis technology was used for color determination, and the color values examined were L^* , a^* , b^* , Chroma, and Whiteness. According to the results, L^* and Whiteness values of fillets were higher than the initial color values of all groups ($p < 0.05$). However, no statistical difference was found between after immersion and one-day storage in L^* values of HBS, CBS, and CBR ($p > 0.05$). The a^* , b^* , and Chroma values were decreased for all groups' fillets after application ($p < 0.05$). There is no effect of solutions on the brightness and yellowness value of seabream skin. However, the changes in seabass skin color upon the applications were statistically different ($p < 0.05$). Although color changes were followed, after immersion, data about the H_2O_2 applications' superiority could not be obtained.

Keywords: Color, Skin, Fillet, Hydrogen Peroxide, Bleaching, Fish.

1. INTRODUCTION

Seafood has a high nutritional value and digestibility¹. However, it is one of the fastest deteriorating food group. Therefore, many applications have been developed throughout history to extend their shelf life. Pre-treatment of fish before processing is one of these methods. It provides surface protection by immersion in various solutions. It is preferred that these solutions do not leave residues and do not negatively affect human health. Hydrogen peroxide (H_2O_2) is one of these substances. In addition to leaving no residual material, it is cheap and easy to find. H_2O_2 solutions are also known as strong oxidants, bleaching agents, and mild antibacterial chemicals^{2,3}.

In 2018, the United Nations Food and Agriculture Organization announced that the world's total Gilthead seabream and European seabass production were 237 049 and 240 673 tons, respectively⁴. The fillet of these species with large amounts of production is quite valuable. Therefore, it is essential to improve and maintain fillet quality with pre-applications to minimize production loss. The most important one of these quality parameters is color. Color is the first noticed quality parameter by the consumer before buying a food product such as seafood. The color of seafood skin and fillet varies according to the fish species. Seabream and seabass have partly white flesh while their skins are silver. Improving the whiteness of this flesh is essential for consumer appreciation. There is some research about H_2O_2 as a bleaching agent in processed seafood^{5,6}. However, there are no researches about the effect of H_2O_2 on seafood color in the literature.

Instrumental methods perform color analysis. For the last forty years, color analysis has been carried out by the computer-based color analysis method. This method gives the most effective results on surfaces that do not have a homogeneous color distribution, especially in seafood ⁷. Computer-based color analysis is another advantage of providing fast, objective, and reliable data ⁸.

2. MATERIAL AND METHODS

2.1. Raw Material

In this study, 16 seabreams and 16 seabass with an average weight of 290 ± 150 g were used. Whole fish samples were transported from a local fish supplier for 12 hours after catching in ice-filled styrofoam boxes and carried out for analysis at the laboratory. Cold storage conditions were maintained before and during transportation.

2.2. Chemicals

The chemicals used in the study were H_2O_2 (30%, aqueous) (Tekkim Inc., İstanbul, Turkey) and colloidal silver added H_2O_2 (50%, aqueous) (SilverTech Chemistry Inc., Ankara, Turkey). Eight hundred ppm solutions of these chemicals were prepared separately. These solutions were applied according to the groups described in the experimental design section.

2.3. Experimental Design

Fresh fish samples were eviscerated and filleted immediately after transportation to the laboratory. The experimental designs of groups were determined as;

SBS: colloidal silver added H_2O_2 group for seabass.

SBR: colloidal silver added H_2O_2 group for sea bream.

HBS: H_2O_2 group for seabass.

HBR: H_2O_2 group for sea bream.

CBS: the control group for seabass.

CBR: a control group for sea bream.

The SBS and SBR were immersed in the 800 ppm silver added H_2O_2 solution, and HBS and HBR groups were immersed in the 800 ppm H_2O_2 solution for 24 hours in a refrigerator (Emsaş Inc, Manisa, Turkey) at $4 \pm 3^\circ C$. Tap water was used as an immersion solution for the CBS and CBR. After 24 hours of immersion, the samples were drained on the laboratory desk for 10 mins and then stored one more day in a plastic box at $4^\circ C$. Every piece of samples' pictures of the fillet and skin parts was taken in the computer-based image analysis system before immersion (fresh), after immersion, and one-day storage.

2.4. Computer-based Image Analysis

The illumination system (lightbox) was used to take images. The system was 122 cm high, 61 cm wide, 46 cm deep, and had 2 LED light sources (one up and one bottom) (ForLife FLP-15, India) with color temperature 6500K (D65 illumination). The upper light source surface was covered with a polarizing sheet (Rosco, Stamford, CT, USA). A color reference (Gretag Color Checker, X-Rite Inc., Grand Rapids, MI, USA) was placed in the box to determine the right color. A Nikon D300 digital camera (Nikon Corp., Tokyo, Japan) with an 18–200 mm zoom Nikkor lens with a 72 mm diameter circular polarizing filter attached was used to take pictures in the lightbox USB cable. The Nikon Camera Control Pro (Nikon Corp., Tokyo, Japan) was used to control camera settings. The camera was set to manual mode, with an ISO setting of 200. Two image method described by Alçiçek & Balaban ⁸ was used. The pictures (2144*1424 pixels) were transferred to a PC immediately after image acquisition. LensEye software (Gainesville, FL, USA) was used to analyze the visual attributes of samples.

2.5. Statistical Analysis

The normally distributed data⁹ were analyzed by one-way ANOVA using SPSS v23.0 (IBM Statistical Package for the Social Sciences Inc., Chicago, IL, USA). Tukey's multiple range test was used to compare the differences at $p < 0.05$. The bar graphs were generated in the same program, and the error bars showed 95% Confidence Interval.

3. RESULTS AND DISCUSSION

L^* , a^* , b^* , Chroma, and Whiteness values changes of the fillet and skin part of the groups during the initial (raw), after immersion and one-day storage were shown in Table 1.

3.1. Fillet Color Values Results

The results showed that the L^* value of SBR was increased ($p < 0.05$), then after one-day storage, the L^* value of SBR slightly decreased ($p > 0.05$) (Figure 1). These results show that colloidal silver added H_2O_2 showed a bleaching effect on the sea bream fillet. The same trend was observed in the HBR group (Table 1). This trend was probably due to H_2O_2 ingredients in two of the immersion solutions. However, no statistically significant difference was found between SBR, HBR, and CBR groups after application ($p > 0.05$). This result shows that H_2O_2 solutions do not have efficacy on the L^* value of sea bream fillets.⁵ found a decreased L^* value trend for carp surimi, but this was not statistically significant. These different results probably due to the different color assessment process used.⁶ were also reported the H_2O_2 has a whitening effect on the fish. L^* value of seabass fillet groups increased significantly after application ($p < 0.05$) (Figure 1). However, the L^* value decreased slightly after one-day storage without any significance ($p > 0.05$) (Table 1). The decreased L^* value, also reported by Kim et. al.². Our study results showed no significance between SBS, HBS, and CBS groups in initial, after applications, and after one-day storage, respectively ($p > 0.05$). The L^* value decreasing of the fish samples was recorded as a result of lipid oxidation by Kim et. al.².

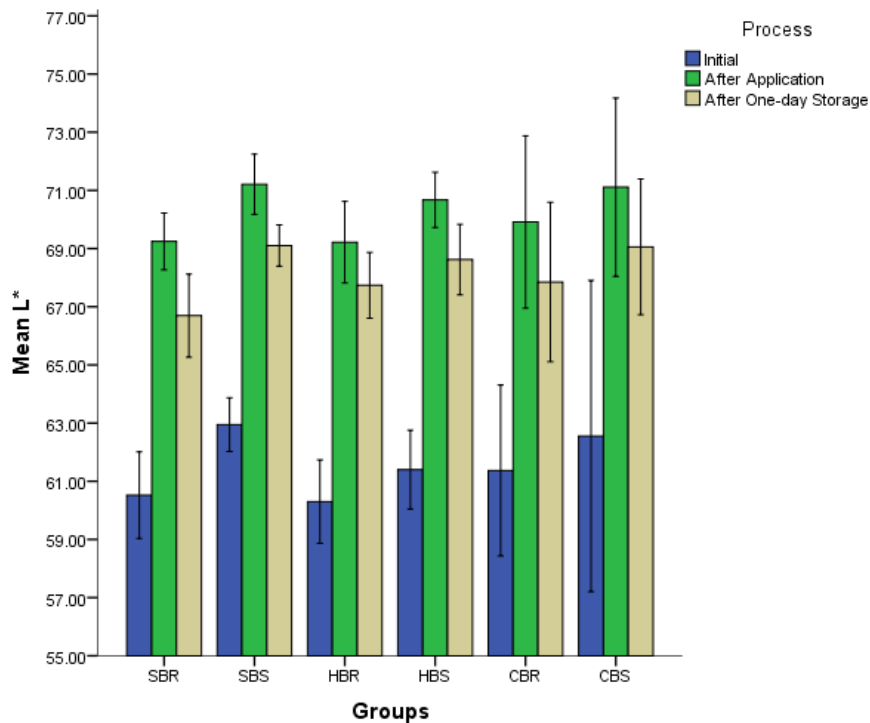


FIGURE 1: The mean L^* values of the sample groups' fillet part (The error bar showing a 95% confidence interval).

The initial a^* value of the SBR was 8.75 ± 0.74 . However, after application, the fillets a^* value was dramatically decreased ($p < 0.05$) (Figure 2). This change was slightly changed after one-day storage, but there was no significance between after application and one-day storage processes ($p > 0.05$). The similarities of these changes were observed in both HBR and CBR groups (Table 1). When the groups were compared during the processes, there was a difference between the HBR and CBR groups in the "after the application" process. Besides, a^* value was not statistically different between the groups at all times. ⁵ found an increase for the carp surimi samples' a^* value, but there was also not found significance between initial and H_2O_2 applied groups statistically.

The initial a^* value of SBS was 9.01 ± 1.62 (Figure 2). The a^* value of the SBS dramatically decreased after the application process. This decrease was statistically different ($p < 0.05$), then slightly increased after one-day storage ($p > 0.05$). HBS followed the same trend. Although CBS a^* value has the same trend, the statistical significance was not observed during the process ($p > 0.05$) (Table 1). The a^* value of SBS, HBS, and CBS were not statistically significant after application ($p > 0.05$). The same results were found in the "after one-day storage" process ($p > 0.05$).

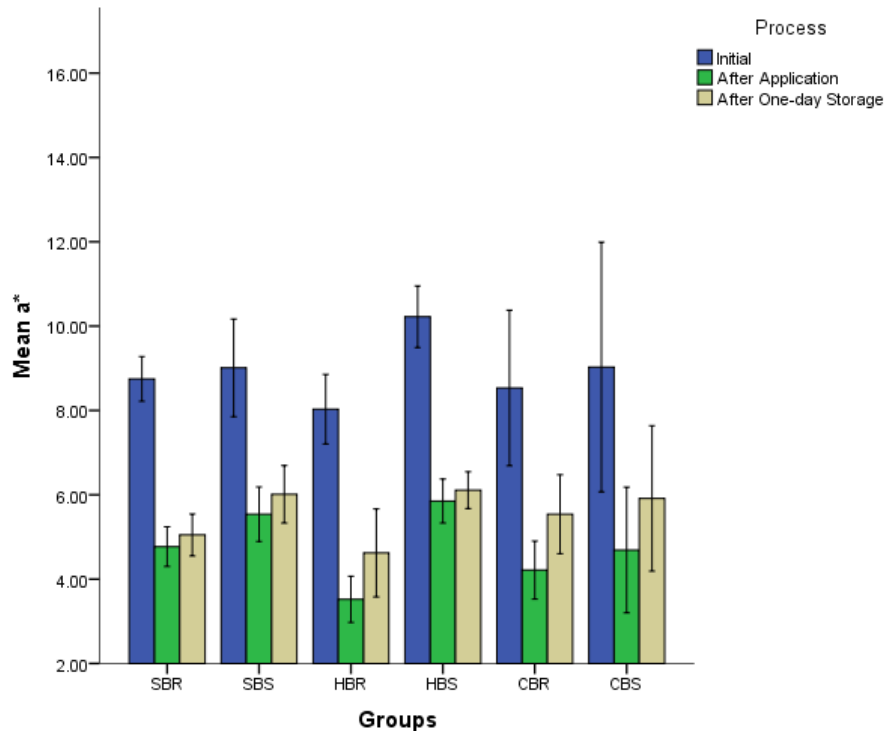


FIGURE 2: The mean a^* values of the sample groups' fillet part (The error bar showing a 95% confidence interval).

The b^* value of the SBR, HBR, and CBR groups decreased significantly after application. After one day of storage, a little increase was observed. This increase is not statistically significant in all groups ($p > 0.05$) (Table 1) (Figure 3). However, there was a similarity between the b^* value of the SBR and HBR groups after application ($p > 0.05$). However, it was concluded that the difference between the CBR group and SBR and HBR groups was statistically different ($p < 0.05$). Also, there was no statistical difference between the groups' b^* values after one-day storage ($p > 0.05$).

SBS, HBS, and CBS groups initial b^* values decreased after application. This change was statistically significant ($p < 0.05$) (Table 1) (Figure 3). However, there were no statistical differences

found after one-day storage between initial and after application processes for all groups ($p>0.05$). There was a statistical difference between HBS and CBS after application ($p<0.05$). This decrease may be related to brown color, which occurred during lipid oxidation ².

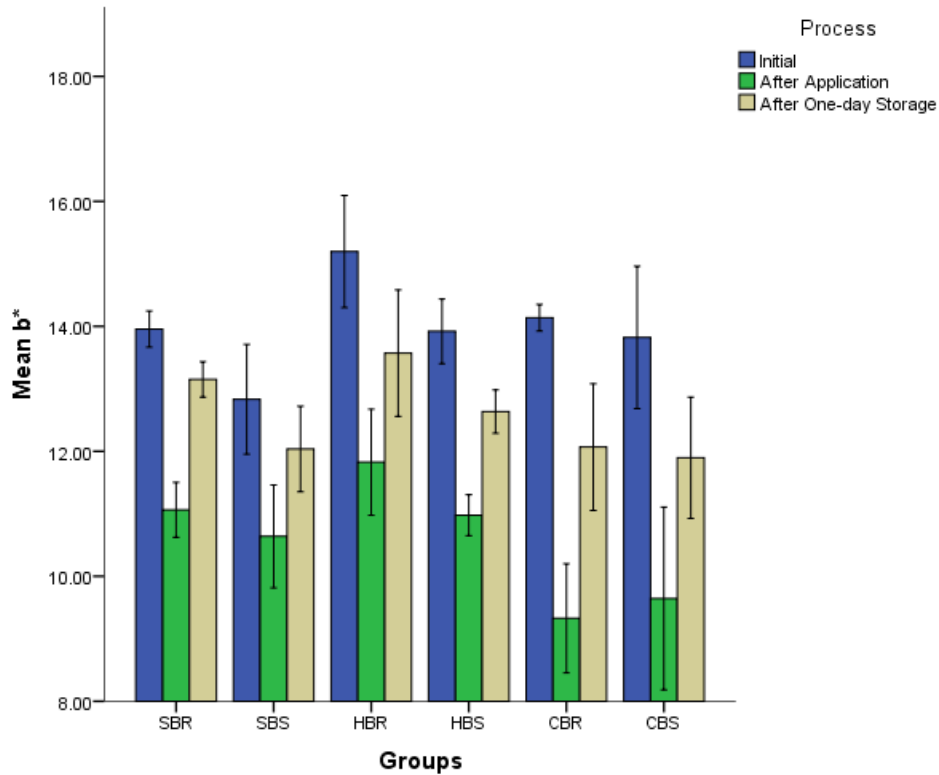


FIGURE 3: The mean b^* values of the sample groups' fillet part (The error bar showing a 95% confidence interval).

The Chroma values of the seabream fillet groups were decreased after application ($p<0.05$) and slightly increased after one-day storage ($p<0.05$). Chroma of the SBR and HBR groups were statistically similar after application ($p>0.05$) (Table 1) (Figure 4). However, the CBR group was statistically different from these two groups after application ($p<0.05$). After one-day storage, this difference was observed only in the HBR group. There was no statistical difference between SBR and CBR after one-day storage ($p>0.05$).

Chroma values of the initial seabass fillets changed after application. This change was found statistically different ($p<0.05$) (Table 1) (Figure 4). After one-day storage, the samples' Chroma value was moderately increased, but it was not significantly different ($p>0.05$). According to the results, SBS and HBS Chroma values have a statistically same trend ($p>0.05$), but these two groups were statistically different from the CBS group ($p<0.05$) after the application process.

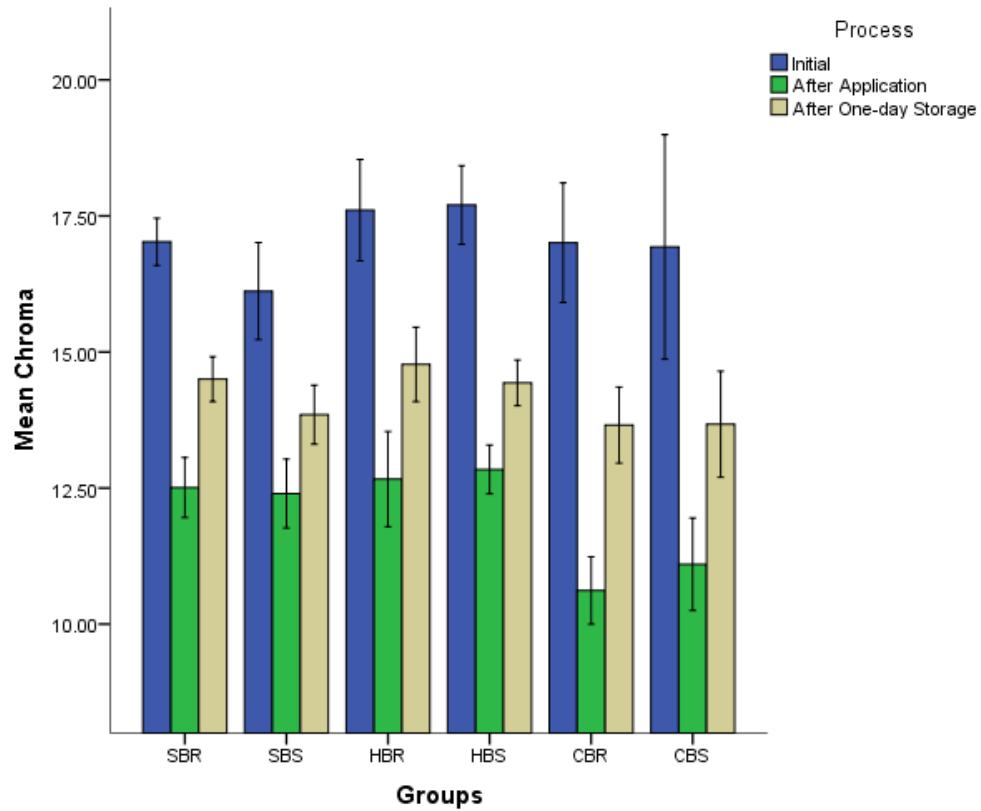


FIGURE 4: The mean Chroma values of the sample groups' fillet part (The error bar showing a 95% confidence interval).

Initial Whiteness values of the SBR, HBR, and CBR were 56.68, 56.21, and 57.49, respectively (Table 1) (Figure 5). All Whiteness values of the groups significantly increased after application ($p < 0.05$), then slightly decreased after one-day storage ($p > 0.05$). There was no significance between groups during processes ($p > 0.05$).

SBS, CBS, and HBS initial Whiteness values showed a statistically significant change after application processes ($p < 0.05$) (Table 1) (Figure 5). All groups Whiteness values decreased slightly after one-day storage ($p > 0.05$). The results showed that CBS and HBS groups were significantly different after the application process ($p < 0.05$). However, there was no significant change between SBS and HBS, and SBS and CBS groups Whiteness value ($p > 0.05$). This value did not show any significance between all groups after one-day storage ($p > 0.05$).

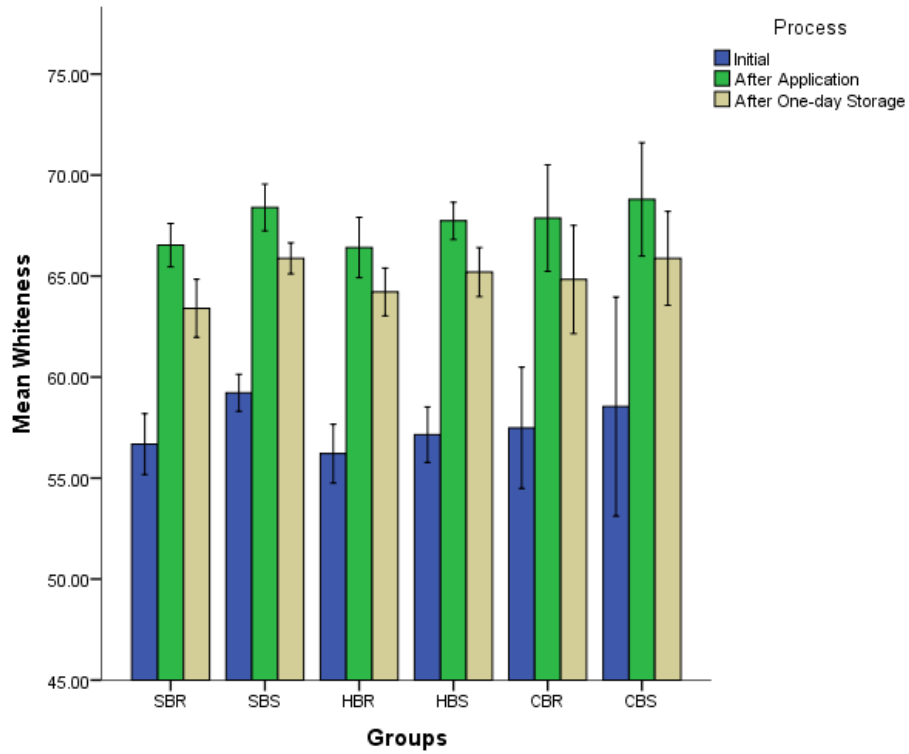


FIGURE 5: The mean Whiteness values of the sample groups' fillet part (The error bar showing a 95% confidence interval).

Table 1. The color values of fillet and skin of the samples during the process

		Groups						
	Process	SBR*	SBS	HBR	HBS	CBR	CBS	
Fillet	L*	Initial	60.52±2.08 ^{Aa**}	62.95±1.29 ^{Aa}	60.30±2.01 ^{Aa}	61.40±1.90 ^{Aa}	61.37±1.85 ^{Aa}	62.55±3.36 ^{Aa}
		After App	69.25±1.36 ^{Ab}	71.21±1.45 ^{Ab}	69.22±1.96 ^{Ab}	70.67±1.32 ^{Ab}	69.91±1.86 ^{Ab}	71.11±1.93 ^{Ab}
		Day 1	66.69±2.00 ^{Ac}	69.10±0.99 ^{Ac}	67.74±1.58 ^{Ab}	68.62±1.70 ^{Ac}	67.85±1.72 ^{Ab}	69.06±1.47 ^{Ab}
	a*	Initial	8.75±0.74 ^{Aa}	9.01±1.62 ^{Aa}	8.03±1.15 ^{Aa}	10.22±1.02 ^{Aa}	8.54±1.16 ^{Aa}	9.03±1.86 ^{Aa}
		After App	4.77±0.66 ^{Ab}	5.54±0.90 ^{Ab}	3.52±0.77 ^{Bb}	5.85±0.73 ^{Ab}	4.22±0.43 ^{Bb}	4.69±0.94 ^{Ab}
		Day 1	5.05±0.69 ^{Ab}	6.01±0.95 ^{Ab}	4.62±1.46 ^{Ab}	6.11±0.61 ^{Ab}	5.54±0.59 ^{Ab}	5.92±1.08 ^{Ab}
	b*	Initial	13.96±0.41 ^{Aa}	12.83±1.23 ^{Aa}	15.20±1.26 ^{Ba}	13.92±0.72 ^{ABa}	14.14±0.13 ^{ABa}	13.82±0.72 ^{Ba}
		After App	11.06±0.62 ^{Ab}	10.64±1.15 ^{ABb}	11.83±1.19 ^{Ab}	10.98±0.46 ^{Ab}	9.33±0.55 ^{Bb}	9.65±0.92 ^{Bb}
		Day 1	13.15±0.40 ^{Ac}	12.04±0.96 ^{Aa}	13.57±1.41 ^{Ac}	12.64±0.48 ^{Ac}	12.07±0.64 ^{Ac}	11.90±0.61 ^{Ac}
	Chroma	Initial	17.02±0.60 ^{Aa}	16.12±1.25 ^{Aa}	17.61±1.30 ^{Aa}	17.70±1.01 ^{Ba}	17.01±0.69 ^{Aa}	16.93±1.30 ^{ABa}
		After App	12.51±0.77 ^{Ab}	12.40±0.88 ^{Ab}	12.67±1.22 ^{Ab}	12.84±0.63 ^{Ab}	10.62±0.39 ^{Bb}	11.10±0.53 ^{Bb}
		Day 1	14.50±0.58 ^{ABc}	13.85±0.76 ^{Ac}	14.77±0.96 ^{Ac}	14.43±0.59 ^{Ac}	13.66±0.44 ^{Bc}	13.67±0.61 ^{Ac}
Whiteness	Initial	56.68±2.11 ^{Aa}	59.22±1.28 ^{Aa}	56.21±2.03 ^{Aa}	57.15±1.92 ^{Aa}	57.49±1.89 ^{Aa}	58.54±3.41 ^{Aa}	
	After App	66.53±1.50 ^{Ab}	68.40±1.61 ^{ABb}	66.41±2.09 ^{Ab}	67.73±1.29 ^{Ab}	67.87±1.66 ^{Ab}	68.80±1.76 ^{Bb}	
	Day 1	63.41±2.01 ^{Ac}	65.88±1.07 ^{Ac}	64.22±1.65 ^{Ac}	65.20±1.70 ^{Ac}	64.83±1.68 ^{Ab}	65.88±1.46 ^{Ab}	
Skin	L*	Initial	60.16±2.86 ^{Aa}	62.79±2.33 ^{Aa}	56.09±6.12 ^{Aa}	62.58±3.47 ^{Aa}	59.93±1.43 ^{Aa}	63.75±6.40 ^{Aa}
		After App	63.45±2.17 ^{Ab}	64.89±3.59 ^{Aa}	63.09±2.51 ^{Ab}	65.28±3.67 ^{Aa}	62.67±2.80 ^{Ab}	64.95±4.88 ^{Aa}
		Day 1	63.26±1.85 ^{Ab}	65.50±5.22 ^{Aa}	63.20±2.37 ^{Ab}	65.45±2.74 ^{Aa}	64.14±1.19 ^{Ab}	63.43±5.57 ^{Aa}
	a*	Initial	0.77±0.36 ^{Aa}	1.34±0.62 ^{Aa}	0.19±0.69 ^{Ba}	1.70±0.52 ^{Aa}	0.82±0.49 ^{ABa}	2.35±0.82 ^{Aa}
		After App	-0.69±0.17 ^{Ab}	-0.09±0.68 ^{Ab}	-0.94±0.19 ^{Bb}	0.24±0.39 ^{Ab}	-0.42±0.19 ^{Ab}	0.25±0.47 ^{Ab}
		Day 1	-0.48±0.19 ^{Ab}	-0.01±0.66 ^{Ab}	-0.68±0.22 ^{Ab}	0.06±0.31 ^{Ab}	0.00±0.03 ^{Bb}	0.65±0.50 ^{Ab}
	b*	Initial	7.66±0.65 ^{ABa}	5.47±1.37 ^{Aa}	7.71±0.69 ^{Aa}	5.63±1.90 ^{Aa}	8.29±1.11 ^{Ba}	5.65±1.57 ^{Aa}
		After App	6.79±0.78 ^{Ab}	5.35±0.57 ^{Aa}	6.16±0.44 ^{Ab}	4.89±1.49 ^{Aa}	7.72±0.24 ^{Ba}	5.78±0.66 ^{Aa}
		Day 1	6.39±0.88 ^{ABb}	4.32±1.21 ^{Aa}	5.49±0.93 ^{Ab}	4.24±1.66 ^{Aa}	6.83±0.69 ^{Ba}	5.27±1.60 ^{Aa}
	Chroma	Initial	8.08±0.54 ^{Aa}	7.81±0.67 ^{Aa}	7.99±0.69 ^{Aa}	8.16±0.55 ^{Aa}	8.67±0.97 ^{Aa}	8.54±1.09 ^{Aa}
		After App	6.96±0.76 ^{Ab}	6.28±0.31 ^{Ab}	6.30±0.43 ^{Bb}	6.32±0.88 ^{Ab}	7.86±0.23 ^{Cab}	6.72±0.27 ^{Ab}
		Day 1	6.71±0.64 ^{Ab}	6.34±0.46 ^{Ab}	5.84±0.68 ^{Bb}	6.21±0.64 ^{Ab}	7.13±0.55 ^{Ab}	6.68±0.62 ^{Ab}
Whiteness	Initial	58.99±2.83 ^{ABa}	61.44±2.32 ^{Aa}	55.14±6.06 ^{Aa}	61.14±3.43 ^{Aa}	58.73±1.24 ^{Ba}	62.04±6.62 ^{Aa}	
	After App	62.60±2.20 ^{Ab}	64.01±3.46 ^{Aa}	62.44±2.50 ^{Ab}	64.35±3.70 ^{Aa}	61.70±2.76 ^{ABa}	63.93±4.89 ^{Aa}	
	Day 1	62.41±1.76 ^{Ab}	64.55±5.08 ^{Aa}	62.56±2.36 ^{Ab}	64.53±2.76 ^{Aa}	63.22±1.30 ^{Ab}	62.37±5.57 ^{Aa}	

*Mean±Std. Deviation was given. **small letters show the difference in the same fish part, same color value, and different process (p<0.05). Capital letters show the difference in the same fish part, same fish species, same color value, and different group (p<0.05)

3.2. Skin Color Values Results

This study is the first study examining seabass and seabream skins treated with hydrogen peroxide. Skin L^* values of the seabream groups increased after application. This change was significantly different for all groups ($p < 0.05$) (Table 1) (Figure 6). However, there was no significant difference between after application and after one-day storage processes for all groups ($p > 0.05$). SBR, HBR, and CBR groups L^* values were not significantly different after applying ($p > 0.05$). The same trend was determined after a one-day storage process.

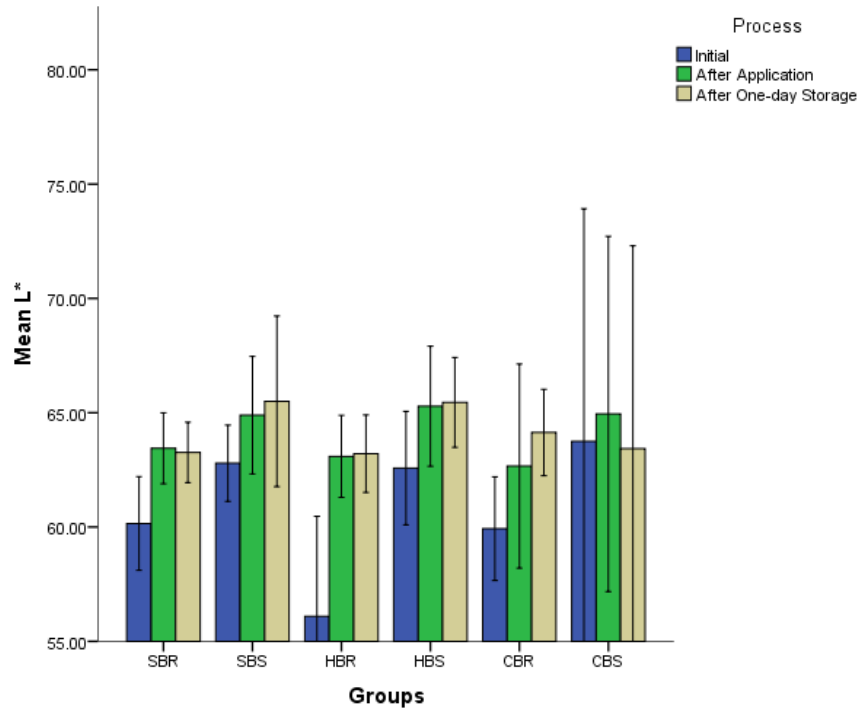


FIGURE 6: The mean L^* values of the sample groups' skin part (The error bar showing a 95% confidence interval).

The a^* values of the SBR, HBR, and CBR groups dramatically decreased after the application process, and the decrease was found statistically significant ($p < 0.05$) (Figure 7) for all groups. After one-day storage, a^* value changes were not significant for SBR and HBR ($p > 0.05$), but for CBR ($p < 0.05$). The same process and different group comparison results showed that SBR and HBR a^* values were significantly different after the application process ($p < 0.05$), but after one-day storage, the significance did not observe yet ($p > 0.05$).

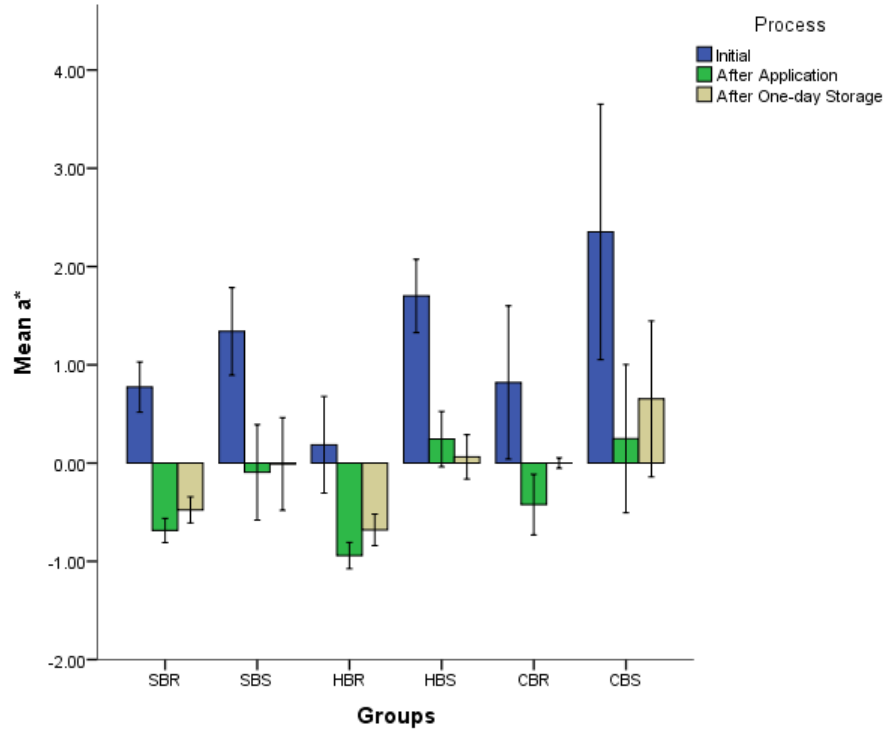


FIGURE 7: The mean a^* values of the sample groups' skin part (The error bar showing a 95% confidence interval).

b^* value of the SBR and HBR showed a significant difference between initial and after application processes ($p < 0.05$) (Figure 8). However, this difference did not determine after one-day storage between these two groups ($p > 0.05$). CBS group's b^* value did not show any significant difference during the process ($p > 0.05$). According to the results, the SBR and HBR did not statistically change after the application process ($p > 0.05$), but there was a significance between these two groups and CBR ($p < 0.05$). This significance continued for HBR and CBR ($p < 0.05$), but not for the SBR group ($p > 0.05$).

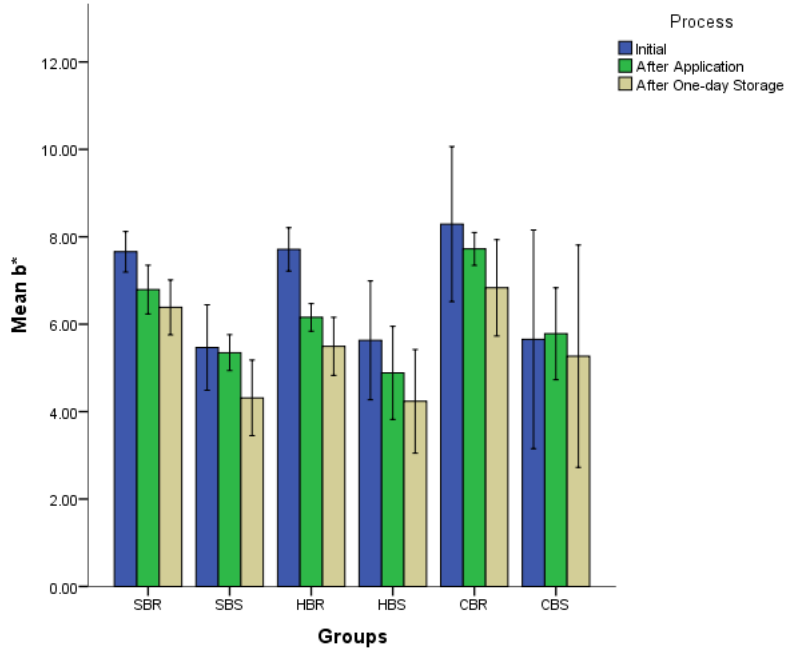


FIGURE 8: The mean b^* values of the sample groups' skin part (The error bars shown the Confidence Interval in 95%).

Chroma values changes in seabream skin samples were determined in Table 1 (Figure 9). This value decreased after the application process for all groups. This decreased result showed a statistically significant difference for all groups after the application process ($p < 0.05$). However, the significance was not observed after one-day storage ($p > 0.05$). The same process and different group comparisons showed that all groups had a statistically significant difference after the application process ($p < 0.05$). However, this significance changed after one-day storage for SBR and CBR ($p > 0.05$).

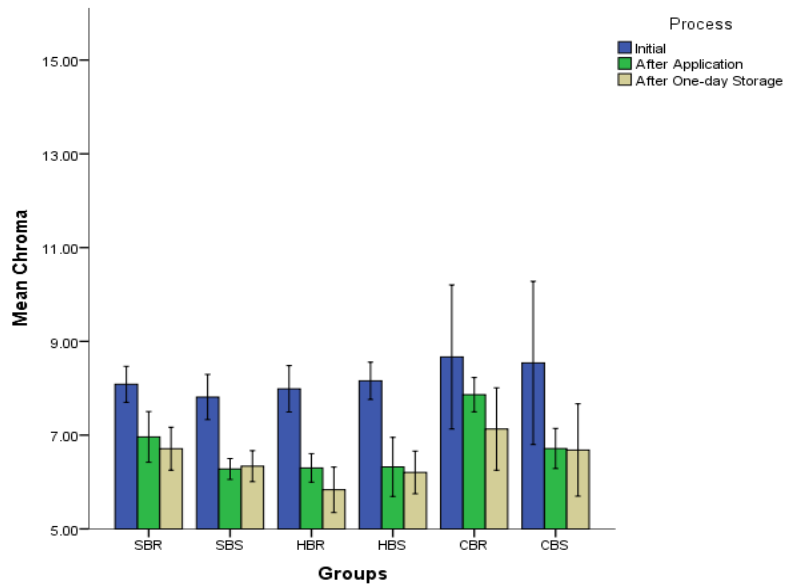


FIGURE 9: The mean Chroma values of the sample groups' skin part (The error bar showing a 95% confidence interval).

SBR, HBR, and CBR groups' skin Whiteness values were shown in Table 1 (Figure 10). The whiteness value of the groups increased after the application process. This increase was only significant for SBR and HBR groups ($p < 0.05$). After one-day storage, the difference did not observe for the groups ($p > 0.05$). The same process and different group comparisons showed no statistically significant difference ($p > 0.05$).

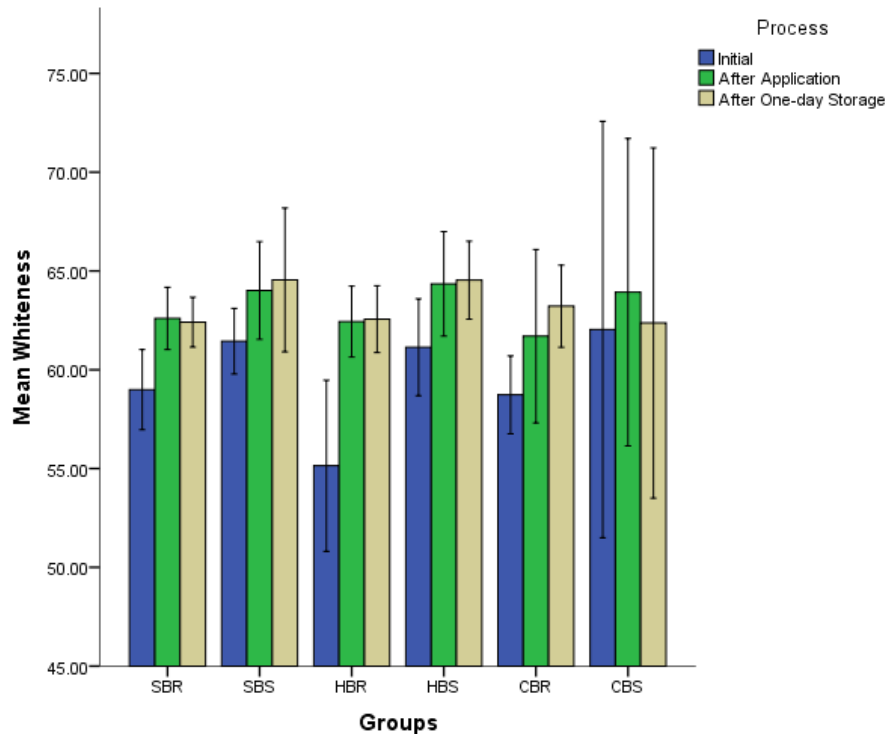


FIGURE 10: The mean Whiteness values of the sample groups' skin part (The error bar showing a 95% confidence interval).

The a^* (Figure 7) and Chroma (Figure 9) values of the SBS, HBS, and CBS skins increased after the application process. This increase showed a statistical difference for all groups ($p < 0.05$). However, after one-day storage and after the application process for all groups ($p > 0.05$), there was no significance between after one-day storage and after the application process. The seabass skin L^* (Figure 6), b^* (Figure 8), and Whiteness (Figure 10) values did not show any statistical significance neither between processes nor the same process and different group comparisons for all groups ($p > 0.05$).

4. DISCUSSION

Colloidal silver added hydrogen peroxide, and H_2O_2 addition in the immersion solution changed sea bream and seabass fillet and skin parts' color parameters. The control group and H_2O_2 solutions group color values were not showed a striking difference. Our results did not show a significant change in L^* and Whiteness values. However, it has been shown that there was no difference in the skins of fish samples, which were not observed in previous studies. According to all our data, if there is a change in the color parameter in the groups with hydrogen peroxide, it is observed that it has a^* or b^* values. It was probably due to the simultaneous appearance of the bleaching effect of H_2O_2 with the blood removal effect of water. However, studies confirm the bleaching effect of H_2O_2 ^{2,6}. Another important reason for this difference is that processed seafood was used in previous studies, but we use fresh fish samples. The oxidation-accelerating effect of H_2O_2 was observed in the decrease in fish samples' color parameter values after a one-day storage process. This result means that the deterioration of the fillet quality suppresses

hydrogen peroxide's effect on color parameters. The most important result supporting this is that this suppression is also clearly observed in the control group samples. This result indicated that the H₂O₂ treatment for the pre-treatment is not suitable for fresh fish.

In our study, striking differences between colloidal silver added H₂O₂ and H₂O₂ groups could not be obtained. Also, many color values and control groups were similar. It is thought that this is due to the use of fresh fish. Therefore, it is recommended for future studies to use processed seafood and use a processing method to reduce oxidation.

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