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유기산으로 처리한 냉장 광어의 미생물 평가

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Microbiological Evaluations of Refrigerated Flatfish Treated with Organic Acids

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Abstract

The effects of 0.5% (v/v) acetic acid (AA), 0.5% (v/v) lactic acid (LA), or 0.5% (v/w) citric acid (CA) on gram-negative bacterial counts (GNC) and aerobic plate counts (APC) in flatfish strips stored at 4° or 10℃ were assessed. Treatments of 0.5% AA were completely ($P<0.05$) inhibited GNC compared to the initial controls for 12 days at 4℃. Treatments of either LA or CA rapidly increased GNC compared to those of AA after 9 days at 4℃. Treatments of AA were microbiological acceptable for 6 days at 10℃, but those of LA and CA did not lasted. AA treatments were completely ($P<0.05$) inhibited aerobic spoilage bacteria (APC) compared to the initial controls for 6 days at 4℃. Treatments containing either 0.5% LA or 0.5% CA were ineffective after 9 days at 4℃ and 3 days at 10℃, respectively.

Based on these results, AA treatment was the most effective for suppresssing the growth of aerobic spoilage bacteria during storage at both 4 and 10℃. Flatfish strips treated with 0.5% AA extended microbiological shelf-life for 12 days at 4℃.

Key words : flatfish, acetic acid, lactic acid, citric acid, aerobic plate counts, gram-negative bacterial counts.

INTRODUCTION

Increasing consumer preferance for fresh fish rather than frozen fish depends on longer shelf-life relating microbiological quality[1~7]. Extension of shelf-life in refrigerated fish and meat is associated with low microbial numbers during storage and handling[4~13], which is resulted in the deterioration of keeping quality. Organic acids such as acetic acid (AA), lactic acid (LA), and citric acid (CA) are known as antimicrobial agents in food and provide inhibitory effects on aerobic spoilage bacteria[6,7,13~16].

Most of microorganisms in food are psychrotrophic bacteria capable of growing and causing spoilage at refrigerated temperature and gram-negative bacteria is the most indicative[6,8]. Ingham[4] noted that aerobic spoilage bacteria in catfish fillets treated with either 1.70% lactic acid or 2.55% lactic acid were significantly lower than controls for 6 days at 2℃. In a pre-

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vious study[11,12] we observed that organic acids were effective in suppressing growth of aerobic spoilage bacteria on refrigerated catfish and red seabream. Kim et al.[6] noted that catfish fillets treated with the combination of 2.5% lactic culture and either 2% lactic acid for 5 min or 3% lactic acid for 1~5 min effectively inhibited gram-negative bacteria for 9 days at 4℃.

Researchers[5,6,11,12,15] noted that antimicrobial effects can be extended if fish and meat are sanitized before refrigerated storage. Previous work[5] in our laboratories found that red seabream strips treated with 3% alginic acid containing 1.5~2.0% AA after 0.5% AA dip for 5 min had a significantly ($P<0.05$) lower levels of aerobic plate counts for 9 days at 4℃. Marshall and Kim[7] reported that catfish fillets treated with either 3.0~4.0% AA or 2.0% AA and 2.0% LA for 30~60 sec suppresssed growth of aerobic microorganisms for 4 days and extended shelf-life to 16 days for storage at 4℃.

However, there are limited studies on the effects of organic acids on fresh flatfish as a surface sanitizer. Therefore, the objective of the present study was to evaluate the microbiological changes on the surface of flatfish treated with AA, LA, and CA during storage at 4 and 10℃.

MATERIALS AND METHODS

1. Flatfish preparation

Fresh flatfish strips were obtained from a commercial source less than 1 hr postmortem, transported to meat laboratory on ice, and used within 2 hr. Three treatment solutions were prepared by mixing 1 L tap water with appropriate amounts (v/v or w/v) of acetic, lactic, or citric acids, respectively. Each one and half kilogram of strips (average weight 20g per strip) was dipped in 1 L sanitizer for 5 min. Strips were allocated to the following experimental trials : (1) 0.5% acetic acid (AA, Sae Won Chemical Co., Korea) dipping for 5 min, (2) 0.5% lactic acid (LA, Moo Jang Ya Chemical Co., Japan) dipping for 5 min, (3) 0.5% citric acid (CA, Dong Yang Glovel Chemical Co., Korea) for 5 min. Strips were submerged in each solution for required times then drained on a sanitized stainless-steel grill for 2 min at room temperature. Control strips were dipped in 1 L tap water for 5 min and drained for 2 min to compensate for possible physical removal of bacteria and for moisture uptake. After dipping, strips were placed in Whirl-Pak sample bags (Fisher Scientific Chemical Co., Norcross, GA, USA), stored at 4° and 10℃.

2. Microbiological analyses

Individual strips were aseptically transferred to Whirl-Pak sample bags, weighed, and diluted 1:1 with 0.1% (w/v) sterile peptone water (Difco Laboratories, Detroit, MI, USA). Samples was shaken for 60 times using standard rinse method[7]. The liquid from each sample was diluted and plated in volumes of 0.1 ml on standard plate count agar (Difco Laboratories, Detroit, MI) for aerobic plate count (APC) and MacConkey agar (Difco Laboratories, Detroit, MI) for gram-negative bacterial count (GNC), respectively. The plates were incubate for 48 hr at 30℃ before colonies were counted. The number of bacteria was expressed as mean Log_{10} colony forming unit (CFU)/g for the duplicate treatments.

3. Statistical analyses

APC and GNC data were analyzed using analysis of variance (ANOVA), and means were seperated by the least significant difference (LSD) test at $P<0.05$[17].

RESULTS AND DISCUSSION

During storage at 4° and 10℃, GNC and APC value of flatfish strips treated with AA, LA, and CA were assessed.

Treatments dipped with 0.5% AA for 5 min completely ($P<0.05$) inhibited the growth of GNC compared to the initial controls for 12 days at 4℃ (Fig. 1). On day 0, flatfish strips had a significantly ($P<0.05$) lower levels of GNC after acid treatments. GNC on flatfish strips treated with both 0.5% LA and 0.5% CA were not significantly different after 9 days compared to the controls. Results show that flatfish strip treated with 0.5% AA had a higher ($P<0.05$) antimicrobial effects than those of 0.5% LA and 0.5% CA. Similarly, Marshall and Kim[7] noted that catfish fillets treated with AA had greater antimicrobial activity than those of LA for storage at 4℃.

Flatfish strips treated with 0.5% AA for 5 min significantly ($P<0.05$) reduced GNC compared to the controls and both LA and CA treatments for 6 days at 10℃. However, there was no difference ($P>0.05$) between LA and CA treatments for storage at 10℃. Both LA and CA treatments were not microbiological acceptable after 6 days at 10℃. Results show that treatments of 0.5% AA significantly ($P<0.05$) inhibited the growth of gram-negative bacteria for 6 days at 10℃ and AA had greater ($P<0.05$) antimicrobial activity than both LA and CA. Ray and Sandine[15] noted that the antimicrobial effects of weak acids is principally produced by the undissociated molecules as well as pH.

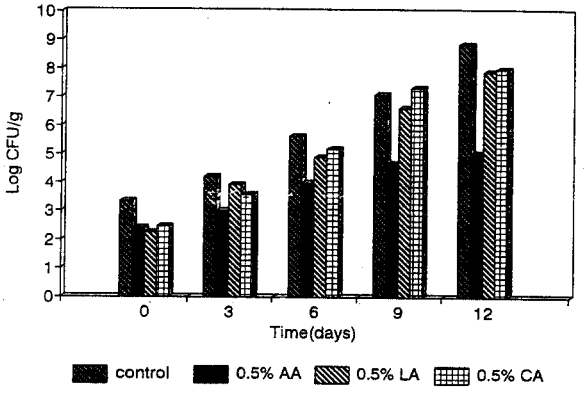

Fig. 1. Gram-negative bacterial counts (GNC) in refrigerated (4℃) flatfish strips dipped in acetic acid (AA), lactic acid (LA), and citric acid (CA) for 5 min. Key: control is tap water-only treatment.

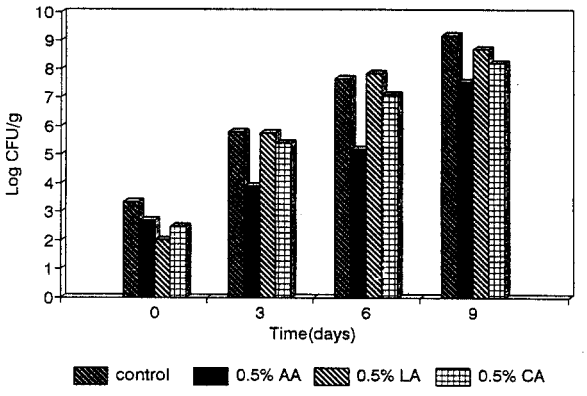

Fig. 2. Gram-negative bacterial counts (GNC) in refrigerated (10℃) flatfish strips dipped in acetic acid (AA), lactic acid (LA), and citric acid (CA) for 5 min. Key: control is tap water-only treatment.

When strips were treated with 0.5% AA for 5 min, APC caused complete inhibition with partial inactivation of aerobic spoilage bacteria for 9 days of storage at 4℃ (Fig. 3). After 12 days of storage, flatfish strips treated with 0.50% AA increased APC by 1.02 log units than those of initial controls. However, treatments of 0.5% LA and 0.5% CA rapidly increased APC after 9 days of storage. The fish industry generally considers that spoilage occurs when APC reaches 10^6 to 10^7 CFU/g. Microbiolo-

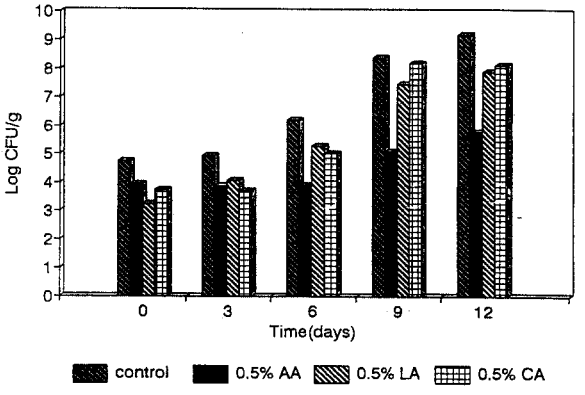

Fig. 3. Aerobic plate counts (APC) in refrigerated (4℃) flatfish strips dipped in acetic acid (AA), lactic acid (LA), and citric acid (CA) for 5 min. Key: control is tap water-only treatment.

gical shelf-life of flatfish treated with 0.5% AA for 5 min could be extended over 12 days of storage.

Marshall and Kim[7] reported that refrigerated (4℃) catfish fillets treated with 2.0~4.0% AA for 30~60 sec suppressed growth of aerobic microorganisms for 4 days and extended shelf-life to 16 days. They noted that antimicrobial activity of AA might have been enhanced as dipping time and concentration increased up to 30~60 sec and 3.0~4.0% AA, respectively.

Fig. 4 shows that flatfish strips treated with 0.5% AA for 5 min completely ($P<0.05$) inhibited growth of APC for 4 days of storage at 10℃. However, there was no difference ($P>0.05$) among LA treatments, CA treatments, and controls after 3 days of storage at 10℃. Results show that treatments of 0.5% AA significantly ($P<0.05$) inhibited the growth of aerobic spoilage bacteria for 6 days at 10℃, while controls and both LA and CA treatments were not microbiologically acceptable. Additionally, microbiological shelf-life of flatfish strips treated with 0.5% AA for 5 min could be extended by 6 days of storage. Ray and Sandine[15] noted that the antimicrobial effects of weak acids is principally produced by the undissociated molecules through the acidification of cytoplasm, destruction of the transmembrane proton motive force, and loss of active transport of nutrient through the membrane. Results show that treatments of 0.5% AA for 5 min colud be extend the microbiological shelf-life by 3-additional days compared to those of 0.5% LA and 0.5% CA. Researshers[7,15,16] reported that acetic acid on equimolar basis generally has greater antimicrobial activity than other organic acids.

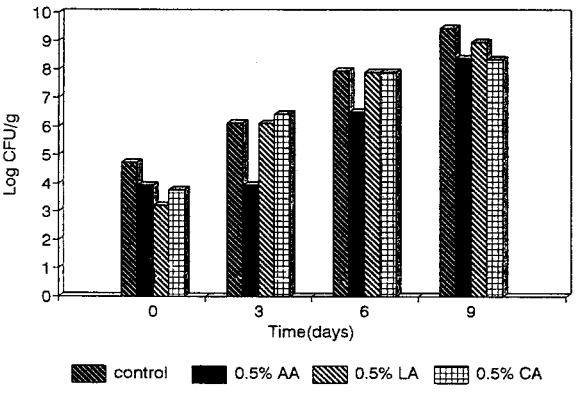

Fig. 4. Aerobic plate counts (APC) in refrigerated (10℃) flatfish strips dipped in acetic acid (AA), lactic acid (LA), and citric acid (CA) for 5 min. Key: control is tap water-only treatment.

CONCLUSIONS

Results of this investigation demonstrate that treatments containing 0.5% AA were more effective than those containing 0.5% LA and 0.5% CA in suppressing gram-negative bacteria on flatfish strips during 12 days of storage at 4℃. During 6 days at 10℃, AA treatments had a antimicrobial effects, but LA and CA treatments did not lasted. Flatfish strips treated with AA had a greater antimicrobial effect than did those treated with LA and CA. Surface sanitizing using 0.5% AA for 5 min effectively reduced aerobic spoilage bacteria during storage at 4~10℃. Since the microbiological shelf-life of flatfish could be extended for 12 days at 4℃, organic acid dip would be extremely desirable.

요 약

본 연구는 4~10℃ 냉장조건에서 0.5% 초산 (AA), 0.5% 유산 (LA) 및 0.5% 구연산 (CA) 침지법으로 5분 동안 처리한 신선한 광어의 미생물학적 저장 안정성에 미치는 영향을 조사하였다. 0.5% 초산 처리구는 4℃, 12일 저장 동안 그람음성세균 (GNC)의 증식을 완전히 억제하는데 효과적이었다. 0.5%의 유산 및 구연산 처리구는 각각 4℃, 저장 9일부터 그람음성세균 (GNC) 의 급속한 증식을 보였다. 10℃에서 6일 저장 동안 0.5% 초산 처리구는 그람음성균의 증식억제에 효과적이었으며, 유산 및 구연산 처리구는 저장 3일 이후 효과가 없었다. 0.5% (v/v) 초산 처리구는 4℃에서 저장 6일 동안 그리고 10℃에서 저장 3일 동안 호기성 부패 세균 (APC)의 증식을 완전히 억제하였다. 0.5% (v/v) 초산 처

리구는 4℃에서 저장 12일 동안, 그리고 10℃에서 저장 6일 동안 미생물학적 저장 안정성을 유지하였다. 본 연구의 결과 4~10℃에서 광어포의 저장 동안 호기성 육부패의 주요 세균인 그람음성세균 (GNC) 억제력이 초산처리구에서 가장 높게 평가되었다. 0.5% 초산처리구의 광어포는 4℃에서 12일동안 미생물학적 저장안정성을 유지하였다.

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