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Microbial Decontamination of Refrigerated Red Seabream by Acetic, Lactic, and Citric Acids

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초산, 유산 및 구연산에 의한 냉장 돔의 오염 미생물 제거

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요 약

본 연구는 4℃ 냉장조건에서 신선한 돔포의 미생물학적 저장 안정성 증진을 목적으로 초산, 유산 및 구연산 침지법을 이용하여 호기성 부패세균의 증식억제에 대한 영향을 조사하였다. 4개의 처리구로 만든 돔포는 각 0.25~1.0%의 초산, 유산 및 구연산의 위생수에서 5분 침지 후 처리구별로 플라스틱 저장백에 넣은 다음 실험에 사용하였다. 대조구는 수도물에 5분간 침지하여 사용하였다. 각각의 2반복 시료는 4℃, 12일 저장하면서 3일 간격으로 취한 다음 분석에 사용하였다. 0.25~1.0%(v/v) 초산 처리구는 4℃, 12일 저장 동안 호기성 부패세균의 증식을 억제하는 데 효과적이었다. 0.25~1.0%(v/v) 유산 처리구 그리고 0.5~1.0%(w/v) 구연산 처리구는 각각 4℃, 저장 9일 동안 호기성 부패세균의 증식을 억제하는데 효과적이었다. 돔포의 냉장 동안 초산처리구의 미생물학적 저장 안정성이 가장 높게 평가되었다.

주요어 : 돔, 초산, 유산, 구연산, 호기성 부패세균

INTRODUCTION

Organic acids such as acetic, lactic, and citric acids are known as antimicrobial agents in food and provide inhibitory effects on aerobic spoilage bacteria[1,5,6,7,8,9,10]. They are generally recognized as safe(GRAS) by the U.S. FDA. Growth of aerobic spoilage bacteria in refrigerated fish and meat is important to the food industry because it is associated with deterioration of keeping quality and safety[7,8,9,11,17]. Studies have shown that antimicrobial effects can be extended if fish and meat are sanitized before refrigerated storage[7,8,9,12,13,18,19]. Ingham[7] noted that aerobic spoilage bacteria in catfish fillets treated with either 1.70% lactic acid or 2.55% lactic acid were lower than controls by 1.3 and 2.3 logs for 6 days at 2℃, respectively. In a previous study we observed that organic acids were effective in suppressing growth of aerobic spoilage bacteria on refrigerated catfish and pork loins. Marshall and Kim[12] reported that catfish fillets treated with either 3.0~4.0% acetic acid or 2.0% acetic acid and 2.0% lactic acid for 30~60sec suppresssed growth of aerobic microorganisms for 4 days and extended shelf-life to 16 days for storage at 4℃. Kim *et al.*[9] reported that pork loins treated with either 1.0 or

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2.0% citric acid for 5min had a significantly($P<0.05$) lower levels of aerobic plate count compared to the controls for 9 days at 4℃.

Although there has been reported work to acetic, lactic, and citric acids in fish and meat products as antimicrobial surface sanitizer, few studies have been directed toward red seabream. Thus, the objective of the present study was to evaluate the antimicrobial effects of refrigerated red seabream treated with those acids.

MATERIALS AND METHODS

1. Red seabream preparation

Fresh red seabream(*Pargus major*) strips were obtained from a commercial source less than 1 hr postmortem, transported to meat laboratory in Seo Kang College 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) 0~1.0% acetic acid(AA, Sae Won Chemical Co., Korea) dipping for 5 min, (2) 0~1.0% lactic acid(LA, Moo Jang Ya Chemical Co., Japan) dipping for 5 min, (3) 0~1.0% 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 staineless-still 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 plastic bags(Clean Zipper Bags, Clean Wrap Co., Busan), stored at 4℃, and periodically removed for analyses.

2. Microbiological analyses

Individual strips were aseptically transferred to sterile "stomacher" bags, weighed, and diluted 1:10 with autoclaved 0.85%(w/v) NaCl. A stomacher LabBlender 400(Tekmar, Cincinnati, OH) was used to homogenize strips for 2 min. Aerobic plate counts(APC) were determined by duplicate spread platings on standard plate count agar(Difco, Detroit, MI), which were incubated at 30℃ for 48 hrs. APC values were expressed as mean Log_{10} CFU/g for samples. Microbial shelf-life was considered the time to reach spoilage levels of 10^7 CFU/g.

3. Statistical analyses

The APC data were analyzed using ANOVA, and means were seperated by LSD[15].

RESULTS AND DISCUSSION

During storage at 4℃, APC of red seabream strips treated with AA, LA, or CA were assessed. When strips were treated with 0.25~1.0% AA for 5 min, APC caused complete inhibition with partial inactivation of aerobic spoilage bacteria for 6 days(Fig. 1). After 12 days of storage, APC of strips treated with 0.25~1.0% AA were lower($P<0.05$) by 3.0~3.9 Log units than those of comparable controls. Marshall and Kim[12] reported that refrigerated(4℃) catfish fillets treated with 2.0~4.0% AA for 30~60sec 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

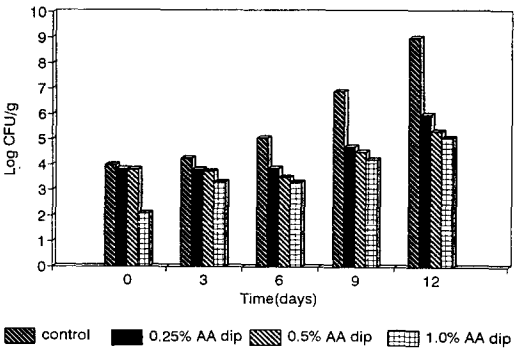

Fig. 1. Aerobic plate count in red seabream strips dipped in acetic acid for 5 min and stored at 4℃. Key: control is no treatment; AA, acetic acid.

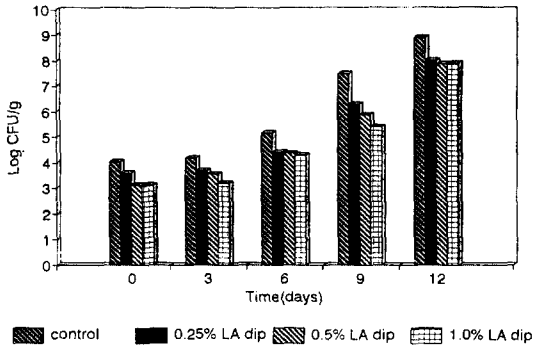

Fig. 2. Aerobic plate count in red seabream strips dipped in lactic acid for 5 min and stored at 4℃. Key: control is no treatment; LA, lactic acid.

increased up to 30~60sec and 3.0~4.0% AA, respectively. The fish industry generally considers that spoilage occurs when APC reaches 10^6 to 10^7 CFU/g. Microbiological shelf-life of red seabreams treated with 0.25~1.0% AA for 5 min could be extended over 12 days of storage.

Fig. 2 shows that strips treated with LA for 5 mins delayed microbial spoilage in comparison to the controls. After 6 days of storage, APC of strips treated with 0.25~1.0% LA were higher than initial control samples by about 0.3 Log units, respectively. All treatments significantly($P<0.05$) inhibited the growth of aerobic spoilage bacteria for 9 days at 4℃, while controls were not microbiologically acceptable. Results show that treatment of 1.0% LA caused the greatest reduction in APC for 9 days of storage, there was significant($P<0.05$) difference between 0.25~0.5% LA and 1.0% LA treatments. Microbiological shelf-life of samples treated with 0.25~1.0% LA for 5 min could be extended by 9 days of storage. Ingham[7] noted that antimicrobial effects of LA related to the concentrations of LA and storage condition. He reported that when catfish fillets were dipped in chilled(7℃) solutions of 1.70% and 2.55%(V/V) LA for 10 min, APC values of those treatments were lower than controls by 1.3 and 2.3 Logs after 3 days at 7℃, respectively.

Treatments dipped with 0.5~1.0% CA for 5 min effectively($P<0.05$) reduced APC by about 2 Log units compared to the controls for 9 days of storage(Fig. 3). However, treatments of 0.25% LA were not microbiologically acceptable after 9 days, there was significant($P<0.05$) difference between 0.25% CA and 0.5~1.0% CA treatments. Microbiological shelf-life of samples treated with 0.5~1.0% CA for 5 min could be extended by 9 days of storage. Results show that inhibitory effects on the growth of aerobic spoilage bacteria are associated with concentration of CA. Similarly, Kim *et al.*[9] noted that pork loins treated with 1.0~2.0% CA for 1~7 min had significantly($P<0.05$) lower APC throughout the experimental period than did those treated with 0.25~0.5% CA for 9 days at 4℃.

The results of this study show that red seabream strips of AA treatments had a higher antimicrobial effect than those of either LA or CA treatments. Several studies have reported that acetic acid on equimolar basis generally has greater antimicrobial activity than other organic acids[3,14,16]. The microbiological shelf-life of red seabream strips treated with AA was prolonged by 3 additional days compared to LA and CA treatments. Ray and Sandine[14] noted that the antimicrobial effect of weak acid was mainly produced by the undissociated molecules as well as the dissociated molecules. Similarly, several studies have reported that LA, with its lower pK, has less effect on neutralizing the proton motive force as compared to acetic acid[2,4,16]. Recent studies confirm that the total antimicrobial effects of

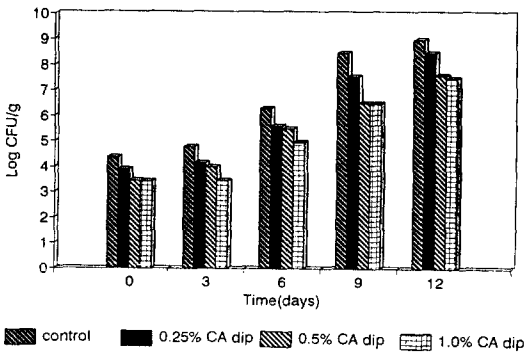

Fig. 3. Aerobic plate count in red seabream strips dipped in citric acid for 5 min and stored at 4℃. Key: control is no treatment; CA, citric acid.

these weak acids are produced by the combined effects of the undissociated and dissociated molecules[2,4,14,20]. It is considered that the effects are produced not by lowering the internal pH and neutralizing the proton motive force, but also by causing sublethal injury and unidentified effects[14].

CONCLUSIONS

The results of this study show that the effectiveness of organic acids at extending shelf-life is dependent on the type and concentration of acidulant used. Red seabream strips treated with AA had a greater antimicrobial effect than did those treated with LA and CA. All samples treated with AA produced microbiologically acceptable for 12 days at 4℃. It is considered that the technique of organic acid dipping may be useful for inhibiting microbial spoilage of refrigerated red seabreams.

ABSTRACT

Red seabream strips were decontaminated by dipping with solutions of 0.25~1.0% acetic, lactic, or citric acids for 5min. Control strips were dipped with tap water only for 5 min. All strips were individually placed in plastic bags and stored at 4℃. Acetic acid(AA) treatments were completely inhibited aerobic spoilage bacteria(areobic plate count : APC) compared to the initial controls for 6 days. Treatments of either lactic acid (LA) or citric acid(CA) completely inhibited APC compared to the initial controls for 3 days. Red seabream strips treated with AA extended microbiological shelf-life for 12 days.

Key words : red seabream, acetic, lactic and citric acids, aerobic spoilage bacteria

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