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Research Article

Effect of Chlorine Dioxide on Quality of Giant Salamander Cutting Meats in Small Modified Atmosphere Packaging

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Abstract: The objective of this study was to study the combined effect of chlorine dioxide and MAP on shelf life extension of giant salamander cutting meats. Salamander cutting meats were treated in five different concentrations (0, 10, 30, 50 and 70 mg/L, respectively) of chlorine dioxide solutions for 10 min, followed by packaged in the modified atmosphere packaging (10% O_2 70% CO_2). Then, the meats were storaged at 0°C for 28 days. Physical (drip loss, whiteness), chemical (TVB-N, TBARS) and Microbiological (TVC) indexes of giant salamander cutting meats were periodically assessed. These indexes were used for evaluation targets to evaluate effect on different treatments. Results show that chlorine dioxide in the process of giant salamander cutting meats preserve has a certain preservation effect. When the concentration was up to 70 mg/L, giant salamander cutting meats has the best effect that storage time can be up to 22 days or so.

Keywords: Chlorine dioxide, cutting meats, giant salamander, preservation, quality

INTRODUCTION

The Chinese giant salamander (*Andrias davidianus*) is living in the mountain streams of the Yellow River and the Pearl River shelter-forest tributaries. It is a kind of extremely high edible valuable economic animal, containing rich amino acids, essential fatty acids, minerals and trace elements (Luo, 2010). These completely accord with human body requirement model, so it is a good source of human nutrition and was known as "ginseng" lived in water (Liu *et al*., 2009; Geng *et al*., 2013). Giant salamander is contained more than 50 natural bioactive substances that to be used in nutrition, health, pharmaceutical and aesthetic (Li *et al*., 2012). In recent years, along with the technique of artificial breeding giant salamander and disease control being more and more mature and the improvement of people's nutritional requirements, customer demand for giant salamander product are on the increase. People mainly took the form of living body to sell on the market at present. Living transportation is too heavy to easy to sell, so it is meaningful to research on the fresh-keeping of giant salamander cutting meats.

Chlorine dioxide is a highly efficient, safe and broad-spectrum disinfection sterilization agent (Benarde *et al*., 1965; Hsu *et al*., 2012). Chlorine dioxide has very good kill and inhibition to general bacteria, the bud spore, sulfate reducing bacteria, iron bacteria and fungi etc. Now chlorine dioxide has been widely used in medical and health care, animal husbandry and food industry and other fields (Lan *et al*., 2010; Luo and Chen, 2005). Antimicrobial ice containing chlorine dioxide was utilized to control food-borne pathogens on fish skin. The initial load of food-borne pathogens was reduced and the lowered microbial level was maintained during treatment (Shin *et al*., 2004). Chlorine dioxide has good killing effect on the water spread of pathogenic microorganisms. It showed an effective conservation effect on fruit, vegetables and meat storage while extending shelf life (Han *et al*., 2004; Wu and Kim, 2007; Chen and Zhu, 2011; Jiang *et al*., 2013).

Modified Atmosphere Packaging (MAP) is the world's food packaging of a popular non-thermal advanced technology. It suitably uses CO_2, O_2 and N_2 mixed gases in food packs to results in inhibition of spoilage microorganisms in perishable foods such as meat, fish and related products (Davies, 1995). An overview about natural compounds combined with MAP storing food shows the good effect on fruit and vegetables, dairy products, meat and fish products (Mastromatteo *et al*., 2010). Pastoriza *et al*. (2002) reported the synergistic effect between Lauric acid and

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MAP on the kinetics of bacterial growth in pre-cooked minced fish products and combined treatment were commercially acceptable after 1 month of storage.

Small packaging is a kind of new situation of modern meat products sales. Because it avoids the pollution of the outside world in a certain extent, to ensure the quality of products, it is very popular with people. This article will be chlorine dioxide as a preservative used in quality control on giant salamander cutting meats in small modified atmosphere packaging. Given that no information exists in the literature on the preservation of giant salamander meats and the application of either chlorine dioxide or MAP to raw giant salamander meats, the objective of the present work was to preliminary study the effect of chlorine dioxide on quality attributes of raw giant salamander cutting meats.

MATERIALS AND METHODS

Materials: Living giant salamander was procured from Hanzhong Longni Biological Engineering Shares (group) Co., Ltd., (Hanzhong, China). CR-10 color reader was procured from Hangzhou Ke Sheng Instrument Co., Ltd. ATM-300 automatic kjeldahl nitrogen determination apparatus was procured from Shanghai HongJi Instrument Equipment Co., Ltd. TU-1810 ultraviolet and visible spectrophotometer was procured from Beijing Puxitongyong instrument Co., Ltd.

Sample preparation: Giant salamander was rinsed clean with tap water and slaughtered. Whole giant salamanders were divided into five lots. Each giant salamander (20°C) was dipped in 45 L of 0 (sterile distilled water), 10, 30, 50 or 70 mg/L of chlorine dioxide (w/v) for 10 min and were marked A, B, C, D, E five groups respectively. After dipping, all were placed in a -40°C condition in order to pre-cool the center temperature to 0°C. After that, they were individually cut into 0.8-1.0 cm thickness. Each group was placed in PP trays with 100 g and individually packed in PE bag for inflatable. MAP (a mixture of gases: 70% CO_2: 20% N_2: 10% O_2) was injected to each group at a product, gas ratio of 1: 2 (w/v) (Mastromatteo *et al*., 2010; Benarde *et al*., 1965). All samples were stored at 0°C for subsequent quality assessment. Physical, chemical and microbiological analyses were performed at 0, 2, 6, 10, 14, 22 and 28 days, respectively.

Determination of whiteness: The surface color of salamander meat was measured using a color reader. L* (brightness), a* (redness) and b* (yellowness) color values together determines the color value of the sample (Du *et al*., 2000). Measurements were taken at three different locations on each sample and directly read from the screen. Whiteness value was calculated using the formula that was put forward by Fuji, Watanabe (Benjakul *et al*., 2003):

$$\text{Whiteness} = 100 - \sqrt{(100 - L^*)^2 + a^{*2} + b^*}$$

Determination of drip loss: The quantity of drip loss after storage was measured by weighing method. The quantity of loss is equal to the difference of initial weight and the reweighing. Drip loss was expressed as a percentage of the initial weight. The drip loss was calculated using the formula as follows:

$$\text{Drip loss} = \frac{W_0 - W_i}{W_0} \times 100\%$$

where,
W_0 = The initial weight of cutting meats
W_i = The reweighing weight of the i sample

Determination of Total Volatile Basic Nitrogen (TVB-N): Giant salamander samples (10 g) were aseptically triturated and soaked 30 min in an erlenmeyer flask. Samples were aseptically on the filter and the filtrate was determined by automatic kjeldahl nitrogen determination apparatus Total Volatile Basic Nitrogen (TVB-N). TVB-N value as freshness measure standards as follows: 5-10 mg/100 g is defined as excellent freshness; 15-25 mg/100 g is defined as the freshness; 30-40 mg/100 g is defined as the early corruption; ≥50 mg/100 g is defined as corruption.

Determination of Thiobarbituric Acid Reactive Substances (TBARS): Fat oxidation in giant salamander samples was determined by thiobarbituric acid colorimetric method. Sample (10 g) was shocked 30 min, adding 50 mL, 7.5% trichloroacetic acid containing 1% EDTA and filtrated with double filter paper. Filtrate (5 mL), adding 5.0 mL, 0.02 mol/L thiobarbituric acid, was placed in a water bath at 95°C for 120 min, then cooled for 60 min. The fluid adding 5.0 mL chloroform shake, let stand after stratification, then to determine the TBARS value. Absorbance (Abs) was measured at 532 and 600 nm by ultraviolet and visible spectrophotometer. TBARS was obtained by the formula:

$$\text{TBARS}\left(\frac{\text{mg}}{100\text{g}}\right) = \frac{\text{Abs}_{532} - \text{Abs}_{600}}{155} \times \frac{1}{10} \times 72.6 \times 100$$

Determination of microbiological: The Total Viable Counts (TVC) were determined by the method of plate counting, Chinese standard GB/T4789.2-2010. Microbiological analysis was carried out on giant

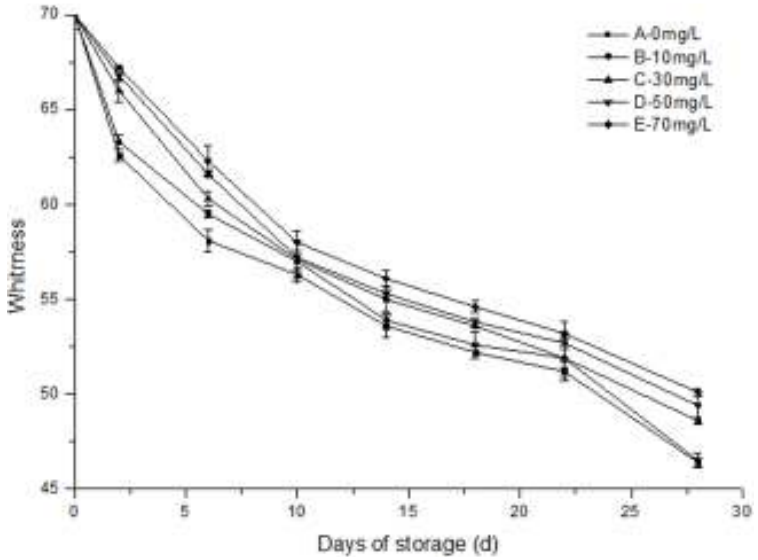

Fig. 1: Whiteness analysis of giant salamander meat with different concentrations of chlorine dioxide

salamander samples in a bacteria-free operating environment. TVC were determined by counting the number of colony-forming units after incubation at 37°C for 48 h. TVC was counted by Colony Forming Units (CFU). Meat product microbial limit standard regulate: TVC Colony Forming Units (CFU) of aquatic products $\leq 10^5$/g was regarded as the first freshness, $\leq 5\times10^5$/g as the secondary freshness, up to 10^6~10^7/g usually indicates the meat has been extremely corrupt.

Statistical analysis: All data were expressed as means ± standard deviations from three independent experiments. Experimental data used statistical software SPSS 17.0 and ORIGIN 8.5 to analyze. These data were subjected to variance analysis, significance test (the significance level is set to ($p<0.05$).

RESULTS AND DISCUSSION

Whiteness analysis: In the process of storage, the color of meat will change due to a series of reaction, such as fat oxidation and degradation reaction of pigment. In addition, some bacteria can produce different colors of pigments which also affect the color of meat (Nuñez de Gonzalez *et al*., 2008). Whiteness value of all treatments is given in Fig. 1. The color of meat became dim slightly yellow during the storage. The whiteness value of raw meat processed by chlorine dioxide was significantly ($p<0.05$) higher than that from the non-processed. Chouliara *et al*. (2008) reported that the color of packaged chicken breast meat became dim due to decrease on L* value. Whiteness value maybe mainly was decided by L* value. The difference of whiteness among groups is due to the inhibitory effect on microorganism by chlorine dioxide. The effect reduces the fat oxidation and the degradation of pigment. At the same time, microbial accumulating pigment metabolism slows down.

Drip loss analysis: The effect on giant salamander meat drip loss by chlorine dioxide is shown in Fig. 2. Experiment on the day drip loss was 0%. Drip loss for giant salamander presented gradually rising trends over time. There was a significant difference between all groups ($p<0.05$). Drip loss varied between 0 and 8.1% for the control group and between 0 and 6.2% for the product of group with 70 mg/L chlorine dioxide after 28 days of storage. Water loss will cause meat color, a series of sensory changes and loss innutrition (Kaale *et al*., 2014).

The activity of protein enzymes was inhibited in low temperature and chlorine dioxide inhibits the activity of microorganism that reduced the muscle cells and muscle protein form and structure changes caused by microbial. So that, the change of muscle protein binding force water molecules was reduced. Meat holding water capacity was protected further.

TVB-N analysis: Changes of TVB-N value in giant salamander samples during storage were presented in Fig. 3. TVB-N reflects the protein aquatic products decompose and produce volatile ammonia and amine compounds caused by endogenous enzymes and the effect of microbial (Fan *et al*., 2009). TVB-N of samples increased progressively ($p<0.05$) from an initial value of 2.8 mg/100 g to final values of 21.2, 20.6, 19.8, 19.8 and 18.4 mg/100 g for the control of chlorine dioxide in 0, 10, 30, 50 and 70 mg/L, respectively. Control group A was the largest of 21.2 mg/100 g and group E was the minimum of 18.4 mg/100 g on the day of 28. TVB-N of control group (4.9 mg/100 g) was beyond the limits of the freshness of the level one at the time of 4 days, but C, D, E, three groups were on the 10^{th} day when beyond its scope. TVB-N rise slowly in the process of storing 0 to 10 day, but increase upward trend up from 10 day.

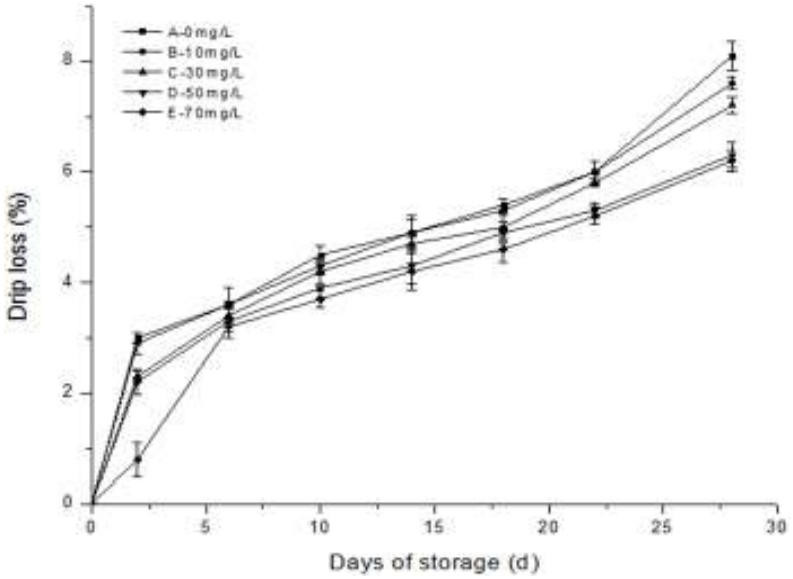

Fig. 2: Drip loss analysis of salamander meat with different concentrations of chlorine dioxide

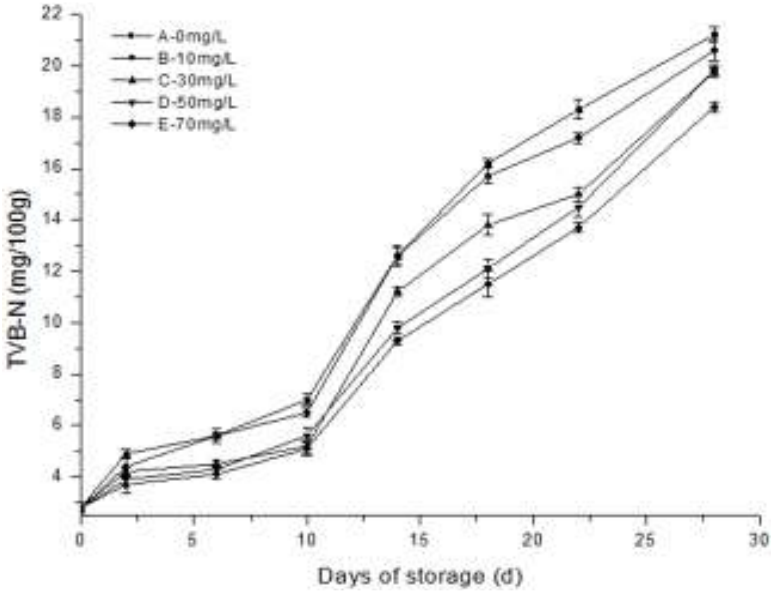

Fig. 3: TVB-N analysis of salamander meat with different concentrations of chlorine dioxide

This shows a high concentration of chlorine dioxide solution has a good fresh-keeping effect on meat. The slow, quick change trend and the change tendency of the microorganism is the same. This indicates that the increase of TVB-N was mainly caused by the growth of microorganisms. In the process of storage, chlorine dioxide had inhibitory effect on bacteria so reduced the decomposition of protein and the rise velocity of TVB-N.

TBARS analysis: TBARS indicate that lipid secondary oxidation product that reflected the stored specimen oxidation of unsaturated fatty acid is inevitable (Nawar, 1996; Cagdas and Kumcuoglu, 2014; Thomas *et al*., 2010; Özden *et al*., 2007). Groups of giant salamander TBARS value showed a trend of increase during the storage. The control group rose from 0.06 to 0.275 mg/100 g and E from 0.094 mg/100 g to 0.201 mg/100 g. At 28th day, control group A compared with group E, a difference of 0.074 mg/100 g. The TBARS curve of A than B, C, D, E more steep as shown in Fig. 4, suggesting that the chlorine dioxide reduces the speed to make specimens of fat oxidation. This and the change of the microbial consistent, chlorine dioxide inhibits the growth of microorganisms, so effectively reduce the specimen fat oxidation and deterioration

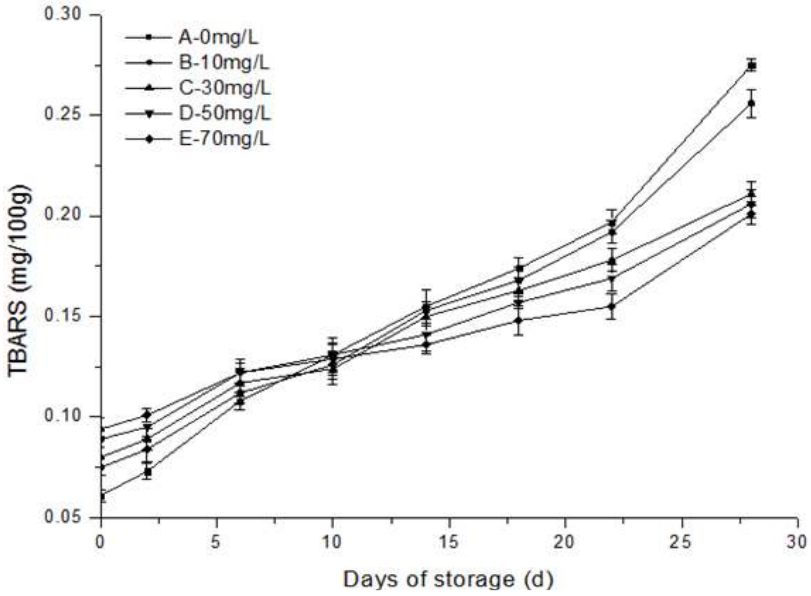

Fig. 4: TBARS analysis of salamander meat with different concentrations of chlorine dioxide

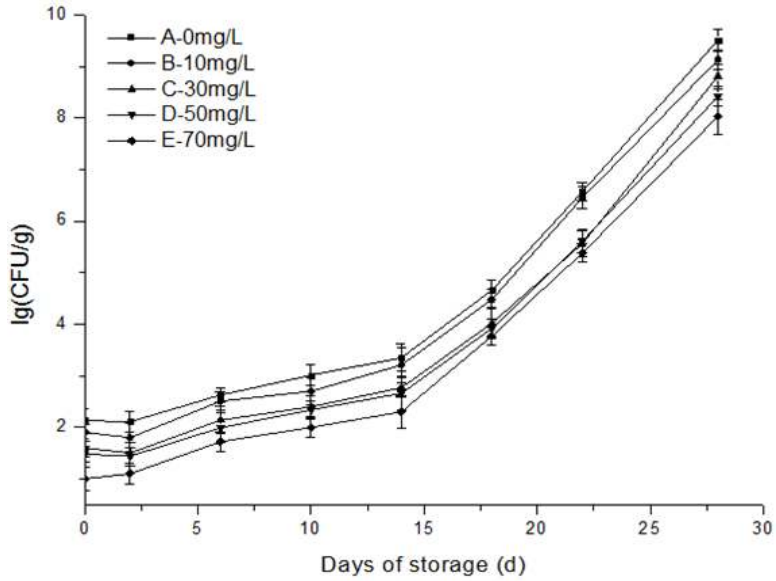

Fig. 5: TVC analysis of salamander meat with different concentrations of chlorine dioxide

process. But, it is interesting to note that it does not exist in the obvious differences between groups.

Giant salamander specimens of initial TBARS increase along with the concentration of chlorine dioxide. By the analysis, chlorine dioxide oxidation to a certain extent, in the initial processing lead to partial fatty acid oxidation was carried out on the specimen.

TVC analysis: Microbial counts in different concentrations of chlorine dioxide are shown in Fig. 5. There was a significant increase on TVC over the period of storage. Bacteria grew more slowly in the first 14 days, then quick. The early is micro-organic adjustment period.

There were significant differences ($p<0.05$) in TVC of giant salamander stored in different concentrations of chlorine dioxide. As shown in Fig. 5: the control group has a higher TVC than the samples with chlorine dioxide. The bacteria growth rate was falling with the increase of the concentration of the chlorine dioxide. On days 21 and 24 of storage, A and E samples, respectively TVC reached 10^6 CFU/g, which is considered as the upper acceptability limit for fresh aquatic product.

Effect of preservation of chlorine dioxide in the experiment performance owe to its inhibitory effect on microorganism. Chlorine dioxide on the cell wall has better adsorption and through performance and can

effectively oxidized mercapto enzyme. Moreover, ClO_2 can react with cysteine, tryptophan and free fatty acids and rapidly control of biological protein synthesis.

CONCLUSION

Chlorine dioxide brings the most obvious impact ($p<0.05$) on drip loss, whiteness, TVB-N and TVC. A high concentration of chlorine dioxide is conducive to the preservation of meats. Giant salamander meat preservation effect mainly is in the role of chlorine dioxide effect on killing microorganism. When the concentration is 70 mg/L, giant salamander cutting meats in small modified atmosphere packaging 10% O_2 70% CO_2) storage at 0°C be up to 22 days or so.

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