Kinetics of the Inactivation of *Vibrio parahaemolyticus* in Weakly Acidic Sodium Chlorite Solution

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The kinetics of the inactivation of *Vibrio parahaemolyticus* in sodium chlorite (NaClO\(_2\)) solution was studied in the weakly acidic pH range of 4.0 to 6.5 and at various temperatures. The logarithmic reduction of the survival ratio depended on the concentration-time product, and all the inactivation curves showed a linear reduction phase. The first-order inactivation rate constant (\(k\)) increased by approximately twice for every 0.44 unit fall in pH. During the inactivation experiments, no formation of chlorine dioxide occurred. These data indicated that undissociated HClO\(_2\) was the active species governing the inactivation of *V. parahaemolyticus*. It was also shown that the use of weakly acidic NaClO\(_2\) solutions containing high concentrations of ionized ClO\(_2^-\) gave slower kinetics of the inactivation, whereas it could achieve the significant reduction of viable cells of more than 4-log. The \(k\) value showed an Arrhenius-type temperature dependence in the temperature range of 5 to 40\(^\circ\)C. The apparent activation energy for the inactivation of *V. parahaemolyticus* was estimated to be 43.5 kJ/mol. The \(k\) value increased by approximately 1.8 times for every 10\(^\circ\)C rise in temperature.

**Key words**: Sodium chlorite / Dissociation of chlorous acid / *Vibrio parahaemolyticus* / Inactivation kinetics / Apparent activation energy.

**INTRODUCTION**

*Vibrio parahaemolyticus* has been recognized as a cause of seafood-borne gastroenteritis in Asia and other countries (Katsui et al., 1999; Wong et al., 2001). Food poisoning caused by *V. parahaemolyticus* has been occurring all over the world, especially since 1996 (Fournier and Ogata, 2005). The main cause of this type of food poisoning is eating raw or undercooked seafood contaminated with the pathogen. To prevent an outbreak of such food poisoning, *V. parahaemolyticus* cells on surfaces of seafood and processing equipment should be inactivated by a suitable disinfecting operation for decreasing viable cells.

Sodium hypochlorite (NaClO) has so far been used in a disinfecting operations for potable water, seawater, food stuff, and food-processing equipment because it has high oxidizing power and therefore a broad antimicrobial spectrum and rapid bactericidal action (Fukuzaki, 2006; Rutala and Weber, 1997). However, the disinfecting efficacy of NaClO is reduced markedly in the presence of organic matter (Wei et al., 1985). When NaClO is used in the disinfection of seawater, the free available chlorine (FAC) added is depleted rapidly by the oxidation of bromide ion (Br\(^-\)) to hypobromite ion (BrO\(^-\)). In addition, the repeated use of NaOCl solution often results in the corrosion of metallic materials. Therefore, alternative disinfectants that compensate for the weakness of NaClO are being explored for seafood and processing settings.

Sodium chlorite (NaClO\(_2\)) is an alternative oxidizing agent which avoids the disadvantage of NaClO. NaClO\(_2\) has a lower reactivity with organic matter than NaClO, so that the efficacy of NaClO\(_2\) in disinfecting processes is maintained in the presence of organic matter (Hasegawa, 1989; Hosoi, 1990). Therefore, it is likely that NaClO\(_2\) is more suitable than NaClO for the inactivation of the surfaces of food, such as meat, vegetables, and marine products. It is known that the antimicrobial activity of NaClO\(_2\) depends on the solution pH (Kobayashi, 1990). Especially, acidified sodium chlorite
(ASC), which is produced by the addition of food-grade-acid such as citric acid and phosphoric acid to NaClO₂ solution, has been demonstrated to have an excellent antimicrobial effect (Benford et al., 2008; Horiuchi et al., 2015). Acidification of NaClO₂ solution results in the protonation of the chlorite ion (\(\text{ClO}_2^-\)) to chlorous acid (\(\text{HClO}_2\)). \(\text{HClO}_2\) is an acid with \(pK_a\) of 1.86 (Horváth et al., 2003) and there is an equilibrium between \(\text{HClO}_2\) and \(\text{ClO}_2^-\) depending on the solution pH.

\[
\text{HClO}_2 \rightleftharpoons \text{H}^+ + \text{ClO}_2^- \quad (1)
\]

In ASC solution, \(\text{HClO}_2\) is believed to be the primary antimicrobial species. Depending on the type of food, ASC solution is generally used in the pH range of 2.5 to 3.2, and in the concentration range of 50 to 1,200 mg NaClO₂/l (Benford et al., 2008). On the other hand, under these strong acidic conditions, \(\text{HClO}_2\) is metastable and rapidly converted to chlorine dioxide (\(\text{ClO}_2\)). \(\text{ClO}_2\) is very volatile and it is therefore lost by evaporation. Despite the strong antimicrobial activity of ASC, it is a short-lived disinfectant that has to be prepared at the time of use. Therefore, it is desirable to develop the effective use of sodium chlorite solution under weakly acidic conditions without formation of \(\text{ClO}_2\). However, little is known about the kinetics of the inactivation of bacteria in weakly acidic NaClO₂ solution.

The purpose of this study is to determine the rate of the inactivation of \(V.\) parahaemolyticus in weakly acidic NaClO₂ solutions at various temperatures. Inactivation experiments were conducted in 0.1M phosphate-buffered NaClO₂ solutions of pH 4.0 to 6.5. In this paper, we report that the first-order inactivation rate constant increases exponentially with decreasing pH and shows an Arrhenius-type temperature dependence. The contributions of \(\text{HClO}_2\) and \(\text{ClO}_2^-\) to the inactivation of \(V.\) parahaemolyticus will also be discussed.

**MATERIALS AND METHOD**

**Bacterial strain and culture condition**

\(V.\) parahaemolyticus NBRC 12711 was obtained from the National Institute of Technology and Evaluation (NITE, Chiba). \(V.\) parahaemolyticus was cultured with tryptic soy broth (TSB, Merck KGaA, Darmstadt, Germany) supplemented with 2% NaCl and its pH was adjusted to 7.5. The culture conditions and preparation of the \(V.\) parahaemolyticus suspension (\(\text{OD}_{660} = 2.0\) in 0.9% NaCl) were the same as those previously described (Takahashi et al., 2016).

**Materials and chemicals**

NaClO₂ (purity of > 79%) was purchased from Kanto Chemical Co. Ltd. (Tokyo). All other chemicals were of analytical grade and purchased from commercial sources.

**Preparation and quantification of NaClO₂ solution**

A 13-g of the reagent NaClO₂ was dissolved in 200 ml of ultra-pure water. The NaClO₂ solution was passed through a filter (0.2-μm pore size; Advantec Co., Ltd.) and stored as the stock solution at 4°C.

The concentration of NaClO₂ was measured by iodo-metric titration. A 0.1-ml aliquot of the NaClO₂ stock solution was diluted in a measuring flask to 30 ml with deionized water, and the diluted NaClO₂ solution was introduced to a 50-ml glass beaker. Then, 3.5 ml of 1.7 M sulfuric acid solution and 5 ml of 20% potassium iodide solution were added into the diluted NaClO₂ solution, which was then titrated with 0.1 M sodium thiosulfate (\(\text{Na}_2\text{S}_2\text{O}_3\)) using a potentiometric titrator (COM-1600, Hiranuma Sangyo, Co., Ltd., Ibaraki). The concentration of the stock solution of NaClO₂ was 563 mM (50,900 mg/l).

**Inactivation of \(V.\) parahaemolyticus by NaClO₂ solution**

A 50-ml portion of a 0.1 M phosphate-buffered saline (PBS) solution of pH 4.0 to 6.5 was put in a 50-ml glass beaker. The beaker was placed in the water bath at 5 to 40°C and the PBS solution was agitated with a magnetic mixer (400 rpm). During the inactivation experiment, the pH and temperature were monitored. A 0.5-ml aliquot of \(V.\) parahaemolyticus suspension (\(\text{OD}_{660} = 2.0\) ) was added into the PBS solution. After the cell suspension was mixed thoroughly, the inactivation reaction was started by adding the stock solution of NaClO₂ (563 mM) to make final concentrations of 1.12 mM at pH 4.0, 2.24 mM at pH 4.5, 5.57 mM at pH 5.2, and 21.7 mM at pH 5.6 and 6.5. During the course of the NaClO₂ treatment, samples (0.5 ml) were withdrawn at appropriate intervals and immediately transferred to a glass tube containing 4.5 ml of 0.1 M PBS solution (pH 8.3) with 0.1 M \(\text{Na}_2\text{S}_2\text{O}_3\) to terminate the inactivation reaction. The number of viable \(V.\) parahaemolyticus cells \((N)\) was estimated by microbial calorimetry as described previously (Takahashi and Fukuzaki, 2012; Takahashi et al., 2016). In a preliminary study, it was confirmed that the \(N\) estimated by microbial calorimetry coincided with that obtained from the number of colonies formed in the dilution plate method for 24h (data not shown). Therefore, cells which could not form colonies within 24h were recognized as inactivated cells. Each experiment was repeated two or three times for each inactivation condition.

The Chick-Watson law (Chick, 1908; Watson, 1908) was used to determine the rate of inactivation of \(V.\) parahaemolyticus.

\[
\log (N/N_0) = -kCT \quad (2)
\]

where \(N\) and \(N_0\) are the number of surviving cells at time \(T\) and time zero, min, respectively; \(C\) is the NaClO₂ concentration (mM), and \(k\) is the first-order inactivation
rate constant, $l$/mmol-min. The logarithmic relative reduction of viable cells ($\log N/N_0$) was plotted against the $CT$ value to obtain the $k$ value.

**Measurement of the formation of ClO$_2$**

An experiment was conducted to assess whether ClO$_2$ was formed in NaClO$_2$ solution during the inactivation experiment. A 3.5-ml portion of a 0.1M PBS solution of pH 4.0 to 6.5 was put in a 10 mm quartz cuvette, which was then set in cuvette holder kept at 25°C in a spectrophotometer (UV-3100PC, Shimadzu Co., Ltd., Kyoto). First, a 35 µl-aliquot of the cell suspension was added into the PBS solution in the cuvette, which was mixed thoroughly using a magnetic stirrer during the experiment. Secondly, the stock solution of NaClO$_2$ solution was added in the cuvette at final concentrations of 1.12 to 21.7 mM as described above. The monitoring of ClO$_2$ and HClO$_2$/ClO$_2$ was conducted by measuring absorbance at 360 nm ($Abs_{360}$) and 260 nm ($Abs_{260}$), respectively, (Hong and Rapson; 1967, Horváth, 2003) at an interval of 12.5 s during 5 min of the inactivation reaction.

**RESULTS**

**Effect of pH**

Figure 1 shows the logarithmic relative reduction of viable *V. parahaemolyticus* in NaClO$_2$ solutions of different pH as a function of $CT$ value (25°C). Each inactivation curve obtained at pH 4.0 to 5.6 started with a short lag phase (ca. 30 s) followed by a linear reduction phase. In the linear phase, the inactivation followed pseudo-first-order kinetics (eq. 2). The inactivation curve at pH 6.5 was characterized by a gradual and slow decline without a lag phase. At all pH values, a reduction in viable cells greater than 4-log could be achieved within 1 to 5 min. It was clearly indicated that the rate of inactivation increased markedly by decreasing the solution pH. The $k$ values were determined by drawing a straight line through the linear phase by using the linear regression method. The $k$ values obtained and the estimated minimum $CT$ values required for at least a 3-log reduction in viable cells are summarized in Table 1. The $k$ value apparently depended on the solution pH. At pH 6.5, the $k$ of 0.107 $l$/mmol-min was extremely low, whereas it increased up to 3.54 $l$/mmol-min at pH 4.0, which corresponded to a 33-fold increase. The $CT$ values to achieve at least a 3-log reduction also decreased markedly from 44.1 mmol-min/l at pH 6.5 to 1.29 mmol-min/l at pH 4.0 (34-fold decrease). The data in Table 1 demonstrate the known fact that weakly acidified NaClO$_2$ solution is a more effective disinfectant against bacteria than a neutral NaClO$_2$ solution.

Figure 2 shows the relationships between the pH of the NaClO$_2$ solution and the $k$ values. The $k$ value increased exponentially by decreasing the solution pH. The graph of the $k$ versus pH gave the following relationship ($R^2$: correlation coefficient):

![FIG. 1](image-url)  
**FIG. 1.** Effect of pH on the inactivation of *V. parahaemolyticus* in NaClO$_2$ solutions of various pH at 25°C. The results were representative of a set of two to four independent experiments. Symbols: (A) □, pH 4.0; ●, pH 4.5; (B) △, pH 5.2; ▲, pH 5.6; (C) □, pH 6.5.

![FIG. 2](image-url)  
**FIG. 2.** The relationship between the solution pH and the $k$ values at 25°C.

**TABLE 1.** Kinetics of the inactivation of *V. parahaemolyticus* in NaClO$_2$ solutions of various pH.

| pH  | $k$  | Minimum CT to achieve at least a 3-log reduction |
|-----|-----|-----------------------------------------------|
| 4.0 | 3.54| 1.29                                           |
| 4.5 | 1.15| 3.67                                           |
| 5.2 | 0.425| 10.8                                           |
| 5.6 | 0.228| 15.5                                           |
| 6.5 | 0.107| 44.1                                           |

The $k$ values and minimum $CT$ values to achieve at least a 3-log reduction were averages of data from two to four independent experiments. Inactivation experiments were conducted at 25°C.
While the CT values decreased by 1/7.6. These results clearly indicated that temperature of NaClO₂ solution was also an important factor in accelerating the inactivation.

The relationship between temperature and the k values was represented by the Arrhenius plot (Fig. 4):

\[
\ln k = \frac{-E_a}{RT} + \ln A
\]

where \(E_a\) is the apparent activation energy, J/mol; \(R\) is the gas constant, 8.314 J/K·mol; \(T\) is the temperature, K; and \(A\) is the frequency factor, l/mmol·min. A linear relationship between \(\ln k\) and \(10^{3/T}\) was obtained, and the \(E_a\) was estimated to be 43.5 kJ/mol from the slope of the graph. These findings indicated that \(k\) increased by approximately 1.8 times for every 10°C rise in temperature.

Effect of temperature

Figure 3 shows the representative results of the effect of temperature on the inactivation of V. parahaemolyticus in the NaClO₂ solution of 21.7 mM at pH 5.6. Each inactivation curve had a lag phase followed by a linear reduction phase. With increasing temperatures from 5 to 35°C, the lag phase duration was shortened and the rate of inactivation increased markedly. The \(k\) values were determined from the slope of the straight line in the linear phase. All the \(k\) values obtained and estimated CT values to achieve at least a 3-log reduction are summarized in Table 2. The \(k\) increased by 8.3 times with the increase in temperature from 5 to 40°C.

FIG. 3. Effect of temperature on the inactivation of V. parahaemolyticus in NaClO₂ solution at pH 5.6. Initial NaClO₂ concentration was 21.7 mM. Symbols: ○, 5°C; ●, 15°C; △, 25°C; ▲, 35°C.

TABLE 2. Kinetics of the inactivation of V. parahaemolyticus in NaClO₂ solutions at various temperatures.

| Temperature (°C) | \(k\) (l/mmol·min) | Minimum CT to achieve at least a 3-log reduction (mmol·min/l) |
|------------------|------------------|-----------------------------|
| 5                | 0.0733           | 53.7                        |
| 10               | 0.0918           | 43.1                        |
| 15               | 0.144            | 25.9                        |
| 20               | 0.169            | 24.9                        |
| 25               | 0.228            | 15.5                        |
| 30               | 0.287            | 14.4                        |
| 35               | 0.467            | 8.14                        |
| 40               | 0.606            | 7.05                        |

The \(k\) values and minimum CT values to achieve at least a 3-log reduction. Inactivation experiments were conducted in the NaOCl₂ solution of 21.7 mM and pH 5.6.

while the CT values decreased by 1/7.6. These results clearly indicated that temperature of NaClO₂ solution was also an important factor in accelerating the inactivation.

The relationship between temperature and the \(k\) values was represented by the Arrhenius plot (Fig. 4):

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where \(E_a\) is the apparent activation energy, J/mol; \(R\) is the gas constant, 8.314 J/K·mol; \(T\) is the temperature, K; and \(A\) is the frequency factor, l/mmol·min. A linear relationship between \(\ln k\) and \(10^{3/T}\) was obtained, and the \(E_a\) was estimated to be 43.5 kJ/mol from the slope of the graph. These findings indicated that \(k\) increased by approximately 1.8 times for every 10°C rise in temperature.

Measurement of ClO₂ formation

Figure 5 shows typical time courses of concentrations of ClO₂ (Abs. 360) and HClO₂/ClO₂- (Abs. 260) under inactivation conditions at pH 4.0 and 25°C. A slight decrease

FIG. 3. Effect of temperature on the inactivation of V. parahaemolyticus in NaClO₂ solution at pH 5.6. Initial NaClO₂ concentration was 21.7 mM. Symbols: ○, 5°C; ●, 15°C; △, 25°C; ▲, 35°C.

FIG. 4. Arrhenius plot of the \(k\) values. For inactivation conditions, see the legend of Fig. 3.

FIG. 5. The time course of concentrations of ClO₂ (Abs. 360) and HClO₂/ClO₂- (Abs. 260) under the inactivation conditions. Experiment was conducted at pH 4.0 and 25°C. Symbols: ○, ClO₂ (Abs. 360); △, HClO₂/ClO₂- (Abs. 260).
in the $\text{Abs}_{290}$ was observed during 5 min of the inactivation reaction, whereas there was no significant change in the $\text{Abs}_{300}$. Similar results to those at pH 4.0 were also observed at pH 4.5 to 6.5 (data not shown). These results indicated that no formation of ClO$_2^-$ occurred during the inactivation experiments in this study.

**DISCUSSION**

It was confirmed that the efficacy of weakly acidic NaClO$_2$ solution in inactivating *V. parahaemolyticus* depended strongly on the solution pH. The $k$ value increased markedly with the decreasing solution pH. In this study, ClO$_2^-$ was not formed under the inactivation conditions, indicating that the action of ClO$_2^-$ could be ignored. It is likely that the pH dependence of the efficacy of NaClO$_2$ in inactivating *V. parahaemolyticus* can be explained on the basis of the HClO$_2$ – ClO$_2$ equilibrium. The proportion of HClO$_2$ in the NaClO$_2$ solution increases from 0.0023% (0.50 µM) at pH 6.5 to 0.72% (8.1 µM) at pH 4.0 (calculated by $pK_a = 1.86$). These findings indicated that HClO$_2$ was the active species governing the inactivation of *V. parahaemolyticus*. It is believed that the strong antimicrobial activity of the electrically neutral undissociated form of the oxyclohal compounds, such as HClO and HClO$_2$, can be attributed to its ability to penetrate into the microbial cell across the cell membrane, i.e., the lipid bilayer (Fukuzaki, 2006; Horiguchi et al., 2015; Kemp, 2000). Consequently, HClO$_2$ can attack cellular components inside the cell, thereby giving a strong antimicrobial activity.

On the other hand, the possible contribution of ClO$_2^-$ to the inactivation of *V. parahaemolyticus* cannot be excluded completely. For instance, the NaClO$_2$ solution of 21.7 mM at pH 6.5 contains only 0.50 µM of HOCl$_2$. Nevertheless, the inactivation of *V. parahaemolyticus* occurred slowly but significantly (> 4 log). This suggested that ClO$_2^-$ at high concentrations would be also involved in the inactivation of *V. parahaemolyticus*. ClO$_2^-$ is an oxidizing agent with a standard reduction potential ($E^0$) of 0.76 V (Weast, 1988). Ionized ClO$_2^-$ is not able to penetrate the bacterial cell membrane and therefore it exerts an oxidizing action from outside the cell. In a previous study, it was reported that NaClO$_2$ could oxidize glutathione preferentially rather than membrane phospholipids, both in vitro and in cells at pH 7.2 to 8.0, where the ratio of ClO$_2^-$ to HClO$_2$ was extremely high (Ingram et al., 2003). In this previous study, it was also confirmed that the effects of NaClO$_2$ in vitro were similar to those observed in vivo. In connection with this, for *Escherichia coli* during HClO stress, loss of physiological function resulting from sulfhydryl oxidation has been suggested to be the antimicrobial event (Albrich et al., 1981; Dukan et al., 1999; Thomas, 1979). At present, although the mode of the antibacterial action of ClO$_2^-$ is unclear, it is presumed that high concentrations of ClO$_2^-$ might exert an oxidative damage to thiol-containing proteins of the cell membrane or cell surface of *V. parahaemolyticus*.

In general, the rate of chemical reactions, including oxidation, is enhanced with the increase in temperature. In this study, $k$ shows an Arrhenius-type temperature dependence and the $E_a$ was estimated to be 43.5 kJ/mol. To our knowledge, this is the first estimation of the $E_a$ value for NaClO$_2$ disinfection. The $E_a$ values for the inactivation of *E. coli* in aqueous solution were reported to be 50.4 kJ/mol in ClO$_2$ disinfection (Benarde, 1967) and 37 kJ/mol in ozone disinfection (Gunten, 2003). The authors reported the $E_a$ values of 56.8 to 59.9 kJ/mol for the inactivation of *Pseudomonas fluorescens* by pH-controlled NaClO$_2$ solutions (Fukuzaki et al., 2009). These values in the literature are in good agreement with the $E_a$ values obtained in this study. Commonly, reactions that are controlled by diffusion in aqueous solution have low $E_a$ values of less than about 20 kJ/mol. Therefore, relatively high $E_a$ values of approximately 40 to 60 kJ/mol obtained for the action of these oxychloro compounds are in the same order as most chemical reactions (Gunten, 2003). It seems that the deleterious oxidative reaction by ClO$_2^-$ is perhaps the rate-determining process in the inactivation of *V. parahaemolyticus*.

In conclusion, the rate of inactivation of *V. parahaemolyticus* increased exponentially by decreasing the pH of the NaClO$_2$ solution. HClO$_2$ was the potent antibacterial species even though its concentration was very low. It was also shown that the use of weakly acidic NaClO$_2$ solution containing high concentrations of ionized ClO$_2^-$ could also achieve a significant reduction of viable cells by more than 4-log. Furthermore, the antibacterial action of NaClO$_2$ solution could be accelerated by elevated temperature. These features confirm that weakly acidic NaClO$_2$ solution is easy to use and applicable as a moderate disinfectant at high concentrations.

**REFERENCES**

Albrich, J. M., McCarthy, C. A., and Hurst, J. (1981) Biological reactivity of hypochlorous acid: implications for microbialicidal mechanisms of leukocyte myeloperoxidase. *Proc. Natl. Acad. Sci. USA*, **78**, 210-214.

Benarde, M. A., Snow, B., Olivieri, V. P., and Davidson, B. (1967) Kinetics and mechanism of bacterial disinfection by chlorine dioxide. *Appl. Microbiol.*, **15**, 257-265.

Benford, D. J., Hill, M. F., Jackson, M. P., and Larsen, J. C. (2008) Acidified sodium chlorite. In Safety evaluation of certain food additives and contaminants, pp. 3-54, World Health Organization, Geneva.

Dukan, S., Belkin, S., and Touati, D. (1999) Reactive oxygen
species are partially involved in the bacteriocidal action of hypochlorous acid. Arch. Biochem. Biophys., 367, 311-316.

Fournier, P. E., and Ogata, H. (2005) Pandemic Vibrio parahaemolyticus O3: K6, Europe. Emerg. Infect. Dis., 11, 1317-1318.

Fukuzaki, S. (2006) Mechanisms of actions of sodium hypochlorite in cleaning and inactivation processes. Biocontrol Sci., 11, 147-157.

Fukuzaki, S., Urano, H., Takahashi, K., Yamada, S., and Takagi, A. (2009) Kinetic study of the effect of temperature on the cleaning and disinfecting actions of sodium hypochlorite (in Japanese). Bokin Bobai, 37, 253-262.

Hasegawa, Y., Nakamura, Y., Tonagai, Y., Kobata, M., and Ito, T. (1989) Antimicrobial effect of sodium hypochlorite on bacteria and yeasts (in Japanese). J. Food Hyg. Soc. Jap., 30, 240-249.

Horiuchi, I., Kawata, H., Nagao, T., Imahoji, H., Murakami, K., Kino, Y., Yamasaki, H., Koyama, H., Fujita, Y., Goda, H., and Kuwahara, T. (2015) Antimicrobial activity and stability of weakly acidified chloric acid water. Biocontrol Sci., 20, 43-51.

Horváth, A. K., Nagyával, I., Peintler, G., Epstein, I. R., and Kustin, K. (2003) Kinetics and mechanism of the decomposition of chloric acid. J. Phy. Chem. A, 107, 6966-6973.

Hosoi, M., Yoshida, M., Takahata, T., Hoshino, T., and Imada, K. (1990) Growth-inhibitory activity of sodium chlorite on several food poisoning bacteria (in Japanese). J. Food Hyg. Soc. Jap., 31, 469-473.

Ingram, P. R., Homer, N. Z., Smith, R. A., Pitt, A. R., Wilson, G. G., Oleinik, O., and Spickett, C. M. (2003) The interaction of sodium chlorite with phospholipids and glutathione: a comparison of effects in vitro, in mammalian and in microbial cells. Arch. Biochem. Biophys., 410, 121-133.

Katsui, N., Kato, N., Asada, S., and Kita, E. (1999) Bleb formation on the cell envelope of osmotic shocked cells of Vibrio parahaemolyticus. Biocontrol Sci., 4, 31-34.

Kemp, G. K., Aldrich, M. L., and Waldroup, A. L. (2000) Acidified sodium chloride antimicrobial treatment of broiler carcasses. J. Food Prot., 63, 1087-1092.

Kobayashi, M., Akiyama, S., Iwashita, M., Suzuki, A., and Nakajima, H. (1990) A study on the bactericidal effect of sodium chlorite (in Japanese). J. Food Hyg. Soc. Jap., 31, 491-498.

Rutala, W. A., and Weber, D. J. (1997) Uses of inorganic hypochlorite (bleach) in health-care facilities. Clin. Microbiol. Rev., 10, 597-610.

Takahashi, K., and Fukuzaki, S. (2012) Removal and inactivation of bacterial cells adhering to polyethylene terephthalate surfaces during a combined alkali-chlorine cleaning (in Japanese). J. Antibact. Antifung. Agents, 40, 405-413.

Takahashi, K., Tanaka, R., and Fukuzaki, S. (2016) Inactivation of Vibrio parahaemolyticus unattached and attached to a solid surface in pH-controlled sodium hypochlorite solutions. Biocontrol Sci., 21, 265-268.

Thomas, E. L. (1979) Myeloperoxidase-hydrogen peroxide-chloride antimicrobial system: effect of exogenous amines on antibacterial action against Escherichia coli. Infect. Immun., 25, 110-116.

von Gunten, U. (2003) Ozonation of drinking water: Part II. Disinfection and by-product formation in presence of bromide, iodide or chlorine. Water Res., 37, 1469-1487.

Watson, H. E. (1908) A note on the variation of the rate of disinfection with change in the concentration of the disinfectant. J. Hyg., 8, 536-542.

Weast, R. C. (1988) CRC handbook of chemistry and physics, 1st student edition, CRC Press Inc., Boca Raton, Florida.

Wei, C. I., Cook, D. L., and Kirk, J. R. (1985) Use of chlorine compounds in the food industry. Food Technology, 39, 107-115.

Wong, H. C., Chun, Y. C., and Yu, J. A. (2002) Attachment and inactivation of Vibrio parahaemolyticus on stainless steel and glass surface. Food Microbiol., 19, 341-350.