

Figure 2.5.02. Changes in numbers of aerobic bacteria in the kimchi prepared with ozone-treated cabbage and inoculation of *L. acidophilus* during fermentation at 25°C (Kim et al., 1993).

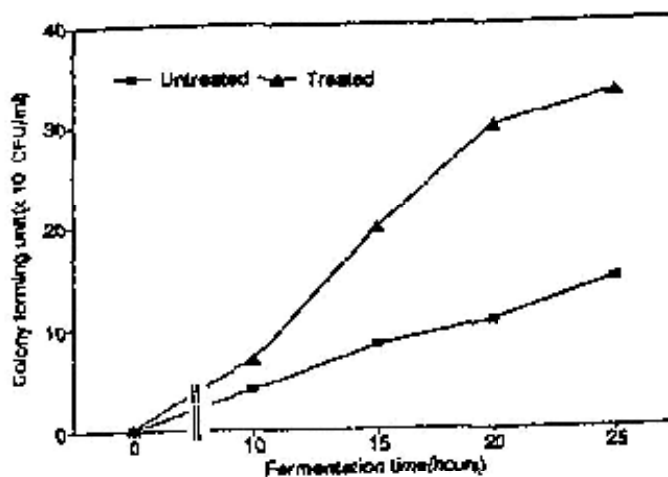


Figure 2.5.03. Changes in numbers of lactic acid bacteria in the kimchi prepared with ozone-treated cabbage and inoculation of *L. acidophilus* during fermentation at 25°C (Kim et al., 1993).

2.5.4 Hampson et al. (1994) – Broccoli, Broccoli and Carrots

Abstract: Using existing and newly acquired equipment, a system was constructed for the testing of ozone (O₃) for use as a sanitizing agent in food processing. The intended use of the system is to demonstrate that ozone is an effective and efficient germicide for the treatment of raw agricultural commodities. The system has the capacity to ozonate 200 gallons of water which then can be used to wash produce, reducing the microbial load through direct contact

between the ozone in the water and the microbes on the produce. The system is designed to model a flume wash system with a cascading water supply. A one horsepower motor powers the water flow and ozone levels are monitored continuously via in-line ORP probes. Ozone levels are confirmed using the HACH colorimetric assay. The corona discharge ozone generator is combined with an oxygen generator yielding ca. 15 grams O₃ per hour. This allows for sufficient variation in test parameters, such as organic loading, incoming microbial load, the decay rate of O₃, specific surface area of the food, water flow, and temperature. All experimental samples are tested immediately on-site for Aerobic Plate Count, Yeast and Mold, Coliforms, or Mesophilic Spores. Initial studies indicate that a three log-fold reduction in microbial load is possible with a ten minute contact time (CT: where CT = mg/L O₃ x minutes). In order for the process to be commercially feasible, the CT must be reduced by at least 50% without sacrificing the germicidal efficacy. From an engineering standpoint, the goal is to develop specific CT values for different commodities based upon the above-mentioned variables. These studies are of significance to the government for the approval of O₃ as a food-contact sanitizing agent; to the industry as a chlorine alternative; and to the consumer, and everyone, from a food safety perspective.

Sample Treatment: Approximately 1000 grams of each sample (broccoli, broccoflower and carrots) was weighed and placed into presanitized nylon mesh bags. These samples were treated in ozonated water for one, five or ten minutes. The control sample was washed for a period of five minutes in non-ozonated water. After treatment, samples were cut into small segments using a sanitized knife. A random 50 gram sample was blended with 450 mL of peptone broth (1:10 dilution) and scheduled microbial tests were performed within 30 minutes of washing.

Microbial Tests: Aerobic Plate Count (APC) and yeast and mold (using DRBC agar; Oxoid) tests were performed using standard microbiological methods (FDA BAM, APHA).

Results: The following figures depict the results of using ozone in the range of 0.64 to 1.11 ppm on broccoli, carrots and broccoflower: Figure 2.5.04 -- Reduction of microorganisms on broccoli using ozone at a concentration of 1.11 ppm; Figure 2.5.05 -- Reduction of microorganisms on carrots using ozone at a concentration of 0.64 ppm; Figure 2.5.06 -- Reduction of microorganisms on broccoflower using ozone at a concentration of 1.08 ppm. Under typical experimental conditions, anywhere from 1 to 3 microbial load reductions were seen. Broccoli shows the best reduction.

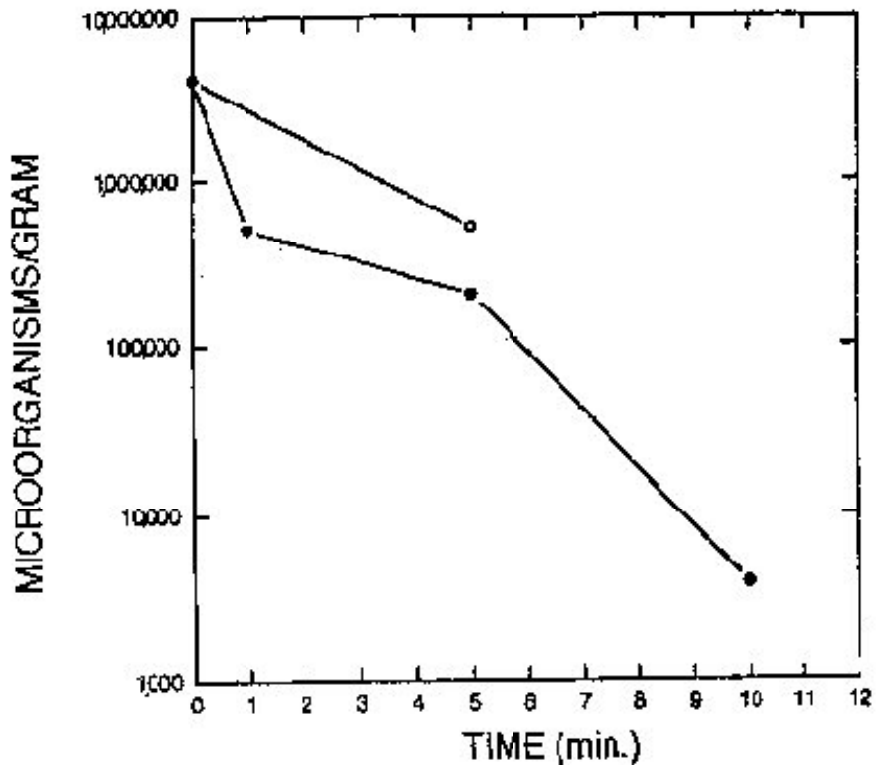


Figure 2.5.04. Reduction of microorganisms on broccoli using ozone at a concentration of 1.11 ppm (Hampson et al., 1994).

From the experimental data obtained, CT value equations were derived for broccoli, carrots and broccoflower. The following three figures show this information: Figure 2.5.07 -- Plot of CT values vs. log reduction for broccoli; Figure 2.5.08 -- Plot of CT values vs. log reduction for carrots; Figure 2.5.09 -- Plot of CT values vs. log reduction for broccoflower. The broccoli gave the smallest CT value per given log reduction. Each graph shows a high degree of correlation among the plotted CT points.

Using the same experimental protocol, chlorine was substituted for ozone as the germicidal agent. Comparing chlorination (Figure 2.5.10) to the results obtained using ozone, an additional 2-log reduction was realized for APC when ozone was used as the germicidal agent.

Conclusions: The results from this study support the following conclusions:

- Ozonation of raw agricultural commodities is a first order disinfection process.
- The results of this study give positive indications that ozone is an effective disinfectant.

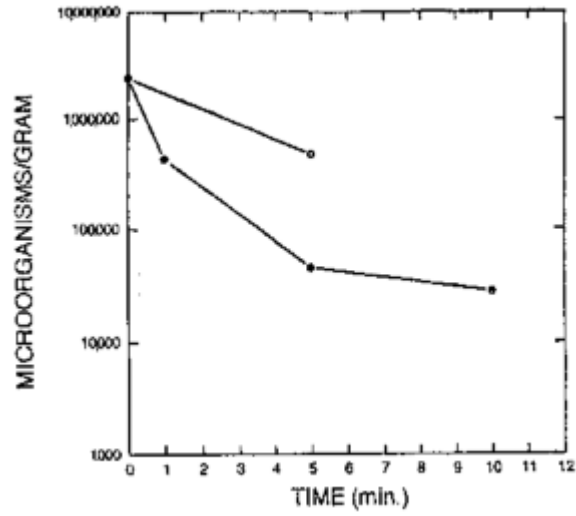


Figure 2.5.05. Reduction of microorganisms on carrots using ozone at a concentration of 0.64 ppm (Hampson et al., 1994).

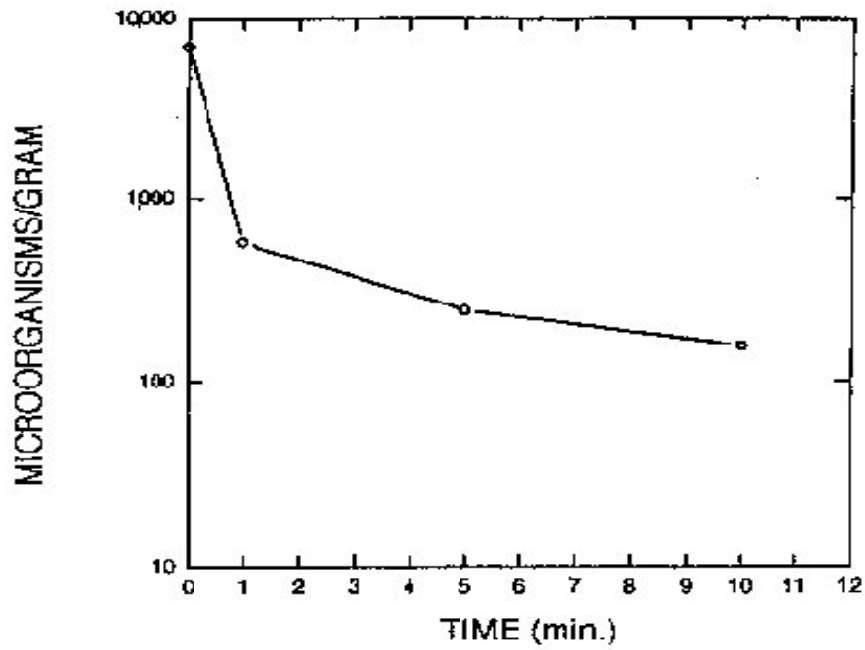


Figure 2.5.06. Reduction of microorganisms on broccoflower using ozone at a concentration of 1.08 ppm. (Hampson et al., 1994).

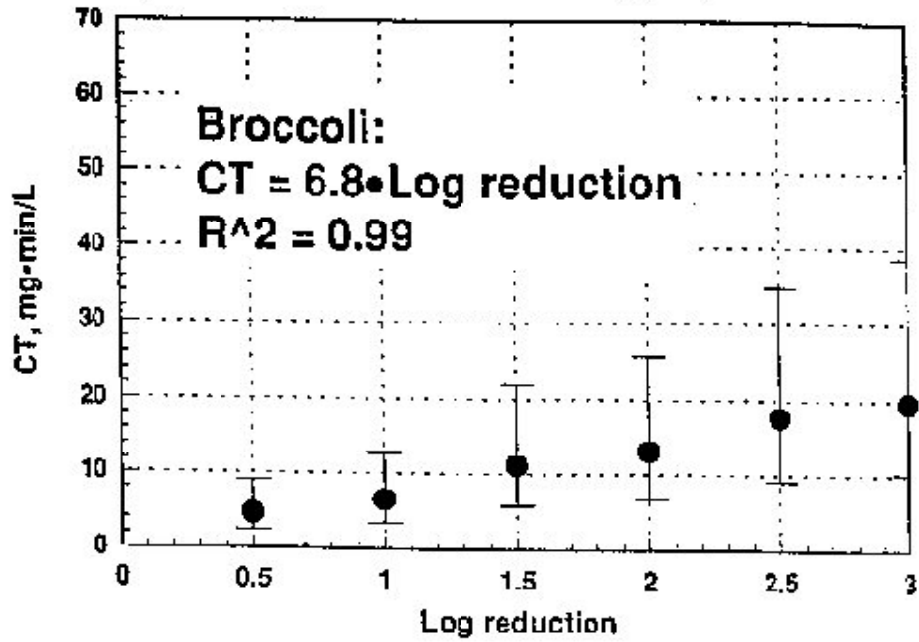


Figure 2.5.07. Plot of CT values vs. log reduction for broccoli (Hampson et al., 1994).

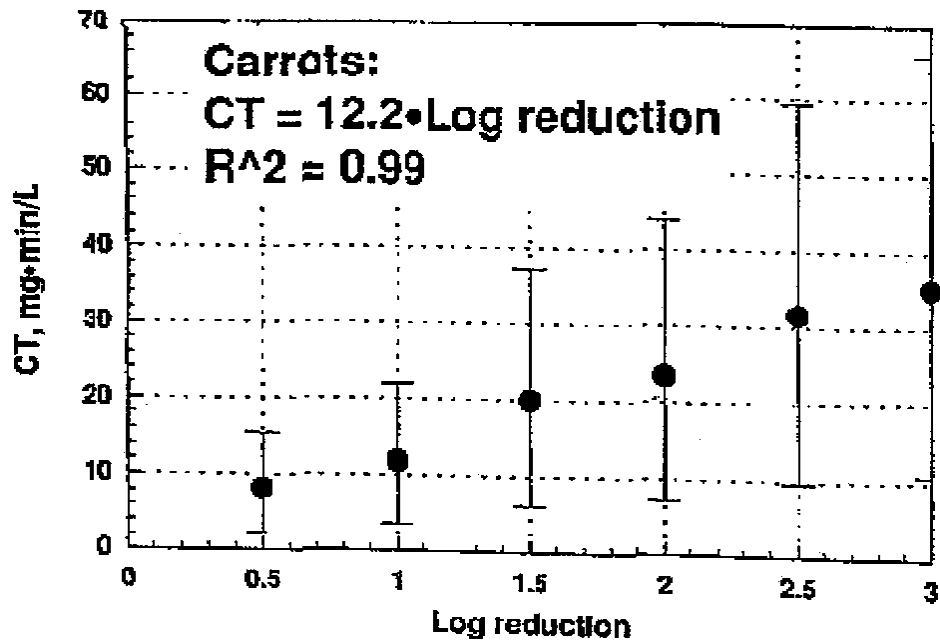


Figure 2.5.08. Plot of CT values vs. log reduction for carrots (Hampson et al., 1994).

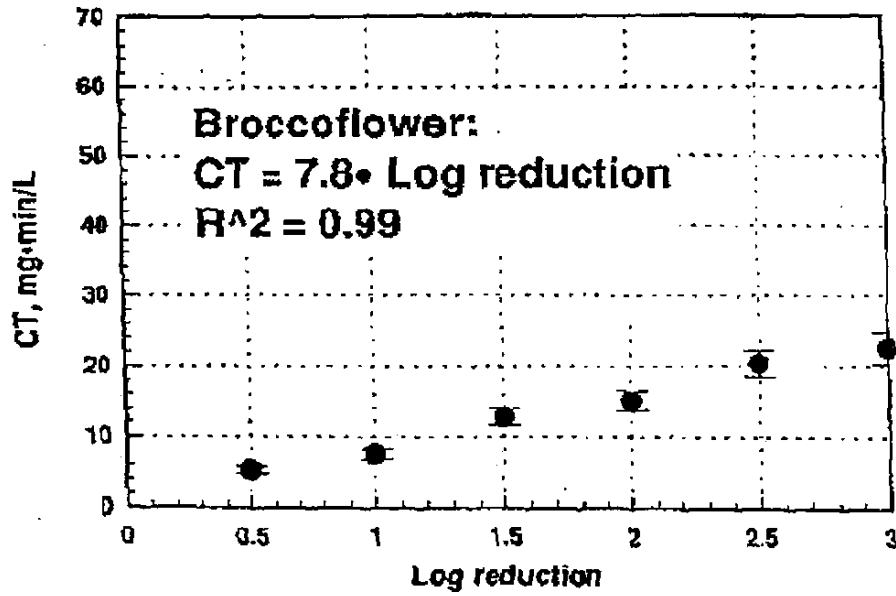


Figure 2.5.09. Plot of CT values vs. log reduction for broccoflower (Hampson et al., 1994).

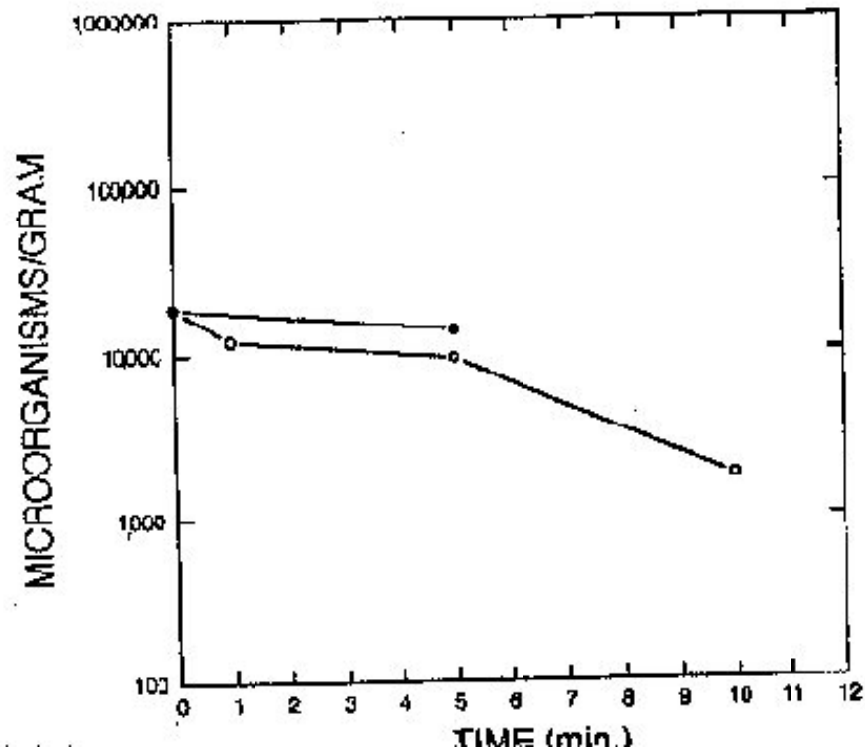


Figure 2.5.10. Chlorination of broccoli using 100 ppm of chlorine (Hampson et al., 1994).

- A number of independent factors influence the overall effectiveness of ozone as a disinfectant, e.g., water temperature, pH, ozone concentration, non-target demand substances, and specific surface area.
- Broccoli had the lowest CT values to achieve a given log reduction followed by broccoflower, then carrots.
- Preliminary results indicate that ozone proves to be a better disinfectant than chlorine under the conditions noted above.

2.5.5 Liew and Prange (1994) – Carrots

Abstract: Effects of ozone and storage temperature on carrots and two post-harvest pathogens, *Botrytis cinerea* Pers. and *Sclerotinia sclerotiorum* de Bary -- were investigated. Pathogen-inoculated and uninoculated whole carrots were exposed to an ozone concentration of 0 (control), 7.5, 15, 30, or 60 $\Phi\text{L-liter}^{-1}$. Treatment chambers were flushed with a total flow rate of 0.5 L-min^{-1} (air and ozone) for 8 h daily for 28 days. The experiment was repeated twice at storage temperatures of 2, 8, and 16EC. The residual ozone concentration (ozone supplied--exhausted and reacted ozone) increased with ozone supply concentration but was less at higher storage temperatures. A 50% reduction of daily growth rates of both fungi at the highest ozone concentration indicated that ozone was fungistatic. Carrot respiration rate, electrolyte leakage, and total color differences increased with ozone concentration. Ozone-treated carrots were lighter (higher L^* values) and less intense (lower chroma values) in color than control carrots.

The objectives of this study were 1) to determine the residual concentration of ozone, 2) to determine the effect of ozone on the two major storage pathogens of carrots, and 3) to observe ozone-induced changes in carrot physiology and quality during storage.

Methods and Materials:

'Vitabrite' carrots, obtained from a local grower (Berwick, N.S.), were hand-washed and stored at 0EC until use. Crown diameters of the carrots were from 3 to 4 cm.. An ozone generator (Tri-Ox Swindon, England) was set to produce 76.5 $\Phi\text{L-liter}^{-1}$ of ozone in air. Air containing ozone at flow rates of 0 (control), 0.05, 0.1, 0.2, or 0.4 liter-min^{-1} were blended with compressed air to produce ozone concentrations of 0, 7.5, 15, 30, or 60 $\Phi\text{L-liter}^{-1}$, in a total flow of 0.5 liter-min^{-1} for each treatment. Ozone and compressed air flows were controlled with needle valves. Treatment chambers consisted of airtight 64-liter polyvinyl chloride containers placed in storage rooms set at 2, 8, or 16EC. The chambers were flushed continuously for 8 h daily for 28 days.

Ozone concentrations in the chambers were monitored during the treatment period with an ultraviolet-based detector with a measurement range of 0 to 100 $\Phi\text{L liter}^{-1}$ at 253.7 nm.

Disease: Isolates of *S. sclerotiorum* and *B. cinerea* were obtained from infected carrots in local storage. Fungal stock cultures and inoculum were maintained on potato dextrose agar (PDA). A 1.0-cm-diameter mycelial plug, obtained near the margin of a 4- to 5-day-old fungal culture, was placed in a wound of each carrot 1.0 ∇ 0.5 cm from the crown. The wound was a 1.0-cm diameter x 0.5-cm-deep depression created with a 1.0-cm diameter cork borer. Fungal growth