Evaluation of InvisiShield™ technology to reduce E. Coli, Salmonella and Listeria monocytogenes using the antimicrobial Chlorine Dioxide

Objective

To determine the influence of Aptar's InvisiShield™ technology against multiple strains of foodborne pathogens *Listeria monocytogenes, Salmonella,* and pathogenic *E. coli* on commercially packaged tomato slices.

InvisiShield™

Research Overview

Sliced tomatoes were inoculated with three foodborne pathogen cocktails and treated in Aptar's InvisiShield™ technology trays in temperature-controlled storage (7°C) for up to 14 days. Inoculated tomato slices experienced significant reductions of 3.6 logs, 4.5 logs and 4.2 logs for *Salmonella,* and pathenogenic *E. coli*, and *L. Monocytogenes* after 7 days in the high inoculation test and 2.58, 2.67, and 1.82 log reductions for the low inoculation study respectively. Findings suggest that the InvisiShield™ technology demonstrated antimicrobial activity against *L. monocytogenes, Salmonella,* and pathenogenic *E. coli* as it was significantly different from the controls with p-values of 0.00 when compared per day. The levels of chlorine dioxide used were safe for food and did not negatively impact the tomatoes as demonstrated in the sensory panel results. This technology will offer a differentiated active packaging solution, protecting high-risk products such as fresh-cut produce.

Highlights

- Technology can reduce risk of foodborne illness
- Chlorine dioxide released from the InvisiShield™ system inhibits major pathogens up to 3 logs
- Sensory properties were not statistically different between treated and untreated tomatoes

Introduction

While chlorine and chlorine dioxide has been used for many years as an antimicrobial on food and water, there have been many challenges for both safety and quality that have prevented chlorine dioxide gas from being utilized commercially.[1] Chlorine dioxide gas is an antimicrobial of choice because it is very effective and is broad-spectrum, demonstrating efficacy against both gram-negative and gram-positive microorganisms. Aptar's novel InvisiShield™ technology is able to fill a gap and offer this effective antimicrobial to the industry due to the specially-engineered delivery system, which can safely create ClO2 and control the dosage in the package in order to reduce negative organoleptic properties. The InvisiShield™ material is extruded and remains stable throughout the supply chain distribution. It contains a base polymer, a channeling agent and the active ingredient (ClO2). The release kinetics of ClO2 is triggered by relative humidity in the package. ClO2 then migrates through the same channels or through the polymer blend itself into the surrounding environment in a controlled manner.

Results and Discussion

Sensory Analysis

Forty-two panelists were asked to rate the appearance, flavor and texture of sliced tomatoes stored at 4°C for 3 days. Treated tomatoes were rated as not significantly different in appearance, flavor, and texture attributes compared to the other sample. The data was analyzed by tabulating the number of correct responses and compared to values in tables for the minimum number of "correct" responses needed to conclude that a perceptible difference exists. For n=42 panelists, the number is 22 (a=0.01).[2]

Microbial Counts in Low Inoculation Study

Salmonella, L. monocytogenes, and pathogenic *E. coli* saw similar levels of inoculation onto the sliced tomatoes in both the treatment and control trays on day 0 shown by no statistical difference in the Tukey test and a p-value of 0.156, 0.204, and 0.258 respectively. *Salmonella, L. monocytogenes,* and pathogenic *E. coli* levels on the surface of sliced tomatoes in the Control trays were stable over the course of storage at 7°C, with an initial average reduction of 0.27 log CFU/g after 2 days, followed by an average increase of 0.87 log after 14 total days of storage (Figures 1, 2, and 3). *Salmonella* and *E. coli* levels were reduced on the surface of tomatoes in the InvisiShield™ Test trays by 1.55 and 1.52 logs respectively after 2 days of storage and were not recoverable (after overnight enrichment and streak plating) from that point forward (Figures 1 and 2). The treatment trays were statistically different from controls after the initial day 0 testing point on all sampling days up to the end of the study on day 14. *L. monocytogenes* levels were also reduced in the InvisiShield™ Test trays by 1.08 log after day 2, but at subsequent test points was recovered sporadically near the limit of detection, with an average reduction of 1.58 logs from that point forward (Figure 3). All treatment trays vs. control trays were statistically different after day 0 on each sampling day up to the end of the experiment on day 14.

Figure 1: Pathenogenic E. coli Low Inoculation Study

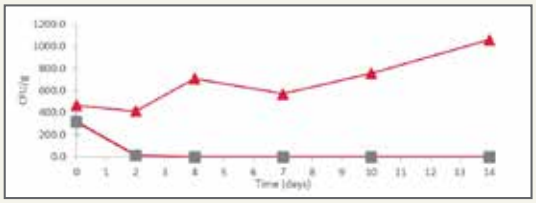


Figure 2: Salmonella Low Inoculation Study

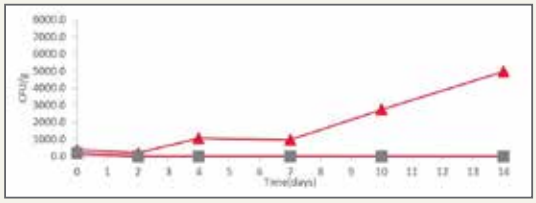
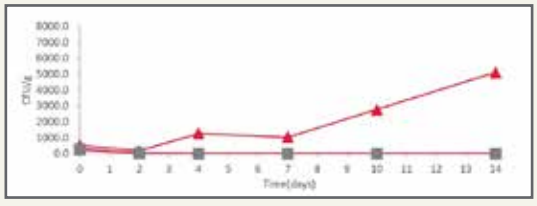


Figure 3: L. monocytogenes Low Inoculation Study



Changes in mean CFU/g counts in control trays (▲) and treatment trays (■) over time in Low Inoculation Study.

Extending Freshness. Enhancing Safety.™

Aptar food + beverage

Results and Discussion

Microbial Counts in High Inoculation Study

Salmonella, *L. monocytogenes*, and pathogenic *E. coli* saw similar levels of inoculation onto the sliced tomatoes in both the treatment and control trays on day 0 shown by no statistical difference in the Tukey test and a p-value of 0.149, 0.183, and 0.283 respectively. *Salmonella*, *L. monocytogenes*, and pathogenic *E.coli* levels on the surface of sliced tomatoes in the control trays were relatively stable with an initial average inoculation of 9 logs and reduction of 0.5 log over the 7 day span for *E.coli* and *L. monocytogenes*, and a 1.5 logs reduction for *Salmonella* over the same 7 day span with a final count of 7.5 logs respectively on day 7 (Figures 4, 5, and 6). The InvisiShield™ Test trays showed a reduction to 3.8 logs for *Salmonella* and *E. coli*, with an average log reduction of 3.6 logs and 4.5 logs respectively (Figures 4 and 5). *L. monocytogenes* showed a similar reduction in the InvisiShield™ Test trays with a count of 4.3 logs on day 7 and a reduction compared to the Control trays of 4.2 logs (Figure 6).

Figure 4: E. coli High Inoculation Study

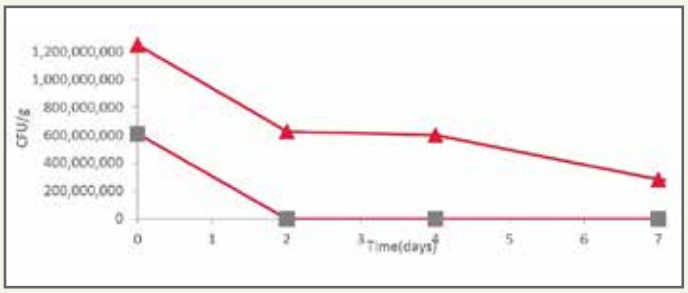


Figure 5: Salmonella High Inoculation Study

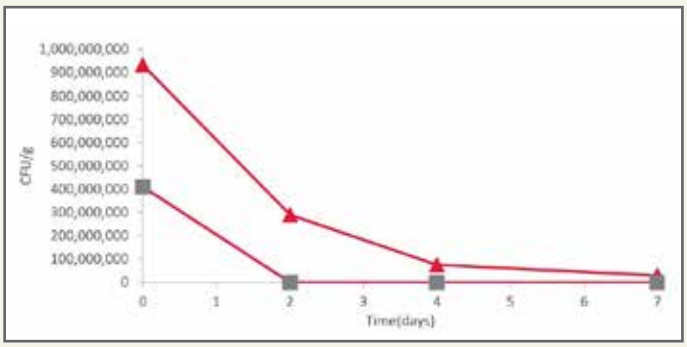
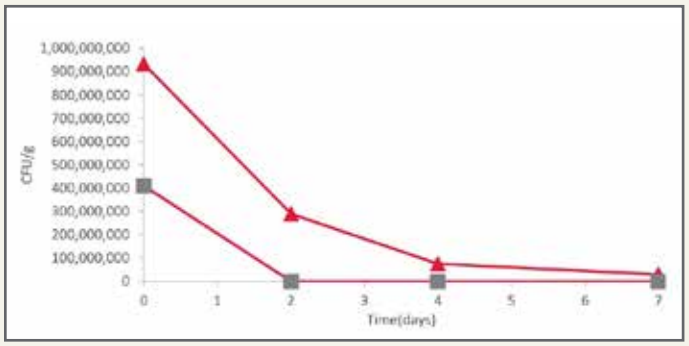


Figure 6: L. monocytogenes High Inoculation Study



Changes in mean CFU/g counts in control trays (▲) and treatment trays (■) over time in High Inoculation Study.

InvisiShield™

Conclusion

The lack of an adverse effect on sensory quality make this treatment promising for sliced tomato commercial application. This is similar to what Sy et al. saw in an earlier study but only had 20-30 min. exposure in a manual exposure tank with whole tomatoes compared to a sealed commercial product with sliced tomatoes in this study.[3] In a similar study, also by Sy, there was a whitening of the strawberries that decreased the sensory quality that was not seen with this controlled release technology on sliced tomatoes.[4] This further shows the advantage of this system over other methods of exposure to chlorine dioxide.

The data in this study indicated that the InvisiShield™ material can reduce the level of *L. monocytogenes, Salmonella* and pathenogenic *E. coli* on sliced tomatoes during elevated refrigerated storage. In the low inoculation study, InvisiShield™ test trays with sliced tomatoes surface inoculated with each pathogen and held at 7°C for up to 14 days showed complete inhibition of the *Salmonella* and *E. coli* during storage after 2 days, and suppression to nearly the limit of detection for the *L. monocytogenes* inoculum after 4 days of storage. Control trays showed a minor initial reduction after 2 days of storage, but by Day 14 had shown a minor outgrowth of approximately 1 log. In the high inoculation study, InvisiShield™ Test trays showed at least a 4 log reduction over the study with the most being a 4.6 log reduction, on pathenogenic *E. coli* on Day 7, and the least being a 3.6 log reduction on *Salmonella* on Day 7, compared to Control trays.

The ability to control the kinetics and customize the release rate of the chlorine dioxide was vital to maintaining the tomato's color, flavor, aroma and texture while achieving efficacy. The technology allows for this specialized engineering in order to strike the perfect balance between safety and quality.

Aptar is actively seeking pilot partners to introduce this in your market!
If you are interested, please contact:

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770-845-6077

1 Ridenour, G. M., and R. S. Ingols. 1947. Bactericidal properties of chlorine dioxide. *J. American Water Works Association* 39.6:561-567.
2 Meilgaard, M. C., G. V. Civille, and B. T. Carr. 2016. Sensory Evaluation Techniques. CRC Press, Boca Raton, Fla. https://doi.org/10.1201/9781439832271.
3 Sy, K. V., M. B. Murray, M. D. Harrison, and L. R. Beuchat. 2005. Evaluation of gaseous chlorine dioxide as a sanitizer for killing Salmonella, Escherichia coli O157: H7, Listeria monocytogenes, and yeasts and molds on fresh and fresh-cut produce. *J. Food Prot.* 68.6:1176-1187.
4 Sy, K. V., K. H. McWatters, and L. R. Beuchat. 2005. Efficacy of gaseous chlorine dioxide as a sanitizer for killing Salmonella, yeasts, and molds on blueberries, strawberries, and raspberries. *J. Food Prot.* 68.6:1165-1175