



## BioIonix White Paper No. 250

### BioIonix Meat and Poultry Industry RTE Products Pre-slicing and Pre-Packaging Microbial Control Intervention

#### Summary and Conclusions:

##### Pilot Scale Research Results

The research reported herein evaluated the performance of the BioIonix process to provide effective microbial control on RTE products through a spray application immediately prior to slicing and packaging. A high bolus of *Listeria innocua* was used as a surrogate for pathogens. The findings were:

- **BioIonix-treated water with low levels of salt provided effective pre-slicing microbial control on the tested RTE products.** No chemicals or other microbial interventions were used. Full disinfection was achieved at free available chlorine levels of approximately 1.0 ppm.
- **BioIonix also maintained proper microbial control as this water was recycled and reused for extended periods of time.** This can permit a significant reduction in plant water use for this microbial intervention step.

##### Full Scale Commercial Installation

Following successful pilot scale testing, a full-size commercial system was installed in the customer's plant in May, 2012. Three spray tunnels passing through the walls of the RTE slicing and packaging room shower water containing BioIonix SuperOxidants over the company's RTE products. One BioIonix system feeds all three spray tunnels. The BioIonix system has been in daily operation since that time, with the following results:

- **Effective continuous microbial control.** The customer reports that it has not detected any pathogenic bacteria on its RTE products or in its entire slicing and packaging room since BioIonix was installed.
- **Major cost savings.** Customer was paying approximately \$165K per year for an acidified sodium chlorite solution to provide microbial control prior to the installation of BioIonix. BioIonix eliminated this expense.
- **Operator complaints stopped.** Eye irritation, breathing issues and other operator health complaints were eliminated with BioIonix.
- **Facilities corrosion prevented.** Damage to concrete wall curbs, wastewater drains and equipment, a problem with the previous chemical treatment, was eliminated with the BioIonix process.

## The BioIonix Advanced Catalytic Disinfection Process

The BioIonix catalytic process disinfects process water containing low levels of salt and brine solutions without the use of chemicals. The platinum catalysts in the BioIonix process take apart the water molecule to create powerful reactive oxygen disinfection species. The sodium chloride salt provides the conductivity and chloride source needed to generate complementary activated chlorine disinfectants.

The process works where other disinfection methods fail or provide inconsistent microbial control results. BioIonix is effective in turbid waters containing color and suspended solids. The BioIonix process automatically compensates for changes in the organic load as this increases over time. In some applications, the process can flocculate solids for easy removal by skimming, eliminating membrane filtration. BioIonix treated process water and brine often can be reused indefinitely when combined with suitable solids filtration.



## White Paper Research Overview

### Test Objective

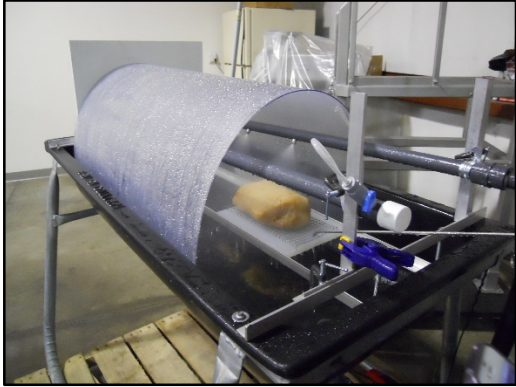



Demonstrate the bacterial reduction of a high bolus of *Listeria innocua* sprayed on the surface of customer's RTE deli products and then treated by the Bioionix system, as a surrogate to achieve USDA Alternative 1 post-lethality treatment for control of *Listeria monocytogenes*.

In the company's pilot plant, Bioionix conducted a series of test conditions to demonstrate log reduction of a *Listeria monocytogenes* surrogate (*Listeria innocua*) on the surface of three different RTE deli products, including turkey bologna, turkey pastrami, and smoked turkey breasts. Bioionix treated water was sprayed over the surface of these inoculated products.



*Bioionix Pilot Scale System*

**Test Protocol**

<p><b>Test Apparatus</b></p> <p>Biolonix pilot system circulates a 3 gpm solution of process water containing 5000 ppm sodium chloride salt through the catalytic reactor module to continuously generate disinfecting SuperOxidants. These disinfect bacteria and destroy other microbials on the surface of the treated products and in the process water itself. The Biolonix control system senses organic load and disinfection strength and automatically adjusted the treatment dose to maintain disinfection efficacy. In the test, free available chlorine was measured with an inline meter and verified with a Hach test kit.</p> <p>Individual spray bars were designed with each nozzle providing one gpm coverage over the deli products.</p>	
<p><b>Product</b></p> <p>Three products were selected from the customers RTE deli product line, representing significantly different surface characteristics.</p> <p>Photos show wrapped product as received. This wrap was removed prior to applying the microbial bolus and being treated through the Biolonix disinfectant shower.</p>	 <p><i>Pastrami is covered with a peppercorn rub without casing.</i></p>  <p><i>Bologna is a casing-on product.</i></p>  <p><i>Oil browned turkey breast is a par fried surface without casing.</i></p>

**Inoculum:**

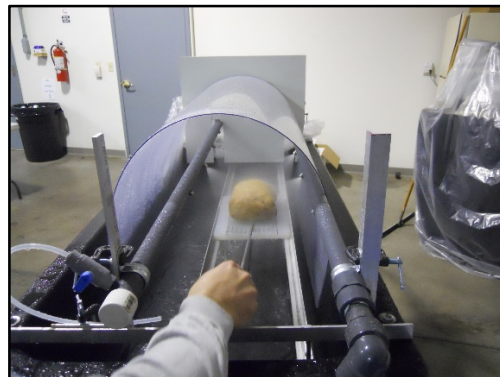
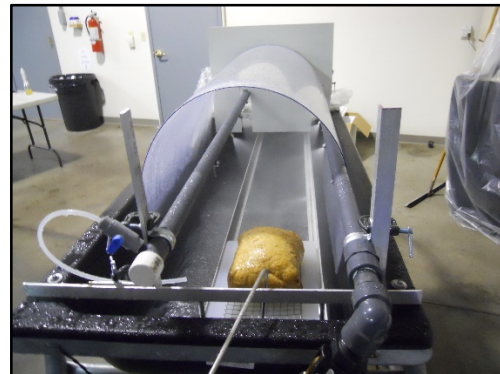
Deibel Laboratories, Madison prepared a *Listeria innocua* cocktail of four strains obtained from Deibel Laboratories, Gainesville, FL. These cultures are identified as L40, L41, L43 and L48. Overnight cultures of each were grown up in 3 ml of tryptic soy broth plus yeast extract. 1ml of each culture was transferred to separate 225ml tryptic soy broths and incubated overnight. About 100ml of each grown culture was combined in a clean, sterile 500ml bottle and given to Bioionix. Deibel Labs also supplied a Preval® aerosol spray gun for the inoculation study

**Inoculation**

Deli product pieces were inoculated by spraying the broth uniformly over the surface of each piece until fully wetted, hoping to achieve a target level of over  $10^6$  *Listeria innocua*. Product pieces were held for a minimum of 15 minutes prior to being placed in the spray tunnel and treated with the Bioionix disinfecting water spray.

**Treatment**

Tested product was placed on a movable tray in the test spray tunnel outside of the nozzle spray area. Timing was started and the tray was moved through the length of the tunnel for the three treatment times in the test protocol (15, 20 and 30 seconds). This treatment time represented the range of times that the product could be in the commercial spray cabinet.



**Sampling**

Product was placed on a sterile surface (bleached bench top-2% bleach solution). Using a pre-moistened sponge provided by Deibel Labs, a 6 inch square area of the product that was not in contact with the bleached bench top was swabbed. It was first swabbed horizontally then vertically making sure the area swabbed was the same for each product. Two swabs were taken for each product piece (marked "left" and "right") to provide multiple data points. The samples were labeled with a number and test condition, refrigerated and sent to Deibel Labs for analysis<sup>1</sup>.



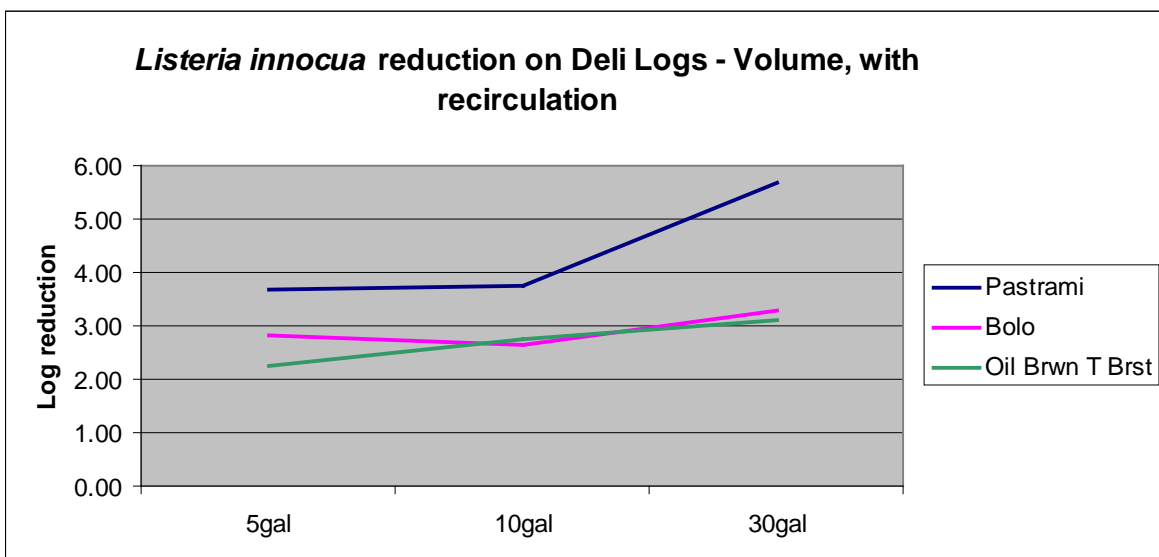
**Analysis**

Deibel Labs technicians added 10ml of a phosphate buffer solution to each sponge and massaged them thoroughly to ensure proper recovery of any *Listeria innocua* present. Dilutions of 10<sup>0</sup> to 10<sup>6</sup> were plated and poured with tryptic soy agar plus yeast extract. The plates were allowed to solidify and were overlayed with modified oxford medium 2. They were incubated at 35°C for two days. Typical colonies were counted and multiplied by the inverse of the dilution to determine the final count (cfu/sponge). These results were communicated to Bioionix.

**Test Results**

**Test 1: Spray Volume vs. Disinfection**

This test measured disinfection performance on the products compared to the volume of water run through the spray nozzles for the test. With multiple nozzles spraying in a relatively long spray cabinet, not all of the water volume contacted the product being treated, but the amount of water contact was directly proportional to the total amount sprayed. Tests were performed with spray volumes of 5, 10 and 30 gallons. Testing was done at relatively low power to maximize the recirculation of the water through the BioIonix system to evaluate water disinfection performance.





**Product A:** Turkey Pastrami with black pepper rub (no casing)

	Left cfu/g	log10	Right cfu/g	log10	Average cfu/g	Ave log10	Log Reduction	Water
Control	420,000	5.62	500,000	5.70	460,000	5.66		
5gal	62	1.79	130	2.11	96	1.98	3.68	<1
10gal	33	1.52	130	2.11	81	1.91	3.75	<1
30gal	1	0.00	1	0.00	1	0.00	5.66	<1

**Product B:** Turkey Bologna (in casing)

	Left cfu/g	log10	Right cfu/g	log10	Average cfu/g	Ave log10	Log Reduction	Water
Control	90,000	4.95	44,000	4.64	67,000	4.83		
5gal	110	2.04	90	1.95	100	2.00	2.83	<1
10gal	41	1.61	270	2.43	155	2.19	2.63	<1
30gal	20	1.30	52	1.72	36	1.56	3.27	<1

**Product C:** Smoked Turkey Breast (no casing)

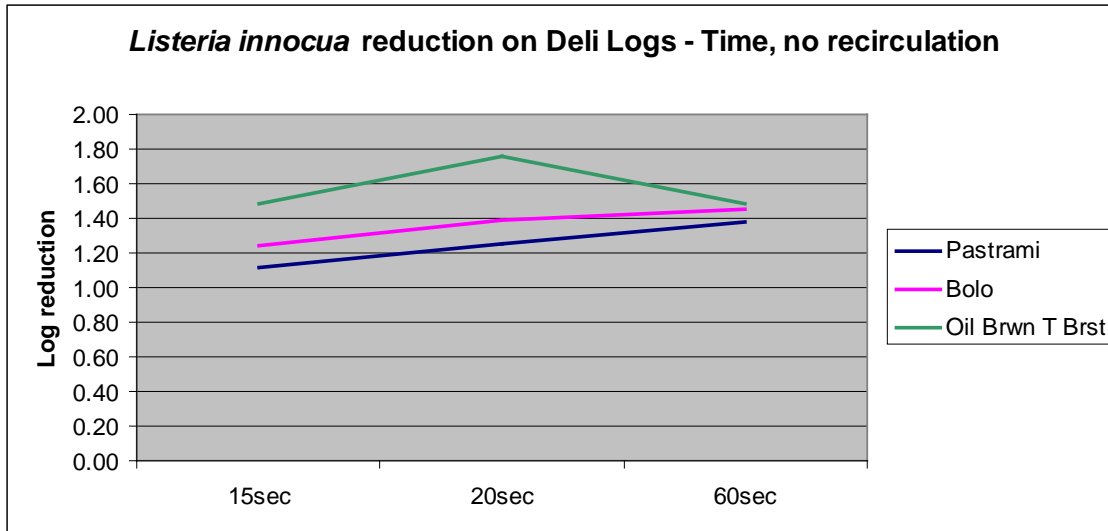
	Left cfu/g	log10	Right cfu/g	log10	Average cfu/g	Ave log10	Log Reduction	Water
Control	300,000	5.48	550,000	5.74	425,000	5.63		
5gal	2000	3.30	2900	3.46	2450	3.39	2.24	<1
10gal	670	2.83	790	2.90	730	2.86	2.77	<1
30gal	150	2.18	520	2.72	335	2.53	3.10	<1

**Test 1 Results**

- **An average reduction of bacteria counts by well over 99.9% was achieved on the product surface itself.** The lower starting counts on the turkey bologna are believed to be caused by the challenge of adhering bacteria to the smooth casing. These lower starting counts limited the maximum log reduction that was obtainable.
- **Complete disinfection of the treatment spray water (<1 cfu/g) was achieved at all treatment volumes.** Complete disinfection will allow this water to be reused for an extended period of time rather than immediately discharged.

**Test 2: Treatment Time vs. Disinfection**

This test measured disinfection performance on the products compared to the treatment time. Test conditions measured the time (15, 20 and 60 seconds) that the water spray contacted the product. A single 1.0 gpm spray nozzle was used. The goal was to duplicate the range of time that the products would likely be in a commercial spray tunnel. This test was conducted in a non-recirculation mode due to the short dwell time.



**Product A:** Turkey Pastrami with black pepper rub (no casing)

	Left cfu/g	log10	Right cfu/g	log10	Average cfu/g	Ave log10	Log Reduction
Control	170,000	5.23	11,000	4.04	90,500	4.96	
15sec	6700	3.83	7,100	3.85	6,900	3.84	1.12
20sec	5400	3.73	4,700	3.67	5,050	3.70	1.25
60sec	3600	3.56	3,900	3.59	3,750	3.57	1.38

**Product B:** Turkey Bologna (in casing)

	Left cfu/g	log10	Right cfu/g	log10	Average cfu/g	Ave log10	Log Reduction
Control	40,000	4.60	120,000	5.08	80,000	4.90	
15sec	3,700	3.57	5,500	3.74	4,600	3.66	1.24
20sec	3,200	3.51	3,400	3.53	3,300	3.52	1.38
60sec	3,300	3.52	2,400	3.38	2,850	3.45	1.45

**Product C:** Smoked Turkey Breast (no casing)

	Left cfu/g	log10	Right cfu/g	log10	Average cfu/g	Ave log10	Log Reduction
Control	730,000	5.86	410,000	5.61	570,000	5.76	
15sec	16,000	4.20	21,000	4.32	18,500	4.27	1.49
20sec	8,000	3.90	12,000	4.08	10,000	4.00	1.76
60sec	24,000	4.38	13,000	4.11	18,500	4.27	1.49

**Test 2 Results**

- **Disinfection takes place very quickly.** While results varied by test, they indicated that a treatment time of 30 seconds was a reasonable target for commercial spray applications. This treatment time was later proven to be sufficient in a full-scale commercial application and allowed for operator efficiency in conveyor loading.
- **Bacteria levels on the product surface were reduced by an average 1.34 log (95.4%).** This lower disinfection level was expected due to the power limitations of the

test power supply. Performance similar to that achieved in Test 1 would be obtained in a commercial system.

## Summary

This test on the spray disinfection of RTE products demonstrated that the BioIonix process can provide effective spray microbial control of RTE deli logs and other deli products. It also demonstrated that BioIonix could totally disinfect this spray water to permit its reuse with proper filtration. This can significantly reduce water use and wastewater discharges.

The customer that partnered with BioIonix on this test purchased a BioIonix commercial system. One BioIonix system provides full microbial control on all three of the customer's RTE product final disinfection tunnels prior to slicing and packaging. This system has been in daily use for over three years with excellent microbial control results.



*BioIonix RTE commercial system in operation.*