

EVALUATION OF BACTERICIDAL ACTIVITY OF A NEW PREPARATION P3 TSUNAMI USED FOR DISINFECTION OF FROZEN FRUIT AND VEGETABLES*

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The aim of the study was to determine, whether the new chemical preparation P3 Tsunami, product of Ecolab, shows bactericidal activity towards *Hafnia alvei*, *Citrobacter freundii*, *Enterobacter cloacae*. The P3 preparation has been suggested for reduction of microbiological contamination in technological water used for washing fruit and vegetables prior to freezing.

The tested bacterial suspension (1.5×10^8 cfu/mL – 5.0×10^8 cfu/mL) was added to prepared samples of tested preparation in four different concentrations. After specified contact time (1 min or 5 min ± 10 sec) bactericidal activity of the preparation was neutralised by a validated method. After 5 min ± 10 sec, 1 mL of neutralised mixture was transferred to Petri plates and 12–15 mL molten TSA medium was added. After 48 h of incubation at $37^\circ\text{C} \pm 1^\circ\text{C}$, the number of bacteria colonies was determined and the decrease of viable count was calculated. Preparation which in the test conditions defined by EN 1040 norm: 1997 caused 10^5 or higher decrease of bacteria count was described as bactericidal.

P3 Tsunami used in the concentration of 0.05% and 0.025% after 5 min contact time showed bactericidal activity towards all 30 bacterial strains used in the study; when used in the concentration of 0.05% after 1 min contact time it caused 10^5 higher decrease of bacteria count in 27 strains.

Bactericidal activity of P3 preparation after such a short contact time (1 min) suggests that it would fulfil the task of disinfecting technological water in the process of freezing fruit and vegetables.

INTRODUCTION

Fresh and frozen fruit and vegetables are required to fulfil microbiological requirements, listed in the Ministry of Health Regulation from 27th of December 2000 [Rozporządzenie Ministra Zdrowia, 2001] and also described by specifications from foreigner customers. Bacteriological requirements are based, among others, on determining the presence of coliform bacteria in a specified amount of product.

Confirming the presence of these bacteria – which is relatively easy – suggests the possibility of presence of pathogenic bacteria, although coliform bacteria group, especially of faecal type, may also comprise enteropathogenic, enterotoxic and enteroinvasive strains of *Escherichia coli*. Coliform bacteria (including faecal coliform) are often present on the surface of fresh fruit and vegetables [Białasiewicz, 2001; Białasiewicz & Królasik, 1999; Białasiewicz & Królasik, 2000; Brackett, 1994]. Therefore, we look for chemical preparations, which could reduce microbiological contamination in technological water and on the surface of fruit and vegetables used for freezing.

The aim of the study was to determine whether the new chemical preparation P3 Tsunami, product of Ecolab, containing peracetic acid, acetic acid, and hydrogen peroxide, shows bactericidal activity towards microorganisms from *Enterobacteriaceae* family.

MATERIAL AND METHODS

For the experiment there were used 30 bacterial strains from species: *Enterobacter cloacae*, *Citrobacter freundii*, *Hafnia alvei* (10 strains from each species), isolated from different food products. From starter bacteria, cultures were obtained on tryptic soy agar (TSA) slants. After 18–24 h of incubation at $37 \pm 1^\circ\text{C}$ first subculture was obtained. Then, second subculture from the first subculture and third subculture from the second subculture were prepared consecutively in the same way. Bacterial test suspension at a density from 1.5×10^8 cfu/mL to 5×10^8 cfu/mL (N) was prepared from the second or third culture. The examined solutions of P3 Tsunami preparation were dissolved in water to obtain required concentration 1.25-times higher than the tested concentration. Prior to testing, all materials (preparation solution, test suspension of bacteria, neutraliser, water) were heated to the test temperature of $20 \pm 1^\circ\text{C}$ in a water bath. Bacterial test suspension (1 mL) was added to each of the tubes containing 8 mL of one of the tested P3 Tsunami preparation solutions and 1 mL of water. Immediately timer was set on, and the contents were mixed in a microshaker ML-1. After specified contact time (1 min ± 10 sec and 5 min ± 10 sec), 1 mL of the tested mixture was transferred by a pipette to the tube containing 8.0 mL of neutraliser (sodium trisulphate) and was filled with water up to 10 mL. Bactericidal activity of the preparation was

immediately neutralised by a validated method [EN 1040, 1997]. After 5 min \pm 10 sec of neutralisation, two samples of the neutralised mixture, 1 mL each, were transferred onto separate Petri plates and 12–15 mL molten TSA ($45^{\circ}\text{C} \pm 1^{\circ}\text{C}$) was added to each plate. The plates were incubated at $37^{\circ}\text{C} \pm 1^{\circ}\text{C}$ for 48 h. Then, the number of cfu/mL (N_a) was calculated from the formula: $c/(n \times d \times v)$, where c is the sum of colonies counted on both plates, n – number of plates taken into account, d – dilution factor (in this experiment 10^{-1}), v – sample volume (in this experiment 1.0 mL).

For all studied bacteria strains and examined concentration of preparation solution the decrease of bacteria count (in cfu/mL) was calculated from the formula $(N \times 10^{-1})/N_a$. For each bacterial strain neutraliser toxicity control and neutraliser dilution control were prepared [EN 1040, 1997]. The preparation, which in the above defined test conditions caused 10^5 or greater decrease in bacteria count within 5 min of contact or less at 20°C , is characterised as bactericidal [EN 1040, 1997].

RESULTS

P3 Tsunami preparation in the concentration of 0.05% and 0.025%, after 5 min contact time with 10 tested strains of *Citrobacter freundii*, showed strong bactericidal activity (Table 1). There was culture growth or only presence of single colonies on TPA medium ($N_a < 1.5 \times 10^2$ cfu/mL). The

decrease of *C. freundii* count in these concentrations was higher than 1.0×10^5 . Lower concentration of the preparation (0.0125% after 5 min contact time) stopped the growth of *C. freundii* culture, but the decrease in bacteria count was higher than 1.0×10^5 only in half of strains. The preparation in the concentration of 0.00625% showed bactericidal activity towards only 2 strains of *C. freundii*.

Reducing the contact time of *C. freundii* with the preparation to 1 min revealed bactericidal activity of P3 Tsunami in the concentration of 0.05% towards 8 strains, in the concentration of 0.025% to 6 strains and in 0.0125% concentration to 2 strains (Table 2).

The analysis of P3 Tsunami preparation effect on *Hafnia alvei* and *Enterobacter cloacae* strains within 5 min showed bactericidal activity in higher concentrations (0.05%, 0.025%) towards all these bacterial strains (decrease in bacteria count was higher than 1.0×10^5), in the concentration of 0.0125% towards 7 strains of *H. alvei* and 8 strains of *E. cloacae*, and in the concentration of 0.00625% towards 3 strains of *H. alvei* and 5 strains of *E. cloacae* (Tables 3 and 5).

At 1 min contact time the evaluated preparation in the concentration of 0.05% showed bactericidal activity towards all the *E. cloacae* strains and 9 *H. alvei* strains, in the concentration of 0.025% towards 6 *H. alvei* strains and 8 *E. cloacae* strains and in the concentration of 0.0125% towards single strains of the tested bacteria.

P3 Tsunami in the concentration of 0.00625% did not show bactericidal activity (Tables 4 and 6).

TABLE 1. P3 Tsunami activity towards *Citrobacter freundii* after 5 min contact time.

Tested suspension (N)	Bacteria count (cfu/mL) in tested mixture (N_a) concentration				Decrease in bacteria count $(N \times 10^{-1})/N_a$ concentration			
	0.05 vol-%	0.025 vol-%	0.0125 vol-%	0.00625 vol-%	0.05 vol-%	0.025 vol-%	0.0125 vol-%	0.00625 vol-%
1.5×10^8	< 1.5×10^2	< 1.5×10^2	9.4×10^2	$> 3.0 \times 10^3$	$> 1.0 \times 10^5$	$> 1.0 \times 10^5$	1.6×10^4	< 1.0×10^4
1.9×10^8	< 1.5×10^2	< 1.5×10^2	7.5×10^2	$> 3.0 \times 10^3$	$> 1.3 \times 10^5$	$> 1.3 \times 10^5$	2.0×10^4	< 1.0×10^4
1.6×10^8	< 1.5×10^2	< 1.5×10^2	4.2×10^2	$> 3.0 \times 10^3$	$> 1.1 \times 10^5$	$> 1.1 \times 10^5$	3.8×10^4	< 1.0×10^4
1.8×10^8	< 1.5×10^2	< 1.5×10^2	< 1.5×10^2	1.7×10^3	$> 1.2 \times 10^5$	$> 1.2 \times 10^5$	$> 1.2 \times 10^5$	1.1×10^5
2.4×10^8	< 1.5×10^2	< 1.5×10^2	5.6×10^2	$> 3.0 \times 10^3$	$> 1.6 \times 10^5$	$> 1.6 \times 10^5$	4.3×10^4	< 1.0×10^4
1.9×10^8	< 1.5×10^2	< 1.5×10^2	< 1.5×10^2	< 1.5×10^2	$> 1.3 \times 10^5$	$> 1.3 \times 10^5$	$> 1.3 \times 10^5$	$> 1.3 \times 10^5$
2.5×10^8	< 1.5×10^2	< 1.5×10^2	< 1.5×10^2	$> 3.0 \times 10^3$	$> 1.7 \times 10^5$	$> 1.7 \times 10^5$	$> 1.7 \times 10^5$	< 1.0×10^4
1.9×10^8	< 1.5×10^2	< 1.5×10^2	< 1.5×10^2	2.8×10^2	$> 1.3 \times 10^5$	$> 1.3 \times 10^5$	$> 1.3 \times 10^5$	6.8×10^4
2.5×10^8	< 1.5×10^2	< 1.5×10^2	2.6×10^2	$> 3.0 \times 10^3$	$> 1.7 \times 10^5$	$> 1.7 \times 10^5$	9.6×10^4	< 1.0×10^4
2.5×10^8	< 1.5×10^2	< 1.5×10^2	< 1.5×10^2	2.3×10^3	$> 1.7 \times 10^5$	$> 1.7 \times 10^5$	1.1×10^4	

TABLE 2. P3 Tsunami activity towards *Citrobacter freundii* after 1 min contact time.

Tested suspension (N)	Bacteria count (cfu/mL) in tested mixture (N_a) concentration				Decrease in bacteria count $(N \times 10^{-1})/N_a$ concentration			
	0.05 vol-%	0.025 vol-%	0.0125 vol-%	0.00625 vol-%	0.05 vol-%	0.025 vol-%	0.0125 vol-%	0.00625 vol-%
1.5×10^8	4.1×10^2	1.3×10^3	$> 3.0 \times 10^3$	$> 3.0 \times 10^3$	3.7×10^4	1.2×10^4	< 1.0×10^4	< 1.0×10^4
1.9×10^8	< 1.5×10^2	2.1×10^2	5.5×10^2	$> 3.0 \times 10^3$	$> 1.3 \times 10^5$	9.0×10^4	3.5×10^4	< 1.0×10^4
1.6×10^8	< 1.5×10^2	< 1.5×10^2	$> 3.0 \times 10^3$	$> 3.0 \times 10^3$	$> 1.1 \times 10^5$	$> 1.1 \times 10^5$	< 1.0×10^4	< 1.0×10^4
1.8×10^8	< 1.5×10^2	< 1.5×10^2	2.0×10^3	$> 3.0 \times 10^3$	$> 1.2 \times 10^5$	$> 1.2 \times 10^5$	9.0×10^4	< 1.0×10^4
2.4×10^8	< 1.5×10^2	2.3×10^2	$> 3.0 \times 10^3$	$> 3.0 \times 10^3$	$> 1.6 \times 10^5$	1.0×10^5	< 1.0×10^4	< 1.0×10^4
1.9×10^8	< 1.5×10^2	< 1.5×10^2	1.0×10^3	$> 3.0 \times 10^3$	$> 1.3 \times 10^5$	$> 1.3 \times 10^5$	1.9×10^5	< 1.0×10^4
2.5×10^8	< 1.5×10^2	< 1.5×10^2	$> 3.0 \times 10^3$	$> 3.0 \times 10^3$	$> 1.7 \times 10^5$	$> 1.7 \times 10^5$	< 1.0×10^4	< 1.0×10^4
1.9×10^8	3.2×10^2	5.4×10^2	1.4×10^3	$> 3.0 \times 10^3$	5.9×10^4	3.5×10^4	1.4×10^4	< 1.0×10^4
2.5×10^8	< 1.5×10^2	4.3×10^2	$> 3.0 \times 10^3$	$> 3.0 \times 10^3$	$> 1.7 \times 10^5$	4.4×10^4	< 1.0×10^4	< 1.0×10^4
2.5×10^8	< 1.5×10^2	< 1.5×10^2	2.2×10^3	$> 3.0 \times 10^3$	$> 1.7 \times 10^5$	$> 1.7 \times 10^5$	1.1×10^4	< 1.0×10^4

TABLE 3. P3 Tsunami activity towards *Enterobacter cloacae* after 5 min contact time.

Tested suspension (N)	Bacteria count (cfu/mL) in tested mixture (N _a) concentration				Decrease in bacteria count (N × 10 ⁻¹)/N _a concentration			
	0.05 vol-%	0.025 vol-%	0.0125 vol-%	0.00625 vol-%	0.05 vol-%	0.025 vol-%	0.0125 vol-%	0.00625 vol-%
3.7 × 10 ⁸	< 1.5 × 10 ²	< 1.5 × 10 ²	1.4 × 10 ³	> 3.0 × 10 ³	> 2.5 × 10 ⁵	> 2.5 × 10 ⁵	2.6 × 10 ⁴	< 1.0 × 10 ⁴
4.0 × 10 ⁸	< 1.5 × 10 ²	< 1.5 × 10 ²	< 1.5 × 10 ²	< 1.5 × 10 ²	> 2.7 × 10 ⁵	> 2.7 × 10 ⁵	> 2.7 × 10 ⁵	> 2.7 × 10 ⁵
4.9 × 10 ⁸	< 1.5 × 10 ²	< 1.5 × 10 ²	< 1.5 × 10 ²	1.0 × 10 ³	> 3.3 × 10 ⁵	> 3.3 × 10 ⁵	> 3.3 × 10 ⁵	4.9 × 10 ⁴
3.5 × 10 ⁸	< 1.5 × 10 ²	< 1.5 × 10 ²	< 1.5 × 10 ²	< 1.5 × 10 ²	> 2.3 × 10 ⁵	> 2.3 × 10 ⁵	> 2.3 × 10 ⁵	> 2.3 × 10 ⁵
1.9 × 10 ⁸	< 1.5 × 10 ²	< 1.5 × 10 ²	< 1.5 × 10 ²	< 1.5 × 10 ²	> 1.3 × 10 ⁵	> 1.3 × 10 ⁵	> 1.3 × 10 ⁵	> 1.3 × 10 ⁵
2.5 × 10 ⁸	< 1.5 × 10 ²	< 1.5 × 10 ²	< 1.5 × 10 ²	< 1.5 × 10 ²	> 1.7 × 10 ⁵	> 1.7 × 10 ⁵	> 1.7 × 10 ⁵	> 1.7 × 10 ⁵
2.2 × 10 ⁸	< 1.5 × 10 ²	< 1.5 × 10 ²	< 1.5 × 10 ²	< 1.5 × 10 ²	> 1.5 × 10 ⁵	> 1.5 × 10 ⁵	> 1.5 × 10 ⁵	> 1.5 × 10 ⁵
3.0 × 10 ⁸	< 1.5 × 10 ²	< 1.5 × 10 ²	< 1.5 × 10 ²	8.1 × 10 ³	> 2.0 × 10 ⁵	> 2.0 × 10 ⁵	> 2.0 × 10 ⁵	< 1.0 × 10 ⁴
2.9 × 10 ⁸	< 1.5 × 10 ²	< 1.5 × 10 ²	6.3 × 10 ²	> 3.0 × 10 ³	> 1.9 × 10 ⁵	> 1.9 × 10 ⁵	4.6 × 10 ⁴	< 1.0 × 10 ⁴
2.6 × 10 ⁸	< 1.5 × 10 ²	< 1.5 × 10 ²	< 1.5 × 10 ²	1.5 × 10 ³	> 1.7 × 10 ⁵	> 1.7 × 10 ⁵	> 1.7 × 10 ⁵	1.7 × 10 ⁴

TABLE 4. P3 Tsunami activity towards *Enterobacter cloacae* after 1 min contact time.

Tested suspension (N)	Bacteria count (cfu/mL) in tested mixture (N _a) concentration				Decrease in bacteria count (N × 10 ⁻¹)/N _a concentration			
	0.05 vol-%	0.025 vol-%	0.0125 vol-%	0.00625 vol-%	0.05 vol-%	0.025 vol-%	0.0125 vol-%	0.00625 vol-%
3.7 × 10 ⁸	< 1.5 × 10 ²	> 3.0 × 10 ³	> 3.0 × 10 ³	> 3.0 × 10 ³	> 2.5 × 10 ⁵	< 1.2 × 10 ⁴	< 1.2 × 10 ⁴	< 1.2 × 10 ⁴
4.0 × 10 ⁸	< 1.5 × 10 ²	< 1.5 × 10 ²	7.1 × 10 ²	> 3.0 × 10 ³	> 2.7 × 10 ⁵	> 2.7 × 10 ⁵	5.6 × 10 ⁴	< 1.3 × 10 ⁴
4.9 × 10 ⁸	< 1.5 × 10 ²	< 1.5 × 10 ²	3.1 × 10 ²	> 3.0 × 10 ³	> 3.3 × 10 ⁵	> 3.3 × 10 ⁵	1.6 × 10 ⁵	< 1.6 × 10 ⁴
3.5 × 10 ⁸	< 1.5 × 10 ²	< 1.5 × 10 ²	1.3 × 10 ³	> 3.0 × 10 ³	> 2.3 × 10 ⁵	> 2.3 × 10 ⁵	2.7 × 10 ⁴	< 1.2 × 10 ⁴
1.9 × 10 ⁸	< 1.5 × 10 ²	3.4 × 10 ²	> 3.0 × 10 ³	> 3.0 × 10 ³	> 1.3 × 10 ⁵	5.6 × 10 ⁴	< 1.0 × 10 ⁴	< 1.0 × 10 ⁴
2.5 × 10 ⁸	< 1.5 × 10 ²	< 1.5 × 10 ²	1.7 × 10 ³	> 3.0 × 10 ³	> 1.7 × 10 ⁵	> 1.7 × 10 ⁵	8.8 × 10 ⁴	< 1.0 × 10 ⁴
2.2 × 10 ⁸	< 1.5 × 10 ²	< 1.5 × 10 ²	> 3.0 × 10 ³	> 3.0 × 10 ³	> 1.5 × 10 ⁵	> 1.5 × 10 ⁵	< 1.0 × 10 ⁴	< 1.0 × 10 ⁴
3.0 × 10 ⁸	< 1.5 × 10 ²	< 1.5 × 10 ²	9.4 × 10 ²	> 3.0 × 10 ³	> 2.0 × 10 ⁵	> 2.0 × 10 ⁵	3.2 × 10 ⁴	< 1.0 × 10 ⁴
2.9 × 10 ⁸	< 1.5 × 10 ²	< 1.5 × 10 ²	1.0 × 10 ³	> 3.0 × 10 ³	> 1.9 × 10 ⁵	> 1.9 × 10 ⁵	2.9 × 10 ⁴	< 1.0 × 10 ⁴
2.6 × 10 ⁸	< 1.5 × 10 ²	< 1.5 × 10 ²	< 1.5 × 10 ²	> 3.0 × 10 ³	> 1.7 × 10 ⁵	> 1.7 × 10 ⁵	> 1.7 × 10 ⁵	< 1.0 × 10 ⁴

TABLE 5. P3 Tsunami activity towards *Hafnia alvei* after 5 min contact time.

Tested suspension (N)	Bacteria count (cfu/mL) in tested mixture (N _a) concentration				Decrease in bacteria count (N × 10 ⁻¹)/N _a concentration			
	0.05 vol-%	0.025 vol-%	0.0125 vol-%	0.00625 vol-%	0.05 vol-%	0.025 vol-%	0.0125 vol-%	0.00625 vol-%
1.8 × 10 ⁸	< 1.5 × 10 ²	< 1.5 × 10 ²	< 1.5 × 10 ²	2.7 × 10 ²	> 1.2 × 10 ⁵	> 1.2 × 10 ⁵	> 1.2 × 10 ⁵	6.7 × 10 ⁴
2.4 × 10 ⁸	< 1.5 × 10 ²	< 1.5 × 10 ²	6.4 × 10 ²	1.6 × 10 ³	> 1.6 × 10 ⁵	> 1.6 × 10 ⁵	3.8 × 10 ⁴	1.5 × 10 ⁴
2.3 × 10 ⁸	< 1.5 × 10 ²	< 1.5 × 10 ²	< 1.5 × 10 ²	5.0 × 10 ²	> 1.5 × 10 ⁵	> 1.5 × 10 ⁵	> 1.5 × 10 ⁵	4.6 × 10 ⁴
3.0 × 10 ⁸	< 1.5 × 10 ²	< 1.5 × 10 ²	< 1.5 × 10 ²	< 1.5 × 10 ²	> 2.0 × 10 ⁵	> 2.0 × 10 ⁵	> 2.0 × 10 ⁵	> 2.0 × 10 ⁵
2.8 × 10 ⁸	< 1.5 × 10 ²	< 1.5 × 10 ²	< 1.5 × 10 ²	> 3.0 × 10 ³	> 1.9 × 10 ⁵	> 1.9 × 10 ⁵	> 1.9 × 10 ⁵	< 1.0 × 10 ⁴
2.6 × 10 ⁸	< 1.5 × 10 ²	< 1.5 × 10 ²	< 1.5 × 10 ²	2.7 × 10 ³	> 1.7 × 10 ⁵	> 1.7 × 10 ⁵	> 1.7 × 10 ⁵	9.6 × 10 ³
2.4 × 10 ⁸	< 1.5 × 10 ²	< 1.5 × 10 ²	< 1.5 × 10 ²	< 1.5 × 10 ²	> 1.6 × 10 ⁵	> 1.6 × 10 ⁵	> 1.6 × 10 ⁵	> 1.6 × 10 ⁵
2.5 × 10 ⁸	< 1.5 × 10 ²	< 1.5 × 10 ²	< 1.5 × 10 ²	< 1.5 × 10 ²	> 1.7 × 10 ⁵	> 1.7 × 10 ⁵	> 1.7 × 10 ⁵	> 1.7 × 10 ⁵
1.9 × 10 ⁸	< 1.5 × 10 ²	< 1.5 × 10 ²	9.8 × 10 ²	> 3.0 × 10 ³	> 1.3 × 10 ⁵	> 1.3 × 10 ⁵	1.9 × 10 ⁴	< 1.0 × 10 ⁴
1.7 × 10 ⁸	< 1.5 × 10 ²	< 1.5 × 10 ²	1.5 × 10 ³	2.9 × 10 ³	> 1.1 × 10 ⁵	> 1.1 × 10 ⁵	1.1 × 10 ⁴	5.9 × 10 ³

DISCUSSION

High degree of contamination with microorganisms (10²–10⁸ cfu/g) [Müller, 1997; Nguyen-the & Carlin, 1994] of fresh fruit and vegetables delivered to a packing house has been attributed to a variety of factors, e.g. contact with soil during growing, contamination during manual harvesting or using combines, as well as transport and storage until processing [Gruda & Postolski, 1999]. Washing treatment, which should significantly decrease the number of microorganisms, is often ineffective. The reason could be the technological process itself, where washing is continued for many hours in the same water. The number of microorganisms

isolated from French beans after washing showed no significant difference to that isolated from the surface of beans received from the transporter before washing [Białasiewicz & Królasik, 1999]. Researches have investigated a variety of chemical preparations added to washing water to decrease the number of bacteria on the surface of fruit and vegetables. Beuchat *et al.* [1998] used hydrogen peroxide and trisodium phosphate separately and in various combinations of concentrations. Raina *et al.* [1995] found, that chlorine dioxide is effective in reducing the number of bacteria in decreasing cucumber hydrocooling wash water, but it did not effectively aid in reducing the number of microorganisms on/in the cucumbers themselves. Similar

TABLE 6. P3 Tsunami activity towards *Hafnia alvei* after 1 min contact time.

Tested suspension (N)	Bacteria count (cfu/mL) in tested mixture (N _a) concentration				Decrease in bacteria count (N x 10 ⁻¹)/N _a concentration			
	0.05 vol-%	0.025 vol-%	0.0125 vol-%	0.00625 vol-%	0.05 vol-%	0.025 vol-%	0.0125 vol-%	0.00625 vol-%
1.3 x 10 ⁸	1.8 x 10 ³	> 3.0 x 10 ³	> 3.0 x 10 ³	> 3.0 x 10 ³	1.3 x 10 ⁴	< 1.0 x 10 ⁴	< 1.0 x 10 ⁴	< 1.0 x 10 ⁴
1.9 x 10 ⁸	< 1.5 x 10 ²	< 1.5 x 10 ²	> 3.0 x 10 ³	> 3.0 x 10 ³	> 1.3 x 10 ⁵	> 1.3 x 10 ⁵	< 1.0 x 10 ⁴	< 1.0 x 10 ⁴
2.3 x 10 ⁸	< 1.5 x 10 ²	2.8 x 10 ²	> 3.0 x 10 ³	> 3.0 x 10 ³	> 1.5 x 10 ⁵	8.2 x 10 ⁴	< 1.0 x 10 ⁴	< 1.0 x 10 ⁴
3.0 x 10 ⁸	< 1.5 x 10 ²	< 1.5 x 10 ²	5.2 x 10 ²	> 3.0 x 10 ³	> 2.0 x 10 ⁵	> 2.0 x 10 ⁵	5.8 x 10 ⁴	< 1.0 x 10 ⁴
2.8 x 10 ⁸	< 1.5 x 10 ²	< 1.5 x 10 ²	> 3.0 x 10 ³	> 3.0 x 10 ³	> 1.3 x 10 ⁵	> 1.3 x 10 ⁵	< 1.0 x 10 ⁴	< 1.0 x 10 ⁴
2.6 x 10 ⁸	< 1.5 x 10 ²	2.3 x 10 ²	4.7 x 10 ²	> 3.0 x 10 ³	> 1.7 x 10 ⁵	> 1.7 x 10 ⁵	1.1 x 10 ⁵	5.5 x 10 ⁴
2.4 x 10 ⁸	< 1.5 x 10 ²	< 1.5 x 10 ²	> 3.0 x 10 ³	> 3.0 x 10 ³	> 1.6 x 10 ⁵	> 1.6 x 10 ⁵	< 1.0 x 10 ⁴	< 1.0 x 10 ⁴
2.5 x 10 ⁸	< 1.5 x 10 ²	1.4 x 10 ³	> 3.0 x 10 ³	> 3.0 x 10 ³	> 1.7 x 10 ⁵	1.8 x 10 ⁴	< 1.0 x 10 ⁴	< 1.0 x 10 ⁴
1.9 x 10 ⁸	< 1.5 x 10 ²	1.3 x 10 ³	> 3.0 x 10 ³	> 3.0 x 10 ³	> 1.3 x 10 ⁵	1.5 x 10 ⁴	< 1.0 x 10 ⁴	< 1.0 x 10 ⁴
1.7 x 10 ⁸	< 1.5 x 10 ²	< 1.5 x 10 ²	> 3.0 x 10 ³	> 3.0 x 10 ³	> 1.1 x 10 ⁵	> 1.1 x 10 ⁵	< 1.0 x 10 ⁴	< 1.0 x 10 ⁴

results were achieved by Brakett [1994] and Senter *et al.* [1985] who tested the influence of chlorine compounds added to the water for washing fruit and vegetables. Brown and Schubert [1997] used sodium orthophenylophenate for disinfection of water used for washing citrus fruit and found it effective against *Xanthomonas campestris*.

It had been notified previously (data not published) that P3 Tsunami preparation showed bactericidal activity towards *Staphylococcus aureus*, *Listeria monocytogenes*, *Enterococcus hirae*, *Pseudomonas aeruginosa*, and *Salmonella typhimurium* strains. Therefore, it could be expected that adding this preparation to technological water would significantly decrease the number of saprophytic and pathogenic bacteria contaminating fruit and vegetables processed in fruit-vegetable plants.

The main component of the tested preparation is peracetic acid, easily and quickly decomposing to acetic acid, which in detected concentration causes no harm to the consumer and no unfavourable changes in organoleptic features in fruit and vegetables. The important advantage of P3 Tsunami is its rapid bactericidal effect in contact time of 1–5 min, *i.e.* within the period of fruit and vegetables washing.

CONCLUSIONS

1. P3 Tsunami in the concentration of 0.05% and 0.025% after 5 min contact time showed bactericidal activity towards all 30 studied bacterial strains from the species: *Hafnia alvei*, *Citrobacter freundii* and *Enterobacter cloacae* from *Enterobacteriaceae* family.

2. The tested preparation in the concentration of 0.05% after 1 min contact time showed bactericidal activity towards 27 bacterial strains from the above-mentioned species.

3. Bactericidal activity of P3 Tsunami in a short contact time (1 and 5 min) suggested that it may be effective in disinfecting technological water during the process of freezing fruit and vegetables.

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OCENA DZIAŁANIA BAKTERIOBÓJCZEGO NOWEGO PREPARATU P3 TSUNAMI PRZEZNACZONEGO DO DEZYNFEKCJI ZAMRAŻANYCH OWOCÓW I WARZYW

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Celem pracy było zbadanie, czy nowy preparat chemiczny P3 Tsunami firmy Ecolab wykazuje działanie bakteriobójcze w stosunku do szczeprów z gatunku *Hafnia alvei*, *Citrobacter freundii*, *Enterobacter cloacae*. Preparat ten został zaproponowany jako środek dezynfekujący do obniżenia poziomu zanieczyszczeń mikrobiologicznych w wodzie technologicznej używanej do mycia owoców i warzyw przeznaczonych do mrożenia.

Testową zawiesinę bakterii ($1,5 \times 10^8$ cfu/mL – $5,0 \times 10^8$ cfu/mL) dodawano do przygotowanej w czterech różnych stężeniach próbki badanego preparatu. Po określonym czasie kontaktu (1 min, 5 min \pm 10 s) neutralizowano bakteriobójcze działanie preparatu zgodnie ze zwalidowaną metodą. Po 5 min \pm 10 s, 1mL neutralizowanej mieszaniny przenoszono do płytka Petriego i dodawano 12–15 mL upłychnionej pożywki TSA. Po 48 godz. inkubacji w $37 \pm 1^\circ\text{C}$ w każdej próbce liczono kolonie i ustalano liczbę bakterii, które przeżyły. Na tej podstawie ustalano stopień zmniejszenia liczby bakterii, które przeżyły. Preparat, który w warunkach badania określonych normą EN 1040: 1997 powodował 10^5 -krotny lub większy spadek liczby bakterii określano jako bakteriobójczy.

Preparat P3 w stężeniu 0,05% i 0,025% po 5 minutach kontaktu wykazywał działanie bakteriobójcze w stosunku do wszystkich 30 szczeprów bakterii użytych w doświadczeniu (tab.1, tab. 3, tab. 5); w stężeniu 0,05% po 1 minucie kontaktu powodował 10^5 -krotny spadek liczby 27 szczeprów bakterii (tab. 2, tab. 4, tab. 6).

Stwierdzone bakteriobójcze działanie preparatu P3 już po krótkim czasie kontaktu (1 min) pozwala przypuszczać, że spełni on zadanie dezynfekcji wody technologicznej w procesie zamrażania owoców i warzyw.