



**DETAILED FINAL REPORT**

January 19, 2021

**SHELF-LIFE STUDY**

**Vac Skin Oysters**

Report Number: 20-464687

**PREPARED BY**

Melanie Reid  
Operations Manager  
Food Safety Net Services, Ltd.

**STUDY SPONSOR**

**Southeastern Sea Products**

**Client Code: SOUSEA**

## **1.0 STUDY DATES**

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Study Initiation Date: 11/12/2020

Study Completion Date: 11/18/2020

## **2.0 STUDY SUMMARY**

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Samples of Vac Skin Oysters were held under refrigerated temperature storage of 2 - 5°C for 7 Days and were evaluated on Days 1, 2, 3, 4, 5, 6, and 7 Days. Microbial assessment included Aerobic Plate Count, *E. coli* and Coliform Count, Lactic Acid Bacteria Count, and Psychrotrophic Plate Count. Additionally, the samples will be tested for *Clostridium botulinum* and *Listeria spp.* and the Day 1 and Day 7 set points only. Organoleptic assessment was performed as well.

## **3.0 MATERIALS AND METHODS**

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### **3.1 Test Samples and Sampling Method**

Samples of product were removed from storage and submitted for analysis on each day of testing. Testing procedures are summarized in Sections 3.2 – 3.8.

### **3.2 Aerobic Plate Count Testing**

Samples were tested for aerobic microorganisms at the time points outlined above by plating onto 3M™ Petrifilm™ Aerobic Count (AC) Plate. The AC plates were incubated under aerobic conditions for 48 ± 3 h at 35 ± 2°C.

### **3.3 Coliform and *E. coli* Count Testing**

Samples were tested for Coliforms and *E. coli* at the time points outlined above by plating onto 3M™ Petrifilm™ *E. coli* and Coliform Count (EC/CC) Plate. The EC/CC plates were incubated under aerobic conditions for 48 ± 2 h at 35 ± 2°C.

### **3.4 Lactic Acid Bacteria Testing**

Samples were tested for lactic acid bacteria at the time points outlined above by plating onto 3M™ Petrifilm™ Lactic Acid Bacteria (LA) Plates. The LA plates were incubated under aerobic conditions for 48 ± 3 h at 28 - 37 ± 1°C.

### **3.5 Psychrotrophic Plate Count Testing**

Samples were tested for Psychrotrophic bacteria at the time points outlined above by plating onto 3M™ Petrifilm™ Aerobic Count (AC) Plate. The AC plates were incubated under aerobic conditions for 10 d at 7 ± 1°C.

### **3.6 *Listeria spp.* Testing**

Samples were tested for *Listeria spp.* bacteria at the time points outlined above by Enzyme Linked Fluorescent Assay (ELFA). The samples were incubated in a primary

enrichment for 20 – 26 h at  $30 \pm 1^\circ\text{C}$ . After incubation the samples were screened on the VIDAS<sup>®</sup> instrument.

### **3.7 *Clostridium botulinum* Testing**

Samples were tested for *Clostridium botulinum* bacteria at the time points outlined above using the FDA method. The samples were cultured using three different primary enrichment mediums stored under anaerobic conditions for 5 days at  $28 \pm 1^\circ\text{C}$  and  $35 \pm 1^\circ\text{C}$ .

### **3.8 Organoleptic Analysis**

At the time points outlined above, the samples were evaluated for organoleptic characteristics such as aroma, texture, appearance, and color. Emphasis was placed on the quality of the product and the determination of the shelf life of the product was based on these results. Organoleptic analysis will reported on a hedonic scale of 1-5; where 1 is unacceptable, 2 is below satisfactory, 3 is satisfactory, 4 is above satisfactory and 5 is good. Organoleptic analysis will be conducted by a panel of three laboratory technicians and will take place before the samples are subjected to microbiological analysis.

### **3.9 Data Analysis**

The results obtained at each time point were analyzed for an overall assessment of shelf life based on the changes in the levels of the microbiological indicators that were observed over the course of testing. For microbiological data, the “index of spoilage” established by Fung et al. 1980 (1), which consists of an aerobic plate count of  $\geq 7.0 \log_{10}$  CFU/g, was used as the “index of spoilage” for this study. All results were analyzed and included in the report, as well as any shelf life conclusions that were drawn from the observed data.

#### 4.0 RESULTS

Log<sub>10</sub> CFU/g transformed Microbiological and Organoleptic data for samples are shown in Tables 1 - 2.

**Table 1: Microbiological (Log<sub>10</sub> CFU/g) Shelf Life Results for Vac Skin Oysters**

Test Date	Aerobic Plate Count (CFU/g)	Coliform Plate Count (CFU/g)	<i>E. coli</i> Count (CFU/g)	Lactic Acid Bacteria Count (CFU/g)	Psychrotrophic Plate Count (CFU/g)	<i>Listeria spp.</i> (VIDAS)	<i>Clostridium botulinum</i> (FDA-BAM)
Day 1	2.2	< 1.0	< 1.0	< 1.0	2.2	Negative	Negative
Day 2	2.1	< 1.0	< 1.0	< 1.0	2.6		
Day 3	2.7	< 1.0	< 1.0	< 1.0	3.3		
Day 4	2.1	1.0	1.0	< 1.0	3.8		
Day 5	2.0	< 1.0	< 1.0	1.0	4.2		
Day 6	2.4	1.7	1.7	1.0	5.0		
Day 7	2.3	1.3	< 1.0	< 1.0	5.0	Negative	Negative

**Table 2: Organoleptic Shelf Life Results for Vac Skin Oysters**

Test Date	ORGANOLEPTIC				
	Appearance	Color	Odor	Texture	Packaging Integrity
Day 1	5	5	5	5	5
Day 2	4	4	3	3	5
Day 3	5	5	5	5	5
Day 4	5	5	5	5	5
Day 5	4	5	3	4	5
Day 6	3	3	2	3	5
Day 7	4	4	3	4	5

#### 5.0 DISCUSSION

The “index of spoilage” of 7.0 log<sub>10</sub> CFU/g aerobic plate count established by Fung et al. 1980 is the microbiological indicator level at which signs of spoilage (such as odor and slime development) will begin to be observed. This index was used to determine the predicted shelf life of the products.

Microbiological data obtained for the Vac Skin Oysters indicated that the shelf life of this product, when stored under refrigerated temperature storage (2 - 5°C), is likely 7 Days of

product age, as  $\geq 7.0 \log_{10}$  CFU/g aerobic plate count was not achieved at 7 Days of product age and all other counts were acceptable. Organoleptic data showed that the product was acceptable through 5 Days of product age.

Upon review of the data, one may notice that there are intermittent increases in the values for certain microbiological parameters that are then followed by a decrease in value upon the next consecutive testing day. This is due in part to the inherent heterogeneity of the samples where bacterial populations are not distributed evenly throughout the mass of the sample. Thus, it is not unlikely to observe these sorts of occurrences throughout the shelf life of a sample, and FSNS has, in its best knowledge, based the shelf life values for each product on the data that are available, taking into account these intermittent increases in values.

## 6.0 REFERENCES

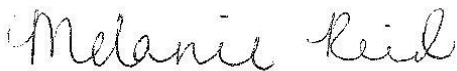
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1. **Fung D. Y. C., J. R. Edwards, and B. A. Crozier-Dodson** (2008). At-line methods for controlling microbial growth and spoilage in meat processing abattoirs. In F. Toldra (ed) Meat Biotechnology. Retrieved From: [https://books.google.com/books?id=XXQqIOIZoHcC&printsec=frontcover&source=gbs\\_ge\\_summary\\_r&cad=0#v=onepage&q=fung&f=false](https://books.google.com/books?id=XXQqIOIZoHcC&printsec=frontcover&source=gbs_ge_summary_r&cad=0#v=onepage&q=fung&f=false)

## 7.0 FINAL REPORT APPROVAL

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**Prepared By:**



01/19/2021

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Melanie Reid  
Operations Manager  
Food Safety Net Services, Ltd.

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Date

**Approved By:**

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Kathryn Leedy  
Southeastern Sea Products

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Date