

Fred Nolte,^{1*} D. Glenn Black,²
John DeBeer³ and Elena Enache²

¹Clover Leaf Seafoods, 13071 Vanier Place, Unit 230
Richmond, BC, Canada V6V 2J1

²Grocery Manufacturers Association, 1350 I St. NW,
Suite 300,
Washington, D.C. 20005, USA³; Chicken of the Sea
International, Suite 500, 9330 Scranton Road, San
Diego, CA 92121, USA



*Tuna End Point Internal Product
Temperatures (EPIPT) are measured
upon discharge from the pre-cooker.*

Use of End Point Internal Product Temperature to Control Histamine Formation in Tuna at Pre-cooking Step

ABSTRACT

A linear heating and cooling model was developed to determine a critical limit for precooking End Point Internal Product Temperature (EPIPT) in order to achieve a 5-log reduction of the most prolific histamine-forming bacterium, (HFB) *Morganella morganii*. The thermal death time values used in a General Method calculation employed the Trapezoidal rule, where $D_{60^{\circ}\text{C}}$ of 0.26m and $z = 4.1^{\circ}\text{C}$. Based on the thermal death time values and the General Method calculations, a 60°C pre-cooking EPIPT for tuna of any size will stop histamine formation during and after pre-cooking, until the de-skinning step is reached, thus providing the canned tuna industry with significant extra time to process tuna, especially with the larger sizes. It is also possible to use process lethality as a tool to evaluate a deviation from the process schedule, where a 5-log reduction of HFB can be demonstrated for a pre-cook batch. This information should be of interest to the tuna processing industry and regulatory officials interested in controlling histamine production in these products.

INTRODUCTION

Histamine is the primary causative agent for scombroid fish poisoning (SFP) (11). In fish species containing high levels of the free or unbound amino acid histidine, including tunas, histamine is formed by decarboxylation of the free histidine. The decarboxylation is catalyzed by the enzyme histidine decarboxylase (HDC) produced during the growth of histamine-forming bacteria HFB (2, 15, 23). The most significant HFB, in relation to SFP, are *Morganella morganii*, *Raoultella planticola*, *Hafnia alvei*, *Enterobacter aerogenes*, and *Photobacterium damsela* (4, 7, 13, 14). Histamine formation takes place primarily at temperatures above 25°C (2, 11). For processes in which any portion of the exposure is above 21°C , the U.S. Food and Drug Administration (FDA) has set a maximum time limit of 12 hours for previously frozen fish and 4 hours for fish that have not been previously frozen (26).

Tuna for the canned trade are almost exclusively processed from raw material frozen at sea. During processing, tuna is thawed, butchered, partially cooked (or “pre-cooked”), cooled, cleaned and canned. The cooking and cooling processes facilitate cleaning (the separation of skin, bones,

* Author for correspondence: Phone: +1 604.838.4961; E-mail: nolte@shaw.ca

and dark meat from the desirable white or light meat) (16). The separated white or light meat is either canned directly or vacuum-packed into plastic “loin bags” and frozen for subsequent thawing and canning (6, 21). The total time from the start of thawing the whole tuna to when the finished product (either cans or loins) reaches inhibitory temperatures in retorting or freezing frequently exceeds 12 hours, thereby exceeding FDA regulatory time limits.

Tuna processors have always believed that the pre-cook step provides a “histamine pause,” that halts histamine formation, allowing sufficient time to complete subsequent process steps without risk of histamine formation (19), and for this reason, this industry belief was the focus of major validation effort during 2011 and 2012 (17). Thermal Death Time studies established that *M. morgani* was the most heat resistant bacterium of the HFB (7, 8). Industry validation studies showed that pre-cook processes that achieve slightly less than a 5-log reduction of *M. morgani* gain an additional 12 hours in available holding time at temperatures above 21°C without a measurable increase in histamine (17). Therefore, a 5-log reduction of *M. morgani* would be more than sufficient to achieve a histamine pause sufficient to permit conversion of tuna into canned product. A 4-log reduction is currently in use by the Almond Board of California to control *Salmonella* in almonds (1), and a 5-log pathogen reduction is used as a regulatory reduction criterion in the U.S. FDA’s HACCP regulation for juice found at Title 21 Code of Federal Regulations, Part 120. (27). A 5-log reduction of HFB in the tuna pre-cooking process is conservative, because histamine pause has been validated

with less than a 5-log reduction (17). Along these lines, Fletcher et al. concluded that hot-smoking was sufficient to eliminate histamine-forming bacteria and published processing guidelines (8).

Real time monitoring of *M. morgani* lethality has been attempted but remains impractical (21). EPIPT monitoring (backbone temperatures) has been the standard practice for decades and remains so (19, 22).

Before processing begins, tuna must be sorted into size categories so that cooking is uniform for the same sized fish and to ensure that none of the fish are under-cooked or over-cooked. As a naturally occurring material, tuna are variable in size; a catch of tuna will normally contain 2 or more year classes, resulting in a polymodal size distribution (10, 21). Within each mode or year class, individual tuna will also vary in size. Finally, tuna grow indeterminately, so that, in living tuna, these size categories are continually changing. As a consequence, cook schedules based on standard size categories are only approximations, and the resultant times needed to achieve the desired cook are only approximations. The cold spot temperature of the fish continues to rise for some time after the end of the heating phase, and cook schedules are designed with this in mind (6, 21). This aspect is a natural phenomenon when cooking any portion of meat or fish where the external temperature is much higher than the target internal temperature (12).

Tuna processors have historically relied on internal product temperature, measured at the coldest spot in the largest fish or fish piece, as the best means to regulate cooking. This site could be the backbone of a whole fish or

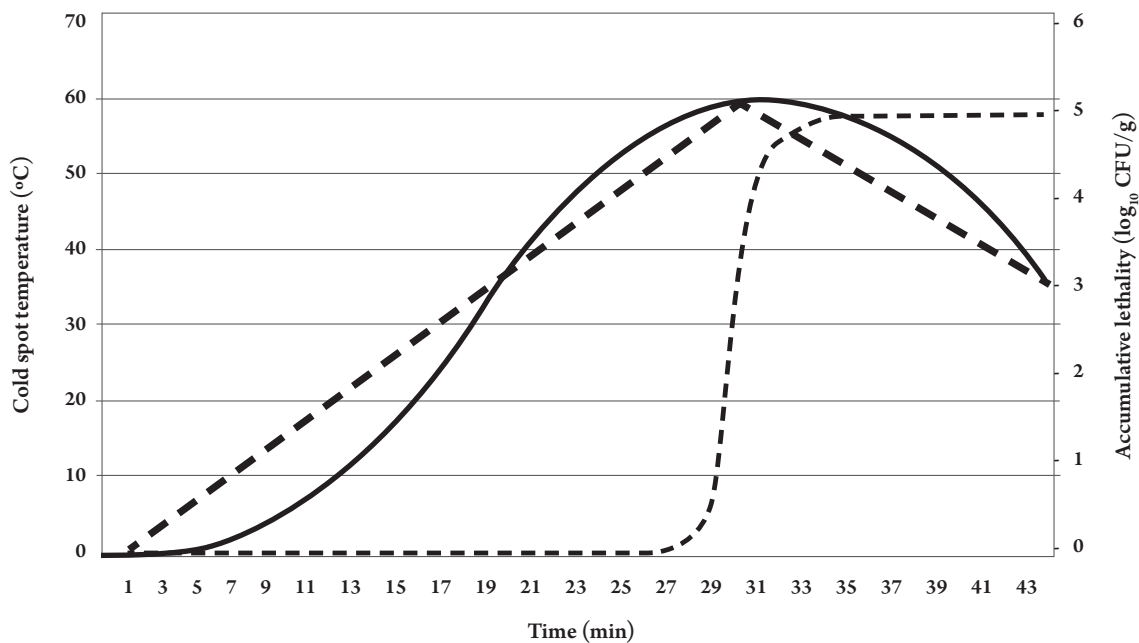


FIGURE 1. Linear heating model used for EPIPT critical limit estimation. Solid line represents sigmoid heating, dashed line linear heating and dotted line cumulative log-reduction.

the geometric center of the largest piece of a split fish (21, 22). Thus, Standard Operating Practices (SOPs) for fish sizing and cook scheduling are essential for reliably achieving the minimum cold spot temperature in all fish or fish pieces in every pre-cook batch (6, 21).

Industry pre-cooking practices have been standardized in the National Fisheries Institute HACCP Guidebook to ensure adequate process procedures (20). Pre-cookers of tuna must deliver an even temperature throughout the pre-cooker vessel, be adequately maintained, be equipped with adequate calibrated instruments, be supplied with sufficient steam to achieve desired results, and be available in sufficient numbers to handle the production volume. Fish must be consistently sorted by size before processing, and the standard industry practice is to sort fish at intervals of 2.5 Kg or less (6). The EPIPT must be measured using adequate sample sizes, methods, and equipment to ensure accuracy (20). If any cook process results in EPIPT falling below the minimum critical limit, corrective actions can and must be

taken to complete the necessary cooking for that batch of fish, as well as a review of procedures to prevent recurrences of any EPIPT below the critical limit on subsequent batches for similar sized fish.

The objectives of this paper are to establish a pre-cook critical limit, based on an EPIPT or peak minimum internal product temperature reached during the pre-cook step that would ensure a 5-log reduction of *M. organii* is reliably achieved and to demonstrate that EPIPT can be effectively used to evaluate and clear a deviation from a pre-cooking process schedule.

MATERIALS AND METHODS

It was necessary to develop a conservative model for the heating and cooling curves at the coldest spot in a fish during the pre-cooking process, based on the worst case scenario. A normal sigmoid curve could not be used in these types of situations since the exact shape of the curve would vary, depending on a number of factors. Instead, heating

TABLE 1. Lethality calculations for *M. organii* in tuna fish

| Time (min) | Temperature (°C) | Log-reduction/min | Cumulative lethality |
|------------|------------------|-------------------|----------------------|
| 0 | -1.1 | 0 | 0 |
| 1 | 2.9 | 0 | 0 |
| 3 | 6.9 | 0 | 0 |
| 5 | 10.9 | 0 | 0 |
| 7 | 14.9 | 0 | 0 |
| 9 | 18.9 | 0 | 0 |
| 11 | 22.9 | 0 | 0 |
| 13 | 26.9 | 0 | 0 |
| 15 | 30.9 | 0 | 0 |
| 17 | 34.9 | 0 | 0 |
| 19 | 38.9 | 0 | 0 |
| 21 | 42.9 | 0 | 0 |
| 23 | 46.9 | 0.0030 | 0.005 |
| 25 | 50.9 | 0.0287 | 0.043 |
| 27 | 54.9 | 0.2716 | 0.402 |
| 29 | 58.9 | 2.5674 | 3.805 |
| 31 | 54.9 | 0.2716 | 4.911 |
| 34 | 48.9 | 0.0093 | 5.038 |
| 36 | 44.9 | 0.0010 | 5.042 |
| 38 | 40.9 | 0 | 5.042 |
| 40 | 36.9 | 0 | 5.042 |
| 42 | 32.9 | 0 | 5.042 |

and cooling were assumed to be linear in order to achieve the most conservative model (Fig. 1). To determine the slope of the heating and cooling model, the fastest average heating and cooling rate observed in industry was employed, namely 2°C per minute (6). Linear heating and cooling are assumed to be more conservative than the commonly seen sigmoid curve, as actual cold spot temperatures will rise and fall more gradually near the peak temperature achieved. In the idealized model presented in Fig. 1, the linear model temperatures are consistently lower than the expected actual temperatures in the range of 50 – 60°C, where *M. morgani* lethality will occur (7). This linear heating and cooling model, together with thermal death time values $D_{60^{\circ}\text{C}}$ of 0.26 m, $z = 4.1^{\circ}\text{C}$ (7) were used in a General Method calculation (3, 5, 18) employing the Trapezoidal rule (5, 24). A similar calculation was used by the American Meat Institute Foundation for validating cook processes for meat and poultry products (9, 25).

RESULTS AND DISCUSSION

A 5-log reduction of *M. morgani* would be achieved when a peak cold spot temperature of 59.3°C was reached (Table 1). These results are compared to real data in Figures 2 and 3 using information supplied by Munshi et al. (17). This approach can be used to establish a critical limit for end point temperature, which can be used in conjunction with cook schedules. Cook schedules are designed to achieve the end point temperature as closely as possible, and the EPIPT

measurement ensures this critical control point is achieved. These calculations are useful in validating that this procedure achieves a safe level of control, thereby preventing histamine formation during processing.

Figure 2 provides an example of fish cooked for 95 minutes, including a come-up time to an initial ambient steam cooking temperature of 90°C, and a cook hold ambient temperature of 70°C. An EPIPT of 58.1°C was reached, resulting in an 8.2-log reduction of *M. morgani*. If the fish being cooked is heated more slowly than presented in the model, the time that the cold spot was exposed to lethal temperatures was longer than in the idealized model, resulting in greater than minimum lethality. Also, as expected, the cold spot heating curve was convex in that portion of the cook where the cold spot temperature was above 50°C, which also contributed to a higher than minimum lethality. In Figure 3, two fish were cooked for 182 minutes, including the come-up time to an initial ambient steam cooking temperature of 90°C and a cook-hold ambient temperature of 70°C. When an ambient temperature drop occurs as a type of process deviation, fish A achieved a cold spot temperature of 58.4°C, resulting in a 4.3-log reduction of *M. morgani*. In comparison, fish B achieved a cold spot temperature of 59.3°C and a 5.7-log reduction of *M. morgani*. These data demonstrate: (i) a convex heating curve at temperatures above 50°C, as expected; (ii) failure to achieve a temperature of at least 59.3°C results in less than a 5-log reduction of *M. morgani*; and (iii) the usefulness of using an EPIPT critical limit in assessing a process deviation,

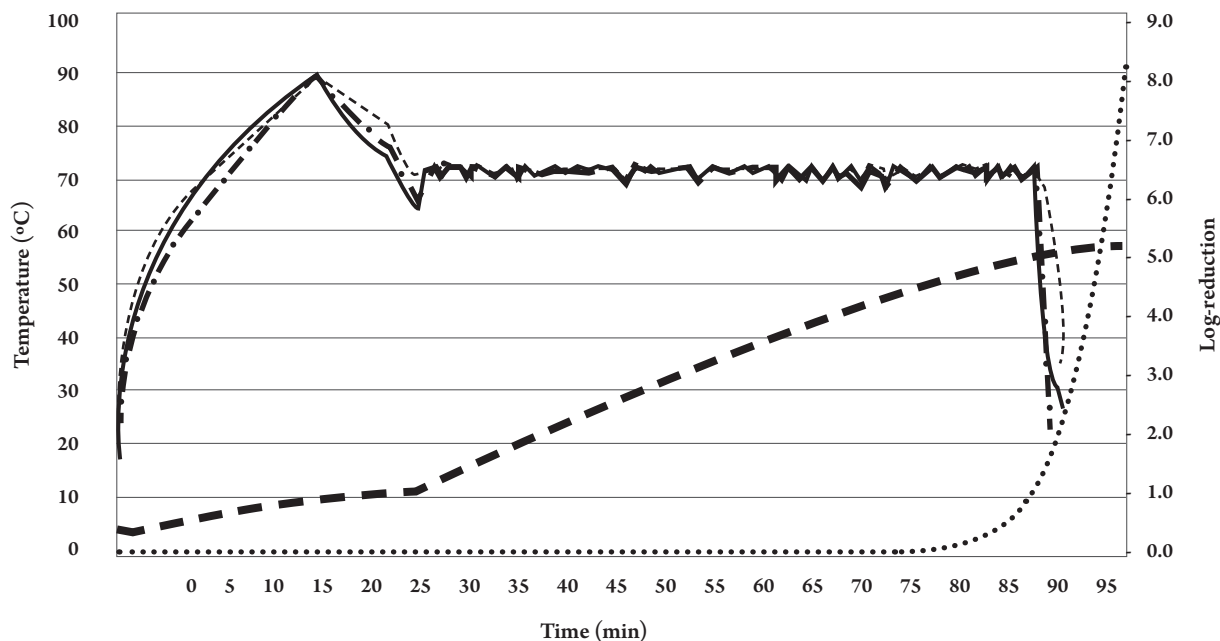


FIGURE 2. Inactivation of *M. morgani* in tuna fish during pre-cooking process under ideal conditions (General Method calculation). Short dash represents cooker temperature Top of fish trolley, dash-dot-dash line is cooker temperature Middle of fish trolley, solid line cooker temperature Bottom of fish trolley, long dash Cold spot of fish, dot line Log-reduction.

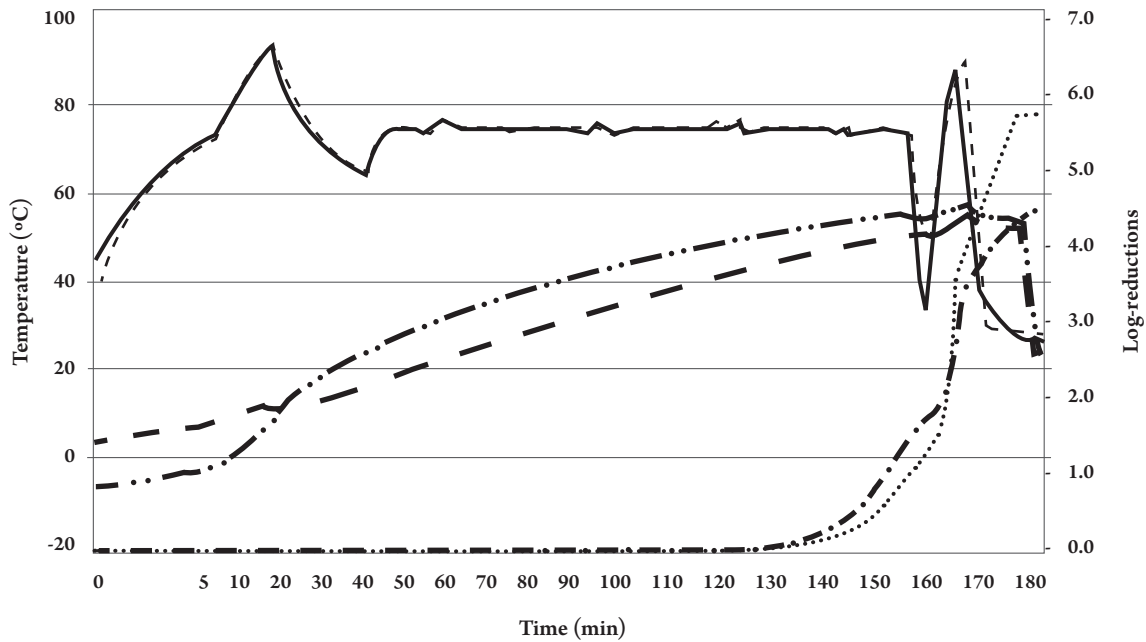


FIGURE 3. EPIPT used to evaluate individual processes for inactivation of *M. morganii* in tuna fish during pre-cooking process (General Method calculation). Short dash represents cooker temperature Top of fish trolley, solid line cooker temperature Bottom of fish trolley, long dash Cold spot of fish A, dash-dot-dot-dash Cold spot of fish B, dash-dot-dash-dot Log-reduction of fish A, dot line Log-reduction of fish B.

thereby facilitating rapid decisions when a failure to achieve the full process schedule for that fish or pre-cooker batch occurs. Upon observing any fish cold spot temperatures below the critical limit, the corrective action would be to place the fish back into the cooker and cook for additional time. Finally, *Fig. 3* demonstrates the usefulness of using a lethality calculation to assess a deviation in which there is a failure to achieve the minimum end point temperature, provided that a full temperature history is available for the largest and coldest fish in a pre-cook batch.

A critical limit based on EPIPT is conservative because: (i) it is based on internal product cold spot temperatures, which are colder than actual in the range of 50 – 60°C, where *M. morganii* lethality occurs; (ii) it is based on the fastest known heating and cooling rate, but slower heating rates will result in more time accumulated at over 50°C in the internal product cold spot, hence higher lethality; and (iii) slower heating rates will occur for larger fish, lower ambient cook temperatures, and other heating media, besides saturated steam.

The EPIPT model assumes that half of the lethality is achieved during cooling, and the results presented in *Figures 2 and 3* both confirm the model. Since cooling makes such an important contribution to *M. morganii* lethality, an extreme worst case simulation of rapid cooling was done, and presented in *Fig. 4* with propriety software known to be reliable (6). In this simulation, cooking was done at 100°C, while the cooling temperature dropped to 0°C over

12 seconds at the end of the cook. The authors are unaware of any cooking and cooling equipment that could achieve such a rapid cool under real world conditions. Fish in this simulation scenario heated product from 0°C to 60°C in 15 minutes, which is faster than what is assumed to derive a 59.3°C critical limit. The heating curve was convex at temperatures above 50 °C. The fish cold spot temperature continued to increase during the cool phase, as is normal. All of the lethality was accumulated while the cold spot was above 50°C, just as the heating and cooling model predicts. As such, all of the lethality was achieved during the cooling phase. Despite heating and cooling more rapidly than the linear model assumes, and despite cooling more rapidly than would be possible under real world conditions, a reduction of 6.8 log of *M. morganii* could be calculated using the convex heating and cooling curve, with a peak temperature of 59.7 °C, which remains below the proposed 60°C critical limit.

The tuna industry has already developed practical recommendations for using the 60°C critical limit for the pre-cook step (20). These include temperature distribution testing, instrument calibration, adequate record keeping, acceptable hazard analysis and critical control points (HACCP), acceptable good manufacturing practices (GMPs), acceptable safe sanitation operating procedures (SSOPs), process schedules designed to achieve a 60°C peak temperature, a heating rate that achieves peak temperature no faster than 2°C per minute, an adequate sampling or control plan to validate a 60°C peak temperature, and an adequate method to measure the EPIPT.

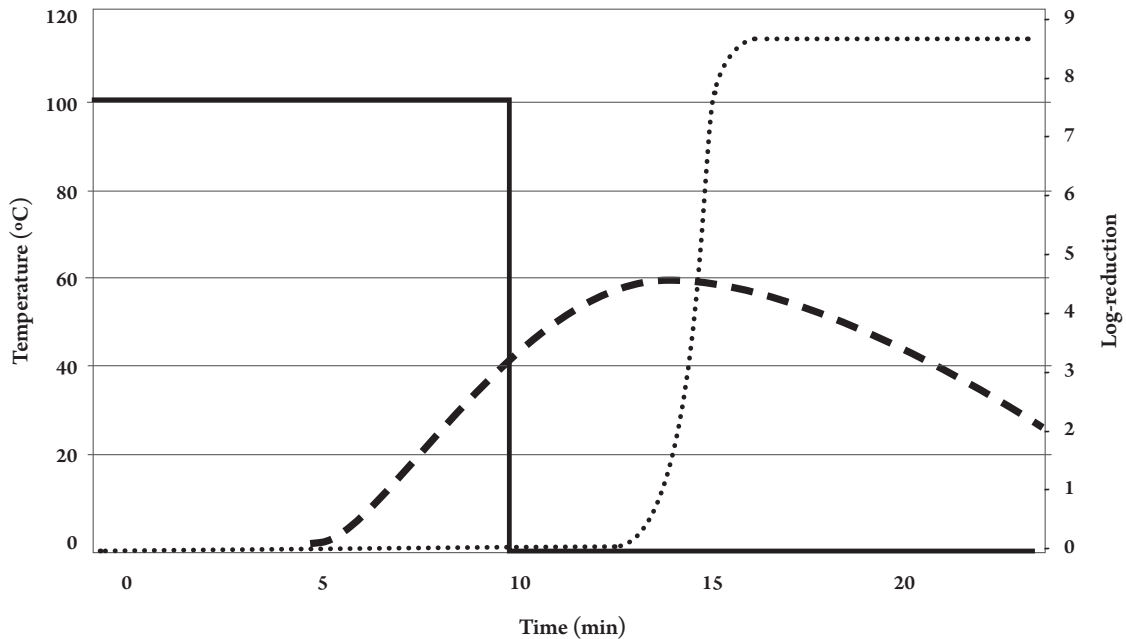


FIGURE 4. Worst case scenario software simulation of the impact of extremely rapid cooking on inactivation of *M. morgani* in tuna fish during pre-cooking process (General Method calculation). In this simulation, cooking was done at 100°C, and the cooling temperature dropped to 0°C over 12 seconds at the end of the cook. Solid line represents cooker temperature, dotted line log-reduction, and dashed line cooking time.

Other pre-cook CCP strategies are possible, even without additional biological validation as per Munshi et al. (17). These strategies include (i) validating that a minimum peak temperature of 60°C is reliably reached without measuring exit temperatures, such as when cooling takes place inside the cooker; (ii) direct monitoring of lethality using probes, where the ability to do so can be successfully demonstrated; or (iii) establishing a critical limit of less than 60°C, if it can be demonstrated that a reduction in *M. morgani* of at least 5 logs will be achieved reliably. Finally, it should also be possible to establish a process critical limit using less than a 5-log reduction of *M. morgani*, although this approach would require further validation, as per Munshi et al. (17).

It is concluded that a 60°C EPIPT for pre-cooking tuna of any size will stop histamine formation during and after pre-cooking, until further contact at the de-skinning step is reached. This approach has the potential to provide the tuna industry with extra time to process tuna, especially the larger sizes. It is also possible to use process lethality as a tool to evaluate a deviation from the process schedule, where a 5-log reduction can be demonstrated for a pre-cook batch.

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