

## Short Communication

## Sensory acceptability of squid rings gamma irradiated for shelf-life extension

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## ARTICLE INFO

## Keywords:

Radiation processing  
Preservation  
Squid  
Sensory acceptability  
Shelf-life

## ABSTRACT

The feasibility of extending the shelf-life of a squid product by gamma irradiation was analyzed. *Illex argentinus* rings were irradiated at 4 and 8 kGy; and stored at  $4 \pm 1$  °C during 77 days. No mesophilic bacteria, enterobacteriaceae and coliforms were detected in irradiated rings during storage. Psychrotrophic bacteria were significantly reduced by irradiation; their counts were fitted to a growth model which was further used for shelf-life estimations: 3 and 27 days for 0 and 4 kGy, respectively. Initially, non-irradiated as well as irradiated rings had very good sensory scores. The overall acceptability of 4 and 8 kGy rings did not decrease during 27 and 64 days, respectively, while control samples spoiled after 3 days. A radiation dose range for squid rings preservation was defined, which attained the technological shelf-life extension objective, without impairing sensory quality.

## 1. Introduction

*Illex argentinus* is the most abundant squid species of the Southwest Atlantic Ocean. It is a natural resource of economic importance for the region. Squid has a short shelf-life under refrigeration that does not exceed 8 days (Melaj et al., 1997; Tomac et al., 2013). Squid rings are obtained by cross-cuts of the peeled mantle muscle. During processing, rings can be contaminated by human manipulation with bacteria different from its native microbial flora, including potential pathogenic bacteria such as *Staphylococcus aureus* and *E. coli*. So, gamma irradiation could be used to extend squid rings shelf-life, improving its quality and reducing risks for human health. Though the preservation of a wide variety of fishery products by irradiation is well documented (IAEA, 1969; Kilgen, 2001; Lescano et al., 1990; Nickerson et al., 1983), not much information regarding squid was found (Kim et al., 2012). At the moment, fish and other seafood irradiation is on path to approval by the Argentine Alimentary Code. When exposing a product to a radiation field, it will absorb a dose gradient. According to Codex Alimentarius (2003), the minimum absorbed dose should be sufficient to achieve the technological purpose, and the maximum absorbed dose should be less than that which would compromise consumer safety, wholesomeness or would adversely affect structural integrity, functional properties, or sensory

attributes. A maximum dose will unavoidably be received by a small part of the product bulk as a consequence of its exposure to an industrial radiation field (ISO, 2011). In a previous work, a dose of 4 kGy was proved to be efficient to extend the shelf-life of squid rings (Tomac, 2013). In the present study, sensory squid quality after irradiating at 8 kGy, presumably the maximum dose attained during an industrial treatment where the minimum is 4 kGy, was studied.

## 2. Materials and methods

## 2.1. Sample preparation and irradiation

*Illex argentinus* caught in the Southwest Atlantic Ocean were beheaded, gutted, peeled, fins removed, and peeled mantles crosscut into rings ( $1.2 \pm 0.2$  cm width). Samples of  $200 \pm 3$  g (40 rings approximately) were packed in polyethylene/polyamide bags. One lot of sixty samples was kept at  $4 \pm 1$  °C as control (0 kGy), while two lots of sixty samples were gamma irradiated at target doses of 4 and 8 kGy, in a cobalt-60 facility at a dose rate of 10.9 kGy/h. The real minimum and maximum absorbed doses were 4.8 and 4.9 kGy, with dose uniformity of 1.02; and 8.4 and 9.6 kGy, with dose uniformity of 1.14, respectively. Silver dichromate dosimeters were used (ISO/ASTM 51401-2003 (E), 2003). Samples were kept at  $4 \pm 1$  °C during transportation, irradiation

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and storage (77 days).

## 2.2. Microbiological analysis

Ten grams of rings in saline solution (0.85%) with 0.1% w/v peptone were homogenized. Aerobic psychrotrophic and mesophilic bacteria, Enterobacteriaceae, total coliforms, fecal coliforms, *Staphylococcus* spp., and sulphite-reducing clostridia were analyzed (ICMSF, 1983).

## 2.3. Sensory analyses

Fifty consumers (60% women; age: 20–60 years) evaluated the overall acceptability, appearance, color, odor, taste and texture using the nine-point hedonic scale (Peryam and Pilgrim, 1957; ASTM, 1977), at four dates. Rings were prepared according to Tomac (2013). Two rings (12 ± 3 g) of each sample with rosemary and black pepper spiced sunflower oil (15 ± 2 g) were served to consumers (20 ± 2 °C). The samples order was randomized. A sample was considered acceptable if more than 70% of consumers rated the overall acceptability ≥ 6 (positive degree of liking).

## 2.4. Statistical analysis and mathematical modeling

The microbiological results were analyzed by the two-ways ANOVA test ( $p < 0.05$ ; main factors: radiation dose and storage days). The Tukey test was used to compare means ( $p < 0.05$ ). The sensory results were tested by Kruskal-Wallis ( $p < 0.05$ ). The growth curves of psychrotrophic bacteria were fitted to the modified Gompertz model (Zwietering et al., 1990).

$$\log N = \log N_0 + A \exp [-\exp (\mu e/A (L-t)) + 1] \quad (1)$$

where  $\log N$  is the decimal logarithm of the number of psychrotrophic bacteria counts at time  $t$  and  $\log N_0$  the psychrotrophic counts the day after irradiation treatment ( $\log CFU/g$ ),  $\mu$  the specific growth rate ( $\log CFU/g \text{ day}^{-1}$ ),  $L$  the lag phase (days),  $A$  the logarithmic bacterial population increase and  $e$  the Euler number. Afterwards, Gompertz model parameters  $\mu$ ,  $L$  and  $A$  were fitted to dose dependent second order polynomials, in order to find an expression that predicts psychrotrophic bacterial counts for different radiation doses. So, a full model was developed by introducing these secondary models into the modified Gompertz equation (Eq. (1)). This complete model is useful to predict psychrotrophic populations for a determined radiation dose applied, enabling shelf-life estimation ( $t$ , days) when the microbiological recommended count value is reached, according to Tomac et al. (2013) considerations. The fitting and the estimations accuracy were evaluated by the Root Mean Square Error (RMSE) and the determination coefficient ( $R^2$ ).

## 3. Results and discussion

### 3.1. Microbiological analyses

The initial mesophilic bacterial counts ( $1.3 \times 10^5$  CFU/g) were inactivated by 4 and 8 kGy to values under the detection limit ( $< 10$  CFU/g). Similar results were observed by Kim et al. (2012) in squid. No mesophiles were detected in irradiated samples during 77 days, whereas they significantly increased in controls (Fig. 1). Within the group of mesophilic bacteria, initial counts of enterobacteriaceae and total coliforms were  $1.2 \times 10^4$  and  $8.1 \times 10^3$  CFU/g, respectively. These were not detected in irradiated rings during storage, in accordance with the results of mesophilic bacteria. The initial values significantly increased in non-irradiated rings during storage. These results agree with the inactivation of enterobacteriaceae caused by 2 kGy observed by Badr (2012) in smoked salmon fillets, during 50 days.

Fecal coliforms, *Staphylococcus* spp. and sulphite-reducing clostridia

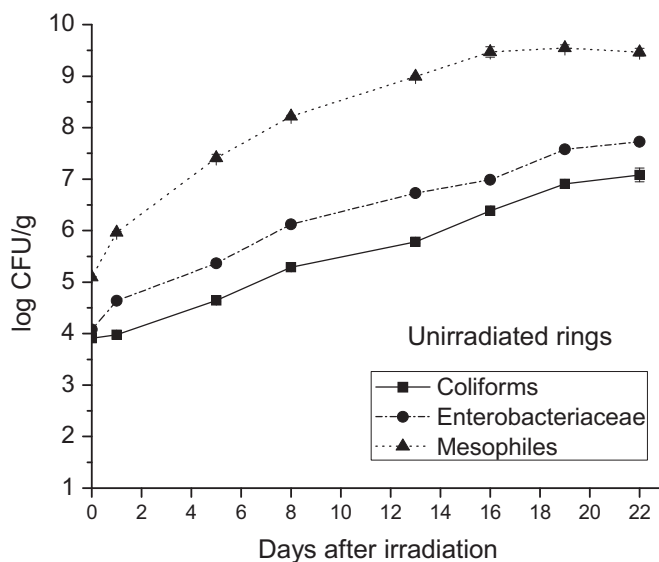


Fig. 1. Coliform, enterobacteriaceae and mesophilic bacterial counts in control rings during storage at  $4 \pm 1$  °C ( $n=2$ ).

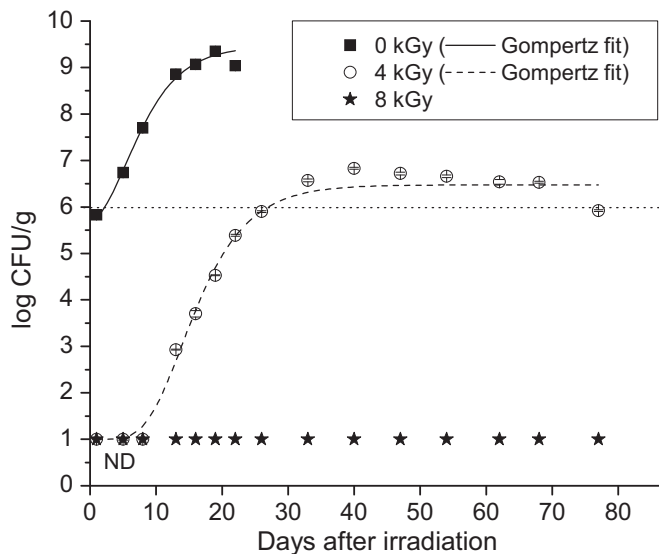


Fig. 2. Psychrotrophic bacterial counts in control and gamma irradiated squid rings during storage at  $4 \pm 1$  °C ( $n=2$ ). ND: not detected.

tridia were detected neither in irradiated nor in non-irradiated rings during storage. The absence of the first two bacterial groups would indicate adequate manipulation during processing.

The growth of psychrotrophic bacteria is shown in Fig. 2. The initial load ( $3.5 \times 10^5$  CFU/g) was inactivated by gamma irradiation at 4 and 8 kGy ( $< 10$  CFU/g). In this way, a population reduction of more than 4 logarithmic cycles was achieved. During storage, control samples counts significantly increased, while in 4 and 8 kGy samples, growth started only at the 8th day after irradiation, and never during 77 days, respectively. These results are in agreement with a previous work in which up to a 4 log cycle reduction of the initial load, and also a growth rate reduction, were achieved in *Illex argentinus* rings pre-treated with poly-phosphates (Tomac et al., 2013).

### 3.2. Shelf-life extension estimation by mathematical modeling of psychrotrophic bacterial growth

Psychrotrophic bacteria are the typical contaminating flora of temperate seawater fish (Huss, 1999). Total bacterial count (TBC) is

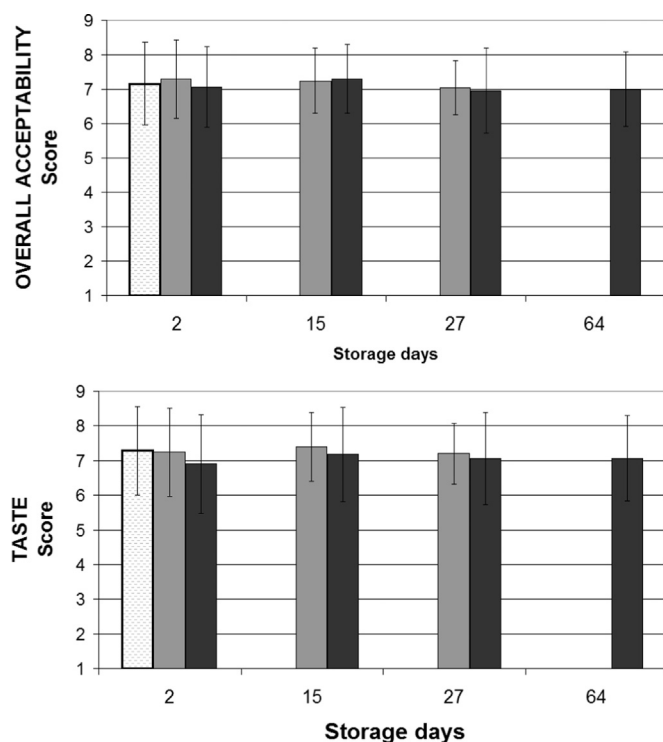


Fig. 3. Overall acceptability and taste of squid rings irradiated at 0 (-), 4 (■) and 8 (■) kGy, during storage at  $4 \pm 1$  °C. Mean  $\pm$  standard deviation. Similar results were found for color, appearance, odor and texture (N=50).

considered one of the best quality indexes for ice-stored fish, so a TBC limit of  $10^6$  CFU/g is recommended for fresh fish and mollusks (ICMSF, 1983). The experimental growth curves of psychrotrophes were accurately fitted to the Gompertz model (Fig. 2), as indicated by the high values of  $R^2$  ( $> 0.98$ ) and low values of RMSE ( $< 0.05$ ). Model parameters obtained for 0 and 4 kGy were:  $\mu=0.35$  and  $0.37$  ( $\pm 0.03$ ) log CFU/g days;  $A=3.54$  and  $5.47$  ( $\pm 0.15$ ) log CFU/g; and  $L=2.56$  ( $\pm 0.52$ ) and  $8.44$  ( $\pm 0.63$ ) days, respectively. So, considering the limit value of  $10^6$  CFU/g, the estimated shelf-life for 0 and 4 kGy rings was 3 and 27 days, respectively. No bacterial growth was detected in 8 kGy samples, so the microbiological limit was not reached during 77 days.

### 3.3. Sensory analyses

The results of overall acceptability and taste are shown in Fig. 3. Similar results were found for color, appearance, odor and texture. The 2nd day after irradiation 0, 4 and 8 kGy samples were evaluated together. The score for each sensory attribute of the three samples was  $\geq 7$ . Overall acceptability of 0, 4 and 8 kGy rings was given scores  $\geq 6$  by 86% of consumers, indicating a high acceptability of the product either irradiated or not.

On days 15 and 27, only 4 and 8 kGy rings were analyzed since psychrotrophic bacterial counts of controls exceeded  $1 \times 10^6$  CFU/g (Fig. 2) and showed spoilage signs (ammoniacal and putrid odor and discoloration). The scores of both samples were  $\geq 7$ , with no significant differences between them. The overall acceptability of both samples was rated  $\geq 6$  by 91% of consumers. The acceptability of irradiated rings did not decrease during 27 days.

On day 64 only rings irradiated at 8 kGy were analyzed since 4 kGy rings had surpassed their shelf-life after the 27th day and suffered evident modifications of their sensory characteristics. Every score was greater than 6.8. The 86% of consumers rated the overall acceptability with 6 or more, indicating that it was maintained during storage.

These results show that 4 and 8 kGy of gamma radiation extended squid rings sensory acceptability 8 and 20 times, respectively, as

compared to control; and that 8 kGy did not impair the product acceptability. This product could tolerate a relatively high radiation dose of 8 kGy due possibly to its low lipid content of about 1% (Sugiyama et al., 1989). Irradiation of fatty fish and seafood could be limited in certain cases by the appearance of off-odors and flavors related with lipid oxidation, which could affect the sensory acceptability.

## 4. Conclusions

It was possible to observe an agreement between the recommended microbiological value for squid rings and the behavior of the sensory characteristics modifications during storage. Gamma irradiation reduced bacterial loads and caused a considerable shelf-life extension of a highly perishable squid product under refrigeration. A dose of 4.8 kGy increased the shelf-life by 24 days, and the good sensory acceptability of the product was not affected even at 8.4 kGy. It was demonstrated that the technological goal of shelf-life extension was met, with no impact on the product sensory attributes. Hopefully these results drive the implementation of this technology in fish and seafood products.

## Acknowledgments

Authors are thankful to the Universidad Nacional de Mar del Plata (Project 15/G264/09) and Consejo Nacional de Investigaciones Científicas y Técnicas (PIP 0403) for economic support, and also to Techn. Maritza Guerrero, Techn. Irene Amezttoy, staffs of the Semi Industrial Gamma Irradiation Facility and High Dose Dosimetry Section of the Ezeiza Atomic Center, and colleagues at this Center and at the Faculty of Engineering (UNMDP) performing sensory evaluation.

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