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Incidence of *Listeria* spp. in domestic refrigerators in Portugal

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Abstract

The main objectives of this study were to determine the incidence of *Listeria* spp. in Portuguese domestic refrigerators and to evaluate some of the hygienic practices in the domestic environment that might contribute to the persistence of the organisms. It was found that *L. monocytogenes* was present in 3 domestic refrigerators of the 86 investigated. *L. grayi* and *L. innocua* were also isolated from 4 and 1 refrigerators, respectively.

Overall, the information obtained from our survey demonstrates the need for consumer education in Portugal regarding safe food handling practices. For the refrigerators investigated, ≈71% were operating at a temperature higher than 6.1 °C, 87% were cleaned only monthly or less frequently, and only 8% were cleaned with appropriate proprietary cleaning products available in supermarkets.

Keywords: *L. monocytogenes*; Incidence; Food safety

1. Introduction

In order to answer consumers' demands, e.g. for minimally processed foods and products with longer shelf-life, the use of refrigeration has increased considerably during the past several years. A potential hazard in refrigerated foods, particularly in products which are eaten without further cooking, is *L. monocytogenes*. This pathogen is widely distributed in many environments and often found in foods (Farber & Peterkin, 1991; Jørgensen & Huss, 1998), and is well known for its survival and growth at refrigeration temperatures. *L. monocytogenes* has also been shown to adhere to various surface materials normally in contact with foods such as stainless steel, rubber, glass and polypropylene (Blackman & Frank, 1996; Mafu, Roy, Goulet, & Magny, 1990) and colonisation of refrigerators by *L. monocytogenes* has been previously demonstrated (Cox et al., 1989; Sergelidis et al., 1997).

The work presented here had as its main objectives to determine the incidence of *Listeria* spp. in Portuguese domestic refrigerators and to evaluate some of the hygienic practices in the domestic environment that might contribute to the persistence of the organisms. To our knowledge, this is the first survey into domestic practices that may affect the safety of food products in Portugal, although similar studies have been conducted in other countries (Cox et al., 1989; Jackson et al., 1993; Sergelidis et al., 1997).

2. Materials and methods

During the period October 2001–May 2002, 86 refrigerators located at private homes in the North of Portugal (around Porto) were sampled for the presence of *Listeria* spp. Before sampling, the temperature of each refrigerator was measured and recorded using a portable digital thermometer (HD 9214, Delta OHM, Caselle di Selvazzano, Italy) and the refrigerator's owners were asked to answer a simple questionnaire, including the following:

How do you clean your refrigerator?
How often do you clean your refrigerator?
When was the last time you cleaned your refrigerator?
Do you pack all your foods before storage in the refrigerator?

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What are the foods you normally store in the refrigerator without any packaging?

From each refrigerator two surface samples were collected (≈100 cm^2 from locations where vegetables were stored and ≈100 cm^2 from locations where cheese or meats were stored) by swabbing at various points of the selected location with sterile cotton swabs, previously immersed in sterile Ringer's solution. The swabs were transferred to 10 ml of half-Fraser broth (Biokar Diagnostics, Beauvais, France) and incubated at 30 °C for 48 h. Aliquots (1 ml) of these primary enrichments were transferred to 10 ml of secondary enrichment Fraser broth (Biokar Diagnostics) and incubated at 30 °C for 48 h. A loopfull of each primary enrichment culture and of the secondary enrichments after 24 h and 48 h of incubation, was streaked separately onto PALCAM and Oxford agar plates (Merck, Darmstadt, Germany). After incubation at 37 °C for 48 h, five typical colonies per plate (when possible) were transferred onto Plate Count Agar and incubated at 37 °C for 48 h. Pure cultures were tested for the Gram reaction, catalase, oxidase, fermentation of the sugars mannitol (0.5%w/v), rhamnose (1%w/v) and xylose (0.5%w/v), CAMP with *Staphylococcus aureus* NCTC 1621 and *Rhodococcus equi* NCTC 25923, and API *Listeria* (BioMérieux 10300).

3. Results and discussion

In the present study, it was found that *L. monocytogenes* was present in 3 domestic refrigerators out of the 86 investigated. *L. grayi* and *L. innocua* were also isolated from 4 and 1 refrigerators, respectively (Table 1). Since the selective media PALCAM and Oxford do not allow a clear distinction between colonies of different *Listeria* spp. it is possible that the incidence of *L. monocytogenes* presented in this work might be underestimated. Johansson (1998) demonstrated that the selection of five colonies for confirmation from the standard selective plating media (Anonymous, 1996) may not be sufficient if other *Listeria* species are present. Additionally, the higher growth rate of *L. innocua* in selective liquid media (Curiale & Lewus, 1994; MacDonald & Sutherland, 1994) compared with *L. monocytogenes*, can result in false negative results on the PALCAM and Oxford media, and Yokoyama, Maruyama, and Katsube (1998) concluded that most *L. innocua* strains produce a bacteriocin-like substance against *L. monocytogenes* that may inhibit growth of the latter organism during enrichment culture. It is also important to comment that the presence of any *Listeria* spp. may be indicative of poor hygiene and cross-contamination scenarios which could favour the persistence of *L. monocytogenes*.

Colonisation of refrigerators by *L. monocytogenes* has already been demonstrated. Sergelidis et al. (1997) in Greece and Cox et al. (1989) in Holland recovered the organism from 2 of 136 and 1 of 35 household refrigerators tested, respectively. However, Jackson et al. (1993) in the USA did not recover *L. monocytogenes* from any of the 195 domestic refrigerators sampled.

According to the results presented here and to those previously published (Cox et al., 1989; Sergelidis et al., 1997), the potential for ready-to-eat products to be cross-contaminated via contaminated surfaces in the refrigerator, must be recognised. Studies in northern Europe and Australia indicate that most of the populations of these countries are not aware of this organism and hygienic practices in domestic environments often pose a risk to the safety of food (Jay, Comar, & Govenlock, 1999; Scott, 1996). There is no data to suggest that the level of awareness of the Portuguese consumer is higher than in these reported studies.

Although no correlation was found between the presence of *L. monocytogenes* in refrigerators and their temperature (Table 1), the results obtained in this study demonstrate that a significant number of the refrigerators investigated were operating at a temperature that

Table 1
Listeria spp. in domestic refrigerators

Organism	Place of isolation	Frequency of cleaning	Last cleaning prior to sampling	Product used for cleaning	Products stored without packaging	Temperature (°C)
L. monocytogenes	Cheese shelf	Monthly	2 weeks	Water	Cheese, fermented sausages	6.0
L. monocytogenes	Meat shelf	Monthly	2 weeks	Water	Cheese, fermented sausages	4.8
L. grayi	Vegetables shelf	Each 6 months	>3 months	Water/vinegar	Meat	8.2
L. grayi	Vegetables shelf	Weekly	1 week	Detergent	Vegetables	10.0
L. grayi	Vegetables shelf	Each 2 months	1 month	Water	Fermented sausages	6.1
L. inoccua	Vegetables shelf	Monthly	3 weeks	Detergent	Fermented sausages	8.5
L. grayi	Vegetables shelf	Each 3 months	1 month	Water/vinegar	Vegetables	5.4
L. monocytogenes	Meat shelf	Monthly	1 month	Water	Vegetables	10.0