

## Evaluation of maximum growth rate of *Listeria monocytogenes* in ready-to-eat fresh-cut papaya and melon

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### Abstract

*Listeria monocytogenes* has been identified on minimally processed fruit and vegetables, and can transmit a severe foodborne disease through the consumption of ready-to-eat (RTE) fresh-cut produce, mostly if the storage conditions are incorrect, and the cold chain is not abided to. The aim of the present work was to evaluate the behaviour of *L. monocytogenes* in RTE fresh-cut papaya and melon, two low-acid fruits stored in temperature-abuse conditions (10 and 8°C, respectively) during the shelf-life indicated by the producer (7 d). A microbiological challenge test was performed to assess the maximum growth rate (MGR) and the duplication time (Td) of the pathogen, testing three strains separately. We also assessed the behaviour of the indigenous microflora of the fruits. Results demonstrated that papaya and melon were two favourable substrates for *L. monocytogenes*. The MGR was 0.044 log CFU/g/h with a Td of 6.77 h for papaya, and 0.029 log CFU/g/h with a Td of 10.24 h for melon. The storage of RTE fresh-cut papaya and melon at an abusive temperature above 8°C could encourage the growth of *L. monocytogenes* with a possible risk to public health.

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### Introduction

*Listeria monocytogenes* is a Gram-positive invasive microorganism which is widely distributed in the environment. In humans, this pathogen is responsible for listeriosis, a foodborne illness that occurs with different degrees of severity: mild gastroenteritis, severe blood and central nervous system infections, and even causing death especially in the immunocompromised and the elderly, with a case-fatality rate of 20% (Angelo *et al.*, 2016). In pregnant women, *L. monocytogenes* can cause miscarriages, premature births, and infections in newborns such as pneumonia and meningitis (Matle *et al.*, 2020).

The European Food Safety Authority (EFSA) reported around 1,900 confirmed invasive human cases of listeriosis in Europe in 2020, with 780 hospitalisations and 167 deaths in Europe (EFSA and ECDC, 2021). Centers for Disease Control and

Prevention (CDC) reported two listeriosis outbreaks in the U.S.; one associated with ready-to-eat (RTE) soft raw milk cheeses in 2017 (CDC, 2017), and the other associated with deli-sliced meats and cheeses from 2016 to 2019 (CDC, 2020). Fruit has also been documented as an important cause of listeriosis outbreaks: in 2011, contaminated melons of the cantaloupe variety caused 33 deaths and a miscarriage in the U.S. (CDC, 2011), and although no outbreaks associated with papaya contaminated by *L. monocytogenes* have been reported so far in literature, papaya has proven to be a favourable substrate for the growth of *L. monocytogenes* since it is an ubiquitarian pathogen (Luciano *et al.*, 2021).

Fresh-cut fruit is increasingly popular on the global market because it can be consumed without preparation or cooking, and because it is generally considered by consumers to be a healthy snack. However, fruit is susceptible to spoilage and pathogenic bacterial growth for various reasons. Fruit

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that grows on the ground, such as melon, can be contaminated by microorganisms from soil, water, animals, and insects before harvesting (Yousuf and Qadri, 2020). After harvesting, contaminations can be linked to handling operations during cutting and packaging, and incorrect storage methods adopted by producers and consumers.

Factors affecting the survival and growth of *L. monocytogenes* in fresh-cut fruits include the pH and water activity ( $a_w$ ) of the product, the storage temperature, and the shelf-life duration. According to community legislation (EC, 2005), products with  $\text{pH} \leq 4.4$  or  $a_w \leq 0.92$ , products with  $\text{pH} \leq 5.0$  and  $a_w \leq 0.94$ , and products with a shelf-life of less than five days are not considered to be foods that promote the growth of *L. monocytogenes*. Papaya and melon can be considered favourable substrates for *L. monocytogenes* growth, considering their usual pH values around 5 - 6,  $a_w$  values around 0.98 - 0.99, and the shelf-life period of seven days.

Some studies have suggested that the growth of *L. monocytogenes* is favoured in fresh-cut produce rather than whole produce because of the protective outer barrier of whole fruit that reduces the availability of moisture and nutrients (Marik et al., 2020). A study on *L. monocytogenes* kinetics in eight whole and 12 fresh-cut products demonstrated that whilst growth was largely supported by the fresh-cut fruit, all the whole products were unable to support growth of the pathogen (Kroft et al., 2022). *Listeria monocytogenes* is also able to survive or grow on the surfaces of other fresh-cut fruit such as apple, papaya, and mango (Penteado and Leitao, 2004; Penteado et al., 2014).

Commission Regulation No. 2073/2005 (EC, 2005) specifies that food business operators (FBOs) must conduct studies, as necessary, to evaluate the growth of *L. monocytogenes* that may be present in the product during the shelf-life under reasonably foreseeable storage conditions. In particular, they may carry out durability studies and challenge tests, or use predictive mathematical modelling to evaluate the *L. monocytogenes* critical or survival growth factors of concern in the product.

The aim of the present work was, therefore, to evaluate the behaviour of *L. monocytogenes* on papaya and melon marketed as RTE fresh-cut products by performing challenge tests and determining the maximum growth rate (MGR). We also assessed, for each product, the indigenous microfloral load and the physical and chemical

parameters (pH and  $a_w$ ) in order to observe possible interactions occurring between the microflora and *L. monocytogenes*.

## Materials and methods

### Products

The RTE fresh-cut papaya (*Carica papaya* L.) and cantaloupe melon (*Cucumis melo* L.) were acquired from a company based in the Emilia Romagna region (Italy) specialised in the processing of fresh fruits, and vegetables marketed within large-scale distribution chains. Traditional and tropical fresh fruits were washed, with chlorine-based disinfectant solution, peeled, cut, diced, stored, and marketed in heat-sealed packages. The various processing steps were carried out both mechanically and manually based on the type of fruit at a temperature of up to 11 - 12°C. The fresh-cut melon and papaya were provided specifically for the present work, in 60 g trays heat-sealed with micro-perforated polyethylene terephthalate (PET), free from additives, and in an unmodified atmosphere. The shelf-life declared by the manufacturer for this type of product was seven days (including the day of production) at a recommended storage temperature of  $5 \pm 1^\circ\text{C}$ , as indicated on the label. The trays were transported from the factory to the laboratory in which the experiments were carried out on the same day of production, following the cold chain at 5°C.

### Strains and inoculum preparation

The experimental protocols were carried out using different strains of *L. monocytogenes*; ATCC® 19115<sup>TM</sup> (reference strain); IZSLER 283337/2010 (wild strain) for papaya; ATCC® 19115<sup>TM</sup> and IZSLER 293317/10 (wild strain) for melon. The wild strains were previously isolated in RTE fresh-cut pineapple and fruit salad containing pineapple. These strains, kept frozen at  $-80^\circ\text{C}$  in brain heart infusion (BHI; Oxoid, Milan, Italy), supplemented with 20% glycerol, were defrosted and inoculated onto 5 mL of BHI and incubated at  $37 \pm 1^\circ\text{C}$  for 24 h. In order to adapt the strains to the storage conditions of the challenge test, 1 mL of the broth culture of each strain was transferred to bottles each containing 100 mL of BHI broth and incubated at  $8 \pm 1^\circ\text{C}$  for 5 d. At the end of the incubation period, each bacterial suspension was enumerated by incubating the plates at  $37 \pm 1^\circ\text{C}$  for 48 h in Agar *Listeria* according to Ottaviani and Agosti (ALOA; Microbiol Diagnostic, Cagliari,

Italy). After the enumeration, the bacterial suspension was diluted in sterile physiological saline solution (PSS; Microbiol Diagnostic) in order to have a starting inoculum concentration of approximately 3.5 - 4 Log CFU/g for papaya, and between 1.5 and 3 Log CFU/g for melon.

#### *Experimental protocol*

The study was conducted on three different batches of the same product. For each batch, we used 35 trays of fresh-cut fruit. Ten trays were spiked with the *L. monocytogenes* reference strain, and ten trays with wild strains. The fruit trays were contaminated using a sterile 1-mL syringe by piercing the lid of the tray without opening the package; 0.2 and 0.5 mL of inoculum was distributed on the surface of the diced melon and papaya, respectively. After contamination, the lid was resealed with a label. The sampling time (t) was defined as 0, 5, 24, 29, 48, 53, 72, 77, 144, and 149 h after inoculation; two times a day in the exponential phase, and once a day in the stationary phase. Six uninoculated trays were used to detect the possible natural contamination of the product by *L. monocytogenes* using a qPCR validated according to AFNOR BRD 07/10–04/05; and to check the  $a_w$  and pH values, with t defined as 0 and 149 h of storage. Lastly, nine trays (blank) were inoculated with PSS in the same quantity as the contaminated inoculum to check for possible effects of the inoculum on the substrate. These trays were tested for  $a_w$ , pH, and the behaviour of the indigenous microflora, with t defined as 0, 24, 48, 72, and 149 h of storage. The storage temperature for assessing the MGR was fixed. For this reason, the trays with diced papaya were stored for 7 d at 10°C, and the trays with diced melon for 7 d at 8°C.

#### *qPCR method*

Before the artificial contamination with *L. monocytogenes*, the fruit was tested using a qPCR method to check for natural contamination.

Initially, 25 g of fruit was homogenised with 225 mL of half Fraser enrichment broth (home-made preparation) followed by incubation at 30°C for 24 h. The DNA extraction was performed using a commercial kit (iQ-Check™ *Listeria monocytogenes* II PCR Detection Kit; Bio-Rad, Milan, Italy): after centrifuging 1.5 mL of the homogenate, the supernatant was discarded, the pellet was suspended in 200 µL of buffer with beads. After mechanical lysis

for 3 min in tissue lyser, the samples were incubated at 95°C for 15 min. The amplification was carried out using a CFX96™ Real-Time System (Bio-Rad) and CFX Manager Software, with the following temperature profile repeated for 50 cycles: initial denaturation at 95°C for 10 min, denaturation at 95°C for 15 s, annealing at 58°C for 20 s, and extension at 72°C for 30 s. For each run, two negative and positive controls were used (one extraction control and one amplification control). We also added an internal control (non-target DNA) to detect inhibitions and avoid false negative results.

#### *Microbiological enumeration methods*

The enumeration of *L. monocytogenes* in artificially inoculated fruit was performed following international standards (ISO, 2017c). All contents of the spiked tray were mixed, then 10 g of sample was homogenised with 90 mL of sterile Buffered Peptone Water (BPW; Biolife) in a filter bag using Stomacher (BagMIXER CC400, Interscience; Saint Nom la Bretèche, France).

The samples were homogenised for 3 min, then 2 mL (0.33 mL in six plates) and 0.1 mL of the undiluted homogenate of ten-fold dilutions, respectively, were plated on ALOA medium. The plates were incubated at 37°C for 48 h.

In addition, the indigenous microflora was detected and enumerated using the following methods: total viable count at 30°C (TVC\_M) (ISO, 2013), Enterobacteriaceae (Ent) (ISO, 2017a), and mesophilic lactic acid bacteria (LAB) (ISO, 1998). We enumerated the yeasts and moulds, then performed the total viable count at 5°C (TVC\_P). Briefly, the enumeration for TVC\_M was carried out on plate count agar (PCA, Biolife) incubated for 3 d at 30 ± 2°C, Ent on violet red bile glucose agar (VRBGA, Thermo Fisher Diagnostics, Monza, Italy) incubated for 1 d at 30 ± 1°C, and LAB on MRS agar (Biolife) incubated for 3 d at 30 ± 2°C. For yeasts and moulds and TVC\_P, we used the same method described previously for the enumeration of *L. monocytogenes*, with different media: for yeast and moulds, we used oxytetracycline glucose yeast extract agar (OGYE agar base; Microbiol Diagnostics) and for TVC\_P, the medium PCA. The petri dishes were incubated for 5 d at 20 ± 4°C, and for 10 d at 5 ± 2°C, respectively, in a sealed container to prevent the medium from drying out.

### Physical and chemical characteristics

For each batch, the pH and  $a_w$  of fresh-cut fruit were measured using a calibrated pH meter (Hanna Instruments HI 5221; Woonsocket, USA) and a water activity meter Aqualab4TE (Meter; Washington, USA), respectively, following standard procedures (ISO, 2017b). All measurements at the sampling times of 0 and 149 h were conducted on three different blanks and uninoculated trays.

### Statistical analysis

A sigmoidal Baranyi model (Baranyi and Roberts, 1994) was fitted to the *L. monocytogenes* experimental data using DMFit software (available at: <http://www.ifr.ac.uk/safety/DMFit/>), obtaining growth rates (GRs) associated with their standard errors (SEs) and the R-squared ( $R^2$ ). The highest GR values calculated were defined as the MGR (maximum growth rate, expressed as Log CFU/g/h). In addition, we also calculated the doubling time of *L. monocytogenes* (Td). The pH and  $a_w$  data taken from the blank and uninoculated samples were compared

using Student's *t*-test, available in R statistical software version 4.1.2.

## Results

### *Listeria monocytogenes* behaviour and maximum growth rate

Two microbiological challenge tests were carried out on RTE fresh-cut fruit (papaya and melon) in order to calculate the MGR of artificially inoculated *L. monocytogenes*, simulating conditions that can occur throughout the cold chain, using different storage temperatures.

Papaya and melon are considered to be favourable substrates for the growth of *L. monocytogenes* because of their pH and  $a_w$  values. Table 1 shows the pH and  $a_w$  values obtained in the present work. In papaya, the pH decreased slightly during the shelf-life period, whilst the  $a_w$  was around 0.97 - 0.98. In melon, there was a greater decrease in pH than in papaya, and the  $a_w$  was around 0.99.

**Table 1.** pH and  $a_w$  values in melon and papaya blank, and uninoculated test units during storage.

			Sampling time (h)						
			0		24	48	72	149	
			Blank*	Uninoculated*	Blank	Blank	Blank	Blank*	Uninoculated*
Papaya	pH	Batch 1	5.26 ± 0.01	5.27 ± 0.02	5.38	5.18	5.17	4.83 ± 0.33	4.62 ± 0.12
		Batch 2	5.20 ± 0.01	5.24 ± 0.02	5.21	5.29	5.07	4.42 ± 0.14	4.38 ± 0.26
		Batch 3	5.47 ± 0.07	5.34 ± 0.07	5.22	5.31	5.18	4.99 ± 0.10	4.91 ± 0.16
	$a_w$	Batch 1	0.98	0.98	0.98	0.97	0.98	0.97	0.97
		Batch 2	0.97	0.97	0.97	0.97	0.97	0.97	0.97
		Batch 3	0.97	0.97	0.97	0.97	0.97	0.97	0.97
Melon	pH	Batch 1	6.52 ± 0.14	6.54 ± 0.10	6.42	6.35	6.20	5.13 ± 0.35	5.20 ± 0.35
		Batch 2	6.50 ± 0.17	6.53 ± 0.05	6.73	6.39	6.30	4.79 ± 0.06	4.72 ± 0.07
		Batch 3	6.56 ± 0.16	6.52 ± 0.06	6.46	6.4	6.27	5.08 ± 0.07	5.07 ± 0.05
	$a_w$	Batch 1	0.99	0.99	0.99	0.99	0.99	0.99	0.99
		Batch 2	0.99	0.99	0.99	0.99	0.99	0.99	0.99
		Batch 3	0.99	0.99	0.99	0.99	0.99	0.99	0.99

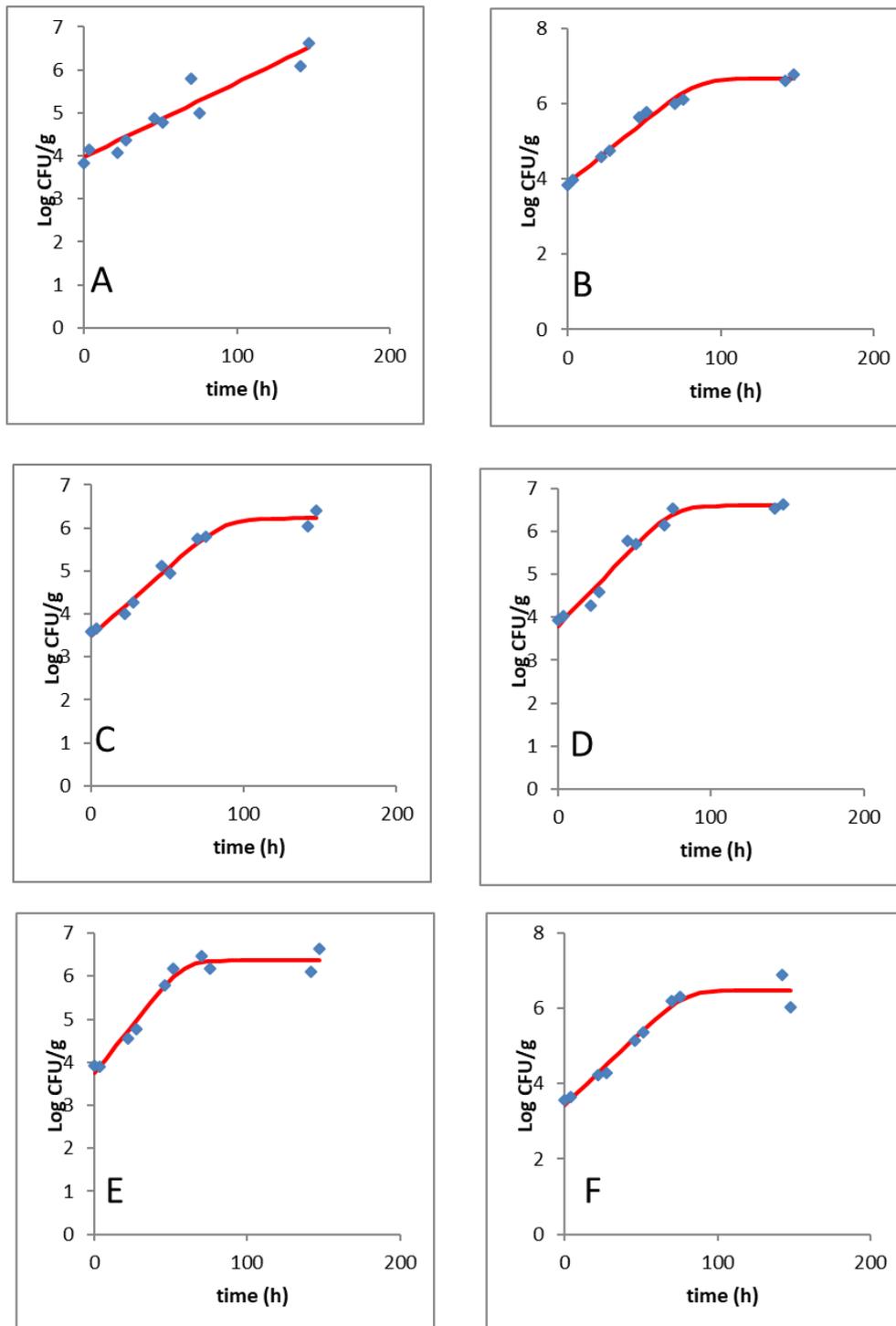
\*mean of three different trays.

Student's *t*-test analysis comparing the  $a_w$  and pH values of uninoculated and blanks test units did not reveal statistically significant differences, and therefore, the inoculum did not appear to influence the behaviour of the *L. monocytogenes* strains used for the challenge.

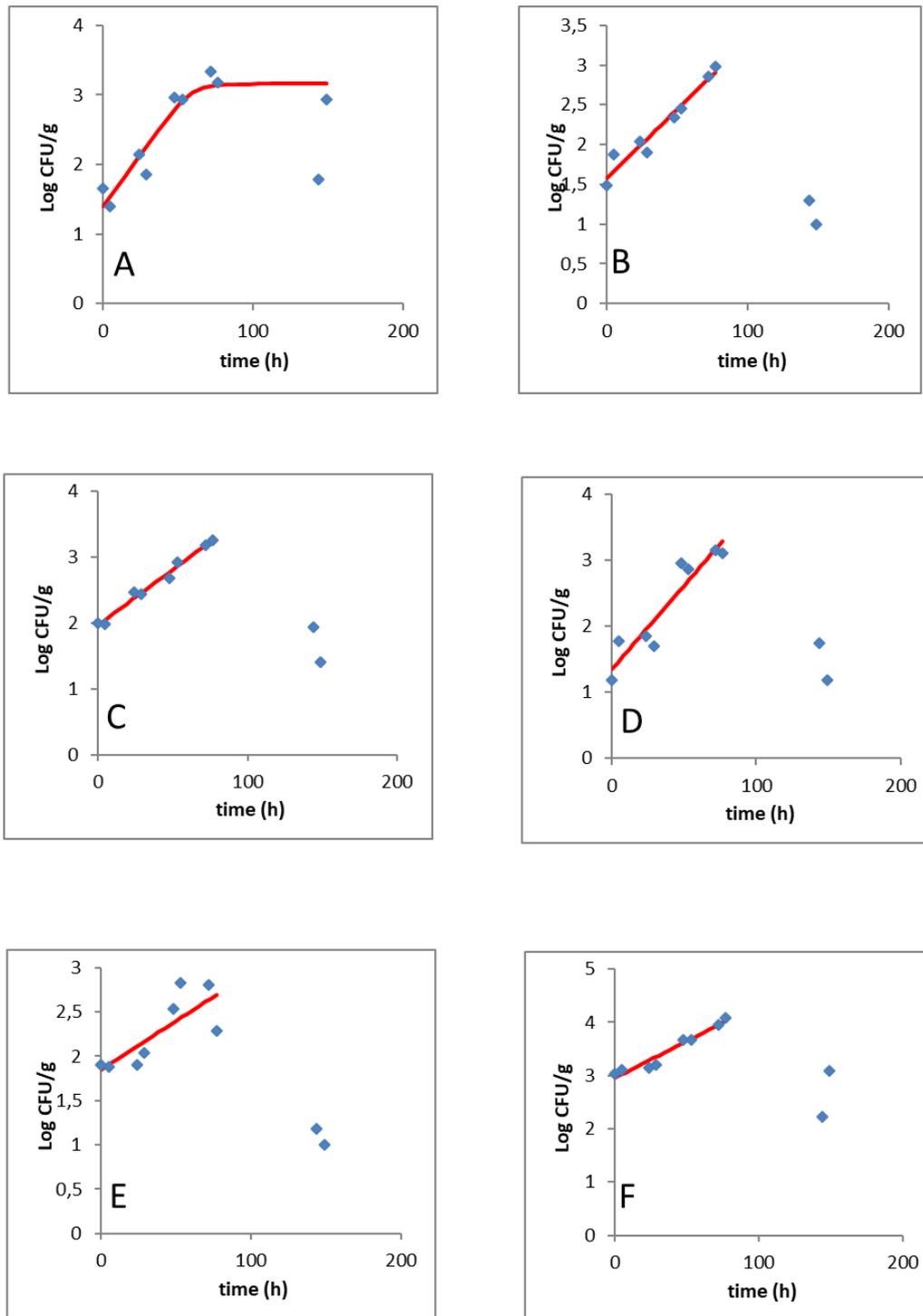
The growth curves for the reference and wild strains of *L. monocytogenes* were fitted using the DMFit software based on the Baranyi and Roberts models.

For papaya, starting from a contamination of 4 log CFU/g, *L. monocytogenes* grew until 6.5 log CFU/g in 149 h (Figure 1). In batch 3, the growth was from 3.5 to around 6 log CFU/g for both strains.

For melon, starting from a contamination of approximately 1.5 - 2 log CFU/g, *L. monocytogenes* grew until 3 log CFU/g in 77 h (Figure 2). In batch 3, the starting inoculum was 3 and reached 4 log CFU/g in 72 h.



**Figure 1.** Growth curves of reference (A, B, C) and wild (D, E, F) *L. monocytogenes* strains on three batches of fresh-cut papaya. Growth rates were calculated using data observed until 77 h.



**Figure 2.** Growth curves of reference (A, B, C) and wild (D, E, F) *L. monocytogenes* strains on three batches of fresh-cut melon. Growth rates were calculated using data observed until 77 h.

The GRs for *L. monocytogenes* were calculated with SE and duplication time (Table 2). The MGR was  $0.045 \pm 0.005$  log CFU/g/h with a Td of 6.77 h

for papaya, and  $0.029 \pm 0.006$  log CFU/g/h with a Td of 10.24 h for melon.

**Table 2.** Output parameters estimated by the DMFit program for each growth curve of two different *L. monocytogenes* strains (R, reference; and W, wild) in three batches of papaya and melon.

Strain		Batch 1			Batch 2			Batch 3		
		GR $\pm$ se (log CFU/g/h)	R <sup>2</sup>	Td (h)	GR $\pm$ se (log CFU/g/h)	R <sup>2</sup>	Td (h)	GR $\pm$ se (log CFU/g/h)	R <sup>2</sup>	Td (h)
Papaya	R	$0.017 \pm 0.002$	0.90	17.34	$0.033 \pm 0.002$	0.98	9.15	$0.031 \pm 0.002$	0.97	9.59
	W	$0.037 \pm 0.004$	0.96	8.10	$0.045 \pm 0.005$	0.96	6.77	$0.039 \pm 0.004$	0.95	7.78
Melon	R	$0.029 \pm 0.006$	0.88	10.24	$0.017 \pm 0.002$	0.93	17.37	$0.017 \pm 0.001$	0.98	17.89
	W	$0.025 \pm 0.004$	0.86	11.91	$0.011 \pm 0.004$	0.54	27.52	$0.014 \pm 0.001$	0.94	21.72

### Indigenous microfloral enumeration

For each batch, the indigenous microflora of the fruit was enumerated in blank test units (Figure 3). For papaya at 149 h, TVC\_M was around 8 log CFU/g, TVC\_P and yeast concentration around 7 - 7.5 log CFU/g, and Ent around 6.5 log CFU/g in batches 2 and 3, and 7.5 in batch 1. LAB concentration reached 6 log CFU/g in batches 1 and 3, and 7 log CFU/g in batch 2. Mould growth did not exceed 2 log CFU/g. For melon, TVC\_P was around 9 log CFU/g, TVC\_M and LAB concentration was around 8 log CFU/g. Ent grew steadily in batches 1 and 2 to 6.5 and 5.5 log CFU/g, respectively, while in batch 3, the concentration stopped at around 4.5 log CFU/g at 48 h. Yeast concentration reached values around 6.5 - 7.5 log CFU/g, as moulds reached a concentration of 7 log CFU/g at day 7 only in batch 2 unlike the other batches, in which the concentration at the last day was around 3 - 3.5 log CFU/g.

### Discussion

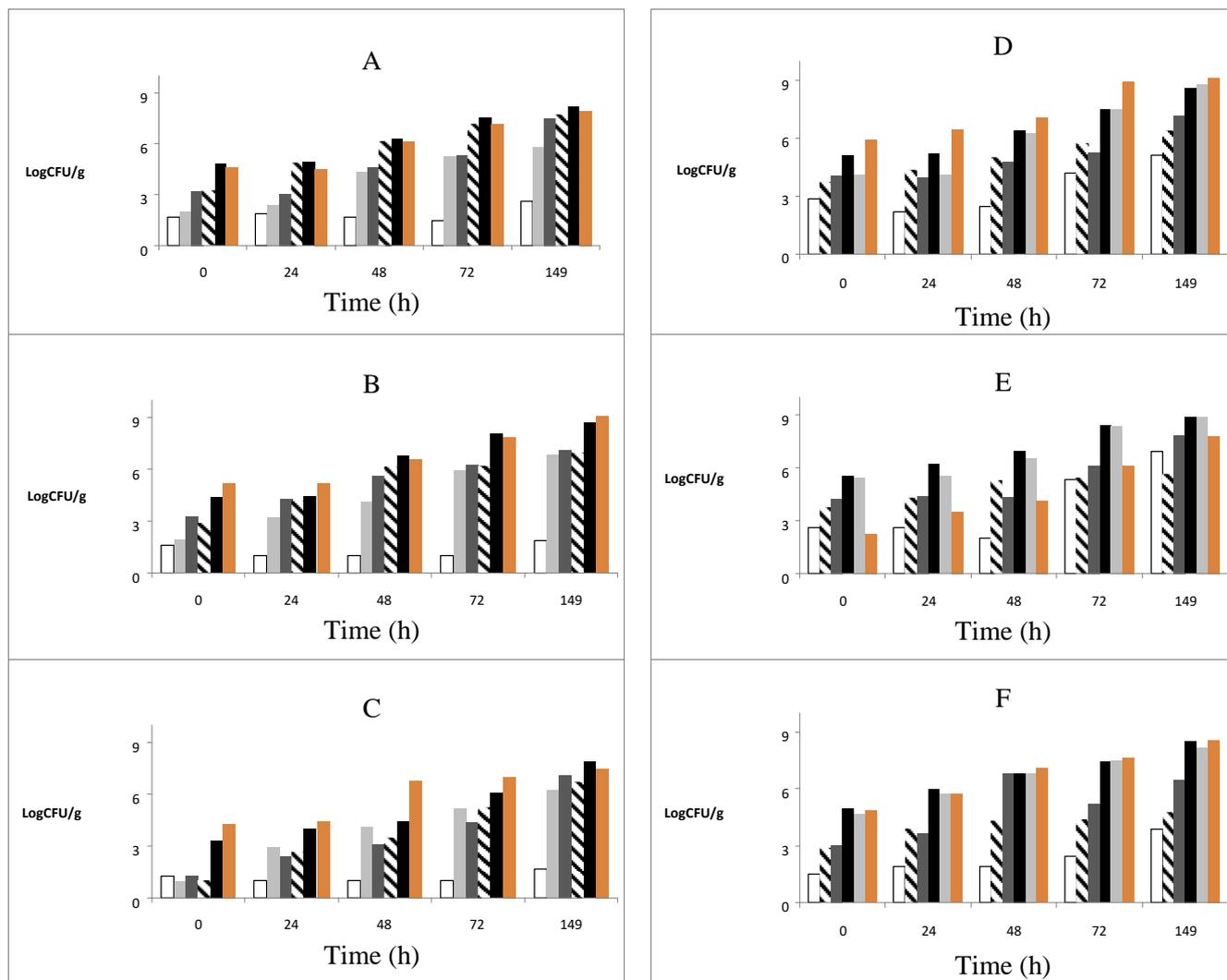
The reasoning behind the present work is that diseases related to the consumption of fresh and minimally processed fruits and vegetables have raised concerns (Nüesch-Inderbinen and Stephan, 2016; Yoon and Lee, 2018; Carstens *et al.*, 2019).

The present work evaluated the growth of *L. monocytogenes* on fresh-cut fruit papaya and melon during the storage period, under moderate temperature-abuse conditions. This evaluation was based on an experimental challenge test in order to calculate the maximum growth rate of two strains of the pathogen for each type of fruit. The challenge test was performed in an unmodified atmosphere, on the same fruit trays that the producer distributed to retailers.

In order to simulate the domestic refrigeration condition, we selected two different incubation temperatures, 8 and 10°C. We wanted to use temperature of slight thermal abuse, similarly to that which was reported in other studies about the growth of *L. monocytogenes* in fruit (Penteado and Leitao, 2004; Huang *et al.*, 2019; Luciano *et al.*, 2021).

The choice of the inoculum level, slightly higher than those reported in other studies (Penteado and Leitao, 2004; Huang *et al.*, 2019), was because of the condition of the packaging. In fact, these fresh samples were packaged without additives, in an unmodified atmosphere, and the indigenous microbial flora could have interfered with the development of *L. monocytogenes* to the point of not allowing assessment of the behaviour (Mellefont *et al.*, 2008). However, the use of a high initial inoculum does not affect the growth of the pathogen. Papaya, stored at 10°C for 7 d, proved to be a favourable substrate for the growth of *L. monocytogenes*, with a growth from 3.5 - 4 to 6 - 6.5 log CFU/g in all the batches for 7 d. Similarly in another study, the growth on papaya pulp artificially inoculated with 2 log CFU/g of *L. monocytogenes* reached about 5 log CFU/g at the end of a 7-d incubation at 10°C (Penteado and Leitao, 2004).

The survival and growth of *L. monocytogenes* on fresh-cut melon stored at an abuse temperature of 8°C have been observed in previous studies: Luciano *et al.* (2021) showed that *L. monocytogenes* populations reached approximately 2 log CFU/g in 7 d starting from an inoculum of 1 - 4 cells/sample; Huang *et al.* (2019) spiked fresh-cut cantaloupe with 2.5 log CFU/g of *L. monocytogenes* and stored it at 8°C for 7 d. The pathogen population increased to as high as 6.72 log CFU/g in 7 d. In the present work,



**Figure 3.** Indigenous microflora in papaya and melon. (A) batch 1 papaya, (B) batch 2 papaya; (C) batch 3 papaya; (D) batch 1 melon; (E) batch 2 melon; and (F) batch 3 melon. White bar = moulds; light grey bar = LAB; dark grey bar = yeasts; black bar = TCV\_M; striped bar = Ent, and orange bar = TCV\_P.

the concentration of *L. monocytogenes* in the fresh-cut melon increased in the first 72 h at 8°C, and then, interestingly, we observed a decrease in growth, except for the reference strain in batch 1. Similar behaviour was also observed in a study on two different varieties of fresh-cut melon (honeydew and cantaloupe) stored at 8°C for 10 d (Collu *et al.*, 2021). Starting from two different degrees of inoculum (0 - 1 and 1 - 2 log CFU/g), *L. monocytogenes* populations increased until day 8 in cantaloupe melon, and until day 2 in honeydew melon, before decreasing. A possible cause of the trend observed in the present work could have been related to the high concentration of indigenous microflora, which might have competed with *L. monocytogenes* for nutrients, in addition to causing the partial acidification of the substrate (average decrease of about 1.5 pH units). A

study found that interactions with the natural indigenous microflora may influence the dynamics of *L. monocytogenes* populations in fresh-cut produce (Francis and O'Beirne, 1998). Campo *et al.* (2001) carried out a study co-infecting *L. monocytogenes* with Enterobacteriaceae and pseudomonas in a minimal medium described by Premaratne *et al.* (1991), and in a minimum medium with reduced concentrations in glucose and amino acids. The authors observed that Enterobacteriaceae reduced the population of *L. monocytogenes* by 2 or 3 log CFU/mL when inoculated at 10°C in the minimal medium, presumably due to competition for glucose and/or amino acids. Furthermore, LAB are considered to be responsible for pathogen inhibition due to the production of lactic acid, which can decrease the pH of the substrate or by other

mechanisms such as the production of inhibitory molecules such as ethanol and bacteriocins, or redox modifications (Leroi, 2010; Linares-Morales *et al.*, 2020). In the present work, the concentration of indigenous microflora in melon increased during storage, except for moulds in batches 1 and 3, reaching high overall values mainly in LAB (8 log CFU/g) and psychrophilic and mesophilic counts (9 and 8 log CFU/g, respectively), while yeast and Ent concentrations reached around 7 and 6 log CFU/g, respectively.

In papaya, the indigenous microflora reached overall values greater than 7 log CFU/g, except for Enterobacteriaceae and LAB, which remained around 6 - 7 log CFU/g, and moulds. However, the natural indigenous microflora did not appear to influence the survival and growth of *L. monocytogenes* during storage. This may be explained by the higher inoculum concentration (3 Log CFU/g), which increased the ability of the *L. monocytogenes* population to compete with the indigenous microflora and to grow in stressful conditions. Moreover, in fresh-cut fruits, the cutting process could create different microenvironments that enable the growth of *L. monocytogenes*, even if the LAB concentration and pH levels in the product are unfavourable (De Cesare *et al.*, 2018).

Some studies examined the MGR of *L. monocytogenes* on various fresh-cut products such as salads (Tucci *et al.*, 2019; Bernardo *et al.*, 2020). Our challenge test measured the MGR and the duplication time, which confirmed the ability of *L. monocytogenes* to grow in RTE fresh-cut papaya and melon under conditions of moderate temperature abuse. Similarly, another study by Salazar *et al.* (2017) evaluated the MGR of three disease-causing serotypes of *L. monocytogenes* in cut cantaloupe flesh stored at 5°C ( $0.025 \pm 0.017$  log CFU/g/h), 10°C ( $0.074 \pm 0.03$  log CFU/g/h), and 25°C ( $0.138 \pm 0.009$  log CFU/g/h). Our data showed that a few hours (on average 9.79 h for papaya, and 17.77 h for melon) in moderate temperature-abuse conditions could be sufficient to double the *L. monocytogenes* microbial charge on fresh-cut papaya and melon.

## Conclusion

The present work confirmed the growth of *L. monocytogenes* in RTE fresh-cut fruit stored in moderate temperature-abuse conditions, highlighting the importance of preventing contamination with *L.*

*monocytogenes* during the production process, and maintaining the cold chain until the product is fully consumed. Our data on the MGR of *L. monocytogenes* may be useful for producers to calculate the optimum time and temperature storage conditions for determining the shelf-life of these RTE fresh-cut fruits.

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