Response of Some Microorganisms to Ultraviolet Treatment on Fresh-cut Pear

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Abstract Ultraviolet energy at a wavelength of 253.7 nm (UV-C) was investigated for its microbicidal effects on pear slices with and without peel. Effectiveness of UV-C light against *Listeria innocua* ATCC 33090, *Listeria monocytogenes* ATCC 19114 D, *Escherichia coli* ATCC 11229, and *Zygosaccharomyces bailli* NRRL 7256 individual strains was tested in both types of pear slices. In a second experiment, strain cocktails of *Listeria* (*L. innocua* ATCC 33090; *L. innocua* CIP 8011 and *L. welshimeri* BE 313/01), *L. monocytogenes* (*L. monocytogenes* ATCC 19114 D, *L. monocytogenes* ATCC 7644), and yeasts (*Z. bailii* NRRL 7256, *Zygosaccharomyces rouxii* ATCC 52519, and *Debaryomyces hansenii* NRRL 7268) were also used as inocula and compared with single-strain responses. Inoculated pear slices were exposed to a UV-C dose range between 0 and 87 kJ/m^2 and enumerated to determine log reductions of microbial populations. Overall, as UV-C dose was increased (increasing the time of exposure), more inactivation was observed for all species assayed. UV-C irradiation appeared to improve microbial inactivation in pear slices without peel. In most experiments, great log reduction rates were observed at doses between 0 and 15 kJ/m^2. Inactivation kinetics was successfully fitted using a Weibullian distribution of resistances model. Narrow frequency shapes strongly skewed to the right were

obtained. This model offers improved tools for designing and implementing UV-C light treatment and assessing the impact of some microorganisms' disease.

Keywords UV-C light · Microbial inactivation · Fresh-cut pear · Weibull distribution

Introduction

Pear (*Pyrus communis L*) is a versatile fruit popular around the world as a fresh fruit product but consumed also in processed forms such as canned and juice. The fresh use sector accounts for a major part of world pear use. Argentina is the largest supplier of imported pear into the USA (USDA 2004) and the leading exporter in the Southern hemisphere. Because of transportation problems or when pears produced for exportation are rejected because of size, shape, peel stains, or other factors, a considerable amount of pear production is wasted (FDA 2001).

The composition of the fruit tissue, the pH value, and type of acid itself play a major role on the nature and extension of the microbial populations that grow on the product (Ahvenainen 1996; Martinez et al. 2000). Because of their high contents in organic acids, some fruits have low pH values, a fact that protects them from most pathogenic strains. However, some *Listeria* species have demonstrated to survive in fruit produces, like apple, pineapple, white grape, and orange juice concentrates with pH values between 3.6 and 5.5 (Parish and Higgings 1989; Martinez et al. 2000; Oyarzabal et al. 2003). Besides, mechanical operations like peeling, coring, cutting, or slicing may cause cells to rupture, leading to biochemical changes, physiological aging, and microbial spoilage of the minimally processed produce. Rosen and Kader (1989) reported

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a lack of wounding response in sliced pears. In particular, pear fruit is a suitable substrate for various microorganisms including *Listeria* species, *Salmonella* spp, *Aeromona hydrophila*, *Yersinia enterocolitica*, *Escherichia coli*, moulds, and yeasts (Soliva-Fortuny and Martín-Belloso 2003; Martinez et al. 2000).

Minimal processing of fruits appears to have potential markets and opens new possibilities for a better utilization of the fruit (Leistner 2000). It includes a wide range of novel technologies for preserving them while minimizing changes to their fresh like characteristics and improving shelf life. Nonthermal processes applied to food preservation without the collateral effect of heat treatments are being deeply studied and tested (Gould 1995). Fresh-cut fruits are still under study because of the difficulties in preserving fresh-like quality during prolonged periods. The use of techniques to extend the shelf life of fresh-cut produce, such as modified atmosphere packaging, may allow microorganisms more time to grow before the product appears unacceptable for consumption (Sapers and Miller 1998). Therefore, it becomes necessary for an integrated approach aimed at preventing contamination, effective sanitization treatments, and adequate cold chain management to ensure product safety.

A good procedure to reduce the microbial risk involved with the consumption of whole or cut fresh fruits includes the reduction in or elimination of external contamination by using surface decontamination techniques (Yaun et al. 2004). A superficial treatment that can aid refrigeration for preserving fruits is the radiation of foods with short-wave ultraviolet (UV-C) light (Allende et al. 2006). UV light is a radiation in the range of 200 to 400 nm. In particular, radiation between 200 and 280 nm (UV-C) penetrates membrane cells and breaks down the deoxyribonucleic acid (DNA) molecules through dimmer formation, rendering them incapable of reproduction (Shama 1999), which results in a germicidal effect (Bintsis et al. 2000). Because of the wide variety of organisms present on food surfaces, the dose levels required for disinfection vary within food products. It does not produce byproducts or generate chemical residues that could change the sensory characteristics in the final product (Guerrero-Beltrán and Barbosa-Cánovas 2004). Another advantage is that it does not deliver residual radioactivity as ionizing radiation. Furthermore, it is a cold and dry process requiring very low maintenance (Morgan 1990). However, UV radiation has a limited penetration depth (Bintsis et al. 2000). Therefore, it is necessary to apply UV to a thin surface. Several researchers have demonstrated that UV light can be used for the inactivation of pathogens without adversely modify the overall quality of food (Allende and Artes 2003; Smith et al. 2002; Yaun et al. 2004; Geveke 2005). Most research on UV-C light treatments focuses on microorganism and/or produce effects and

not on inactivation kinetics modeling, an issue that has not been reported so far.

The present study was undertaken to investigate the effect of UV-C light at various exposure times on survival of some microorganisms on fresh-cut pear slices with and without peel. Additionally, the response of pooled species was studied. A Weibullian-type model was used to mathematically characterize and compare inactivation curves.

Materials and Methods

Tested Cultures and Preparation

L. innocua ATCC 33090, *L. monocytogenes* ATCC 19114 D, *E. coli* ATCC 11229, and *Z. bailli* NRRL 7256 single inocula were tested. Bacteria strains were subcultured, purified weekly in trypticase soy broth supplemented with 0.1% w/w yeast extract (TSBYE) and trypticase soy plus yeast extract agar (TSAYE) at 37 °C and stored at 4 °C. The initial inoculum was prepared by transferring a loopful of a stock culture maintained on agar slants to 20 mL of TSBYE contained in 50-mL Erlenmeyer flasks. The microorganisms were incubated at 37 °C (±1 °C) until it reached the stationary phase (≈24 h). A similar procedure was repeated for yeast cultures. The initial inoculum was prepared by transferring a loopful of a stock culture maintained on potato dextrose agar slants to flasks with 20 mL of potato dextrose broth. The organism was grown at 27 °C (±1 °C) until it reached the stationary phase (≈36 h).

In a second experiment, equal aliquots of each individual strain in the stationary phase were vortexed and then aseptically combined into a sterile dilution blank to produce a cocktail of three strains of *Listeria* (*L. innocua* ATCC 33090, *L. innocua* CIP 8011, and *L. welshimeri* BE 313/ 01), two trains of *L. monocytogenes* (*L. monocytogenes* ATCC 19114 D, *L. monocytogenes* ATCC 7644), and three strains of yeasts (*Z. bailii* NRRL 7256, *Z. rouxii* ATCC 52519, and *D. hansenii* NRRL 7268). All microbiological media were purchased from Britania (Laboratorios Britania S.A., Argentina).

Preparation of Produce Samples

Ripened pears (*William* cv, pH ~4.2) were purchased in a local market and processed within 1 day of purchase. They were rinsed with 0.02% sodium hypochlorite for 2 min and sterile water to eliminate the surface microbial load before cutting and gently dried with a sterile cloth. Pears were sliced into 3-mm-thick pieces. A household machine specially adapted to obtain slices of equal thickness was disinfected with 1% hypochlorite solution and equipped with backed stainless steel razor blades. A stainless steel

sharp cutting disk was used for obtaining pear disks (3 cm in diameter). Disks with peel were obtained from the external slices. Finally, disks with or without peel (~2.5 g in weight) were transferred into a sterile Petri dish. The overall preparation of produce samples was made inside a Class II Security Cabinet (Nuaire, Plymouth, MA) to prevent postcontamination.

Produce Inoculation

Mixed or single inocula (0.1 mL) were widespread with an alcohol-flamed glass spreader onto the surface of each pear disk with or without peel to mimic mid-/postprocessing contamination at initial levels of approximately 10^5–10^6 CFU/g of slice pear (~4×10^4–4×10^5 CFU/cm^2). The inoculated pear disks were immediately treated in the UV-C chamber. Inoculated and nonirradiated pear disks were used as controls.

Ultraviolet Chamber and Treatment

The UV-C irradiation device consisted of one bank of two reflectors with unfiltered germicidal emitting lamps (TUV-15W G13 T8 55V germicidal lamp, Philips, Holland) that emit 253.7-nm UV light, located 10 cm above the produce tray. The UV-C lamps and treatment area were enclosed in a wooden box covered with aluminum foil with a cover protection for the operator. A ventilation device was installed in a corner of the box to avoid temperature increase because of UV radiation. The mean temperature during the treatments was 27 ± 1 °C. Before use, the UV-C lamps were allowed to stabilize by turning them on at least 15 min.

The UV-C dose emitted from the lamps was determined by using the iodide/iodate chemical actinometer according to the technique proposed by Rhan (1997). The UV-C radiation doses 0, 15, 31, 35, 44, 56, 66, 79, and 87 kJ/m^2 were obtained by altering the exposure time up to 20 min.

For the UV-C treatments, a Petri dish with three inoculated disks was placed over the center line of the treatment tray for each treatment.

Enumeration

After UV-C irradiation, the disks contained in the Petri dish were immediately put into stomacher bags (Whirl-Pak, Nasco, USA) containing 15 mL of sterile peptone water and were pummeled in a Laboratory blender (AES Laboratoires, France) at high speed for 3 min. Tenfold dilutions of homogenate samples were made in 0.1% w/v peptone water, and 0.1 mL sample suspension was surface plated using tryptone soy agar (bacteria) or potato dextrose agar (yeasts). Two plates were used for each dilution. When UV-C irradiation treatment resulted in low counts, up to 3 mL of homogenate was directly pour plated using four plates as replicates. Plates were

incubated at 37 °C, in the case of bacteria, or 27 °C for the yeasts for 72 h. A colony counter (Comecta S.A., Spain) was used for enumeration of colonies. Survival curves were generated from experimental data by plotting log N/N_0 (where N is the number of CFU/cm^2 at a given time and N_0 the initial number of CFU/cm^2) vs time of treatment.

Statistical Analysis

Trials were replicated at least three times with three samples for each ultraviolet dose. Nonlinear regressions were carried out using the STATGRAPHICS PLUS for Windows 3.0® Package (Statistical Graphics, Washington, USA). Internal validation of the model was carried out through the comparison between the observed and the predicted values and the evaluation of the adjusted coefficient of determination $\left(R^2_{\mathrm{adj}} \right)$.

Mathematical Modeling

Survival curves ($S(t)$) were assumed to reflect lethal events having a Weibull distribution (Peleg and Cole 1998):

$$\mathrm{Log}_{10} S(t) = \mathrm{Log}_{10}(N/N_0) = -bt^n \qquad (1)$$

where b and n are constants. The values of b and n were then used to generate the frequency distributions of resistances using the following equation:

$$\frac{\mathrm{d}\phi}{\mathrm{d}t_\mathrm{c}} = bnt_\mathrm{c}^{n-1} \exp\left(-bt_\mathrm{c}^n\right) \qquad (2)$$

where t_c is a measure of the organism's resistance or sensitivity and $\frac{\mathrm{d}\phi}{\mathrm{d}t_\mathrm{c}}$ is the Weibull distribution corresponding to t_c. Other statistical parameters that characterize the distribution (distribution mode, t_cm; mean, t_c; variance, σ^2_{tc}; and coefficient of "skewness", v_1) were calculated from the following equations (Peleg and Cole 1998):

$$t_\mathrm{cm} = [(n-1)/nb]^{1/n} \qquad (3)$$

$$\bar{t}c = \left\{ \Gamma[(n+1)/n] \right\} \big/ b^{1/n} \qquad (4)$$

$$\sigma^2_{tc} = \left\{ \Gamma[(n+2)/n] - (\Gamma[(n+1)/n])^2 \right\} \big/ b^{2/n} \qquad (5)$$

$$v_1 = \frac{\left\lfloor \Gamma(n+3/n) \big/ b^{3/n} \right\rfloor}{[\Gamma(n+2/n)/b^{2/n}]^{3/2}} \qquad (6)$$

$$\Gamma(z) = \int_0^\infty t^{z-1} \times e^{-t} \mathrm{d}t \qquad (7)$$

where Γ is the gamma function (Eq. 7), an extension of the factorial function to real and complex numbers. The distribution mode, t_{cm}, represents the treatment time at which the majority of population dies or inactivates. The mean, t_c, corresponds to the inactivation time on average with its variance, σ_{tc}^2. The "skewness" coefficient, v_1, represents the skew of the distribution.

Results and Discussion

L. innocua, *L. monocytogenes*, *E. coli*, and *Z. bailii* counts after 1 h of being inoculated in fresh cut pear slices without UV-C irradiation (controls) showed in average 0.5 log reduction (data not shown), revealing that these microorganisms can survive in this fruit.

The first series of experiments were carried out to determine the inactivation of some species on pear by UV-C light. Figure 1 showed the destruction kinetics of *L. innocua*; *L. monocytogenes*; *E. coli* and *Z. bailii* in fresh cut pear slices with and without peel (Fig. 1). UV-C treatment significantly reduced microbial load in all cases. Semilogarithmic survival curves were clearly nonlinear. The presence of peel greatly modified the curve profiles. For all studied species, the UV-C treatment was more effective in pear slices without the presence of peel. The log reduction ranges varied between 2.6 and 3.4 log cycles for pear slices without peel (Fig. 1a) and 1.8 and 2.5 log cycles for treated samples with peel (Fig. 1b) after 20-min treatments (corresponding to 87-kJ/m^2 UV-C dose).

In pear slices without peel subjected to doses larger than 44 kJ/m^2, *L. innocua* and *L. monocytogenes* were the most sensible microorganisms, showing similar survival profiles (Fig. 1a). In contrast, at lower doses, *E. coli* and *Z. bailii* survival curves were the most sensible ones. However, for irradiation times larger than ≈7 min (33 kJ/m^2), the inactivation rate significantly fell down. This fact would suggest that the remaining members of the population were or became more resistant.

When the species were inoculated onto pear slices with peel (Fig. 1b), the survival patterns changed. Overall, microorganisms showed a less but constant decrease in population as UV-C dose increased. *E. coli* and *L. innocua* were the most sensible species, while scarce differences in survival patterns were observed for *L. monocytogenes* and *Z. bailii*.

There were notorious differences in survival curves of *Listeria* species depending on the presence of peel. In pear slices with peel (Fig. 1b), *L. innocua* showed no adequacy to mimic *L. monocytogenes* response staying in discrepancy with the findings reported by many authors for other inactivation techniques (Moseley 1990; Coma et al. 2001).

In this study, the use of UV-C light to reduce microbial populations led to the formation of a curve with upward
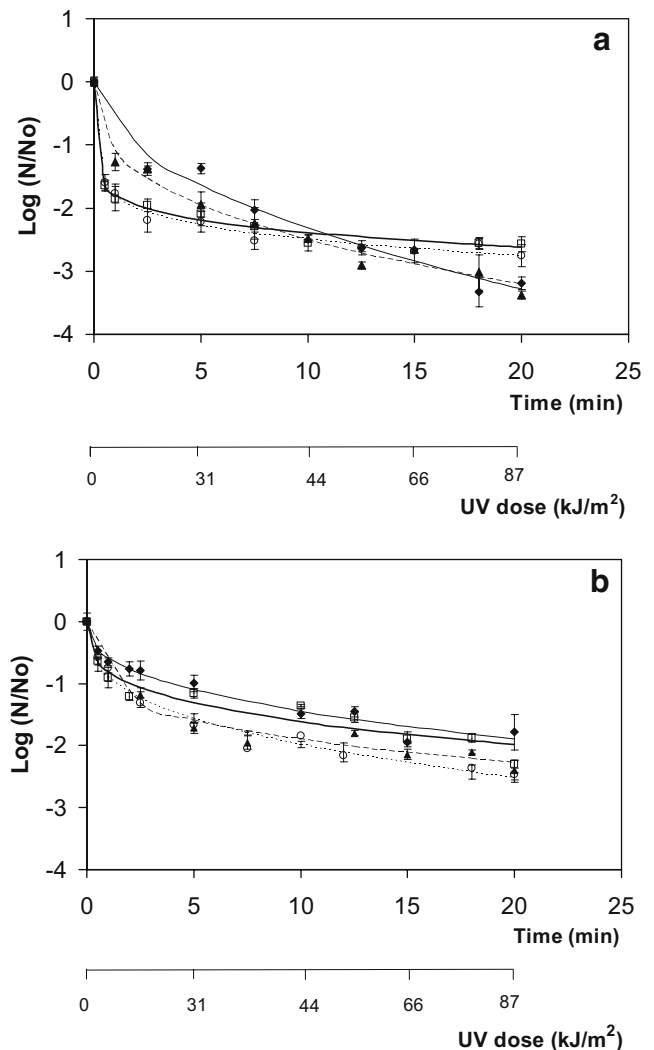

Fig. 1 Semilogarithmic survival curves of some microorganisms in pear slices irradiated with UV-C light. Experimental (*points*) and fitted values derived from the Weibullian model (*lines*): a Pear slices without peel, b pear slices with peel. L. *innocua* ATCC 33090 (*triangles on dashed line*), L. *monocytogenes* ATCC 19114 D (*diamonds on thin solid line*), E. *coli* ATCC 11229 (*circles on dotted line*), and Z. *bailli* NRRL 7256 (*squares on thick solid line*); standard deviation (*I*)

concavity and pronounced tailing effect. Other researchers have also reported that UV energy inactivates bacteria exponentially with a notorious tail (Yousef and Marth 1988; FDA 2000). Yaun et al. (2004) studied the inhibition of some pathogens by ultraviolet light on the surface of fresh tomatoes, lettuce, and delicious apples. The observed highest log reduction was approximately 3.3 log cycles corresponding to *E. coli* O157:H7 inoculated in apples subjected to a 0.24-kJ/s m^2 dose. As in this study, they observed in all experiments a well-defined tail in the survival curves. On the other side, results here obtained do not support the presence of a shoulder, as mentioned by other authors for UV-C inactivation (Yousef and Marth 1988). This behavior could be attributed to the fact that the minimum

Table 1 Estimates of Weibullian distribution parameters[a, b]; standard errors, and adjusted R^2 for microorganism survival in UV-C light irradiated pear slices without peel

Microorganism	b	n	R^2_{adj}	$\bar{t}_c$ (min)	σ^2_{tc} (min^2)	$v_1(-)$
Single strain						
L. monocytogenes ATCC 7644	0.73±0.11	0.51±0.06	98.0	3.7±0.1	65±5	6±1
L. innocua ATCC 33090	1.10±0.09	0.36±0.03	98.0	3.6±0.1	190±5	15±1
Z. bailii NRRL 7256	1.79±0.04	0.13±0.01	99.1	+++	+++	+++
E. coli ATCC 11229	1.82±0.05	0.14±0.01	99.0	+++	+++	+++
Mixture of strains[c]						
Cocktail 1	0.97±0.10	0.28±0.04	97.8	15.2±0.3	+++	38±2
Cocktail 2	1.33±0.09	0.16±0.03	96.7	+++	+++	+++
Cocktail 3	1.40±0.07	0.26±0.02	98.8	5.1±0.2	+++	49±2

[a] b, n Constants of Weibullian model, R^2_{adj} adjusted determination coefficient, $\bar{t}_c$ distribution's mean, σ^2_{tc} variance, v_1 coefficient of skewness
[b] +++ Value of the statistic greater than 100
[c] Cocktail 1 L. monocytogenes ATCC 19114 D and L. monocytogenes ATCC 7644, Cocktail 2 L. innocua ATCC 33090, L. innocua CIP 8011, and L. welshimeri BE 313/01, Cocktail 3 Z. bailii NRRL 7256, Z. rouxii ATCC 52519, and D. hansenii NRRL 7268

employed dose exceeded that necessary for initial cellular injury. Initial exposure of bacteria to UV-C light is believed to injure cells (FDA 2000). As increasing dose levels of UV-C are received, mutations arise in the DNA code where neighboring pyrimidine bases begin to form cross-linkages that impede cellular replication. Cellular death occurs after the threshold of cross-linked DNA is exceeded (EPA 1999).

The tail in UV-C inactivation curves has been explained in several different ways. One explanation is the multiple hit phenomena described by Yousef and Marth (1988), which states that the survival curve was accounted for on the basis of multiple UV hits on a single cell or single UV hits on multiple cells. In this scenario, the less resistant cells are inactivated first, leaving the more resistant cells to form a tail. The tailing has also been interpreted as caused by the presence of matrix solids that may block the UV irradiation (FDA 2000). Some such solids may include particles from the pear slice and other environmental contaminants such as

dust or dirt. The heterogeneity in the resistances of the population to UV-C irradiation (Block 2000) and the presence of suspended solids (FDA 2000) are other reasons for the tail existence.

Figure 1 also shows the fitting of experimental inactivation data using the cumulative Weibull distribution function described by Eq. 1. Experimental curves were adequately correlated to predicted data, with very significant determination coefficients R^2_{adj} (Tables 1 and 2), showing that between 93.1 and 99.8% of variation in the experimental data could be explained by the selected model. These results could be accepted as a good correlation considering the heterogeneity of the experimental system assayed and the inoculation technique employed.

The Weibull distribution is a flexible nonlinear model that has been proved to describe well the inactivation of microorganisms by several factors, such as heat (Peleg and Cole 1998; Peleg 1999), pulsed electric fields (Peleg 1995), radiation (Anellis and Werkowski 1968), high pressure (Heinz and Knorr

Table 2 Estimates of Weibullian distribution parameters[a, b]; standard errors, and adjusted R^2 for microorganism survival in UV-C light irradiated pear slices with peel

Microorganism	b	n	R^2_{adj}	$\bar{t}_c$ (min)	σ^2_{tc} (min^2)	$v_1(-)$
Single strain						
L. monocytogenes ATCC 7644	0.58±0.06	0.39±0.04	96.7	13.7±0.5	+++	11±2
L. innocua ATCC 33090	1.05± 0.14	0.26±0.05	96.3	16.2±0.5	+++	50±3
Z. bailii NRRL 7256	0.81±0.10	0.30±0.05	93.1	18.5±0.6	+++	27±4
E. coli ATCC 11229	0.90±0.07	0.35±0.03	97.7	7.2±0.3	+++	17±2
Mixture of strains[c]						
Cocktail 1	0.76±0.02	0.37±0.01	99.8	9.2±0.1	+++	14±1
Cocktail 2	0.74±0.03	0.26±0.02	99.1	66.2±0.1	+++	53±1
Cocktail 3	0.84±0.07	0.31±0.03	98.1	14.6±0.2	+++	25±2

[a] b, n Constants of Weibullian model, R^2_{adj} adjusted determination coefficient, $\bar{t}_c$ distribution's mean, σ^2_{tc} variance, v_1 coefficient of skewness
[b] +++ Value of the statistic greater than 100
[c] Cocktail 1 L. monocytogenes ATCC 19114 D and L. monocytogenes ATCC 7644, Cocktail 2 L. innocua ATCC 33090, L. innocua CIP 8011, and L. welshimeri BE 313/01, Cocktail 3 Z. bailii NRRL 7256, Z. rouxii ATCC 52519, and D. hansenii NRRL 7268
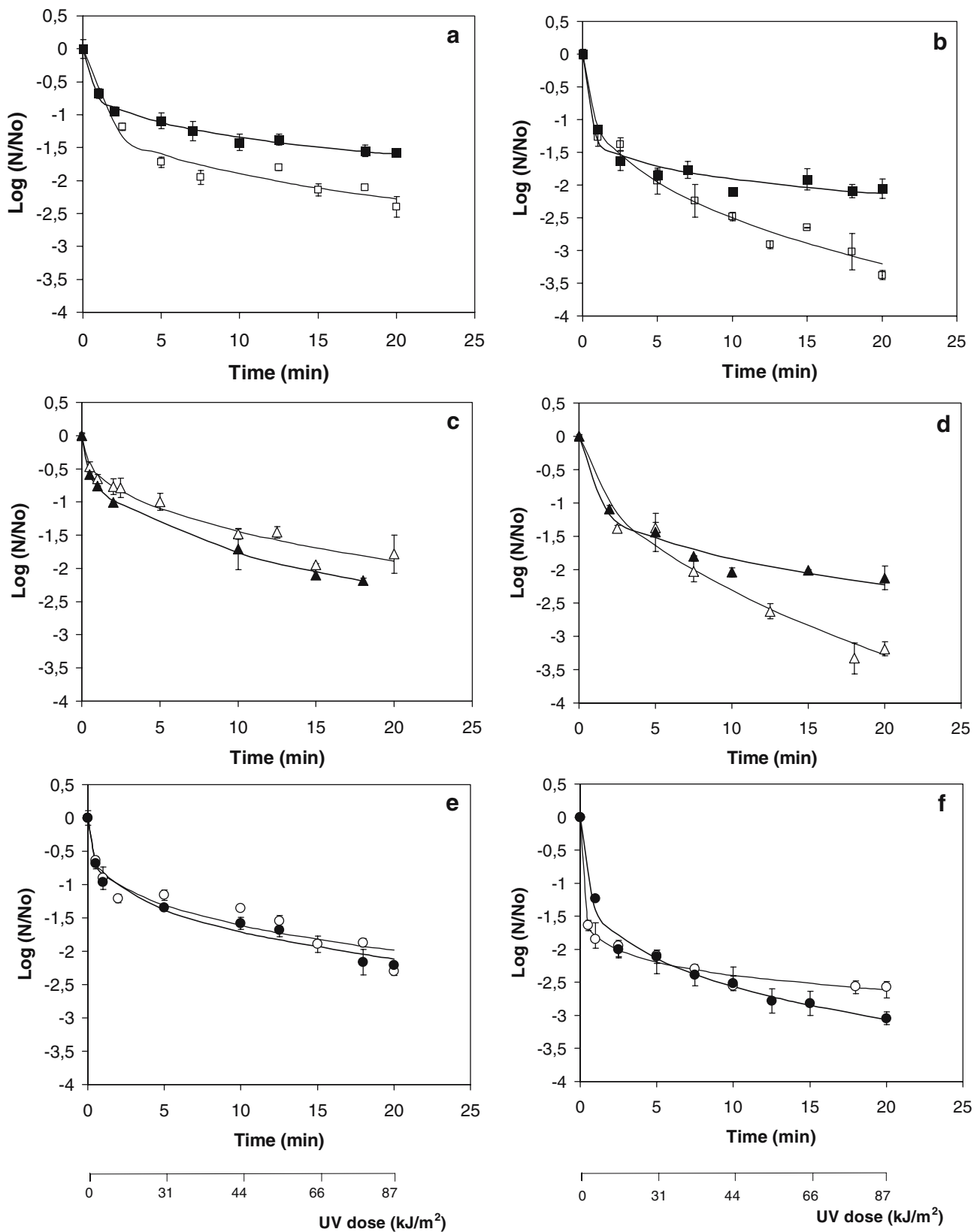
Fig. 2 Effect of UV-C irradiation on semilogarithmic survival curves of single (*open symbols*) and mixed strains (*filled symbols*) inoculated on pear slices with (**a**, **c**, **e**) or without (**b**, **d**, **f**) peel. Experimental (*points*) and fitted values derived from the Weibullian model (*lines*). **a**, **b** *L. innocua* ATCC 33090 (*open squares*) and Cocktail 2 (*filled squares*); **c**, **d** *L. monocytogenes* ATCC 19114D (*open triangles*) and Cocktail 1 (*filled triangles*) and **e**, **f** *Z. bailii* NRRL 7256 (*open circles*) and Cocktail 3 (*filled circles*); standard deviation (*I*)
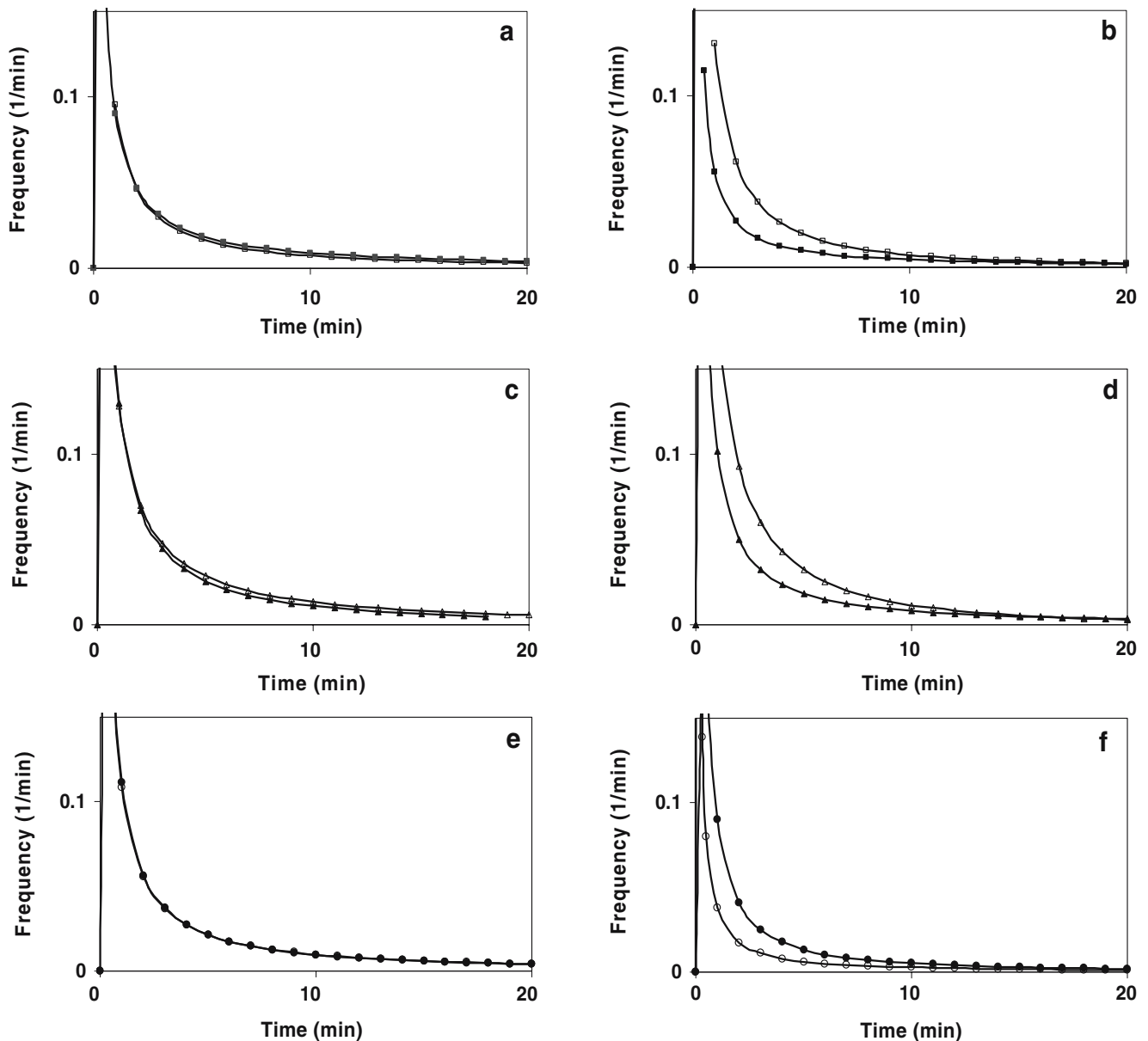
Fig. 3 Weibull frequency distributions of resistances corresponding to survival curves of different microorganisms in UV-C-irradiated pear slices with (a, c, e) or without (b, d, f) peel. a, b *L. innocua* ATCC 33090 (*open squares*) and Cocktail 2 (*filled squares*); c, d *L. monocytogenes* ATCC 19114D (*open triangles*) and Cocktail 1 (*filled triangles*) and e, f *Z. bailii* NRRL 7256 (*open circles*) and Cocktail 3 (*filled circles*)

1996), natural antimicrobials (Corte et al. 2004), and high-intensity ultrasound (Guerrero et al. 2005; Ferrante et al. 2007). This model considers that there is heterogeneity between microbial cells of a population. A single microorganism is either alive or it dies because of lethal agents such as heat, disinfectants, pressure, and radiation. It is unlikely that all cells behave in the same way. Rather, the inactivation time varies to some extent for each microorganism in a population, even if the population is pure (van Boekel 2002). Consequently, the survival curve could be assumed as the cumulative form of the underlying distribution of the individual inactivation times (Peleg and Cole 1998).

Tables 1 and 2 show the underlying b and n regression parameters derived from applying this model to the experimental data. The statistical parameters associated to each frequency distribution (mode, mean, variance, and skewness coefficient) calculated according to Eqs. 3 to 6 are also consigned. The parameter n or shape parameter was less than 1 for all the inactivation curves, as expected according to the notorious upward concavity.

Experiments using mixtures of strains were also carried out to analyze the change in the inactivation response because of the presence of other strains. Figure 2 shows the experimental inactivation data and the predicted ones using

the Weibullian model for different cocktails of strains inoculated onto pear slices with or without peel. Survival curves of individual strains obtained in the first part of this study were also plotted to make a better comparison. The regression parameters corresponding to the inactivation of strain mixtures are also shown in Tables 1 and 2.

The frequency distributions generated according Eq. 2 are shown in Fig. 3. All frequency distributions exhibited similar right-skewed shapes with a considerable spread of data, with tails, without mode, and with large variance values. These frequency shapes evidenced that the majority of the organisms were destroyed in a short time during UV-C exposure while a fraction of the population survived after the treatment (Peleg and Cole 1998). The anomalous high values of variance and skewness could be related to the fact that the microorganisms were attached to the pear surface during the UV-C treatment. There is no information about the application of a Weibullian-type model to survival curves corresponding to microorganisms inoculated onto a solid surface. Some studies made with biofilms mentioned the difficulty of observing a homogeneous response because of several factors like different distribution of cells in the biofilm and the existence of a nonuniform biolayer onto the considered surface (Ganesh and Anand 1998). Furthermore, the cells could suffer a nonequal UV-C treatment because of the irregularities in the surface that may shield microorganisms from the UV rays. This heterogeneity, in conjunction with an intrinsic distribution of resistances to UV-C within the population, could be responsible of the great values assumed by the statistics associated to the distributions.

The cocktails of strains showed survival curves similar in shape ($n<1$) to the ones of individual strains, but the resistance patterns (especially at UV-C doses greater than 10–15 kJ/m^2) were highly dependent on the considered microorganism and the presence of peel. In pear slices without peel (Fig. 2b, d), L. monocytogenes and L. innocua cocktails showed similar behavior being markedly more resistant than individual cultures at high UV-C doses. This fact was confirmed by the analysis of the n parameter values (Table 1). If $n\ll1$, the remaining cells would have less probability of dying, indicating that the remaining cells were the sturdy ones, or perhaps adapting to the stress (van Boeckel 2002), or not receiving UV rays because of the topography of the surface.

Figure 2a, c depicts the inactivation curves for the same cultures inoculated on pear slices with peel. Again, the mixture of strains of L. monocytogenes and L. innocua showed different inactivation rates than the individual cultures. The two-strain mixture of L. monocytogenes was a little more sensible to the UV-C treatment than L. monocytogenes ATCC 19114 (Fig. 2c). This fact was also evidenced by the analysis of the correspondent frequency distribution of resistances and its related statistics (Fig. 3c

and Table 2). Both distributions were strongly skewed to the right with large variances and similar skewness coefficient, but the mixture of strains of L. monocytogenes showed a lower value of mean death time, t_c (9.2 against 13.7 min), indicating in the case of the mixture that a greater proportion of cells died before (Table 2).

The observed inactivation curves corresponding to the three-strain mixture of yeasts and Z. bailii to UV-C irradiation were very different from bacteria responses (Fig. 2e, f). In pear slices without peel, the pooled yeast culture was more resistant than Z. bailii at very low times of treatment (<1 min, UV-C dose=7 kJ/m^2). However, as the irradiation process continued, the opposite response was observed. UV-C rays provoked three log reductions in the three-strain mixture at the end of treatment (20 min, 87 kJ/m^2), while the population of Z. bailii remained almost constant (~2.5 log reductions; Fig. 2f). This type of inactivation curve with a tailing effect generated a frequency distribution with very large variance and mean death time values (Table 1). When the yeasts were inoculated onto pear slices with peel, lesser inactivation was observed (~2 log reductions), with no or scarce differences between mixed and individual cultures (Fig. 2e).

Conclusions

The use of UV-C has proved to be effective at reducing microbial populations on the surface of fresh cut pear slices. UV-C was more effective on the surface of pears without peel than in pears with peel. Logarithmic reductions between 2 and 3.4 logs were possible with an appropriate dose of radiation. The different survival patterns were successfully described in terms of the Weibullian distribution. This model led to a better explanation about the influence of the UV-C radiation on microorganism inactivation evidencing differences between survival patterns that did not possibly surge by applying traditional survival models.

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