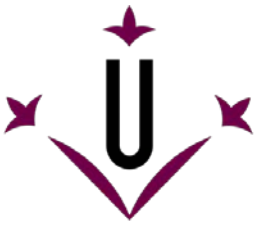

Relevance of the main postharvest handling operations on the development of brown rot disease on stone fruits

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Abstract

BACKGROUND: Brown rot caused by *Monilinia* spp. is one of the most important postharvest diseases of stone fruit. The aim of this study was evaluate the relevance of the main postharvest operations of fruit; hydrocooling, cold room, water dump, sorting and cooling tunnel in the development of *M. laxa* on peaches and nectarines artificially infected 48, 24 or 2 hours before postharvest operations.

RESULTS: Commercial hydrocooling operation reduced incidence to 10 % in 'Pp 100' nectarine inoculated 2 and 24 hours before this operation however in 'Fantasia' nectarine incidence was not reduced, although lesion diameter was decreased in all studied varieties. Hydrocooling operation during 10 min and 40 mg L^{-1} of sodium hypochlorite reduced brown rot incidence between 50 to 77 % in nectarines inoculated 2 hours before operation, however in peaches varieties was not reduced. Water dump operation showed reduction of incidence on nectarine infected 2 hours before immersion during 30 seconds in clean water at 4 ºC and 40 mg L^{-1} of sodium hypochlorite however in peaches varieties was not reduced. Cold room, sorting and cooling tunnel operation did not reduce brown rot incidence.

CONCLUSION: From all studied handling operations on stone fruits packinghouses, hydroccoling is the most relevant on the development of brown rot disease. Duration of the treatment seems to be more important than chlorine concentration. In addition, hydrocooling and water dump were less relevant on peaches than in nectarines. As a general trend, hydrocooling and water dump reduced incidence on fruit with recent infections (2 or 24 hours before operation) however when infections have been established (48 hours before operation) diseases was not reduced.

Keywords: *Monilinia laxa*, water dump, hydrocooling, cooling tunnel, sorting, cold room

1. Introduction

Brown rot caused by *Monilinia* spp is the main disease of stone fruit in the Ebro Valley (Spain) and many other stone fruit production areas around the world. Brown rot in peaches (*Prunus persica*) or nectarines (*P. persica* var. *nectarina*) is caused mainly by two species, *Monilinia laxa* (Aderh.et Rulh.) and *Monilinia fructicola* (G. Wint.) Honey (De Cal et al., 2009). In the Ebro Valley both *Monilinia* species coexist at field (Villarino et al., 2013).

Monilinia spp. conidia can infect flowers and both immature and mature fruit (Biggs & Northover, 1985, Gell et al., 2009). Fruit infected can remain asymptomatic (latent), and visual decay symptoms only develop during the late ripening period and during postharvest (Gell et al., 2008, Luo & Michailides, 2003). Postharvest losses routinely occur during handling, storage and transport (Tian & Bertolini, 1999) and when conditions are favourable for disease development, brown rot losses may be more severe in postharvest than in field (Larena et al., 2005).

Fruit is transported from the orchard to the packinghouse and where the objective is maintaining fruit quality and extending their shelf life (Brosnan & Sun, 2001). The temperature of harvested peaches and nectarines in Lleida (Catalonia) field can reach 30 ºC or higher when maximum heat. Field heat can cause rapid deterioration of products and therefore it is desirable to remove this heat as quickly as possible after harvesting (Dennis, 1984). On packinghouse, fruit is cooled as soon as possible using hydrocooling or pre-cooling room in order to slow down metabolism and reduce fruit deterioration. Chilled water circulating around and through stacks is used for this purpose and has been described as economical and effective for stone fruit (Dincer et al., 1992). In additions, this postharvest operation has also been reported as useful to improve other aspects as for example internal breakdown (Crisosto et al., 2004).

There are several different commercially hydrocooler designs depending of food type. Food are conveyed in harvest bins through a tunnel and chilled water is sprayed over the product for a certain length of time, depending on the season and the incoming product temperature (Brosnan & Sun, 2001). On the other hand, a simple cooling method commonly used is the cooling room which discharge cold air into a cooling room just below the ceiling. The air sweeps the ceiling and returns cooling food on the floor (Mitchell et al., 1972). Once fruit is cold, they can keep during some hours or days at cold room until sorting. Optimum temperature of peaches for storage is -1 to 0 ºC and RH should be 90 and 95%. (Crisosto & Kader, 2014).

During sorting operations, fruits are selected in order to eliminate fruit defects and sometimes to select fruit with a range of colour, size and shapes. Immersion dumping operation is usually used during fruit sorting, and it consist in submerged a pallet full of fruit into a tank of water and fruit are released. Peaches and nectarines float out so that fruit are transported from tank to lines through conveyor belt. Generally, sodium hypochlorite is added to sanitize water tank of spores of numerous fungi species from field (Bertrand & Saulie-Carter, 1979, Suslow, 2005).

During sorting and packaging operation, fruit temperature increase and the use of forced air cooling tunnel is a common practice before shipping. Cooling tunnel provides a high flow rate to pull refrigerated air through pallet or packing fruit during the itinerating through the tunnel (Fraser, 1992). The air is humidified to 90% RH or higher to minimize water loss during cooling. Then, packed fruit are kept in cold room until transport.

Peaches or nectarines could reach stone packinghouse with recent field infections of *Monilinia* spp. produced before or during harvest but without visible symptom and immediately fruit begin to be treated through different postharvest operations. The aim of this study was to evaluate the effect of hydrocooling, cold room, immersion tank, sorting and cooling tunnel on *Monilinia laxa* development on fruit 48, 24 and 2 hours previously inoculated to simulated fruit with recent infections coming from orchards.

2. Material and Methods

2.1 Fruit

Freshly harvest peaches (*Prunus persica* (L) Batch) and nectarines (*P. persica* var. Nectarine (Ait.) Maxim) were selected by hand from fruit bins on orchards without visible wounds and similar size. Fruit were harvested at the commercial mature stage and were grown in organic commercial orchards located in Lleida (Catalonia). Fruit not used at the time of harvest were stored at 0 ºC for a maximum of 5 days until use.

2.2 Fungal isolate and inoculum preparation

The isolate of *M. laxa* (CPML2) come from the collection of the Pathology Unit, IRTA Centre of Lleida (Catalonia) and this strain was isolated and classified at the Department of Plant protection, INIA (Madrid, Spain). The strain was kept in our

laboratory on potato dextrose agar (PDA) amended with 5% of tomato sauce Petri dishes and stored at 4 ºC in the dark.

The isolate of *M. laxa* were subculture onto PDA with 5% of tomato sauce Petri dishes and incubated in the dark at 25 ºC approximately 1 week. Then to ensure conidia production for the experiment, the pathogens were inoculated onto peaches or nectarines, by wounding fruit with a sterilized steel rod (1mm wide and 2 mm long) and transferring conidia and mycelium from the PDA culture to the wound site with a sterilized pipette tip. Fruit inoculated were incubated at 25 ºC and 85% RH in a photoperiod incubator with 12 h light and 12 h dark for 5-7 days.

Conidia from infected fruit were scarped with a sterile loop and transferred to a test tube with 5ml of sterile distilled water amended with one droplet of 80% tween per litter. The concentration of spore solution was adjusted to 10^3 conidia ml^{-1} with a haemocytometer.

2.3 Fruit inoculation and data recorded during postharvest fruit handling

Peaches and nectarines were organized in plastic fruit trays of 20 fruits and each fruit was wounded once in the equatorial section of the fruit with a sterilized steel rod (1mm wide and 2 mm long) and inoculated with 15µl of the conidia suspension of 10^3 conidia ml^{-1}. Fruit were wounded and inoculated 48, 24 or 2 hours before being subjected to each of the different postharvest operations. Inoculated fruit were incubated at 20 ºC and 85% RH until treated, in order to simulate different time of infection on fruit.

After each postharvest fruit operation, fruit were stored at 0 ºC and 85% RH during 8 days and then at 20 ºC and 85% RH during 5 days. Every inoculated fruit was assessed at 8 days at 0 ºC and at 3 and 5 days at 20 ºC. In each assessment, the number of brown rot infected fruit was recorded and its decay diameter were measured with a malleable ruler to take into account the curvature of the fruit surface.

2.4 Effect of different postharvest fruit operations on brown rot development

Five experiments were carried out to simulated the main postharvest stone fruit operations (Fig. 1.): (i) hydrocooling, (ii) cold room, (iii) water dump, (iv) sorting and (v) cooling tunnel.

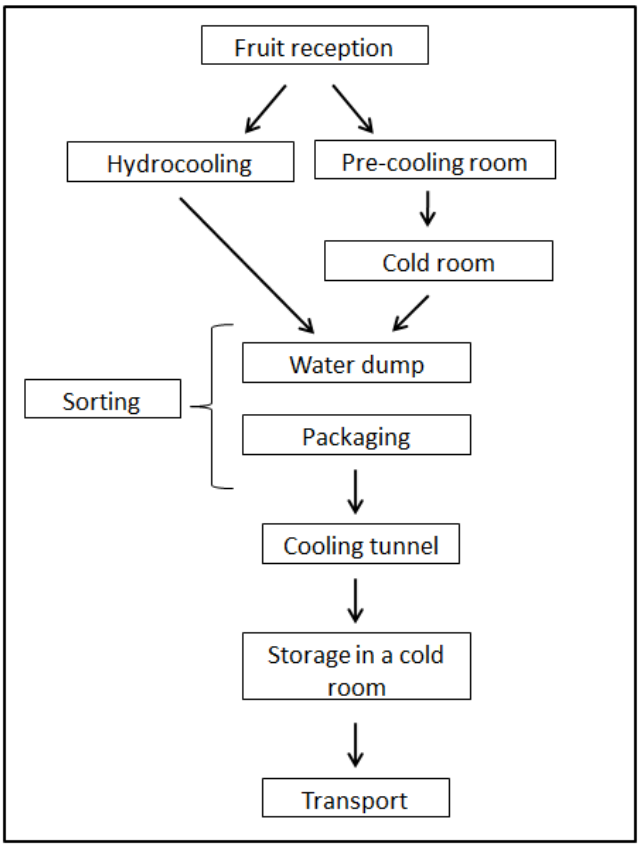

Figure 1. Diagram of a typical postharvest fruit handling operation through which peaches and nectarines are treated while they are in a packinghouse.

All the experiment were performed twice and were carried out with four replicates of 10 fruits each. Experiments were performed each of them with one variety of nectarine and other of peach.

Hydrocooling

'Pp100' and 'Fantasia' nectarines and 'Pollero' and 'Rome Star' peaches were artificially inoculated 48, 24 or 2 hours before to be placed in a commercial hydocooling tunnel during approximately 17 min with a cold water between 2-5 ºC. Sodium hypochlorite was recorded at concentration of 14 mg L^{-1} in 'Pp100' and 'Pollero' varieties and 7 mg L^{-1} in 'Fantasia' and 'Rome Star' varieties. Hydrocooling experiment was performed in a commercial packinghouse of Lleida (Catalonia, Spain). A set of fruit artificially infected was untreated and stored at 0 ºC as a control.

In order to study the effect of time treatment and sodium hypochlorite concentration a semi-commercial hydrocooling was used. 'Alba Red' and 'Fantasia'

nectarines and 'Very Good' and 'Ermidione' peaches artificially inoculated 48, 24 or 2 hours before to be treated during: (a) 1 min with a water solution of 40 mg L^{-1} of sodium hypochlorite 10% (w/v) (Panreac Química, S.A.U., Barcelona, Catalonia), (b) 10 min with a water solution of 10 mg L^{-1} of sodium hypochlorite and (c) 10 min with a water solution of 40 mg L^{-1} of sodium hypochlorite all of them at 4 ºC. A set of artificially infected fruit was untreated and stored at 0 ºC as a control. Free chlorine concentrations were measured using the Free and Total Chlorine HR colorimetry (HANNA Instruments) and pH was measured with an electrode.

Cold room

'Big Bang' and 'Alba Red' nectarines and 'Crimson Lady' and 'Baby Gold 9' peaches were artificially inoculated 48, 24 or 2 hours before stored for 3 days at 4 or 0 ºC in a cold room.

Water dump

To simulate the water dump, 'Big Bang' and 'Alba Red' nectarines and 'Crimson Lady' and 'Baby Gold 9' peaches artificially inoculated 48, 24 or 2 hours before submersion for 30 second in water at 4 or 15 ºC combined with a solution of 40 or 0 mg L^{-1} of sodium hypochlorite 10% (w/v) (Panreac Química, S.A.U., Barcelona, Catalonia). A set of fruit artificially inoculated was untreated and stored at 0 ºC as a control. Free chlorine concentrations were measured using the Free and Total Chlorine HR chlorimeter (HANNA Instruments) and pH was measured with an electrode.

Sorting

'Big Bang' and 'Alba Red' nectarines and 'Crimson Lady' and 'Baby Gold 9' peaches were artificially inoculated 48, 24 or 2 hours before postharvest fruit sorting. To simulated the period between harvest and sorting, fruit was stored for 24 hours at 0 ºC and then stored during 5 hours at 15 or 25 ºC to simulate the period of sorting. After this time, fruit were stored again at 0 ºC until assessment. A set of fruit artificially infected was constantly stored at 0 ºC as a control.

Cooling tunnel

'Pp 100' and 'Fantasia' nectarines and 'Pollero' and 'Rome Star' peaches were artificially inoculated 48, 24 or 2 hours before to be placed in a commercial cooling tunnel for 15 min. A set of fruit previously inoculated as describe above was constantly stored at 0 ºC and was used as a control.

2.5 Statistical analysis

The incidence and severity of brown rot for each postharvest handling treatment were analysed with JMP® 8 statistical software (SAS Institute, Cary, NC, USA). Brown rot incidence was analysed with a non-parametric test Kruskal-Wallis since data are discrete due experimental design of the fruit handling. The lesion diameter followed a normal distribution (confirmed by Shapiro-Wilk test) and an analysis of variance (ANOVA) was performed to each treatment. When both incidence and severity of brown rot were statistically significant, the least significant difference (LSD) test at the level $P<0.05$ for separation of mean was performed.

3. Results

3.1 Effect of hydrocooling on M. laxa development

Lesion diameter was reduced to 3.8, 0.03 and 0.2 cm compared with untreated fruit (9.9, 8.1 and 5.5 cm, respectively) on 'Pp 100' nectarines infected 48, 24 and 2 hours, respectively (Fig. 2 A). In addition, the hydrocooling operation reduced brown rot incidence more than 90% on 'Pp100' nectarine infected 24 and 2 hours before operation.

On 'Fantasia' nectarine, the hydrocooling reduced to 3.5 and 2.2 cm lesion diameter compared with untreated fruit (5 and 4 cm, respectively) infected 24 and 2 hours, respectively before operation (Fig. 2 B). However, hydrocooling did not reduce incidence of brown rot. Hydrocooling tested with 'Pollero' and 'Rome Star' peaches had the same trend that 'Fantasia' nectarine (data not shown).

3.2 Effect of time treatment and sodium hypochlorite concentration of hydrocooling operation on M. laxa development

Time treatment and sodium hypochlorite concentration of hydrocooling operation had different results on peaches and nectarines and therefore results are represented in independent figures (Fig. 3 and Fig. 4). In addition, results on 'Fantasia' nectarine artificially infected 48 hours before operation and 'Very Good' and 'Ermidione' peaches 24 hours before operation were not considered because some problems during the performance of the trials.

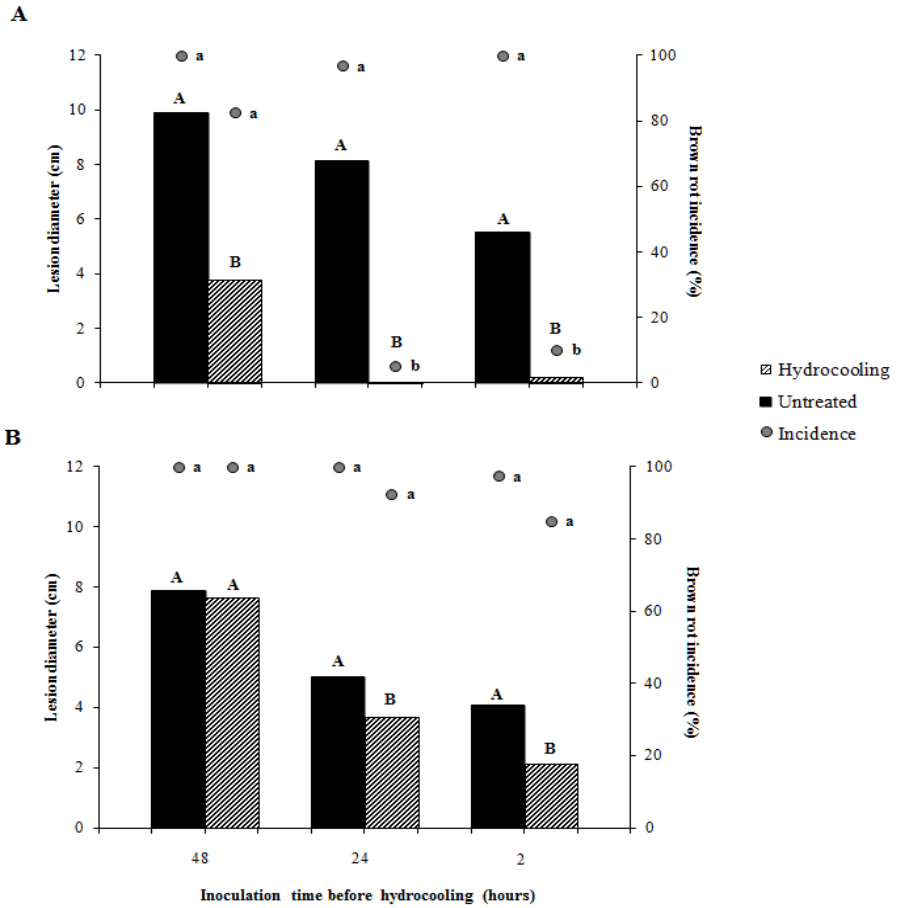

Figure 2. Lesion diameter and brown rot incidence on 'PP100' (A) and 'Fantasia' (B) nectarines artificially inoculated with *Monilinia laxa* at 10^3 conidia mL^{-1} 48, 24 or 2 hours before the hydrocooling operation. Columns show brown rot lesion diameter (cm) and circles above corresponding columns show incidence (%) on fruit. After operation, fruit were incubated for 8 days at 0 ºC and 85% RH plus for 5 days at 20 ºC and 85% RH. Each value is the mean of 40 fruits. Means with the same lowercase letter for brown rot incidence and the same uppercase letter for lesion diameter are not significantly different ($P<0.05$) according to LSD test for each infection time.

On 'Fantasia' nectarine, hydrocooling operation treated for 10 min with a water solution of 40 mg L^{-1} of sodium hypochlorite reduced significantly lesion diameter for the infection time of 24 and 2 hours (Fig. 3 A). However, on 'Alba Red' nectarine, lesion diameter was reduced in all studied treatments compared with untreated fruit for infection time at 24 and 2 hours (Fig. 3 B). The smaller lesion diameters was registered on 'Alba Red' nectarine when was treated for 10 min both 10 and 40 mg L^{-1} of sodium hypochlorite for 24 and 2 hours infection time. Lesion diameter was reduced from 7.4 cm of untreated fruit to 1.4 and 2.1 cm on fruit treated during 10

min with 40 and 10 mg L^{-1} of sodium hypochlorite, respectively on fruit inoculated 24 hours before operation. For infection time of 2 hours before treatment, lesion diameter was reduced from 2.3 cm of untreated fruit to 1.4 and 2 cm on fruit treated during 10 min with 40 and 10 mg L^{-1} of sodium hypochlorite, respectively.

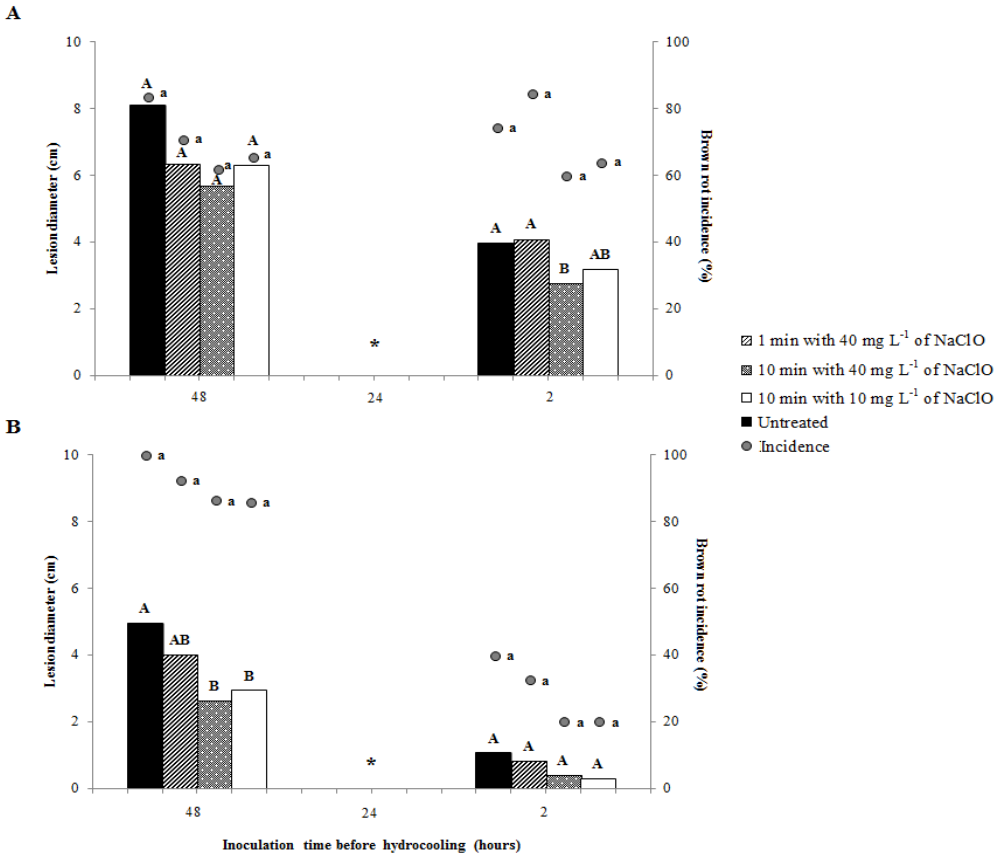

Figure 3. Lesion diameter and brown rot incidence on 'Alba Red' (A) and 'Fantasia' (B) nectarines artificially inoculated *with Monilinia laxa* at 10^3 conidia mL^{-1} 48, 24 or 2 hours before the hydrocooling operation. Columns show brown rot lesion diameter (cm) and circles above corresponding columns show incidence (%) on fruit. After operation, fruit were incubated for 8 days at 0 ºC and 85% RH plus 5 days at 20 ºC and 85% RH. Each value is the mean of 40 fruits. Means with the same lowercase letter for brown rot incidence and the same uppercase letter for lesion diameter are not significantly different ($P<0.05$) according to LSD test for each infection time.

* *Indicate brown rot diameter and incidence was not determinate.*

Brown rot incidence were significantly reduced from 100% on untreated fruit to less than 80% and 60% on ‘Alba Red’ and ‘Fantasia’ nectarines, respectively inoculated 2 hours before hydrocooling operation during 10 min and 40 mg L-1 of sodium hypochlorite (Fig. 3). In addition, brown rot incidence was also significantly reduced from 100% on untreated fruit to less than 60% on ‘Fantasia’ nectarine inoculated 24 hours before hydrocooling operation during 10 min and 40 mg L-1 of sodium hypochlorite (Fig. 3 B).

On ‘Very Good’ peach, hydrocooling operation during 10 min and 40 mg L^{-1} of sodium hypochlorite reduced significantly lesion diameter to 2.7 cm (compared with 4 cm of untreated fruit) for infection time of 2 hours (Fig. 4 A). However, on ‘Ermidione’ peach, lesion diameter was reduced from 5 cm on untreated fruit to 2.6 and 3 cm on fruit treated during 10 min with 40 and 10 mg L^{-1} of sodium hypochlorite, respectively for 48 hours before hydrocooling treatments (Fig. 4 B). For infection time of 2 hours, lesion diameter was not reduced for any of the treatment tested. In addition, on both ‘Very Good’ and ‘Ermidione’ peaches, brown rot incidence was not reduced in any hydrocooling treatment at 48 and 2 hours of infection time.

3.3 Effect of cold room temperature on M. laxa development

On ‘Crimson Lady’ peach, lesion diameter of fruit infected 2 hours before storage in cold rooms was statistically lower at 0 ºC (5.2 cm) than fruit stored at 4 ºC (6.2 cm). However, no differences were found in fruit infected 48 and 24 hours before storage (Table 1). Brown rot incidence was not different on Crimson Lady’ peach stored at 0 or 4º C in any of the infection time studied. Cold room storage tested with ‘Big Bang’ and ‘Alba Red’ nectarines and ‘Baby Gold 9’ peach had the same trend that ‘Crimson Lady’ peach (data not shown).

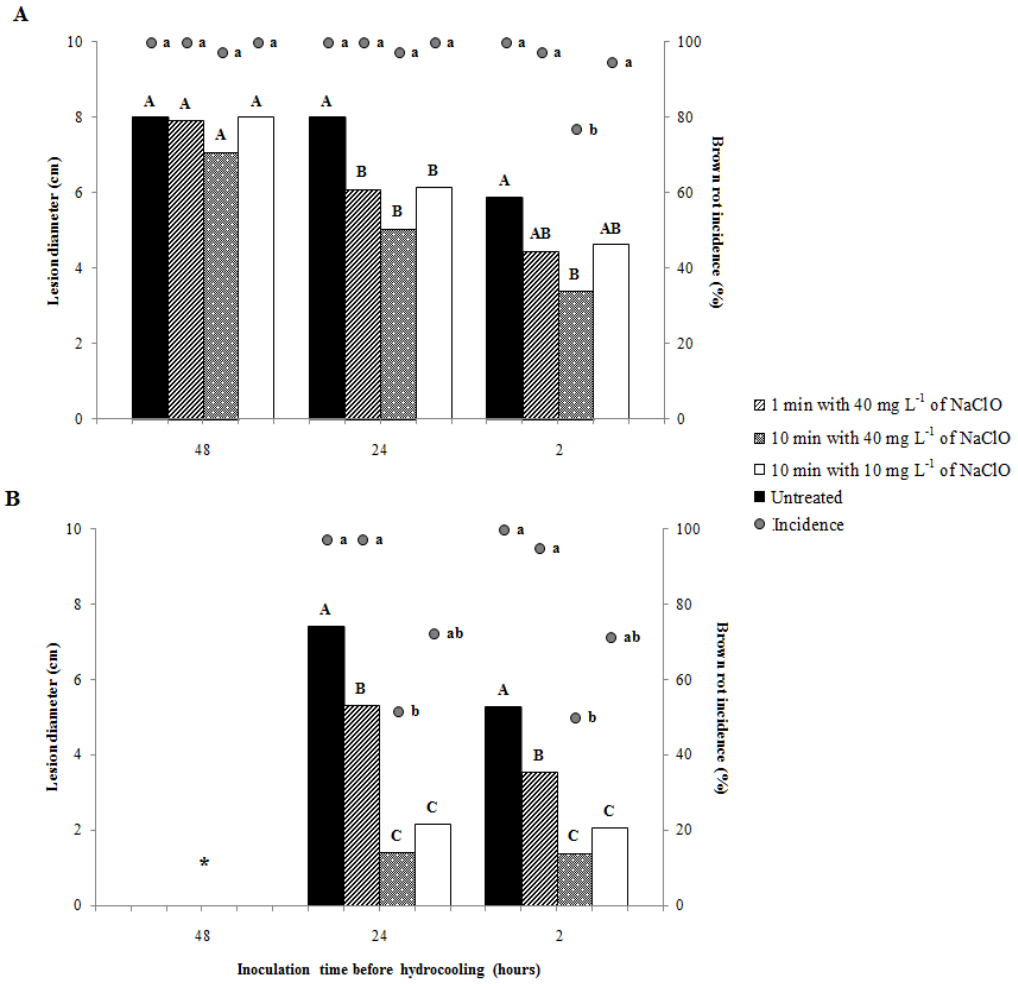

Figure 4. Lesion diameter and brown rot incidence on 'Very Good' (A) and 'Ermidione' (B) peaches artificially inoculated with *Monilinia laxa* at 10^3 conidia mL^1 48, 24 or 2 hours before the hydrocooling operation. Columns show brown rot lesion diameter (cm) and circles above corresponding columns show incidence (%) on fruit. After operation, fruit were incubated for 8 days at 0 ºC and 85% RH plus 5 days at 20 ºC and 85% RH. Each value is the mean of 40 fruits. Means with the same lowercase letter for brown rot incidence and the same uppercase letter for lesion diameter are not significantly different ($P<0.05$) according to LSD test for each infection time.

** Indicate brown rot diameter and incidence was not determinate.*

Table 1. Brown rot incidence and lesion diameter on 'Crisom Lady' peaches infected with *Monilinia laxa* at 10^3 conidia mL^{-1} 48, 24 or 2 hours before storage in a cold room at 0 and 4 ºC. After 3 days of storage, fruit were stored for 8 days at 0 ºC and 85% RH plus 5 days at 20 ºC and 85% RH. Means with the same letter for each infection time are not significantly different (P<0.05) according to LSD test. Each value is the mean of 40 fruits.

Cold room temperature	Diameter (cm)			Incidence (%)		
	48h[1]	*24h*[1]	*2h*[1]	*48h*[1]	*24h*[1]	*2h*[1]
0 ºC	7.4 a	6.7 a	5.2 b	91.4 a	95 a	95 a
4 ºC	7.6 a	7.2 a	6.2 a	94.4 a	97.5 a	100 a

[1] *Infection time (hours)*

3.4 Effect of water dump on M. laxa development

Water dump operation reduced brown rot incidence from 100% on untreated fruit to 57.5% on 'Big Bang' nectarine infected 2 hours before immersion for 30 seconds in water at 4 ºC and 40 mg L^{-1} of sodium hypochlorite (Figure 5), and lesion diameter was reduced from 5 cm on untreated fruit to 3 cm for the same treatment. Lesion diameter and brown rot incidence were not reduced for any of the water dump treatments tested on 'Big Bang' nectarine infected 24 and 48 hours before. Lesion diameter of the water dump operation tested with 'Alba Red' nectarine and 'Crimson Lady' and 'Baby Gold 9' peaches had the same trend that 'Big Bang' nectarine (data not shown). However, brown rot incidence was not reduced on 'Alba Red' nectarine and 'Crimson Lady' and 'Baby Gold 9' peaches infected 2 hours before immersion for 30 seconds in water at 4 ºC and 40 mg L^{-1} of sodium hypochlorite (data not shown).

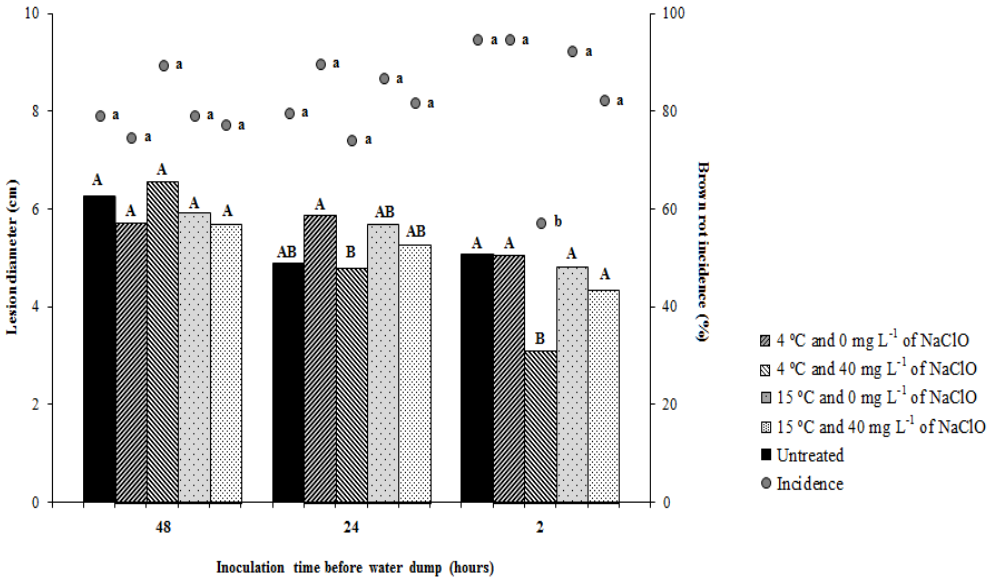

Figure 5. Lesion diameter and brown rot incidence on 'Big Bang' nectarines artificially inoculated with *Monilinia laxa* at 10^3 conidia mL^{-1} 48, 24 or 2 hours before water dump operation. Columns show brown rot lesion diameter (cm) and circles above corresponding columns show incidence (%) on fruit. After operation, fruit were incubated for 8 days at 0 ºC and 85% RH plus 5 days at 20 ºC and 85% RH. Each value is the mean of 40 fruits. Means with the same lowercase letter for brown rot incidence and the same uppercase letter for lesion diameter are not significantly different ($P<0.05$) according to LSD test for each infection time.

3.5 Effect of sorting temperature on M. laxa development

On 'Alba red' nectarine, lesion diameter of fruit infected 2 hours before sorting at 0 ºC (6.2 cm) during 5 hours was lower than fruit storage at 15 and 25 ºC (6.7 and 6.9 cm, respectively) (Table 2). However, no differences were found in fruit infected 48 and 24 hours before sorting at different temperatures. Brown rot incidence was not different on 'Alba red' nectarine in any studied conditions. Sorting operation tested on 'Crimson Lady' and 'Baby Gold 9' peaches and 'Big Bang' nectarine had the same trend that 'Alba Red' nectarine (data not shown).

Table 2. Brown rot incidence and lesion diameter on 'Alba Red' nectarine infected with *Monilinia laxa* at 10^3 conidia mL^{-1} 48, 24 or 2 hours before sorting at 0, 15 and 25 ºC. After 5 hours of sorting, fruit were stored for 8 days at 0 ºC and 85% RH plus for 5 days 20 ºC and 85% RH. Means with the same letter for each infection time are not significantly different (P<0.05) according to LSD test. Each values is the mean of 40 fruits.

Sorting temperature	Diameter (cm)			Incidence (%)		
	48h[1]	*24h*[1]	*2h*[1]	*48h*[1]	*24h*[1]	*2h*[1]
0 ºC	9.3 ab	8.4 a	6.2 b	97.5 a	97.5 a	100 a
15 ºC	10.3 a	8.3 a	6.7 a	100 a	100 a	100 a
25 ºC	10.5 a	8.4 a	6.9 a	100 a	100 a	100 a

[1] *Infection time (hours)*

3.6 Effect of cooling tunnel on M. laxa development

On 'Pollero' peach, lesion diameter of fruit infected 48 hours before cooled with a cooling tunnel (10.1 cm) was lower than fruit cooled in in cold room at 0 ºC (Table 3). However, no differences were found in fruit infected 24 and 2 hours before cold room or cooling tunnel. Brown rot incidence on 'Pollero' peach was the same in all studied conditions. The cooling tunnel operation did not reduce lesion diameter and incidence on 'Pp 100' and 'Fantasia' nectarines and 'Rome Star' peach (data not shown).

Table 3. Brown rot incidence and lesion diameter on 'Pollero' peach infected with *Monilinia laxa* at 10^3 conidia mL^{-1} 48, 24 or 2 hours before cold room or cooling tunnel for 15 min. After this operation, fruit were stored for 8 days at 0 ºC and 85% RH plus 5 days at 20 ºC and 85% RH. Means with the same letter for each infection time are not significantly different (P<0.05) according to LSD test. Each value is the mean of 40 fruits.

Postharvest operation	Diameter (cm)			Incidence (%)		
	48h[1]	*24h*[1]	*2h*[1]	*48h*[1]	*24h*[1]	*2h*[1]
Cold room	10.8 a	8.34 a	5.90 a	100 a	100 a	91.7 a
Cooling tunnel	10.1 b	8.21 a	5.44 a	100 a	100 a	100 a

[1] *Infection time (hours)*

4. Discussion

Hydrocooling and water dump operation were able to reduce brown rot incidence in comparison to direct storage at 0 ºC, although this depend on some specific factors such time of treatment or concentration of sodium hypochlorite in the water. Other postharvest operations as cold room, sorting and cooling tunnel have also some effect on reduced brown rot severity in some cases.

The aim of the hydrocooling operation is reduce field temperatures of fruit as quickly as possible to maintain a high level of quality. Hydrocooling in this work reduced brown rot incidence on 'Pp100' nectarine and disease severity both on 'Pp100' and 'Fantasia' nectarines infected 24 and 2 hours before commercial hydrocooling. However, the difference regards brown rot incidence on both fruit varieties was unexpected. It could be attributed to higher concentration of chlorine registered when 'Pp100' nectarine were processed (about 14 mg L^{-1}) regarding 'Fantasia' nectarine (about 7 mg L^{-1}). During the 60s and 70s several research were carried out studying the effectiveness of hydrocooling operation with chlorinated water on the development of brown rot (Mc Clure, 1958, Smith et al., 1962, Wells & Bennett, 1975) and they reported partly or irregularly reduction of decay on peaches under commercial conditions. In addition, Phillips and Grendahl (1973) reported that chlorine effectively reduce brown rot decay on peaches and nectarines artificially-inoculated within the range of 50-100 mg L^{-1} when were treated during 20 min in a hydrocooling. Chlorine concentration could improve the efficiency of hydrocooling operation, however higher chlorine concentration could have the handicap that produce an unpleasant odor to packinghouse staff.

In the studied hydrocooling operation, time could be varied between 10-20 min regarding to the initial temperature of fruit. To calculate an optimal precooling operation should be considered the heat load of the fruit and the cooling rate due treatment time and water temperature (Brosnan & Sun, 2001) but at commercial scale, optimal conditions could varied depending on workload and availability of staff. The success of commercial hydrocooling on 'Pp100' nectarine could be partially explained by the time of treatment and sodium hypochlorite concentration of hydrocooling. The time treatment could be more important factor than chlorine concentration since fruit treated during 10 min decrease incidence more than fruit treated for 1 min. In addition, brown rot incidence was more reduced on nectarine than on peach fruit infected at 2 hours before hydrocooling and it could be due to the differences between the skins of both fruit varieties. Nectarine skin is smoother than

peach so cleaning effect of water on inoculated conidia could be more notable on nectarine.

Hydrocooling operation was less effective in fruit infected 48 and 24 hours before the treatment, probably due infection is already established although symptoms are not visible. This patters has been observed for all operations studied in this report. Bernat et al. (Bernat et al., 2017) showed that decay development on peaches and nectarines artificially inoculated and incubated at 20 ºC required less than 4 days to reach the half of the asymptote indicating that overall the first symptoms of disease could be appear around this days if inoculation was carried out by wounding the fruit and inoculating 10^{-4} conidia ml^{-1}. In addition, brown rot disease development on wounded fruits is faster than non-wounded fruits for the same fruit variety and inoculum concentration (Martínez-García et al., 2013). On other hand, if fruit reaching packinghouse were infected but without wound probably the effect of the different operations will be different and the effectiveness of some postharvest operations studied in the experiments performed in the present manuscript could not be applied for these cases.

The effect of immerse fruit on water does not mean a greater lesion diameter in our study suggesting that once fruit is infected, humidity has a low influence on decay development, however humidity has a direct influence in conidia germination (Casals et al., 2010, Tamm & Flückiger, 1993) and infection (Xu et al., 2007). Immerse fruit in water with a concentration of 40 mg L^{-1} of sodium hypochlorite reduced severity of brown rot disease in recent infections for all tested cultivars. Nevertheless, incidence only was reduced on 'Big Bang' nectarine infected 2 hours before water dump. In a study carried out by Smilanick et al (2002) on oranges and lemons inoculated with *Penicillium digitatum* 24 hours before immersion in water with 50, 1000, 2000 and 4000 mg L^{-1} of free chlorine for 2 minutes, showed that at higher tested concentration, the incidence of green mold disease was reduced less than 40% compared with the 100% of untreated fruit. Other study (Spotts & Peters, 1980) on pears artificially inoculated with *Botrytis cinerea*, *Mucor piriformis* and *Penicillium expansum* and immersed for 130 seconds in a commercial packinghouse water tank containing 130 ±10 mg L^{-1} of chlorine, showed no effect of the treatment. Those studies agree with our results in that chlorine-water solution have a low effectiveness to reduce incidence of infected fruit. However, the effect of chlorine on water disinfection has been reported effective for different postharvest pathogens. For example to *M. piriformis*, *P. expansum* and *Phialophora malorum*, chlorine at 64 mg L^{-1} inhibited germination from 90 to 100% for all pathogens (Spotts & Cervantes, 1989) and to *Botrytis cinerea*, *M. piriformis* and *P. expansum* 50 mg L^{-1} reduce significantly

conidia germination after for 30 seconds (Spotts & Peters, 1980). Regardless chlorine is not an effective method of control disease on fruit, it is commonly used to prevent the risk associated with water dump and hydrocooling decay due to recirculated contaminated water which resulting in the contamination of the cooled produce leads to the possibility of new infections (Brosnan & Sun, 2001).

Temperature is a factor highly influencing *Monilinia* spp. development (Bernat et al., 2017, Phillips, 1984, Xu et al., 2001). During cold room, sorting and cooling tunnel, the temperature is the main factor involved in these postharvest operations. When fruit were stored at low temperatures during cold room and sorting, decay development was delayed on fruit infected 2 hours before storage at 0 ºC however on fruit with established infections such at 48 and 24 hours, no effect of lesion diameter was observed. Storing fruit at 0 ºC is recommended for keep quality (Garg et al., 2005) and for delay brown rot symptoms on fruit infected (Bernat et al., 2017). Optimal temperature for *M. laxa* development has been reported at 25 ºC (Tamm & Flückiger, 1993, Papavasileiou et al., 2015) although significant differences has been reported between *M. fructicola* and *M. laxa* at 25 ºC (Bernat et al., 2017, Papavasileiou et al., 2015). In addition, reducing the temperature after sorting in cold rooms or treated trough cooling tunnel did not affect to *M. laxa* development. Moreover, temperature involve on the water dump did not shown any effect on *M. laxa* incidence or severity. Nevertheless, Phillips and Grendahl (1973) reported that the toxicity of 3-5 mg L^{-1} chlorine to *M. fructicola* conidia increased with temperature. In general to all postharvest operations, higher temperature develop higher lesion diameter when was compared with low temperature.

Taking appropriated action to reduce brown rot infections at field such as optimal application of conventional fungicides (Rungjindamai et al., 2014, Usall et al., 2010) in synergy with other integrated strategies such picking fruit carefully to reduce wounds (Xu & Robinson, 2000), decrease brown rot incidence. In addition, reducing the period between harvest at field and their transport to the packinghouse in combination with a first hydrocooling treatment during a suitable period of time and chlorine concentration could decreased brown rot incidence when infections have been produced recently at field. Likewise, water dump fruit as soon as possible in an appropriated choline concentration is an encouraged postharvest fruit handling. On other hand, low temperatures during the complete chain of postharvest handling operations reduce fruit decay as well as keep fruit quality during this period. Further economic studies will be necessaries to evaluate the benefit to implement our recommended handling operations and management at packinghouses in relation to the expenses that it could cause.

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