Effects of Heat Treatments on Populations of Four Fruit Decay Fungi in Sodium Ortho Phenylphenate Solutions

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ABSTRACT

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Conidia of *Botrytis cinerea, Mucor piriformis, Penicillium expansum*, or *Phialophora malorum* did not germinate after exposure to temperatures of 54.4 or 60 C for 1–20 min in sodium ortho phenylphenate solutions. *M. piriformis* and *Penicillium expansum* conidia in water from packinghouse dump tanks containing high populations were killed by heating to 54.4 C for 25 min. Heating dump-tank water to 54.4 C for 20 min in a packinghouse lowered populations of *M. piriformis* and *P. expansum*. Costs to heat were 77% less than costs to empty, clean, and refill the 11,355-L tank.

Storage decays of pear (*Pyrus communis* L.) caused by *Botrytis cinerea* Pers. ex Fr., *Mucor piriformis* Fischer, *Penicillium expansum* Lk. ex Thom., and *Phialophora malorum* (Kidd & Beaum.) McColloch are major problems in the Pacific Northwest fruit industry (2,6). Packinghouses use either chlorine or sodium ortho phenylphenate (SOPP) in dump tanks and flumes to reduce populations of decay fungi (1). Dump-tank solutions require frequent changes because of accumulation of debris and increase of fungal spores. High labor and chemical costs are involved in changing solutions, and disposal of dump-tank water containing SOPP is often a problem.

Hot-water treatment has been used to control decay of raspberry (13) caused by *Botrytis* sp. and of tomato (4) caused by *Penicillium expansum*. Decays of blueberry (3) and peach (7,8) have been controlled using hot-water treatment. Decay control may be more effective at lower water temperatures if fungicides are added to the hot water than if hot water is used alone (12). In hot-water treatments, the margin between the temperature effective for decay control and that causing crop injury is often narrow. Injury to pear fruit by hot SOPP solution has been observed (R. Spotts, *unpublished*), and packinghouses in Oregon are careful to keep dump-tank water temperatures under 27 C when SOPP is used.

This study was conducted to determine the effect of hot SOPP-flotation salt solutions on conidial germination of *B. cinerea, M. piriformis, Penicillium expansum,* and *Phialophora malorum*. Research also is reported on heat treatment of dump-tank solutions in a commercial packinghouse to reduce populations of decay spores.

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MATERIALS AND METHODS

Effect of heat treatment of SOPP-salt solutions on conidial germination. Solutions of sodium salts of carbonate, lignin sulfonate, silicate, and sulfate in distilled water were prepared, and the concentration adjusted with a hydrometer to obtain a specific gravity of 1.05. SOPP was added to achieve a concentration of 0.58%. Solutions were heated to 43.3, 48.8, 54.4, and 60 ± 0.1 C. Conidia of *B. cinerea, M. piriformis, Penicillium expansum,* and *Phialophora malorum* were washed from 2- to 3-wk-old cultures growing on potato-dextrose agar acidified with 1.5 ml of 85% lactic acid per liter (APDA) and added to the SOPP-salt solutions at each temperature to obtain 10^5 conidia per milliliter. After 1, 5, 10, and 20 min, 5-ml amounts were removed, conidia were recovered on a Millipore filter (0.45 μm for *Penicillium expansum* and *Phialophora malorum* and 5 μm for *B. cinerea* and *M. piriformis*) and washed with sterile distilled water as described previously (10). Conidia were transferred to APDA and incubated at 17 C for 24 hr. Germination of 100–200 conidia was determined microscopically for each fungus at each temperature-time combination.

Heat treatment of commercial dump-tank water. Dump-tank water was collected from three packinghouses, and each sample was treated independently. Because these houses were handling apples at the time water samples were obtained, no flotation salts or fungicides were in the water. Specific gravity was adjusted to 1.05 with sodium silicate, and SOPP was added to achieve a concentration of 0.35%. Before heating, each water sample was divided into two parts, and conidia of *M. piriformis* and *Penicillium expansum* were added to one portion to obtain 10^5 conidia per milliliter. Both solutions were heated at 54.4 C for 25 min. Natural fungal populations in the dump-tank water samples were determined before and after heating by placing 0.5 ml of a 1:99 dilution of dump-tank water on APDA and counting fungal colonies on three replicate plates after 4 days at 20 C. Germination of 100–200 conidia of *M. piriformis* and *P. expansum* was determined after 20 hr at 15 C in samples to which these fungi were added.

Packinghouse trials with heat treatment of dump-tank water. Three heat-treatment trials were carried out in a commercial packinghouse between January and April 1983. Because hot SOPP solutions are phytotoxic to pears, tests were conducted at night when no fruit was in the tank. Dump-tank volume was 11,355 L, and water contained sodium silicate plus SOPP (trials 1 and 2) or sodium lignin sulfonate plus SOPP (trial 3). Natural fungal populations were determined before and after heating as described before. The tank was covered with Styrofoam to minimize heat loss and heated to 54.4 C for 20 min with two boilers, each producing 158,000 kcal. Water loss and changes in SOPP concentration were monitored. In trial 3, decaying Anjou pear fruits infected with *B. cinerea, M. piriformis,* and *P. expansum* (four fruits per fungus) were placed in the tank solution before heating to study fungal survival in internal tissues of infected fruits. After heating, decayed fruits were sectioned, surface-sterilized with 0.525% NaOCl, and rinsed with sterile distilled water. Sections were removed from outer, middle, and core areas of each fruit, plated on APDA, and observed for fungal growth for 3 wk at 20 C.

Table 1. Effect of flotation salt, temperature, and exposure time on germination of conidia of *Botrytis cinerea, Mucor piriformis, Penicillium expansum*, and *Phialophora malorum* in SOPP solution

Flotation salt[a]	Temperature (C)	Percent germination[b] of *B. cinerea* exposed (min)				*M. piriformis* exposed (min)				*P. expansum* exposed (min)				*P. malorum* exposed (min)			
		1	5	10	20	1	5	10	20	1	5	10	20	1	5	10	20
None	43.3	81	27	2	0	99	96	85	51	98	97	90	48	92	10	4	3
	48.9	0	0	0	0	12	4	2	0	38	27	11	2	5	2	3	3
Sodium carbonate	43.3	14	1	0	0	4	0	1	1	92	11	8	4	5	8	9	3
	48.9	0	0	0	0	3	0	0	0	21	6	1	1	4	1	0	0
Sodium lignin sulfonate	43.3	0	0	0	0	1	2	7	2	2	1	0	0	0	0	0	0
	48.9	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Sodium silicate	43.3	0	0	0	0	6	1	0	0	81	3	4	2	1	0	0	0
	48.9	0	0	0	0	0	0	0	0	70	1	2	0	1	0	0	0
Sodium sulfate	43.3	0	0	0	0	18	5	0	0	79	5	7	2	1	3	1	2
	48.9	0	0	0	0	1	0	0	0	0	0	1	0	0	0	1	2

[a]Salt concentration of solution adjusted to specific gravity of 1.05. All solutions contained 0.58% sodium ortho phenylphenate (SOPP).
[b]Each value based on 100–200 conidia incubated 24 hr at 17 C after treatment and examined microscopically.

RESULTS

Effect of heat treatment of SOPP-salt solutions on conidial germination. As solution temperature and exposure time increased, germination of conidia decreased (Table 1). No conidial germination of any fungus occurred after exposure to 54.4 or 60 C for 1–20 min, so data are not shown. Germination varied from 81 (*B. cinerea*) to 99% (*M. piriformis*) when conidia were treated at 43.3 C for 1 min in SOPP solution without flotation salt. Germination of *P. expansum* conidia after a 1-min exposure to 43.3 and 48.8 C in solutions of sodium carbonate was 92 and 21%, respectively, and germination was 81 and 70%, respectively, after exposure to 43.3 and 48.8 C in sodium silicate. Germination of all fungi was 7% or less after exposure to 43.3 C for 1 min or more in sodium lignin sulfonate-SOPP solution. When plates in which no conidia germinated within 24 hr at 17 C were held for several days, limited germination and fungal growth often occurred, except in plates with spores previously heated to 54.4 or 60 C for 20 min. Germination and growth after these treatments were rare.

Heat treatment of commercial dump-tank water. Water sampled from packinghouse dump tanks contained high natural populations of *M. piriformis* and *P. expansum* (Table 2). After heat treatment, no fungal growth occurred. Similarly, no germination of added *M. piriformis* or *P. expansum* conidia was observed after heat treatment.

Heat treatment in packinghouses. In most trials, heat treatment reduced populations of *M. piriformis* and *P. expansum* to zero (Table 3). Low populations of *M. piriformis* and *P. expansum* were each detected after heat treatment in one of three trials. Evaporative water loss after heat treatment was estimated at about 10%, and SOPP loss, at 24%. No fungal growth occurred from tissue of Anjou pear fruits present in the dump-tank solution during heating that had been infected previously with *B. cinerea, M. piriformis,* or *P. expansum.*

Table 2. Survival of conidia of *Mucor piriformis* and *Penicillium expansum* in heated dump-tank water

Source of dump water[a]	No. of conidia per milliliter[b] *M. piriformis* Before heating	*M. piriformis* After heating[c]	*P. expansum* Before heating	*P. expansum* After heating[c]
House A	1,067	0	2,133	0
House B	0	0	400	0
House C	667	0	5,133	0

[a]Water sampled from packinghouse dump tanks and heat-treated in the laboratory after adjusting specific gravity to 1.05 with sodium silicate and adding sodium ortho phenylphenate to 0.35%.
[b]Natural spore population based on the number of colonies formed on acidified potato-dextrose agar after 4 days at 20 C. Each value represents the mean of three replicate plates.
[c]Water treated at 54.4 C for 25 min.

Table 3. Effect of heat treatment on *Mucor piriformis* and *Penicillium expansum* populations in dump-tank water in a commercial packinghouse

Trial no.[a]	Number of conidia per milliliter[b] *M. piriformis* Before heating	*M. piriformis* After heating[c]	*P. expansum* Before heating	*P. expansum* After heating[c]
1	1,133	0	4,200	0
2	2,000	133	1,067	0
3	267	0	2,133	200

[a]Tank held 11,355 L and contained sodium silicate (trials 1 and 2) or sodium lignin sulfonate (trial 3). Sodium ortho phenylphenate concentration was 0.35 ± 0.05%.
[b]Natural spore population based on the number of colonies formed on acidified potato-dextrose agar after 4 days at 20 C. Each value represents the mean of three replicate plates.
[c]Dump-tank water heated to 54.4 C for 20 min.

DISCUSSION

Conidia of *B. cinerea, M. piriformis, Penicillium expansum,* or *Phialophora malorum* did not germinate after exposure to 54.4 C for 1 min or more in water containing SOPP. Pierson (5) reported slight conidial germination of *Penicillium expansum* after exposure to 54.4 C for 3 min in hot water alone, and Thom and Ayers (11) stated that *P. expansum* was killed by pasteurization at 54.5 C for 30 min. Thus, addition of SOPP to hot water may have increased its effectiveness for reducing spore germination. Wells (12) showed that decay control of peaches, plums, and nectarines was more effective with 2,6-dichloro-4-nitroanaline (DCNA) combined with hot water than with either hot water or DCNA alone. Germination of *P. expansum* conidia after exposure to 43.3 C appeared to be less in sodium lignin sulfonate than in the other pear flotation salts.

Although treatment of fungal spores in SOPP at 54.4 C for 1 min completely prevented germination after 24 hr of incubation at 17 C, limited germination

and fungal growth did occur after an incubation period of several days. Under certain conditions, SOPP previously was shown to act as a fungistat rather than as a fungicide (9). Because of the fungistatic nature of SOPP as well as the presence of large amounts of debris, which may protect spores in dump tank water, a temperature and time of 54.4 C for 20 min was selected for commercial trials.

Previously, SOPP was reported to reduce the spore load in dump tanks and to aid in control of postharvest decay (1). However, phytotoxicity of SOPP to Anjou pear fruit occurred and increased as exposure time, solution temperature, and salt concentration increased. In studies described herein, heat treatment of dump-tank water was used, when fruit was not present, to reduce population levels of decay fungal spores. Viable spores detected in the tank after heating may represent contamination from air or other sources after treatment. In packinghouse trials, 11,355 L of dump-tank water containing SOPP solution was heated from 7 to 54.4 C within 4.8 hr. By the next morning, solution temperature was 18 C, and pear fruits were processed without injury. Cost of heat treatment was calculated to be about 23% of the cost to empty, clean, and refill the tank. Thus, heat treatment appears to be an effective, economical method to reduce dump-tank spore loads, decay, and the number of times tanks must be emptied. However, because debris accumulates in the tanks, emptying and cleaning cannot be eliminated entirely.

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