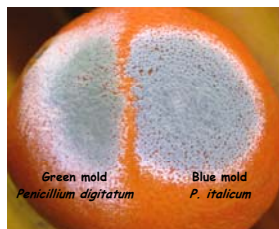


RECENT RESEARCH IN CALIFORNIA TO MINIMIZE POSTHARVEST DECAY OF CITRUS BY PRE- AND POSTHARVEST ACTIONS

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Multiple-decrement concept

Decay losses with no actions:	30%
Action #1 20% effective	24%
Action #2 40% effective	14%
Action #3 50% effective	7%
Action #4 60% effective	3%
Final decay losses:	3%

- Postharvest decay in California and Florida compared
- What can you do before harvest?
- What can you do after harvest?
- Research in progress: Fumigation with ammonia

Multiple-decrement concept

Elements in postharvest decay management actions for citrus:

Sanitation preharvest (grove decay removal, bin cleanliness, minimize harvest to packinghouse interval)

Minimize fruit injuries (minimize drops and impacts, worker training, sorter technology, black light rooms, etc.)

Packinghouse treatments (fungicides, hot water, GRAS substances, biological control, aqueous fungicides, mixtures, temperature management)

Sanitation postharvest (reduce spore populations with chemical sanitizers, low-level constant ozone, facility design, steam or hot water cleaners,)

CA and FL decay pathogens

- **California.** Green/blue molds and sour rot are the major pathogens, others usually minor. Fungicide resistance a major problem, but limited to packinghouses.
- **Florida.** Diplodia stem end rot predominates, with green/blue molds and sour rot following. Fungicide resistance rare.
- **Why?** Longevity of green/blue mold conidia longer under the dry conditions of CA. Sparse CA rainfall minimizes stem end rot inoculum production and infection in groves. Degreening in CA is cooler and favors green/blue molds, while the higher FL degreening temperatures reduce these diseases. Many CA packinghouses operate year round and practice long in-house storage. No fungicides are used before harvest in CA, unlike FL.

What can you do before harvest?

Actions other than chemical control...

Very little research in recent years has been done in this area; most research directed at preharvest actions is old but still valuable...

G. Harold Powell

Research career 1903 -1911

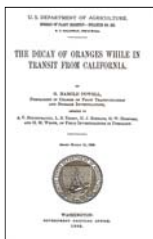


G. Harold Powell, a former U.S. Department of Agriculture official whose investigation into the causes of fruit decay led to radical changes in fruit handling, was general manager of the California Fruit Growers Exchange from 1912-1922.

Postharvest losses from California were up to \$1,500,000 in 1904, from Florida about \$500,000

USDA sent plant pathologists to California and Florida in 1905

In 1908, both groups issued long bulletins that identified rough handling as the source of most of the decay problem



“It was an exception to find a grader who could see and throw out an orange showing slight mechanical injury. It is probable 95% of the fruit injured in handling is included in the packed fruit.” Powell, 1908 California

“The most common type of injury was made by ... the clippers ... many were injured by stem punctures, while others showed scratches from thorns. Other common ... injury... were from gravel and twigs in the bottom of boxes and cuts by the finger nails of the pickers.”

Powell, 1908 Riverside, California

“... it is now accepted without question that the elimination or reduction of the mechanical injury of the orange is one of the largest problems that the California citrus-fruit grower and packer have to overcome.”

Powell, 1908

“The very foundation of decay control is careful handling; in fact, there is no substitute for it.”

Winston, 1949 Florida

“It has been observed by many that this fungus can only infect citrus fruit through an injury. ...as early as 1921 [they were advised] to use the utmost care ... to avoid any possible injury to the fruit.”

Christ, 1966 South Africa

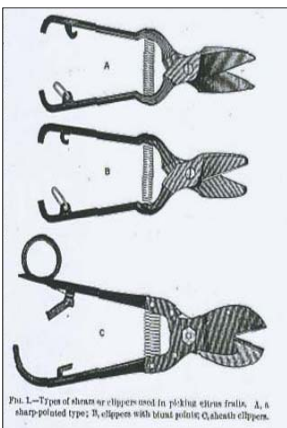


FIG. 1.—Types of stem or clippers used in picking citrus fruits. A, A sharp-pointed type; B, clippers with blunt points; C, sheath clippers.

“... In 1905, the average amount of mechanically injured fruit, based on careful inspection of more than 40,000 oranges ... was 17%.”

Powell, 1908 California

“... abundant data ... show that by using care in the instruction and supervision of labor not more than 2 or 3% of oranges will be injured in handling the fruit on a commercial scale.”

Powell, 1908 California

“The excess in net return has been many times greater than the extra cost of careful handling.”

Lloyd Tenny, G. W. Hosford, and H. M. White, 1908 Florida

Impact of careful handling on the incidence of postharvest decay on oranges after harvest

Season	Hand	Typical
	graded	commercial
1906	2.9	22.9
1907	2.1	18.2

Lloyd Tenny, G. W. Hosford, and H. M. White, 1908 Florida

What can you do before harvest?

Chemical control

In repeated tests where fruit were sprayed with fungicides before harvest, Topsin (2 lbs/ac) was consistently reduced subsequent postharvest decay by 80 to 85% in repeated tests and superior to many other fungicides. **Topsin is not registered in California**

At rates that controlled postharvest decay, preharvest applications of potassium phosphite, potassium sorbate, and similar organic acids or GRAS substances have injured the trees or fruit or both.

Syngenta Crop Protection, Inc. is proposing to register Quadris Top (difenoconazole FRAC group 3 + azoxystrobin FRAC group 11) for preharvest use on citrus to retard postharvest decay. Imazalil is in the same FRAC group as difenoconazole, so efforts to minimize the development of resistant isolates will be particularly important.

Since that work was completed in 2004, pre-harvest use of Topsin remains a reliable treatment to control postharvest green mold

However, it is not registered in California

Pre-harvest applications of Topsin to control postharvest green mold at Lindouse on "Atwood" navel oranges. Asterisk indicates fruit were inoculated after harvest.

Year	Treatment	Green mold (%)	Reduction in losses (%)
2005	Control	3.20	81
	Topsin 2 lb/ac	0.60	
2005	Control	84.5*	85
	Topsin 2 lb/ac	12.9*	
2006	Control	98.9*	86
	Topsin 2 lb/ac	13.8*	
2007	Control	1.95	72
	Topsin 2 lb/ac	0.55	
2008	Control	20.99	83
	Topsin 2 lb/ac	3.53	
2009	Control	4.51	93
	Topsin 2 lb/ac	0.33	

Mean reduction in green mold losses 83%

Review and update on pre-harvest use of Topsin

Table 1. Green mold incidence on Navel oranges and its percent reduction as a result of preharvest application of different fungicides. Fruit were harvested one week after application, inoculated with *P. digitatum* isolate M6R, degreened for three days at 20°C and 5 µl/l ethylene, then stored at 10°C for one additional week.

Treatment	Green mold incidence (%) ^a	Reduction in green mold (%)	Class
Control	98.9 a
Switch	95.8 b	3.1	contact
Pristine	92.0 bc	7.0	contact
Headline	89.4 c	9.6	contact
Abound	79.0 d	20.1	contact
Topsin	19.2 e	80.6	systemic

^aValues followed by unlike letters differ significantly by Fisher's least significant difference ($P=0.05$). An arcsin transform was applied before ANOVA; actual values are shown.

Fungicides in common use to control citrus green and blue molds of citrus

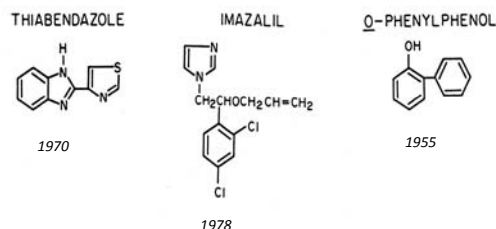


Table 2. The influence of a preharvest application of thiophanate methyl (TM) residues in fruit and on the postharvest incidence of green mold on navel oranges. Immediately after harvest, the fruit were inoculated with *P. digitatum* followed storage for one week at 20°C. Those fruit harvested one week after TM or water (control) application were degreened immediately after harvest for 3 days in 5 µg/ml ethylene at 20°C, then stored at 20°C for an additional 4 days.

Test	Interval ^a	Rainfall ^b	Residue ^c	Green mold incidence (%) ^d		
				TM	Control	Reduction ^e
1	1	1.0	...	17.4	79.4	78.1
2	1	1.0	3.6	17.5	95.1	81.6
3	1	16.5	0.3	8.9	96.9	90.8
4	1	4.1	2.1	13.1	77.0	83.0
	3	28.5	4.8	6.8	80.4	91.5
	5	36.6	4.3	17.0	87.7	80.6
	7	67.6	5.1	26.0	90.5	71.3
5	1	6.1	...	19.2	98.9	80.6
	4	27.5	1.8	41.2	99.1	58.4
	7	51.6	...	31.8	100.0	68.2

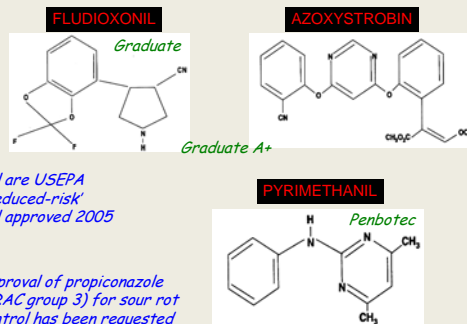
^aInterval (wk) between application of treatments to trees and harvest. ^bRainfall (mm) between application of the treatments and harvest. ^cThiophanate methyl residues in fruit (µg/g fresh wt). Each value is the mean of 2 or 3 replicates of 8 fruit each. ^dGreen mold after seven days at 20°C. Each value is the mean of four replicates containing 60 to 73 fruit each. ^ePercent reduction in green mold compared to the control.

Topsin effective and rainfast

Residue as low as 0.3 ppm effective

Protection persisted for 7 weeks

New citrus postharvest fungicides



Maximum residue tolerances

	Japan	Germany	USA	CODEX
Pyrimethanil	15	10	10	7
Azoxystrobin	1	1	10	...
Fludioxonil	10	7	10	7
Imazalil	5	5	10	5
Thiabendazole	10	5	10	7

From: USDA FAS mrl database <http://www.mrl-database.com/>



Why is the solution heated?

- Pathogen inoculum contamination eliminated in fungicide solution
- Some decay control by heat alone
- Fungicide effectiveness increased
- Fruit cleaning improved, better wax deposition and shorter drying time
- Often fungicide residues can be reduced
- Little line space needed
- Residue on fast and uniformly w/o rotary brushing

State of the art in 1925

Wood boxes with individually wrapped fruit (for soilage control) were the industry standard



Soak tank use common - contained soda ash, sodium bicarbonate, borax-boric acid. Investigators: UC (Klotz and Fawcett) and USDA (Barger CA) (Winston FL)

What can we put in the tanks?

- Sodium bicarbonate/sodium carbonate ('soda ash')
 - Common food components – baking soda
 - Compatible with fungicides, improves their performance
 - Sole 'Organic' option, no residue issues
 - Problems: high pH, weight loss, difficult disposal in some areas, effectiveness can be poor
- Potassium sorbate
 - Very common organic acid food preservative
 - Compatible with fungicides, improves their performance
 - Exempt active ingredient EPA sec25b
 - Problems: weight loss, acceptance in Japan, effectiveness can be poor
- Potassium/calcium phosphite
 - Common fungicide for Oomycetes
 - Registration coming soon
 - Evaluation in progress, effectiveness relatively high
 - Problems: cost, weight loss





Why is the use of heated solutions and tanks still common in California?

Solutions compared

Lemons inoculated 24 h before treatment, 1 min immersion in each solution, followed by 10 days at 20C

Treatment	Green mold incidence (%)				Blue mold incidence (%)			
	Percentage	Stdev	Percentage	Stdev	Percentage	Stdev	Percentage	Stdev
Water	100.0 a	0.0	58.3 a	14.7	32.1 ab	6.0	23.8 a	3.9
2% K-Silicate	77.4 b	8.1	52.4 a	6.7	53.5 a	12	11.9 ab	8.2
2% Na ₂ CO ₃	53.3 cd	15	35.7 ab	13.7	21.9 bc	9	7.1 bc	6.1
2% NaHCO ₃	38.1 cdi	20	32.1 ab	4.6	25.6 bc	6	15.5 abc	12.5
2% K-Phosphite	59.5 bc	4.8	16.7 bc	9.1	5.5 bc	10	2.4 bc	2.7
2% K-Sorbate	23.8 e	5.5	6.0 c	2.4	7.1 c	6.1	3.6 bc	4.6
2% Ca-Phosphite	35.7 de	4.8	4.8 c	4.8	6.0 c	4.6	1.0 c	2.1

* Heated solutions were much more effective

Phosphoric acid Phosphorus	Phosphorous acid Phosphite
Necessary for plants and all life	Occurs in nature but rare
Fertilizer component, no fungicide properties	Fungicide activity
Valence = 5+ Accepts 5 electrons	Valence = 3+ Accepts 3 electrons
	

Ammonia fumigation

Does ammonia fumigation merit re-evaluation?


- Simple, natural compound with known toxicology.
- Commercial sources of NH₃ are many and its price is low (\$1.47 per lb)
- Previously, it was exempt from residue tolerances and may still be.
- High pH of ammonia should show synergy in interactions with modern fungicides and help manage fungicide resistant isolates.
- Insecticidal and microbial activity may be high and useful – this needs evaluation.

Soak tank solutions compared

	Sodium bicarbonate Soda ash	Potassium sorbate	Potassium phosphite
Effectiveness alone for green mold	+ to ++	++	++
When combined with imazalil + heat	++++	++++	++++
pH (1.5% sol'n)	8.3 - 11.5	6.5	6 - 6.5
Conductivity	12.2	8.3	18.7
Sodium	yes	no	no

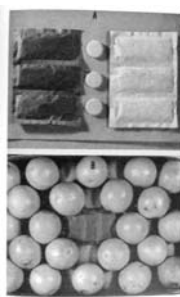
Ammonia fumigation

Steve Tebbets USDA ARS
Clara Montesinos IVIA, Valencia, Spain



Ammonia fumigation

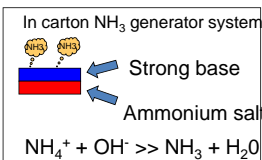
- First report by Bottini (1927), then many others in 1930s; NH₃ sublimed from ammonium bicarbonate reduced *Penicillium* and *Diplodia* decay, some rind injury.
- 1950 to 1960s both in-package generators and whole room fumigation evaluated.
 - NH₃ from in-package generators worked effectively, but under commercial conditions performance was too variable and they were not implemented
 - NH₃ room fumigation was not implemented because it was unconventional and the newly introduced (at that time) fungicides controlled decay effectively and fungicide resistance was then rare.



NH₃ rates:

- 50-200 ppm constantly
- 500-3000 ppm in a single fumigation

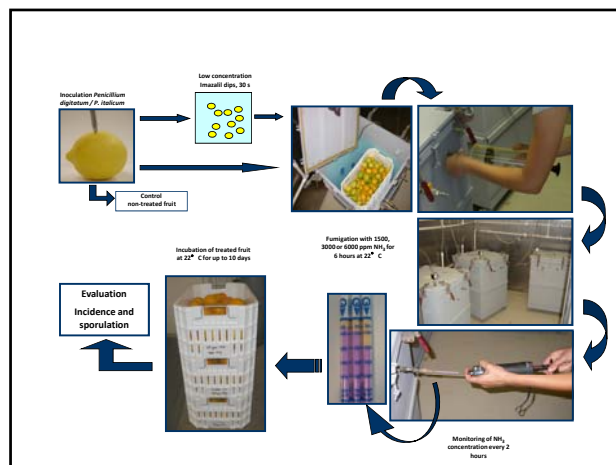
In carton NH₃ generator system

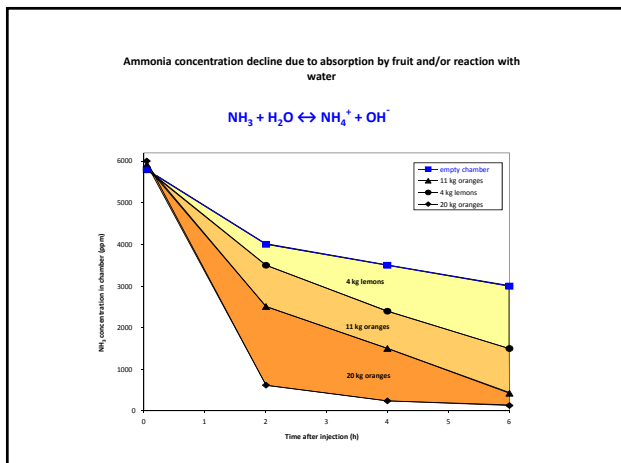


Strong base
Ammonium salt

$$\text{NH}_4^+ + \text{OH}^- \gg \text{NH}_3 + \text{H}_2\text{O}$$

Eckert et al. 1963 Phytopathology 53:140-148.





Ammonia and imazalil interactions

pH greatly influences imazalil toxicity, and ammonia raises pH

Smilanick et al. 2005, Plant Dis. 89:640-648.

NC1=CC=C(Cl)C(Cl)=C1C(C)CNC

$NH_3 +$ Ionized *Weaker fungicide* \rightleftharpoons Neutral *Stronger fungicide* $+ NH_4^+$

$pK_b = 6.53$

Influence of ammonia fumigation on fruit quality

Ammonia fumigation to 6000 ppm did not harm Valencia oranges or Eureka lemons, but caused wounds to darken on the lemons

The pH of the wounds several hours after treatment was increased by 0.6, 0.9, and 1.3 units, respectively, by fumigation with 1500, 3000, or 6000 ppm NH_3 .

Synergy between imazalil and ammonia

Imazalil 30 ppm dip: 53%
 Ammonia 1500 ppm: 73%
 Both: 7%
 CONTROL: 0%

Ammonia fumigation accelerated natural loss of green color in lemons

Post-fumigation storage for 3 weeks at 10°C

L 61.03 C 47.85 Hue 106.7 L 54.78 C 45.96 Hue 113.1

Ammonia fumigated (6000 ppm) Untreated

Ammonia gas fumigation

- Citrus fruit tolerate ammonia gas well and it was a promising method to control postharvest decay.
- Compressed ammonia cylinders are widely available and inexpensive (150 lbs/\$220). Toxicological issues probably few.
- Initial test showed old work repeated well in a preliminary test.
- Rates of 3000 ppm (initial) for 6 hours were effective; injury potential seen at 6000 ppm on lemons. Risk of injury needs evaluation.
- Ammonia darkened and raised pH of wounds – increased pH should improve imazalil activity markedly, perhaps other fungicides as well.
- Ammonia fumigation increased rate of natural degreening of lemons – could it be done before degreening within the same rooms?
- Insecticidal and microbial activity of ammonia applied by fumigation needs evaluation.