

Research Reports

Effect of Cultivar, Ethephon, Flooding, and Storage Duration on Sweetpotato Internal Necrosis

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SUMMARY. The reason for internal necrosis occurrences in sweetpotato (*Ipomoea batatas*) storage roots is not well understood. This disorder begins internally in the storage roots as small light brown spots near the proximal end of the root that eventually can become more enlarged as brown/black regions in the cortex. The objective of this study was to determine the effect of ethephon and flooding on the development of internal necrosis in the sweetpotato cultivars Beaugard, Carolina Ruby, and Covington over storage durations from 9 to 150 days after harvest (DAH) when roots had been cured. Soil moisture treatments were no-flooding, and simulated flooding that was created by applying 10 inches of overhead irrigation during 2 weeks before harvest. Ethephon was applied at 0, 0.75, and 0.98 lb/acre 2 weeks before harvest. Overall, 'Covington' and 'Carolina Ruby' had greater internal necrosis incidence (22% to 65% and 32% to 51%, respectively) followed by 'Beaugard' (9% to 22%) during storage duration from 9 to 150 DAH at both soil moistures. No significant change was observed for either internal necrosis incidence or severity for 'Beaugard' and 'Carolina Ruby' over the storage duration of 9–150 DAH. However, there was an increase of internal necrosis incidence and severity 9–30 DAH in 'Covington', with incidence and severity remaining similar 30–150 DAH. Storage roots in treatments sprayed with 0.75 or 0.98 lb/acre ethephon had higher internal necrosis incidence and severity compared with the nontreated, regardless of cultivars at both soil moistures. This research confirms that sweetpotato cultivars differ in their susceptibility to internal necrosis (incidence and severity), ethephon applied to foliage can contribute to internal necrosis development in storage roots, and internal necrosis incidence reaches a maximum by 30 DAH in 'Covington' and 9 DAH in 'Carolina Ruby' and 'Beaugard'.

North Carolina accounts for 56% of national sweetpotato production in the United States, with 95,000 acres of the crop harvested in 2016 (U.S. Department of Agriculture, 2017). Among all the sweetpotato cultivars, Covington is especially adapted to North Carolina growing conditions, producing high

yielding and high quality storage roots (Yencho et al., 2008), and because of its adaptability is grown on more than 88% of the commercial acreage across the state (Schultheis, 2016). 'Beaugard' is the predominant orange flesh cultivar grown in Louisiana and Mississippi (81% and 70% of acreage, respectively) because

of its high yield and excellent adaptation to the environmental conditions encountered in those states (Meyers, 2016; Rolston et al., 1987; Sistrunk and Smith, 2016). 'Carolina Ruby' is a specialty cultivar released by the North Carolina Agricultural Research Service (Collins et al., 1999). It is sold by many home gardening vendors but is also sold by certified seed growers in North Carolina for production in smaller commercial acreages for its attractive dark ruby skin, orange flesh, and outstanding taste (S. Scott, personal communication).

Internal necrosis occurs in the sweetpotato flesh near the proximal end as small light brown/black areas, but severe symptoms can extend half way longitudinally down the storage root with large brown/black areas across the entire cross section (Clark et al., 2013). Although precise statistics on the economic impact of this issue are not available, more than \$1 million was lost by one grower who had suffered from this problem during one season (J.R. Schultheis, unpublished data). A commercial storage survey of 'Covington' has revealed that internal necrosis is widespread in the North Carolina industry but high incidence and severity only happens in a few farms each year (Jiang et al., 2015). Incidence is erratic across years and among storage rooms, and in a survey, patterns of storage temperature and relative humidity after curing were not associated with the incidence (Jiang, 2013; Jiang et al., 2015). Incidence varies among cultivars, with Covington and Hatteras having significantly greater incidence than other commercially important cultivars, such as Beaugard (Clark et al., 2013). The symptoms of internal necrosis are not transferred over growing seasons/generations (Schultheis and Thornton, 2007) and no microorganisms have been consistently isolated from infected roots suggesting that it might be a stress-induced physiological disorder (Schultheis et al., 2009). No relationship between internal necrosis and registered insecticides or herbicides in sweetpotato production systems have been found (Beam et al., 2017; Jiang et al., 2015).

Preharvest ethephon spray and postharvest curing are two reported factors, and when used alone or interactively, which have caused elevated

incidence of internal necrosis (Clark et al., 2013; Jiang et al., 2015). Although ethephon is not registered for use in sweetpotato, an interest existed in pursuing a registration for this use because of the potential to reduce storage root damage at harvest through tightening the epidermis (Wang et al., 2013). In ‘Covington’ storage roots, ethephon induced–internal necrosis was first seen 6 DAH during curing, and incidence progressively increased to 30 DAH (Jiang et al., 2015). In the study, removing either ethephon or curing reduced necrosis incidence. However, curing is a standard healing process to maintain shelf life and improve quality; thus, growers continue to use this process (Edmunds et al., 2008). In many commercial cases, where roots were cured but had not been sprayed with ethephon (Jiang et al., 2015), necrosis incidence was still detected indicating that additional unknown factors in the field or storage might have contributed to the development of internal necrosis.

In years with heavy rainfall, excessive flooding may occur in sweetpotato fields in North Carolina. For example, in the major sweetpotato growing regions, an accumulated 9.1, 10.1, and 6.9 inches of rainfall was observed between 26 Sept. and 1 Oct. 2010 for Sampson, Nash, and Johnston counties, respectively (State Climate Office of North Carolina, unpublished data). In Sept. 2014, Johnston County experienced an 8.6-inch rainfall within a 1-week period. Depending on the soil texture, these levels of heavy and continuous precipitation could cause flooding in the field. Farmers have suggested that high internal necrosis occurs in storage

roots harvested from fields with heavy rainfall, which led to our hypothesis that excessive moisture may contribute to internal necrosis. With flooding, roots suffer oxygen deficiency (Jackson, 1985), which can increase the concentration of 1-aminocyclopropane-1-carboxylic acid (ACC), the precursor of ethylene, in flooded roots of tomato plants [*Solanum lycopersicum* (Else and Jackson, 1998)]. Delivery of ACC from roots to shoots in flooded plants also increased 6 h after flooding and coincided with epinastic leaf curvature. This increased ethylene synthesis in response to flooding has also been observed in sweetpotato plants (Patterson et al., 1979). Because ethephon, an ethylene-generating compound, has a known effect on the induction of internal necrosis, we hypothesized that a similar reaction to ethephon might be induced by flooding, which could elevate ethylene levels in roots and shoots of sweetpotato and thus cause elevated levels of internal necrosis.

The objective of our studies was to determine the effect of ethephon application rates, simulated flooding, and storage duration on the development of internal necrosis (incidence and severity) in storage roots of three sweetpotato cultivars. Results from these studies test our hypothesis that excessive moisture may contribute to internal necrosis in sweetpotato storage roots and will provide new information on the development of internal necrosis in longer term (9–150 d) storage after curing.

Materials and methods

The study was conducted at the Cunningham Research Station in Kinston, NC (lat. 35.31°N, long. 77.58°W), in a field with two soils containing a Lynchburg loamy fine sand (Fine-loamy, siliceous, semiactive, thermic Aeric Paleaquults) and Goldsboro loamy sand (Fine-loamy, siliceous, subactive, thermic Aquic

Paleudults) in 2009 and at the Horticultural Crops Research Station in Clinton, NC (lat. 35.02°N, long. 78.27°W), the soil was a Norfolk sandy loam (fine-loamy, kaolinitic, thermic typic Kandiudults) in 2011. Treatments included two types of soil moisture (no flooding or simulated flooding), three ethephon rates (0, 0.75, and 0.98 lb/acre), and three sweetpotato cultivars (Beauregard, Carolina Ruby, and Covington). In each study, the field was separated into two large plots with each soil moisture treatment applied to a single large plot, resulting in a design where soil moisture treatments are not replicated because of practical constraints. Within each large plot, ethephon rates and cultivars were arranged according to a split plot design with ethephon rates assigned to whole plots replicated four (in 2009) and three (in 2011) times in randomized complete blocks. Furthermore, cultivars were randomized to subplots within each ethephon rate. Each subplot was four rows wide (three treatment rows and one border row) and 100 ft long. Between row spacing was 3.5 ft and in-row plant spacing was 1 ft. Ethephon (Prep; Bayer CropScience, Research Triangle Park, NC) was applied 2 weeks before harvest on 29 Sept. 2009 and 3 Oct. 2011. Spray solution was delivered with 20 gal/acre using a carbon dioxide–pressurized backpack sprayer fitted with XR8002VS nozzles in 2009 and DG8002VS nozzles in 2011 (Teejet® Technologies, Wheaton, IL). A 6-h rain-free period was achieved after each year’s application, which met the stated conditions on the product label. For the simulated flooding treatment, sweetpotato plants received an average of 10 inches supplemental irrigation applied through a linear overhead irrigation system. Irrigation started the day after ethephon application, with the maximum output of 2 inches per

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Units

To convert U.S. to SI, multiply by	U.S. unit	SI unit	To convert SI to U.S., multiply by
0.4047	acre(s)	ha	2.4711
0.3048	ft	m	3.2808
9.3540	gal/acre	L·ha ⁻¹	0.1069
2.54	inch(es)	cm	0.3937
25.4	inch(es)	mm	0.0394
1.1209	lb/acre	kg·ha ⁻¹	0.8922
(°F – 32) ÷ 1.8	°F	°C	(°C × 1.8) + 32

day for 5–7 d, and then the field was allowed to dry so the harvesting equipment (commercial chain digger) could enter the field. The quantity of irrigation was selected to mimic soil moisture during worst-case scenarios of a tropical depression or hurricane, which can occur in eastern North Carolina. Sweetpotato storage roots were harvested on 14 Oct. 2009 and 18 Oct. 2011. In 2009 and 2011, 0.3 and 0.4 inches of rainfall, respectively, were recorded between ethephon application and harvest.

For internal necrosis evaluations, all storage roots were discarded 5 ft from each end of the plot to avoid cross contamination and 150 no. 1 storage roots (U.S. Department of Agriculture, 2005) were collected from the center, 90 ft of each plot. Roots from the same plant were separated into different subsamples. Samples were cured at 85% relative

humidity and 29 °C for 7 d and then stored at 13 °C (Edmunds et al., 2008). Twenty-five roots were sampled from each plot for internal necrosis evaluation at 9, 30, 60, 90, 120, and 150 DAH by slicing the top 2 inches of the proximal end horizontally into multiple 3-mm-thick segments. The percent of sweetpotato storage roots that showed internal necrosis symptoms at each sampling date is referred to as incidence. Severity was assessed (scale of 0 = no internal necrosis present to 9 = more than 80% necrosis) from the slice with the most internal necrosis symptomology from each no. 1 root, and rating averaged more than 25 roots. Data were analyzed with PROC MIXED (SAS version 9.3; SAS Institute, Cary, NC) and type III analysis of variance tables were obtained for incidence and severity. Fixed effects included year, ethephon rate,

cultivar, storage duration, soil moisture, plus all interactions. Replications within year and soil moisture combinations, and replications by ethephon within year and soil moisture were included as random effects. Tests of significance were not reported for the main effect or any interactions involving the nonreplicated factor soil moisture. Means were separated using the Tukey's honestly significant difference test at the 0.05 significance level.

Results and discussion

Years and year by treatment interactions were not significant ($P > 0.05$) for both internal necrosis incidence and severity, thus, data were pooled across both years. The three-way interactions among ethephon rate, cultivar, and storage duration were not significant ($P > 0.05$); therefore, only significant two-way interactions (cultivar by storage duration and cultivar by ethephon rate) are discussed for each soil moisture treatment to explain the effects of these factors on the development of internal necrosis (Table 1).

NO FLOODING. 'Beauregard' and 'Carolina Ruby' each had a stable incidence and severity of internal necrosis from 9 to 150 DAH whereas internal necrosis in 'Covington' increased within the first 30 DAH of storage, then generally did not increase after 30 DAH (Table 2). 'Carolina Ruby' and 'Covington' had a higher internal necrosis incidence and severity than 'Beauregard' at each storage time except at 9 DAH where

Table 1. Analysis of variance of the effects of cultivar (C), ethephon rate (E), and storage duration (SD) under no flooding (NF) or simulated flooding (SF) on internal necrosis incidence and severity of sweetpotato no. 1 storage roots.^z

Effect	df	Incidence		Severity	
		NF	SF	NF	SF
		P value			
C	2	0.008	0.072	0.060	0.597
E	2	0.147	0.071	<0.0001	0.061
SD	5	0.397	0.310	0.001	0.017
C × E	4	<0.001	<0.0001	0.001	0.508
C × SD	10	<0.001	0.026	0.001	0.015
E × SD	10	0.225	0.655	0.131	0.979
C × E × SD	20	0.811	0.937	0.980	0.779

^zYears and year by treatment interactions were not significant ($P > 0.05$) for both internal necrosis incidence and severity, thus, data were pooled across both years (2009 and 2011).

Table 2. Under no flooding, internal necrosis incidence and severity of sweetpotato no. 1 storage roots at selected time intervals after harvest as affected by interaction between sweetpotato cultivars (Beauregard, Covington, and Carolina Ruby) and storage duration.^z

Storage duration (d)	Incidence ^y			Significance	Severity ^y			Significance
	Beauregard	Carolina Ruby	Covington		Beauregard	Carolina Ruby	Covington	
		Significance			Significance			
9	22	41	36 b ^x	NS	1.9	2.8	2.2 b	NS
30	12 B	32 A	47 abA	*	2.4 B	4.0 A	4.3 aA	*
60	16 B	40 A	55 abA	*	1.8 B	3.4 A	3.8 aA	*
90	9 B	42 A	53 abA	*	2.1 B	3.9 A	4.3 aA	*
120	11 B	51 A	65 aA	*	1.5 B	3.4 A	4.3 aA	*
150	12 B	43 A	57 abA	*	1.2 C	3.2 B	4.4 a A	*
Significance	NS	NS	*		NS	NS	*	

^zData were pooled from year 2009 and 2011. Sweetpotato storage roots were cured at 85% relative humidity and 29 °C (84.2 °F) for 7 d, and then stored at 13 °C (55.4 °F).
^yPercent incidence = (number of roots with internal necrosis symptoms/25) × 100; severity was assessed (scale of 0 = no internal necrosis present to 9 = more than 80% necrosis) from the slice with the most internal necrosis symptomology from each no. 1 root, and rating averaged more than 25 roots.

^xMeans followed by different small letters in column for storage duration for each cultivar or capital letters in the row which compare a specific sampling time across cultivars are significantly different according to Tukey's honestly significant difference ($\alpha = 0.05$).

^{NS} Nonsignificant and significant at $\alpha = 0.05$, respectively.

all cultivars had the same level of internal necrosis incidence and severity.

An ethephon rate response was observed in 'Beauregard' where internal necrosis incidence increased with the increase of ethephon rates (Table 3). However, internal necrosis incidence and severity in 'Carolina Ruby' and 'Covington' were similar for both ethephon rates 0.75 and 0.98 lb/acre as well as higher compared with the nontreated control. Regardless of whether ethephon was applied or not, 'Beauregard' had significantly lower internal necrosis incidence and severity as compared with 'Carolina Ruby' and 'Covington'.

SIMULATED FLOODING. 'Beauregard' and 'Carolina Ruby' had consistent incidence and severity ratings throughout the storage period from 9 to 150 DAH, whereas both incidence and severity of 'Covington' went up

from 9 to 30 DAH and then remained consistent through 150 DAH (Table 4). 'Carolina Ruby' and 'Covington' had a higher incidence of internal necrosis than 'Beauregard' at each storage time except at 9 DAH where the difference between 'Beauregard' and 'Covington' was not significant. All cultivars had a similar level of internal necrosis and severity at each storage time except at 60 and 90 DAH where Carolina Ruby and Covington had higher internal necrosis severity than Beauregard.

There was an increase in internal necrosis incidence when ethephon rates increased from 0 to 0.98 lb/acre, regardless of cultivars (Table 5). In any case whether ethephon was applied or not, 'Carolina Ruby' and 'Covington' had significantly higher internal necrosis incidence as compared with 'Beauregard'. However, for internal necrosis severity only the

main effect of ethephon rate was significant, and severity increased from 1.9 to 3.8 when ethephon rates increased from 0 to 0.98 lb/acre, regardless of cultivars.

Even though the statistical analysis was not conducted to directly compare soil moisture treatments because of the limitation of the experimental design, response to cultivar by storage duration and cultivar by ethephon rate was similar for both soil moisture treatments (Tables 2–5). Significance was reported for both interactions for internal necrosis incidence and severity at both soil moistures except for the cultivar by ethephon rate interaction for severity (Table 5). The internal necrosis severity was higher for 'Carolina Ruby' and 'Covington' as compared with 'Beauregard' for each ethephon rate under no flooding (Table 3); however this difference in cultivars did not

Table 3. Under no flooding, internal necrosis incidence and severity of sweetpotato no. 1 storage roots as affected by interaction between sweetpotato cultivars (Beauregard, Covington, and Carolina Ruby) and ethephon rate applied 2 weeks before harvest.^z

Ethephon rate (lb/acre) ^y	Incidence ^x			Significance	Severity ^x			Significance
	Beauregard	Carolina Ruby (%)	Covington		Beauregard	Carolina Ruby	Covington (0 to 9 scale)	
0.98	23 aB ^w	61 aA	65 aA	*	2.5 aB	4.5 aA	4.6 aA	*
0.75	14 bC	54 aB	67 aA	*	1.9 abB	4.0 aA	4.5 aA	*
0 (nontreated)	3 cC	10 bB	24 bA	*	1.1 bB	1.9 bAB	2.5 bA	*
Significance	*	*	*		*	*	*	

^zData were pooled from year 2009 and 2011. Sweetpotato storage roots were cured at 85% relative humidity and 29 °C (84.2 °F) for 7 d, and then stored at 13 °C (55.4 °F).

^y1 lb/acre = 1.1209 kg·ha⁻¹.

^xPercent incidence = (number of roots with internal necrosis symptoms/25) × 100; severity was assessed (scale of 0 = no internal necrosis present to 9 = more than 80% necrosis) from the slice with the most internal necrosis symptomatology from each no. 1 root, and rating averaged more than 25 roots.

^wMeans followed by different small letters in column for each cultivar in response to ethephon rate or capital letters in the row which compare a specific ethephon rate across cultivars are significantly different according to Tukey's honestly significant difference ($\alpha = 0.05$).

^{ns} *Nonsignificant and significant at $\alpha = 0.05$, respectively.

Table 4. Under simulated flooding, internal necrosis incidence and severity of sweetpotato no. 1 storage roots at selected time intervals after harvest as affected by interaction between sweetpotato cultivars (Beauregard, Covington, and Carolina Ruby) and storage duration.^z

Storage duration (d)	Incidence ^y			Significance	Severity ^y			Significance
	Beauregard	Carolina Ruby (%)	Covington		Beauregard	Carolina Ruby	Covington (0 to 9 scale)	
9	15 B	40 A	22 bB [*]	*	1.7	3.0	1.8 b	NS
30	12 B	32 A	32 abA	*	2.9	4.4	3.4 a	NS
60	16 B	36 A	41 abA	*	1.8 B	3.4 A	3.2 aA	*
90	13 B	40 A	44 abA	*	2.5 B	3.7 A	3.8 aA	*
120	16 B	45 A	49 aA	*	2.3	3.2	3.3 a	NS
150	16 B	48 A	43 abA	*	2.3	3.2	3.4 a	NS
Significance	NS	NS	*		NS	NS	*	

^zData were pooled from year 2009 and 2011. Simulated flooding was created by applying 10 inches (25.4 cm) of overhead irrigation during 2 weeks before harvest. Sweetpotato storage roots were cured at 85% relative humidity and 29 °C (84.2 °F) for 7 d, and then stored at 13 °C (55.4 °F).

^yPercent incidence = (number of roots with internal necrosis symptoms/25) × 100; severity was assessed (scale of 0 = no internal necrosis present to 9 = more than 80% necrosis) from the slice with the most internal necrosis symptomatology from each no. 1 root, and rating averaged more than 25 roots.

^{*}Means followed by different small letters in column for storage duration for each cultivar or capital letters in the row which compare a specific sampling time across cultivars are significantly different according to Tukey's honestly significant difference ($\alpha = 0.05$).

^{ns} *Nonsignificant and significant at $\alpha = 0.05$, respectively.

Table 5. Under simulated flooding, internal necrosis incidence and severity of sweetpotato no. 1 storage roots as affected by sweetpotato cultivars (Beauregard, Covington, and Carolina Ruby) and ethephon rate applied 2 weeks before harvest.^z

Ethephon rate (lb/acre) ^y	Incidence ^x			Significance	Severity ^x (0 to 9 scale)
	Beauregard	Carolina Ruby (%)	Covington		
0.98	23 aB ^w	61 aA	57 aA	*	3.8 a
0.75	16 bB	42 bA	44 bA	*	3.2 b
0 (nontreated)	4 cB	18 cA	14 cA	*	1.9 c
Significance ^y	*	*	*		*

^zData were pooled from year 2009 and 2011. Simulated flooding was created by applying 10 inches (25.4 cm) of overhead irrigation during 2 weeks before harvest. Sweetpotato storage roots were cured at 85% relative humidity and 29 °C (84.2 °F) for 7 d, and then stored at 13 °C (55.4 °F).

^y1 lb/acre = 1.1209 kg·ha⁻¹.

^xPercent incidence = (number of roots with internal necrosis symptoms/25) × 100; severity was assessed (scale of 0 = no internal necrosis present to 9 = more than 80% necrosis) from the slice with the most internal necrosis symptomatology from each no. 1 root, and rating averaged more than 25 roots.

^wMeans followed by different small letters in column for each cultivar in response to ethephon rate or capital letters in the row which compare a specific ethephon rate across cultivars are significantly different according to Tukey's honestly significant difference ($\alpha = 0.05$).

^{ss}*Nonsignificant significant at $\alpha = 0.05$, respectively.

occur under simulated flooding (Table 5). This apparent occasional difference in internal necrosis symptom response between soil moisture indicated that excessive moisture might not be a causal factor for internal necrosis. Instead, the moisture levels in the field in our study may have indirectly changed the accessibility of plants to ethephon chemical spray, thereby affecting the internal necrosis severity. Variations in internal necrosis response may have resulted because of the wilting of plants, which are typical responses of root flooding shown in multiple species; e.g., tomato, wheat (*Triticum aestivum*), and sunflower (*Helianthus annuus*) (Bradford and Hsiao, 1982; Sojka and Stolzy, 1980). The physiological changes of plants due to flooding may reduce the transport of ethephon within plant tissue and thus variation in internal necrosis severity reported in cultivars under both soil moisture conditions.

Overall, these results showed that storage period from 9 to 150 DAH did not change the internal necrosis incidence and severity in 'Beauregard' and 'Carolina Ruby'. However, in 'Covington' both incidence and severity of internal necrosis increased from 9 to 30 DAH. Jiang et al. (2015) reported a progressive increase of internal necrosis incidence and severity in 'Covington' from 0 to 30 DAH, and noted incidence occurred near 6 d in storage, before roots were completely cured. Similarly, samples from North Carolina commercial facilities have revealed that the incidence and severity of 'Covington' does not seem to change over time from the end of curing to 6 months after storage. Our studies

confirm that incidence and severity of internal necrosis increased shortly after harvest, whereas storage longer than 1 month had little effect on the continued development of this disorder. Unlike 'Covington', this study did not identify an increasing amount of internal necrosis for 'Beauregard' and 'Carolina Ruby', suggesting that development of internal necrosis in these two cultivars, might be sooner than Covington as reported by Jiang et al. (2015). Previous research indicated that cultivars differ in internal necrosis incidence and severity. Beam et al. (2017) and Clark et al. (2013) reported lower internal necrosis incidence in 'NC 05-198' as compared with 'Covington'. Similar cultivar differences were observed in our study with greater internal necrosis incidence and severity in Carolina Ruby and Covington compared with Beauregard.

Our studies confirm that incidence and severity of internal necrosis in sweetpotato roots increased with ethephon application. Similar results were reported by other researchers, and found that ethephon application was associated with the development of internal necrosis in several sweetpotato cultivars (Arancibia et al., 2013; Beam et al., 2017; Clark et al., 2013; Jiang et al., 2015). Arancibia et al. (2013) also reported that an increase in internal necrosis incidence was correlated with ethephon rate and timing of application before harvest.

Preharvest spray of ethephon can consistently enhance internal necrosis in all cultivars; however, even without ethephon application, internal necrosis still occurred in all three tested cultivars. This suggests the existence of other contributors of internal

necrosis, such as those that mimic ethephon and ethylene behavior in the field, or those that are related to the curing process after harvest. Future research may focus on the genetic screening for nonsusceptible cultivars, as well as on optimizing curing conditions and lengths for commercially important cultivars.

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