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Chlorophyll fluorescence of Delicious apples at harvest as a potential predictor of superficial scald development during storage

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Abstract

Chlorophyll fluorescence (Fv) of Delicious apples at harvest was evaluated as an indicator of superficial scald development during storage. The Fv of 'Imperial' and 'Sturdeespur' Delicious apples was determined at three weekly harvest intervals, and the apples were later evaluated for scald intensity after 2, 4 and 6 months of storage at 0°C. 'Sturdeespur' apples had an overall higher incidence of scald than 'Imperial' apples. Fv at harvest was found to correlate positively with scald development in 'Sturdeespur' apples from Harvest 1. 'Sturdeespur' apples with low Fv (<10) at Harvest 1 were least likely to develop scald, while those with high Fv (>18) were most likely to develop severe scald. However, no such relationship was observed for 'Imperial' apples, or for some 'Sturdeespur' apples from harvests 2 and 3. The results of this study suggest that Fv at harvest may be a predictor of scald development.

Keywords: Malus domestica Borkh. cv. Delicious; 'Imperial'; 'Sturdeespur'

1. Introduction

Chlorophyll fluorescence techniques are often used to detect stress in plant tissue (Lichtenhaler, 1988; Schreiber and Bilger, 1987). These techniques have been used in postharvest studies of chilling injury in banana and mango (Smillie et al., 1987), in cucumbers (van Kooten et al., 1992) and in green bell peppers (Lurie et al., 1994).

There is presently no industrially adopted method to rapidly and nondestructively detect postharvest stress in apples before associated disorders develop. However, a previous study (DeEll et al., 1995) found chlorophyll fluorescence to be a measure of

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metabolic changes associated with low O_2 and high CO_2 stress in McIntosh apples during storage. The characteristic pattern of fluorescence is altered in apples, due to low O_2 or high CO_2 stress, before any associated visual symptoms are observed.

Superficial scald is a common storage disorder of apples, which is characterized by diffuse browning of the skin (Ingle and D'Souza, 1989). Since scald is considered to be a chilling-related disorder (Watkins et al., 1995), the objective of this study was to evaluate the potential use of chlorophyll fluorescence techniques at harvest as a rapid, nondestructive predictor of scald development in Delicious apples during storage.

2. Materials and methods

Plant material

'Imperial' (standard) and 'Sturdeespur' (spur-type) Delicious apples were harvested from an orchard in the Annapolis Valley, NS. Since both strain and harvest time have been shown to affect scald susceptibility in Delicious apples (Lord and Southwick, 1964; Meheriuk and Porritt, 1971), two 20-kg samples of each strain were harvested on October 6, 13 and 20, 1993, representing Harvests 1, 2 and 3, respectively.

Chlorophyll fluorescence measurements

Chlorophyll fluorescence was determined using a Plant Productivity Fluorometer Model SF-20 (Richard Brancker Research, Ottawa, Ontario). Apples were warmed to 20°C and dark-adapted for at least 4 hours prior to measurements. The SF-20 probe, with a light intensity of 20 μmol m^{-2} s^{-1}, was placed firmly on the side of the apple. The peak (P) and terminal (T) values of the fluorescence induction curve were recorded from the SF-20 digital display. The T value was obtained after 50 s. Each apple was numbered, to ensure that subsequent readings would be from the same apples. Variable fluorescence (Fv), calculated as $\left((P-T)/P\right) \times 100$, was used in the statistical analyses.

Storage treatments

One sample (80 apples) of each strain from each harvest was treated with 1500 ppm diphenylamine (DPA) to control scald development, while another similar sample was not treated with DPA. Chlorophyll fluorescence measurements were then taken on all apples of the 12 samples (2 strains × 3 harvests × 2 treatments). All samples were stored at 0°C in ambient air, with thin perforated polyethylene over the top of the vented plastic container to reduce moisture loss. After 2, 4 and 6 months of storage, the apples were removed and warmed to 20°C. Each apple from each harvest was then assessed for scald development (0–3 scale; 0 = none, 3 = severe).

Statistical analysis

Samples of apples were completely randomized, with a factorial arrangement of treatments. Scald incidence was analyzed using the regression procedure of Genstat 5 (Payne, 1993). A binomial distribution with a logit link function was used, and the terms Strain and Harvest Time were fitted into the model. Separate analyses were performed for data from each storage removal (2, 4 and 6 months).

Correlation coefficients between Fv at harvest and scald development after storage were determined using Genstat 5. DPA-treated and non-DPA apples were combined to provide a range of scald intensities from 0 to 3. The relationship between Fv at harvest and scald after storage was studied further using the data from 'Sturdeespur' apples from Harvest 1 and the regression procedure of Genstat 5.

Changes in the scald ratings, as a function of Fv, were assessed after grouping the apples into six Fv categories with mid points: 8, 11, 13, 15, 17 and 20. A model to describe the probabilities of apple samples having scald ratings of 0, 1 or ≥2 was derived as a function of its measured Fv. Scald ratings of 2 or 3 were combined into one category (≥2) for these analyses, to avoid an empty category for statistical purposes. Since the scald ratings have an ordered structure, a generalized linear model with an assumed multinominal distribution and a logit link function was used to quantify the relationship between increasing scald ratings and Fv at harvest. Analysis of deviance was used to determine whether the effect of Fv could be represented simply as a linear regressor, or covariate, or whether a more complex model was required.

Probabilities γ_{oj}, γ_{1j}, and γ_{2j} are those associated with an apple being in scald category 0, 1, or ≥2 for a given Fv at harvest x_j. The basic relationship was derived on the log-odds scale as given by McCullagh and Nelder (1989),

$$\ln\left(\gamma_{ij}/(1-\gamma_{ij})\right) = \theta_i - \beta x_j \text{ for } i = 0,\ 1; \quad 8 \le x_j \le 20$$

The cut point parameters θ_i, are associated with the cumulative probabilities of a response being in scald categories: 0 alone, and in either 0 or 1. This basic model was compared with more complex models to determine how well the three parameters (θ_0, θ_1, β) described the scald by chlorophyll fluorescence table of counts. For a given value of x_j, the following estimates were derived:

$$\hat{\gamma}_{1j} = \exp(\hat{\theta}_1 - \hat{\beta}x_j)/\left(1 + \exp(\hat{\theta}_1 - \hat{\beta}x_j)\right) - \hat{\gamma}_{oj}$$

$$\hat{\gamma}_{oj} = \exp(\hat{\theta}_0 - \hat{\beta}x_j)/\left(1 + \exp(\hat{\theta}_0 - \hat{\beta}x_j)\right)$$

$$\hat{\gamma}_{2j} = 1 - \hat{\gamma}_{0j} - \gamma_{1j}$$

Only sources of variation that were significant at $P < 0.05$ are discussed.

3. Results and discussion

'Sturdeespur' apples developed more scald than did 'Imperial' apples from each harvest (Table 1). This was expected since spur-type Delicious apples are more susceptible to scald than are non-spur strains (Meheriuk and Porritt, 1971). However, an exception was observed after 6 months of storage, with 'Imperial' apples from Harvest 2 having an unusually high incidence of scald.

'Sturdeespur' apples from Harvest 1 had the highest correlation coefficients (r) of Fv at harvest and scald intensity after storage ($P < 0.01$), while 'Sturdeespur' apples from Harvests 2 and 3 also had significant r values after 4 months of storage (Table 2). On the other hand, there was no significant correlation between Fv at harvest and scald intensity after storage in any 'Imperial' apples, or in 'Sturdeespur' apples from Harvest 2 and 3 after 2 and 6 months of storage.

Table 1
Scald incidence (%) of 'Imperial' and 'Sturdeespur' Delicious from three harvest times after 2, 4 and 6 months of storage at 0°C

Factors	Scald (%)		
Storage time:	2 months	4 months	6 months
Harvest 1			
Imperial	0	1.25	3.13
Sturdeespur	17.50	20.63	26.25
Harvest 2			
Imperial	0	1.90	26.88
Sturdeespur	3.75	5.00	29.38
Harvest 3			
Imperial	0	1.90	6.88
Sturdeespur	1.25	3.13	18.75
Significance [a]	S***, H***	S**, H*	NS*

[a] (n = 240, df = 11), using regression with a binomial distribution and logit link function.
NS, *,**,*** significant at $P < 0.05$, $P < 0.01$ or $P < 0.001$, respectively, where S = strain and H = harvest.

Table 2
Correlation coefficients (r) of Fv at harvest with scald intensity of 'Imperial' and 'Sturdeespur' Delicious from three harvest times, after 2, 4 and 6 months of storage at 0°C

Factors	Correlation coefficients (r) [a]		
Storage time:	2 months	4 months	6 months
Harvest 1			
Imperial	−0.02	−0.05	0.10
Sturdeespur	0.47 **	0.43 **	0.50 **
Harvest 2			
Imperial	−0.10	0.00	0.13
Sturdeespur	0.19	0.31 **	0.11
Harvest 3			
Imperial	0.19	0.21	0.10
Sturdeespur	0.02	0.28 *	0.06

[a] *,**significant at $P < 0.05$ or $P < 0.01$, respectively.

Correlation is only a measure of strength of the relationship between Fv at harvest and scald development, so regression analysis was used to describe the relationship that existed for 'Sturdeespur' apples from Harvest 1. 'Sturdeespur' apples with Fv <14 at Harvest 1 developed less scald after storage than those with Fv >14 which had higher scald ratings (Table 3). In the Analysis of Deviance for these data, the Fv at harvest was significant at $P < 0.001$ as an explanatory variate of scald development after storage (Table 3).

The probabilities of 'Sturdeespur' apples from Harvest 1 having a scald rating of 0, 1 or ≥2 after storage based on Fv at harvest were determined using log-odds (Table 3).

Table 3
Relationship of Fv at harvest to scald rating after 6 months of storage at 0°C for 'Sturdeespur' apples from Harvest 1, and the probability of scald at a particular Fv based on the accumulated analysis of deviance

Scald rating	Fv					
	<10	10–12	12–14	14–16	16–18	>18
	% apples [a]					
0 none	11	6	12	5	1	0
1 mild	5	4	13	10	6	1
≥2 severe	1	2	2	7	10	4
	Probability of scald [b]					
0 none	0.74	0.51	0.34	0.21	0.12	0.05
1 mild	0.22	0.38	0.46	0.46	0.39	0.23
≥2 severe	0.04	0.11	0.20	0.33	0.49	0.73

[a] % = (#/160 apples) × 100
[b] Accumulated analysis of deviance

Source of change	df	Mean deviance	Parameter	Estimate	SE
Fv	1	44.8***	θ_0	3.75	0.740
Residual	21	1.02	θ_1	5.78	0.825
			β	0.34	0.056

*** significant at $P < 0.001$.

The probability of a 'Sturdeespur' apple having a scald rating of ≥2 after 6 months of storage ranged from 4% when Fv at harvest was <10 to 73% when Fv was >18, while the probability of not developing scald ranged from 74% when Fv was <10 to 5% when Fv was >18. This pattern was similar after 2 and 4 months of storage (data not presented).

Scald develops as a result of free radicals oxidizing α-farnesene and producing conjugated trienes, which are toxic to the cells and result in the characteristic brown discoloration (Ingle and D'Souza, 1989). The higher Fv in 'Sturdeespur' apples from Harvest 1 that was associated with scald development, may be due to either an increase in free-radical production or a decrease in free-radical scavenger activities (O. van Kooten, personal communication). van Kooten et al. (1994) and Tijskens et al. (1994) found fluorescence yield to be a measure of radical scavengers in chilling injury in fruits and developed a statistical model based on fluorescence for determining the occurrence of radical damage at the membrane level before any visual symptoms associated with chilling injury occur.

4. Conclusion

The results of this study suggest that scald development in Delicious apples may be predicted by chlorophyll fluorescence techniques at harvest. However, there was no consistency in this method and harvest time and strain appeared to influence its efficacy.

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