



Managing the Flesh
Browning Disorder of

Cripps Pink Apples

A summary of Australian research investigating
the causes and management of the problem

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Managing the Flesh Browning Disorder of Cripps Pink Apples.

A summary of Australian research investigating the causes and management of the problem.



Know-how for Horticulture™

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This publication covers research conducted by research projects HAL AP02009 "Understanding Flesh browning in Cripps Pink apples" and HAL AP04008 "Continuation of the understanding of the flesh browning disorder of Pink Lady™ apples to validate recommendations".

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Introduction

This guide is a summary of the key outcomes from a 6 year study aimed at understanding the flesh browning disorder in Cripps Pink apples. The project involved international collaboration with 6 Institutes from USA, Italy, New Zealand and Australia.

Over the 6 years there have been 2 research projects funded through Horticulture Australia Ltd;

- 1. HAL AP02009** "Understanding Flesh browning in Cripps Pink apples" 2002 - 2004
- 2. HAL AP04008** "Continuation of the understanding [of the flesh browning disorder of Pink Lady™ apples to validate recommendations" 2004 – 2007

Project AP02009 was facilitated by HAL in partnership with APAL. It was funded by the Apple levy and voluntary contributions from industry. The Australian Government provides matched funding for all HAL's R&D activities. Voluntary Contributions were received from Pink Lady Australia, University of California, Davis, HortResearch, New Zealand, and Research Centre for Agriculture and Forestry Laimburg, South Tyrol, Italy.

Project AP04008 was facilitated by HAL in partnership with the Apple Industry. It was funded by voluntary contributions from industry. The Australian Government provides matched funding for all HAL's R&D activities. Voluntary Contributions were received from Maersk Sealand, Department of Agriculture and Food Western Australia, Pink Lady Australia, University of California, Davis, HortResearch, New Zealand, and Valent Biosciences.

The success of this project has been the result of the effective collaboration between the financial and scientific partners and also the commercial growers and exporters.

This work has gone a long way to providing a practical management strategy for the flesh browning disorder in Cripps Pink apples. It is important to make the point that this management strategy is built on a sound scientific understanding of the physiology of the flesh browning disorder.

If you need any more information please consult the sources listed in the reference lists provided or contact the authors.

Naming Convention "Pink Lady™" apples

Pink Lady™ is a trade mark rather than a variety name. The base variety is Cripps Pink. Cripps Pink apples that comply with quality specifications set down by Apple and Pear Australia Limited may be sold as Pink

Lady™ apples. Simply put, Pink Lady™ branded apples are high quality Cripps Pink apples that meet specific quality criteria.

The Cripps Pink Apple

The Cripps Pink apple cultivar was developed in the Western Australian apple breeding program and was released for commercial evaluation in 1986 (Cripps et al., 1993). The Cripps Pink cultivar has since become a major variety around the world and is regarded as a cultivar with premium qualities. It has a distinctive colour and flavour and does not have the tendency to go mealy during long term storage.

The Pink Lady™ trademark is owned by Apple and Pear Australia Ltd and a royalty is paid by growers or licensees to cover licensing and marketing costs associated with the trademark branding and

development. The combination of a unique set of quality characteristics of the apple and a targeted international marketing effort has made this variety of apple one of the most popular around the world.

This project was initiated in response to the first occurrences of flesh browning in 2000 in Australia (Jobling et al., 2003; Brown et al., 2003; Wilkinson, 2000). The industry took the initiative to support a collaborative research effort that would help manage the disorder and in turn ensure that the integrity of the variety, marketing and trademark would be maintained.

References

Brown, G., L. Schimanski, and D. Jannings. 2003. Investigating internal browning of Tasmanian 'Pink Lady' apples. *Acta Horticulturae*. 628, 161-166.

Cripps, J.E.L., L.A. Richards, and A.M. Mairata. 1993. 'Pink Lady' apple. *HortScience*. 28, 1057.

Jobling, J, Brown, G, Mitcham, E, Tanner, D, Tustin, S, Wilkinson, I, Zanella, A (2004) Flesh browning of Pink Lady™ apples: Why do symptoms occur? Results from an international collaborative study. *Act Hort*. 682:851 – 858.

James, H., G. Brown, E. Mitcham, D. Tanner, S. Tustin, I. Wilkinson, A. Zanella, and J. Jobling. (2005). Flesh browning in Pink Lady™ apples: research results have helped to change market specifications for blush colour which is an added bonus for growers. *Proceedings of the International Conference Postharvest Unlimited Downunder 2004*. *Acta Horticulturae*. 687, 175-180.

James, Hannah (2007). Understanding the flesh browning disorder in 'Cripps Pink' apples. PhD Thesis, Faculty of Agriculture, Food and Natural Resources, University of Sydney, Sydney 2006 NSW, Australia.

Studdert, J., 2002. A Pink Lady rides to the rescue, *The Australian*, Sydney.

Wilkinson, I., 2000. Pink Lady apples - maintaining export quality, IHD Media Release, IHD Knoxfield, Knoxfield.



Overall Recommendations

Table 1 summarises the recommendations for radial and diffuse flesh browning in Cripps Pink apples. Please take particular note of the subscripts and disclaimers in the Table.

Table 1 Recommendations for the optimal growth, harvest and storage of ‘Cripps Pink’ apples for the prevention of the diffuse flesh browning (DFB) and radial flesh browning (RFB) disorders. The severity of browning increases in cooler seasons for both diffuse and radial flesh browning.

	Diffuse Flesh Browning	Radial Flesh Browning
Classification	Chilling injury	Senescent breakdown
Climatic range	<1100 GDD	>1100 GDD ^z
Maturity	SPI ^y 3.5	SPI 3.5
Storage temperature	3°C ^x	1°C ^w or stepwise cooling ^v
Storage atmosphere	<1% CO ₂	<1% CO ₂
Orchard management	Ensure calcium levels adequate	Best commercial practice ^u

^z Insufficient data in the climatic range of 1100-1400 growing degree days (GDD) >10°C has currently been collected, the type of flesh browning that develops in this range has not been determined however it is likely that the recommendations for RFB will be suitable as a guide.

^y Starch pattern index (SPI) recommendation is based on the CTIFL 10 point scale.

^x Storage at 3°C will prevent the development of DFB, however storage at 1°C will reduce symptoms. Storage at 3°C will reduce the period of storage time before the loss of quality occurs.

^w Storage at 1°C was found to be successful for the prevention of RFB, however this was in a low risk season, in a high risk season, storage at a higher temperature may be required.

^v Stepwise cooling recommendation is 2 weeks at 3°C followed by 2 weeks at 2°C then the remainder of the storage period at 1°C.

^u Best commercial practice for the management of crop load and fruit nutrition are recommended.

With only three complete seasons, this research has identified and classified Australian regions that are susceptible to DFB and RFB and it has determined the pre and postharvest factors that are involved in the development of both of these disorders. This project has demonstrated how targeted research can lead to the physiological determination of a storage disorder in a relatively short timeframe. A physiological understanding of a disorder can lead to the accurate identification of risk factors allowing for the rapid establishment of commercial recommendations.

It is important to note that these results are still preliminary and more work is required to determine the effectiveness of the strategies outlined in years where the seasonal risk of flesh browning in “Cripps Pink” apples is high.

References

Hannah James (2007). Understanding the flesh browning disorder in ‘Cripps Pink’ apples. PhD Thesis, Faculty of Agriculture, Food and Natural Resources, University of Sydney, Sydney 2006 NSW, Australia.

The Flesh Browning Disorder

Flesh browning was first recognised in Tasmania in fruit that had been stored in controlled atmosphere storage and the disorder was soon reported throughout other growing regions of Australia and around the world (Brown et al., 2003). In the year 2003, 35 containers of Cripps Pink apples that had been exported from Australia to the United Kingdom had been rejected due to the presence of the

flesh browning disorder (Anon., 2004), representing a large economic loss to Australian apple growers and the potential loss of the reputation of the Cripps Pink brand.

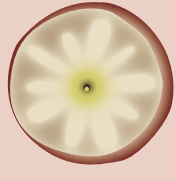
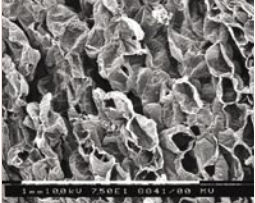

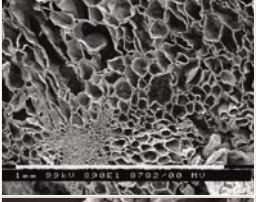

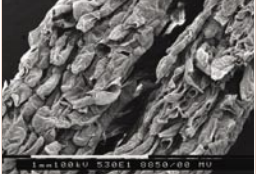
Being such an unpredictable and intermittent disorder it could rapidly undermine the established confidence in the cultivar and erode the market advantage that had been developed.

Classification of the Flesh Browning Disorder

Originally thought of as a single disorder, the results of this project demonstrated that the flesh browning disorder of 'Cripps Pink' apples could be described as three distinct disorders. The flesh browning disorder was defined as, diffuse flesh

browning, radial flesh browning and CO₂ injury based on the visual expression of symptoms. Each disorder has a specific type of physiological damage that occurs resulting in the development of flesh browning (see Table opposite).



	Type	Damage observed	Physiological description	Climatic region where symptoms occur	For more information
Diffuse			Chilling Injury	Cool Regions	Diffuse Flesh Browning, page 10
Radial			Senescent Breakdown	Warm Regions	Radial Flesh Browning, page 13
CO ₂ Injury			High CO ₂ Injury	All Cripps Pink apples susceptible	CO ₂ Injury

References

Anon., 2004. Internal browning problem overcome, HortLink Vol. Spring, pp. 8.

Brown, G., L. Schimanski, and D. Jannings. 2003. Investigating internal browning of Tasmanian 'Pink Lady' apples. *Acta Horticulturae*. 628, 161-166.

James, H., J. Jobling, and D. Tanner (2006). Investigating structural and physiological differences between radial and diffuse types of flesh browning of Cripps Pink apples. *Proceedings of the 27th International Horticultural Congress*. In Press.

James, Hannah (2007). Understanding the flesh browning disorder in 'Cripps Pink' apples. PhD Thesis, Faculty of Agriculture, Food and Natural Resources, University of Sydney, Sydney 2006 NSW, Australia.

Calculating Growing Degree Days for a Region

The project has identified that the risk of developing flesh browning is related to the accumulated heat units over the season.

GDD are calculated by taking the average of the daily maximum and minimum temperatures compared to a base temperature, T_{base} , (usually 10 °C). As an equation:

$$GDD = \frac{T_{max} + T_{min}}{2} - T_{base}$$

$$Total\ Seasonal\ GDD = \sum_{Harvest}^{Full\ bloom} (Daily\ GDD)$$
















(ie. the sum of the daily growing degree days from full bloom until harvest)

In our work the base temperature was 10°C. Over a season the daily GDD value (maximum temperature + minimum temperature)/2 – 10) was the sum of the daily heat unit accumulation from full bloom until harvest. The values calculated ranged from 900 to 1700 GDD depending on the region and the season.

Climatic Determination of Flesh Browning

The different types of flesh browning were found to occur in different climatic conditions. Cool regions such as Tasmania were found to develop the diffuse type of flesh browning whereas warm districts such as the Goulburn Valley were found to develop the radial type of flesh browning. Carbon dioxide injury was not shown to be dependent on climatic conditions during fruit growth and development.

The type of flesh browning occurring as well as the severity of the symptoms have been shown to be determined by climatic conditions during fruit growth and development. Several climatic conditions were examined and it was found that the accumulation of growing degree days (GDD) above 10°C during the growth period from full bloom to harvest had the closest relationship to the type of flesh browning that occurred during storage.

District	GDD _{10°C} (2005 data)		Type of Flesh Browning		
			Radial	Diffuse	CO ₂ Injury
Tasmania, Australia	807	Cool			
Nelson, New Zealand	1026				
Hawkes Bay, New Zealand	1102				
Yarra Valley, Australia	1162				
Manjimup, Australia	1405				
Batlow, Australia	1556				
Goulburn Valley, Australia	1688				
California, USA	1840	Hot			



Future Work

Seasonal data can be used to predict the incidence of flesh browning.

It might be possible to predict the risk of flesh browning for a district or orchard if more data is collected.

The data required would include;

- **Daily maximum and minimum temperatures from full bloom until harvest**

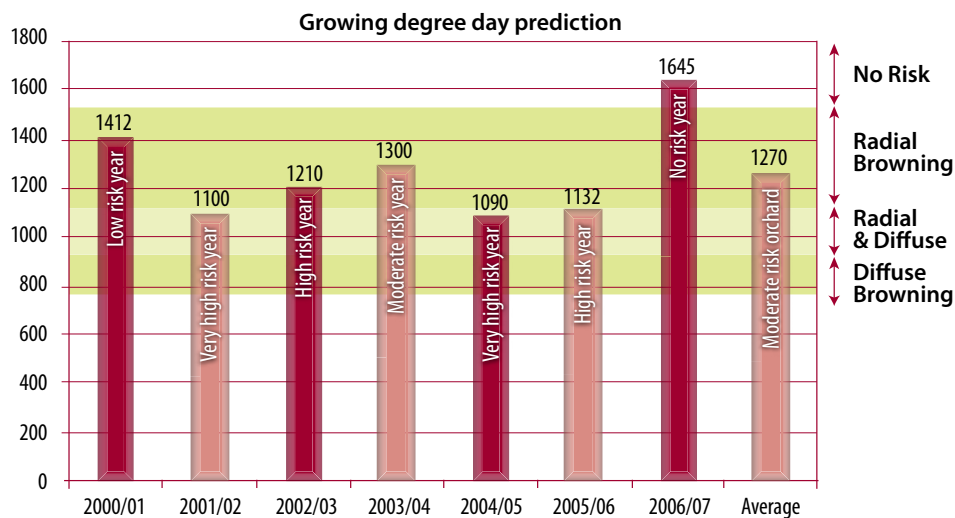
Calculate the seasonal accumulated growing degree days above 10°C.

- **Incidence of browning**

Cut a **minimum of 100 fruit per orchard block** use fruit of the same maturity each season. Must cut fruit at **the same removal time each year** such as 5 months. Leave fruit for 7 days at room temperature after removal from storage and then cut. Calculate % of fruit that are brown.

- **Develop District or Orchard models for browning risk**

Relate seasonal GDD to the incidence of flesh browning.

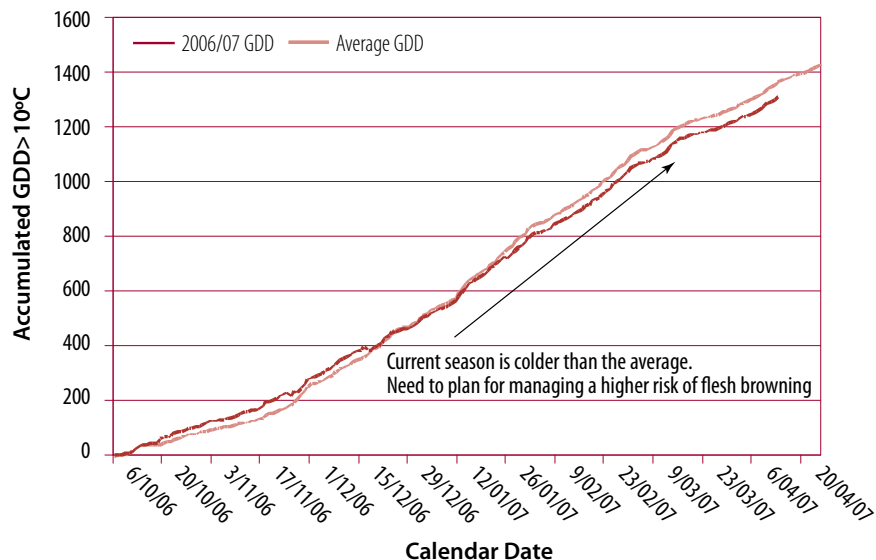


Total seasonal growing degree days could be used to predict the risk of flesh browning. This example shows that this orchard is susceptible to radial flesh browning. However, district and orchard cut off points would need

to be determined from historic or collected data relating incidence to accumulated growing degree days for that district or orchard. The data presented in this report can only be used as a guide.

Comparison of Seasonal Browning Degree Days for a District

Growers could chart the progress on previous data and plan for the level of browning risk



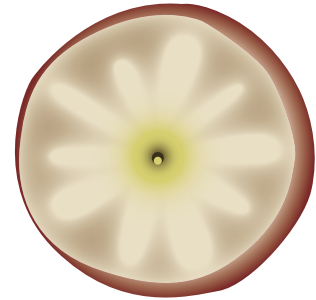
The progress of the season can be plotted and the potential risk can be seen prior to harvest and appropriate marketing and storage strategies put in place.

Diffuse Flesh Browning

Diffuse flesh browning is a chilling injury occurring when susceptible 'Cripps Pink' apples are stored below 3°C for longer than 4 months.

Apple growing regions susceptible to diffuse flesh browning

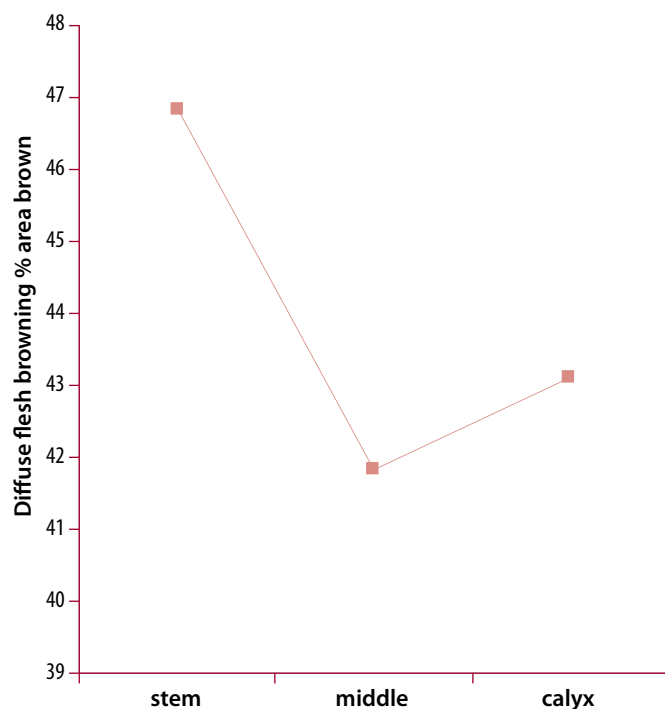
Diffuse flesh browning occurs in 'Cripps Pink' apples grown in districts accumulating less than 1100 GDD_{10°C} during the growing season from full bloom to harvest.



Symptoms

Diffuse flesh browning is characterised by browning throughout the cortex of the fruit with the vascular tissue of the fruit remaining clear and unaffected. The cortex cells of the fruit are larger and have thinner cell walls than the vascular cells, consequently these cells are more prone to collapse resulting from chilling injury and leading to the development of the characteristic browning.

Diffuse flesh browning has a high area of browning at the stem and calyx ends of the fruit, with a lower area of affected tissue in the middle of the fruit. This distribution of affected tissue is supported by the classification of this disorder as a chilling injury.



Factors influencing the development of the diffuse flesh browning disorder

The development of diffuse flesh browning in 'Cripps Pink' apples is influenced by both orchard and storage conditions.

i. Orchard conditions

The key factor influencing the development of the diffuse flesh browning disorder during storage was found to be the accumulation of growing degree days above 10°C during the period from full bloom to harvest.

In a season accumulating greater than 1100 GDD during the period from full bloom to harvest, the likelihood of 'Cripps Pink' apples developing diffuse flesh browning during subsequent storage is relatively low. However, in a cool season where the accumulation of GDD is below 1100 GDD_{10°C}, the likelihood of developing diffuse flesh browning increases substantially. Consequently, the monitoring of seasonal climatic conditions is vital for the determination of optimal storage conditions is vital for the determination of optimal storage conditions and storage time for the minimization of the development of diffuse flesh browning during storage.

Fruit nutrition has also been found to influence the development of diffuse flesh browning of 'Cripps Pink' apples. The incidence of diffuse flesh browning was found to increase with a decreasing ratios of calcium: potassium and calcium:magnesium. Mineral ratios often show a higher correlation to the development of disorders than individual minerals, however it is likely that the concentration of calcium is of most importance to the development of diffuse flesh browning. Calcium has several important functions in the cell and is primarily involved in strengthening the cell walls as well as strengthening the bonds between neighbouring cells.

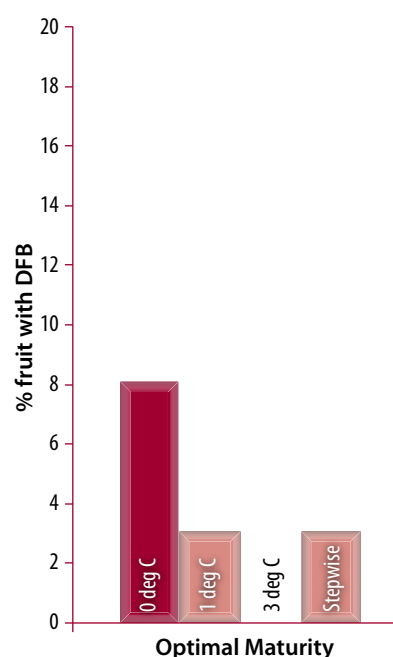
ii. Storage conditions

Diffuse flesh browning was found to reach commercially significant levels following 4-5 months of storage, depending on the seasonal risk. In a high risk season, diffuse flesh browning occurs after a shorter period of time in storage than in a low risk season.

The storage temperature was found to have a strong relationship to the incidence of diffuse flesh browning developing during long term storage.

When stored at 0°C, the incidence of diffuse flesh browning was shown to reach levels of 8-80% following 7 months of storage, depending on the seasonal risk. However, fruit that were stored at 3°C had an incidence of between 0 and 5% depending on the seasonal risk. This indicates that the storage temperature can be modified to reduce the incidence of diffuse flesh browning in 'Cripps Pink' apples. However, while storage at 1°C or the use of a stepwise cooling regime were found to significantly reduce the incidence of diffuse flesh browning, compared to those apples stored at 0°C, these temperature treatments were not able to reduce the incidence of diffuse flesh browning to within commercial threshold levels.

One consequence of long term storage at 3°C is a reduction in fruit quality due to the loss of firmness and the development of skin greasiness. There is a compromise to be made between the development of diffuse flesh browning and the loss of quality. The reduction in quality will shorten the length of effective storage time when the apples are stored at 3°C.



*Data from a low browning risk year.

Recommendations

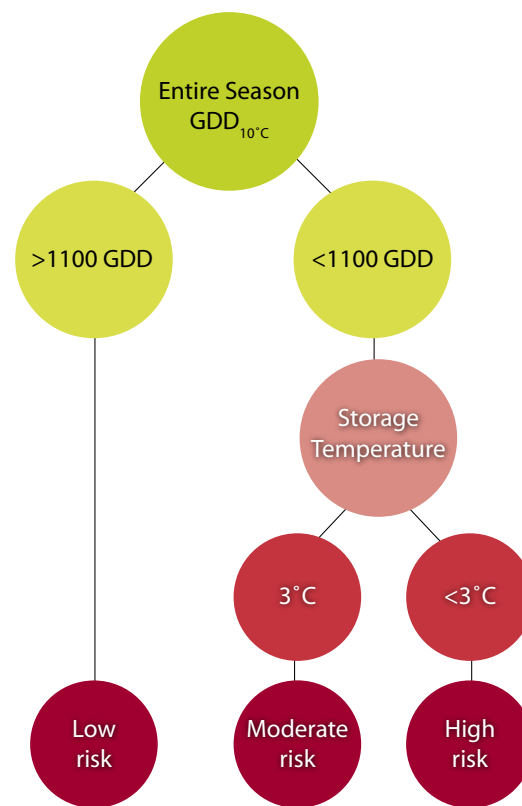
For optimal long term storage and control of the diffuse flesh browning disorder, 'Cripps Pink' apples grown in regions accumulating less than 1100 GDD_{10°C} should be stored at 3°C for a maximum period of 4 months.

Harvest maturity was not consistently shown to increase the risk of 'Cripps Pink' apples developing diffuse flesh browning during storage, however it is recommended that best commercial practices for harvest maturity are employed and that fruit are harvested and placed into ideal storage conditions prior to the ethylene climacteric. It is recommended that 'Cripps Pink' apples

are harvested at an SPI of 3.5 (CTIFL chart, Centre Technique Interprofessionnel des Fruits et Légumes, France).

It is also recommended that calcium levels are adequate during fruit growth and development.

The addition of CO₂ to the storage atmosphere was not found to detrimentally effect the development of the diffuse flesh browning disorder, however it is recommended that the concentration of CO₂ in the storage atmosphere is kept below 1% in order to reduce the likelihood of the fruit developing internal CO₂ injury.



Risk factors for the development of diffuse flesh browning (DFB) of 'Cripps Pink' apples during storage.

Insufficient data in the climatic range 1100 – 1400 growing degree days (GDD_{10°C}) has currently been collected, the type of flesh browning that develops in this range has not been determined however it is likely that the recommendations for radial flesh browning will be suitable as a guide.

Reference

James, H., G. Brown, E. Mitcham, D. Tanner, S. Tustin, I. Wilkinson, A. Zanella, and J. Jobling (2005). Flesh browning in Pink Lady™ apples: maturity at harvest is critical but how accurately can it be measured? Proceedings of the International Symposium on Harnessing the Potential of Horticulture in the Asian-Pacific Region. Acta Horticulturae. 694, 399-403.

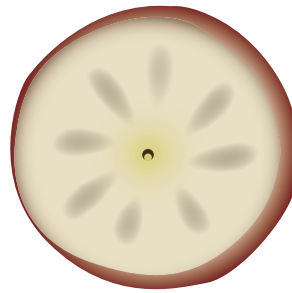
James, H., J. Jobling, D. Tanner, S. Tustin, and I. Wilkinson (2005). Climatic conditions during growth relate to risk of Pink Lady™ apples developing flesh browning during storage. Proceedings of the 9th International Controlled Atmosphere Research Conference. In Press.

James, Hannah (2007). Understanding the flesh browning disorder in 'Cripps Pink' apples. PhD Thesis, Faculty of Agriculture, Food and Natural Resources, University of Sydney, Sydney 2006 NSW, Australia.



Radial Flesh Browning

Radial flesh browning is a combination of a senescent disorder and a chilling injury occurring when 'Cripps Pink' apples are harvested over mature and stored below 1°C and in high CO₂ for longer than 4 months.



Apple growing regions susceptible to radial flesh browning

Radial flesh browning occurs in 'Cripps Pink' apples grown in districts accumulating greater than 1100 GDD_{10°C} during the growing season from full bloom to harvest.

Symptoms

Radial flesh browning is characterised by browning of the vascular tissue of the fruit with the cortex tissue remaining clear and relatively unaffected. This is in direct contrast to diffuse flesh browning which has the opposite pattern of browning. The small vascular cells are thought to limit the diffusion of carbon dioxide through the fruit leading to the build up of toxic quantities of CO₂ aggravating the senescent disorder.

Radial flesh browning has a high area of affected tissue at the stem end of the fruit and the affected area decreases toward the calyx end of the fruit.

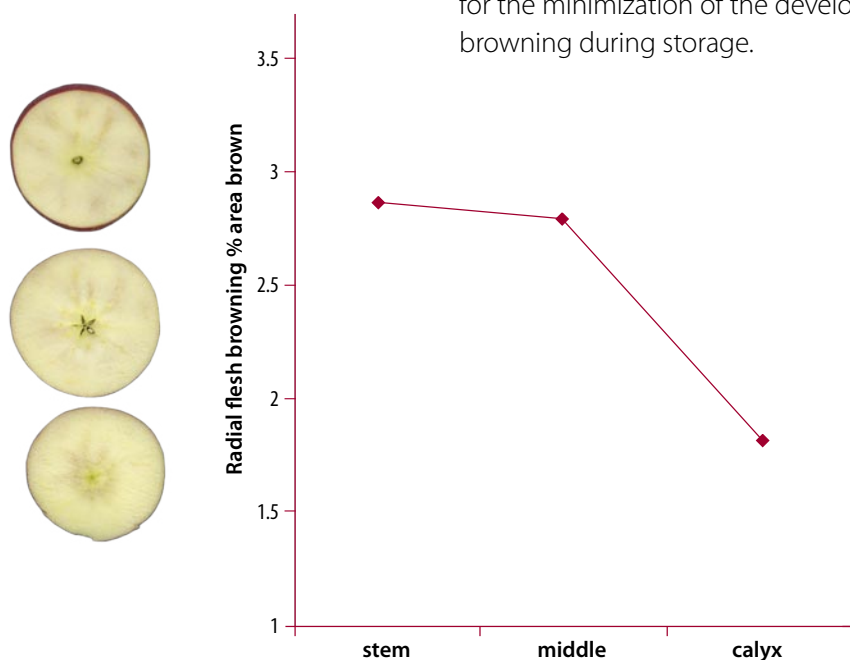
Factors influencing the development of the diffuse flesh browning disorder

The development of the radial flesh browning disorder of 'Cripps Pink' apples is influenced by both orchard and storage conditions.

i. Orchard conditions

Similarly to diffuse flesh browning, the key factor influencing the development of the radial flesh browning disorder during storage was found to be the accumulation of growing degree days above a base temperature of 10°C during the period from full bloom until harvest.

In a season accumulating greater than 1700 GDD during the period from full bloom to harvest, the likelihood of 'Cripps Pink' apples developing radial flesh browning during subsequent storage is relatively low. However, in a cooler season where the accumulation of GDD is between 1100 and 1700 GDD_{10°C}, the likelihood of developing radial flesh browning increases substantially. Consequently, the monitoring of seasonal climatic conditions is vital for the determination of optimal storage conditions and time for the minimization of the development of diffuse flesh browning during storage.



Fruit nutrition has also been found to influence the development of radial flesh browning of 'Cripps Pink' apples. The incidence of radial flesh browning was found to increase with a decreasing tree crop load and an associated decrease in fruit calcium concentration. Low crop load has been linked to the development of similar disorders in other apple varieties. For example, a low cropload in Jonathan apples has been shown to be related to the development of breakdown during storage (Little & Holmes, 2000). Tree crop load influences fruit nutrition as well as fruit size, both of these factors are likely to influence the development of the radial flesh browning disorder during storage.

Fruit maturity at harvest is another important orchard factor that can influence the development of the radial flesh browning disorder of 'Cripps Pink' apples. Late maturity at harvest has been shown to result in an increased incidence of radial flesh browning developing during storage.

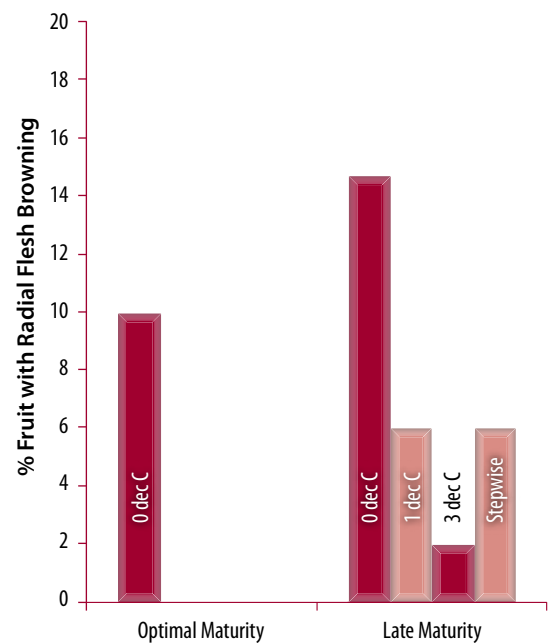
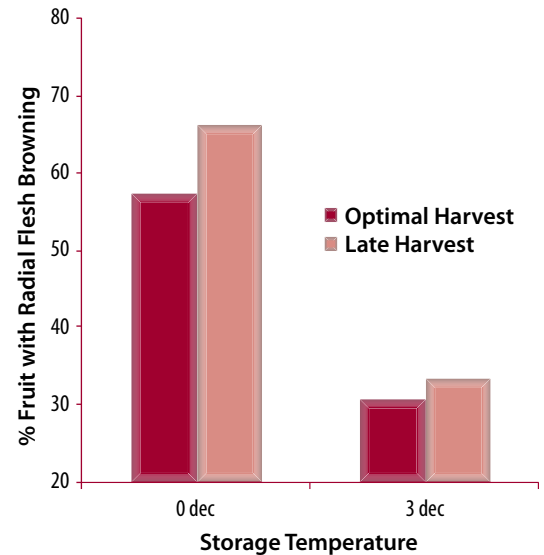
As late harvested apples have an increased risk of developing radial flesh browning during storage, it is important that regular samples are taken to accurately predict fruit maturity before harvest.

ii. Storage conditions

Radial flesh browning was found to reach commercially significant levels following 4-5 months or storage, depending on the seasonal risk. In a high risk season, radial flesh browning occurs after a shorter period of time in storage than in a low risk season.

The storage temperature was found to have a strong relationship to the incidence of radial flesh browning developing during long term storage.

Storing 'Cripps Pink' apples susceptible to radial flesh browning at 1°C was found to significantly reduce the incidence of flesh browning to within commercial threshold levels in low risk years. Storing at 1°C in a high risk year will reduce the incidence of radial flesh browning dramatically but not necessarily to below the commercial threshold.



Storing 'Cripps Pink' apples susceptible to developing radial flesh browning in controlled atmosphere storage with elevated CO₂ in the storage atmosphere was shown to increase the incidence of the disorder during storage.

Increasing the concentration of CO₂ in the storage atmosphere is a useful commercial technique that it used to prolong the storage life of apples and maintain fruit quality. However, this method has been found to increase the likelihood of 'Cripps Pink' apples developing the radial flesh browning disorder during storage. The build up of CO₂ within the apple can lead to the accumulation of toxic quantities of the gas causing damage to the cells and resulting in the development of the characteristic browning.



Recommendations

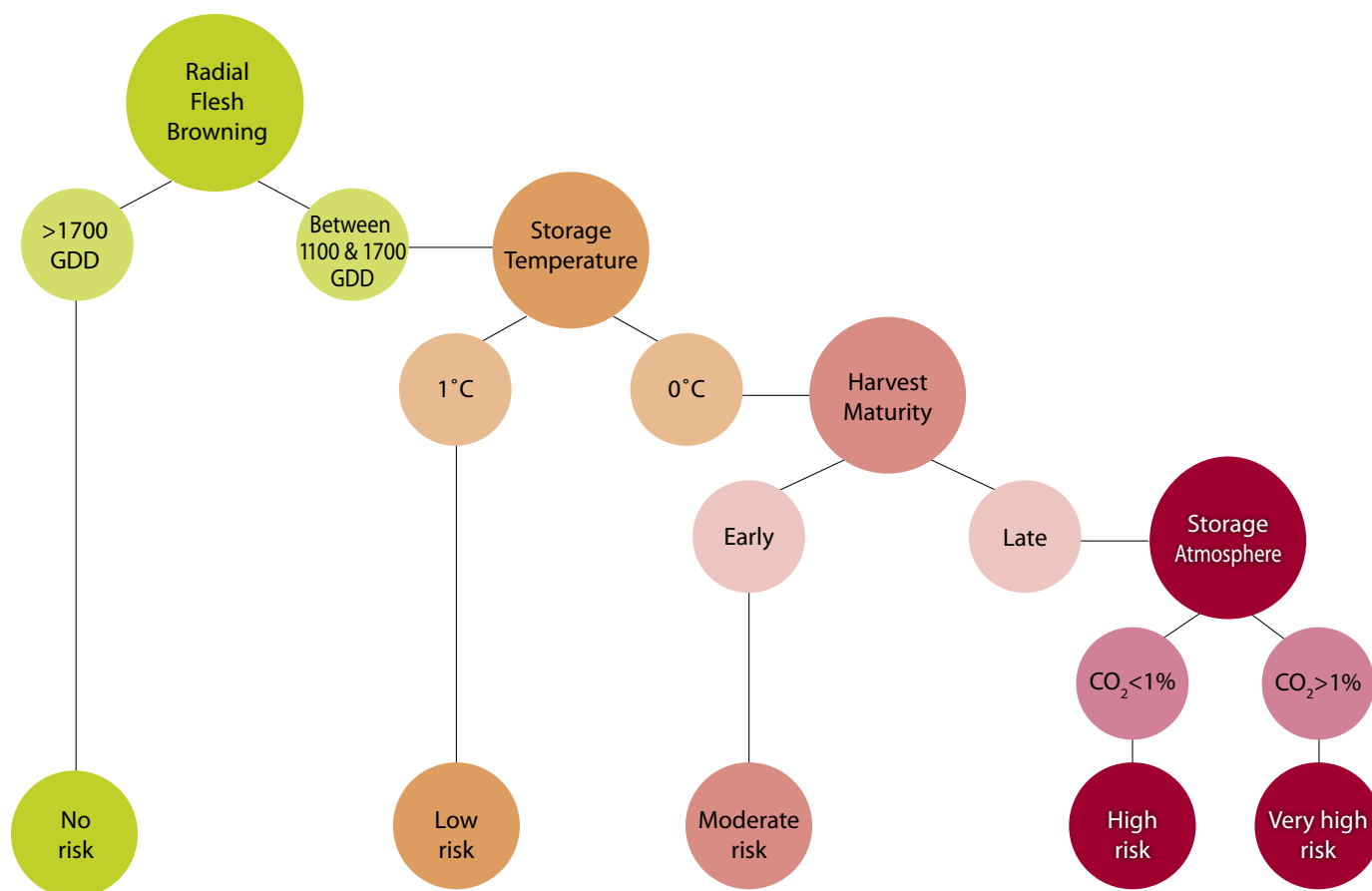
For optimal long term storage and control of the radial flesh browning disorder, 'Cripps Pink' apples grown in regions accumulating greater than 1100 GDD_{10°C} should be stored at 1°C for a maximum period of 9 months.

Harvest maturity was shown to increase the risk of 'Cripps Pink' apples developing radial flesh browning during storage, consequently it is recommended that best commercial practices for harvest maturity are employed and that fruit are harvested and placed into ideal storage conditions prior to the ethylene climacteric. It is recommended that 'Cripps Pink' apples are

harvested at an SPI of 3.5 (CTIFL chart, Centre Technique Interprofessionnel des Fruits et Légumes, France).

It is also recommended that calcium levels are adequate during fruit growth and development and that the tree crop load is maintained at an adequate level.

The addition of CO₂ to the storage atmosphere was also found to detrimentally effect the development of the radial flesh browning disorder. It is recommended that the concentration of CO₂ in the storage atmosphere is kept below 1% in order to reduce the likelihood of the fruit developing both radial flesh browning and internal CO₂ injury.



Risk factors for the development of radial flesh browning (RFB) of 'Cripps Pink' apples during storage.

Insufficient data in the climatic range 1100 – 1400 growing degree days (GDD_{10°C}) has currently been collected, the type of flesh browning that develops in this range has not been determined however it is likely that the recommendations for radial flesh browning will be suitable as a guide.

References

James, H., G. Brown, E. Mitcham, D. Tanner, S. Tustin, I. Wilkinson, A. Zanella, and J. Jobling (2005). Flesh browning in Pink Lady™ apples: maturity at harvest is critical but how accurately can it be measured? Proceedings of the International Symposium on Harnessing the Potential of Horticulture in the Asian-Pacific Region. Acta Horticulturae. 694, 399-403.

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