



# Preharvest factors that affect the development of internal browning in apples cv. Cripp's Pink: Six-years compiled data



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## ABSTRACT

Development of internal flesh browning (IB) of 'Cripp's Pink' (Pink Lady™) apples was studied over 6 years (2004–2011), on stored fruit coming from different orchards. The aim was to determine the effect of geographical location, climatic condition and fruit maturity on incidence of IB in the most important production regions of Chile. Geographical and climatic conditions were based on latitude, longitude, altitude, growing-degree-days (GDD), growing-degree-hours (GDH) and days after full bloom (DAFB). Maturity indexes included flesh firmness, starch index, soluble solids, background and red skin color. Fruits were stored at 0–1 °C for 5 months and IB incidence evaluated after removal plus 7–10 days at 18 °C. The disorder was classified as radial, diffuse or mixed IB. Data were analyzed through ANOVA, principal components analysis (PCA) and logistic regression analysis. Results showed that radial and diffuse IB can appear within the same orchard, and both symptoms may be combined within a fruit (mixed IB), which has not been reported. The different ranges of diffuse, radial and mixed IB found after storage were highly influenced by preharvest conditions (geographic/climatic and maturity variables). Although the three types of IB were found to increase with extended harvests, which were associated with advanced maturity, relative strength of the models to predict IB were higher for radial and mixed IB (Max-rescaled  $R^2 = 0.466$  and  $0.404$ , respectively), but less effective for diffuse IB (Max-rescaled  $R^2 = 0.117$ ). Data from 6 years suggest that radial IB could best be explained by GDH and firmness at harvest; diffuse IB could be accounted for by GDH and background color at harvest, whereas for mixed IB the most meaningful variables would be DAFB and background color.

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## 1. Introduction

Fruit storage for prolonged periods has allowed the development of off-season marketing or shipment to other countries. However, this practice can involve the loss of quality and allow the development of physiological disorders. In apples, internal browning (IB) has been documented in specific cultivars such as 'Braeburn', (Braeburn Browning Disorder) (Elgar et al., 1998; Lau, 1998); 'Fuji' and 'Granny Smith', (Brown Heart and Core Flush, respectively), (Argenta et al., 2002); and 'Cripps's Pink', (internal browning; IB) (Brown et al., 2003; Jobling et al., 2005), among others. The circumstances affecting the occurrence of these have

been associated with biochemical factors (Gong and Mattheis, 2003; de Castro et al., 2008), storage conditions (Volz et al., 1998; Gong and Mattheis, 2003), stage of maturity and preharvest factors (Argenta et al., 2002; Streif and Saquet, 2003; Jobling et al., 2005; Jobling and James, 2008).

This physiological disorder appears to be influenced by the interaction of preharvest and postharvest factors (Ferguson et al., 1999; Jobling et al., 2005,b; de Castro et al., 2007a,b; Jobling and James, 2008). Among factors that have been implicated in determining the degree of susceptibility to the disorder are: preharvest temperatures from full bloom to harvest, the accumulation of growing degree days, (GDD, above a base of 10 °C) (Jobling and James, 2008); the stage of maturity at harvest; nutrition; and handling production (crop load) (Jobling et al., 2005). In addition, the occurrence of IB is not constant between seasons or production areas; therefore, the development of predictive models is not easy (James et al., 2005; de Castro et al., 2007a,b).

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In relation to preharvest factors, Australian experience indicates that fruit developed in areas with an accumulation of less than 1100 GDD by harvest time, tend to show an increased formation of diffuse browning, while radial browning tends to prevail in areas with an accumulation of over 1100 GDD (Jobling and James, 2008; James et al., 2010). In addition, high daytime temperatures before harvest, as well as late harvests, that imply an advanced stage of maturity, lead to greater development of the disorder (Brown et al., 2005).

In relation to postharvest factors, atmosphere gas composition, storage temperature and the length of the storage period, directly affect the incidence of IB in this cultivar (Jobling et al., 2005; James and Jobling, 2008).

As in other production areas of the world, in Chile, the two symptoms related with this disorder, diffuse browning (associated with the outer part of the flesh) and radial browning (associated with vascular bundles) (James and Jobling, 2009) have been seen. The first is correlated to chilling injury during storage, while the second is related with senescence of fruit (Jobling and James, 2008).

The objective of this study was to compile a 6-year dataset to identify preharvest and postharvest factors that predispose the appearance of IB in 'Cripp's Pink' apples.

## 2. Materials and methods

### 2.1. Plant material

During 6 years (2004–2011), apples (*Malus x domestica* Borkh.) cv. Cripp's Pink (MM106) were harvested from different commercial orchards, located within the most important Chilean production areas, from Graneros to Longaví (Table 1), where 88% of this cultivar is grown (ODEPA, 2014). The information was divided in two groups: orchards having one or more years of data and one harvest per year (Table 2), and orchards with one year of data and 6 sequential harvests, representing different maturity stages (Table 3). Each harvest consisted of a sample of 520 fruit, taken randomly from 50 trees of similar size, vigor and crop load. Fruit maturity was assessed at harvest using 5 replicates of 4 fruit each. After 5 months of cold storage (0–1 °C), internal browning (%) was assessed on 5 replicates of 100 fruit each.

### 2.2. Fruit maturity

Flesh firmness, starch index, soluble solids, background and red skin color were evaluated at harvest and following 5 months of cold storage. Equator flesh firmness (N), was determined with a penetrometer (Effegi FT 327, McCormick Fruit Tech, Yakima, USA). Starch degradation was registered using the starch index developed by CTIFL (Centre technique interprofessionnel des fruits et légumes; 1 representing full starch to 10 representing no starch). Soluble solids (%) were measured by refractometry (Atago ATC 1, Tokyo, Japan). Background was assessed visually using a subjective scale from 1 (green) to 3 (yellow), and red skin color as a percentage of fruit coverage.

### 2.3. Internal browning evaluations

After 5 months of cold storage (0–1 °C), 5 replicates of 100 fruit, from each location and harvest date, were removed and placed for 7–10 days at room temperature (20 °C, simulating shelf life conditions). After this period, fruit were cut in half (Brown et al., 2005; James and Jobling, 2009) and internal browning (IB) was categorized as radial, diffuse or mixed (radial+diffuse) (Fig. 1). Damage was assessed, for each replicate, as percentage of incidence among the 100 fruit.

### 2.4. Geographic and climatic conditions

From full bloom to harvest, air temperature was automatically recorded by weather stations settled at each orchard. Growing-degree-days (GDD) were calculated by taking the average of daily maximum and minimum temperatures compared to a base temperature of 10 °C (Jobling and James, 2008). Growing-degree-hours (GDH) were calculated according to Anderson and Seeley (1992) and expressed as GDH/1000. Values were calculated as a total from full bloom to harvest, as well as the amount of GDD or GDH/1000 up to 60 days prior to harvest and the amount of GDD or GDH/1000 from full bloom up to 50 days after full bloom (DAFB). On each plot, a GPS sensor was used to determine longitude, latitude and altitude (m.a.s.l.).

### 2.5. Statistical analysis

Analyses of variance (ANOVA) were performed to compare maturity indexes measured at harvest, using R 3.0.0 (R Development Core Team, 2008) and Sigma Plot (version 12.0, SPSS, Chicago, IL, USA). The significance of the differences was determined by Tukey's *t* test ( $P \leq 0.05$ ). In order to establish relationships between IB incidence and the explanatory variables under study (geographic, climatic, and maturity), principal components analysis (PCA) and multiple logistic regression analysis were performed. For the last, the probability of the occurrence of IB was estimated as the proportion of apples that developed the disorder. Proportions are binomially distributed, and hence bounded between 0 and 1, so logistic regression is an appropriate statistical method for this response (Kleinbaum and Klein, 1994). In order to select an adequate model, a stepwise logistic regression procedure was performed, where variables were entered in the model and removed from the model using the chi-square distributed Wald test statistics at  $P \leq 0.05$ . Additionally coefficient parameters of the explanatory variables were interpreted in terms of odds ratio, a measure of association that has been widely used (Lammertyn et al., 2000). The odds ratio, can range between zero and infinity and expresses by which factor the odds of the event increase ( $>1$ , considered as an induction effect), or decrease ( $<1$ , considered as a protection effect). Therefore, a value of 1 indicates little or no influence of the variable on the incidence of the disorder; likewise if 1 lies within the 95% confidence interval of the odds ratio. In this article the odds ratio was interpreted as % of influence; for instance an odds ratio of 1.1 for the variable GDH/1000 indicates that when

**Table 1**  
Geographic location of orchards, age of trees, planting distance and sampling season of 'Cripp's Pink' apples (MM106).

Orchard	Location	Altitude (m.a.s.l.)	Tree age (years)	Planting distance (m)	Season
Graneros	34°05'S; 70°43'W	484	3	4.0 × 2.3	2004/05, 08/09, 09/10
Rancagua	34°14'S; 70°45'W	334	12	4.5 × 2.5	2007/08
Chimbarongo	34°35'S; 71°00'W	332	10	3.5 × 2.5	2007/08
Molina	35°10'S; 71°16'W	267	10	4.5 × 3.0	2007/08
San Clemente	35°30'S; 71°28'W	204	3	4.0 × 2.3	2004/05, 07/08, 08/09, 10/11
Longaví	35°58'S; 71°41'W	153	6	4.5 × 2.5	2007/08

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