

Article

# Cider Terroir: Influence of Regionality on Australian Apple Cider Quality

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**Abstract:** Understanding how regionality influences the key characteristics of cider will assist the industry to produce a premium and consistent quality product. Three dessert apple varieties were sourced from three (minimum) orchards per region from nine major growing regions across Australia over two seasons. Ciders produced from these apples were analysed for base quality characteristics, including total phenolic content (TPC), pH, total soluble solids (TSS) and titratable acidity (TA). Across both seasons, region had a significant influence on TPC, with the region model explaining approximately 25% of the variation in TPC. TSS and TA were significantly influenced by an interaction between variety and region, with the model accounting for approximately 60% and 75% of the variation, respectively, over both seasons. An interaction between variety and region influenced pH results in the first season, with only mean effects in the second season. A climate model was developed using average rainfall data and growing degree days (GDDs). Differences in climate accounted for most, but not all, of the regional variation observed in cider quality measurements. These results demonstrate that the apple growing region can significantly impact the resulting cider quality. Such findings are critical for supporting cider producers to make informed decisions when sourcing fruit.

**Keywords:** apple; cider; polyphenol; region; climate; quality; terroir



**Citation:** Way, M.L.; Jones, J.E.; Hunt, I.; Damberg, R.G.; Swarts, N.D. Cider Terroir: Influence of Regionality on Australian Apple Cider Quality. *Beverages* **2024**, *10*, 99. <https://doi.org/10.3390/beverages10040099>

Academic Editor: José Sousa Câmara

Received: 30 July 2024

Revised: 28 August 2024

Accepted: 26 September 2024

Published: 19 October 2024



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## 1. Introduction

Terroir is a French term that expresses the influence of environmental factors on the quality and flavour attributes of the product [1]. Terroir is a well-known concept in wine, where the relationship between variety and the specific growing conditions of a region (regionality) have an influence on beverage quality [2]. Specifically, interactions between climate, geology, variety and management practices create a distinctive environment which develops characteristics that influence the quality and sensory attributes of a product [1,3]. Understanding how specific varieties grown in certain conditions combine to create desirable characteristics is critical to the production of a high-quality and consistent consumer-preferred cider [1].

The understanding of how apple growing regions influence the quality characteristics of cider is currently limited. Traditional cider-producing countries, including England, France and Spain, boast hundreds of cider-specific varieties and growing location combinations, which have been developed over time to produce unique chemical profiles [4,5]. This rich history has allowed for the development and selection of varieties matched to a region's growing conditions that subsequently produce a premium quality cider [4]. In comparison, countries relatively new to cider making are facing the challenge of producing enough volumes of cider-specific apple varieties to make varietal cider production viable. To manage this shortfall, cider producers often combine both cider apples and readily available dessert varieties for cider making, with limited knowledge of how the growing region will influence the flavour and quality characteristics of the cider produced [4,6].

Increasingly, studies are demonstrating the relevance and importance of regionality for both cider and dessert apple varieties used in the production of cider [4–7]. Miles et al. [4] compared juice quality characteristics from cider varieties grown in Washington State to juice from apples grown in Bristol, England. The authors also found that overall, the difference in fruit quality was likely due to differences in the climate of the production regions. Furthermore, Thompson-Witrick et al. [5] characterised the polyphenolic composition of 20 apple cider varieties grown in Virginia, U.S.A, finding that polyphenol concentration varied in response to orchard age, agronomic management and differing climates. Similarly, Valois et al. [7] reported that the TPC of cider varieties in New York was noticeably lower than those reported for Western Europe and suggested there is a need for further studies to better understand phenolics for cider apples grown in New York, especially between seasons, regions and varieties. Plotkowski and Cline [8] also suggested further studies are required to compare juice characteristics of varieties grown in different regions, especially those with varying climates like Canada. Cline et al. [3] identified that there is a lack of information on variation in juice attributes between orchards. These observations reflect a similar knowledge gap for the Australian cider industry, which is still in its infancy and relies heavily on dessert apples to fill a significant production gap.

Australia has a range of cool climate apple-growing regions, which produce an average of 280,000 tonnes of fruit per annum [9]. Substantial variation between the growing conditions of each region provides the perfect context for testing the concept of regional influence on cider quality ('terroir'). This is especially relevant with the growing demand for fresh apples from these regions to make consistent premium quality ('craft') ciders, as opposed to relying on imported apple concentrate. Therefore, there is a requirement to understand the influence of regionality and provenance ('terroir') on cider.

Base cider characteristics such as total phenolic content (TPC), sugar and acidity are commonly used quality indicators for cider and wine, as they directly relate to the organoleptic qualities [10–12] responsible for mouthfeel, colour and flavour [11,13–15]. These same characteristics are often correlated with fruit quality and are impacted by climate conditions such as light exposure, growing degree days (GDDs) and rainfall. These climatic conditions are also the factors responsible for differentiating between growing regions and therefore are likely to influence the cider produced [16]. Previous studies have determined that pH is influenced by temperature and solar exposure [17–19] and TA has been shown to be affected by region and climate [16,20,21]. Climatic conditions such as light exposure have been found to impact TSS [16,22,23] and a difference in region has previously been shown to influence TPC [5,7].

This study investigated the influence of growing region on the quality attributes of apple cider. We hypothesise that ciders made from apples grown in geographically different growing regions will vary in base cider quality parameters. We also hypothesise that the varying climatic conditions (rainfall and GDDs) experienced in these growing regions will influence cider quality attributes. To test these hypotheses, three readily available dessert varieties, commonly used to produce bulk ciders, were sourced from locations representing Australia's main apple growing regions over two seasons. The following questions were asked: (i) Does the quality of base cider made using apples from different regions vary (as determined by laboratory chemistry assessments)? (ii) Is there a relationship between cider quality and climate factors? This study aims to provide practical evidence and statistical modelling to determine the influence of growing region on cider quality. It is hoped that such information can then be utilised by producers to make informed decisions when purchasing fruit and making cider.

## 2. Materials and Methods

### 2.1. Fruit

Over the 2017/18 and 2018/19 growing seasons, three dessert apple varieties, ‘Pink Lady’, ‘Fuji’ and ‘Royal Gala’ were collected from nine major apple growing regions across Australia (Table 1). Apples were picked at commercial harvest maturity, as determined by the grower. From each region, 12 kg of each variety were collected from at least three different orchards representative of the region and shipped to the University of Tasmania, Sandy Bay campus (Hobart, Tasmania, Australia). Apples grown outside of Tasmania were transported via refrigerated freight to Melbourne, Australia where they received required quarantine treatment before entering Tasmania. Each box of apples was sampled and tested following a standard starch index test for apple maturity.

**Table 1.** Location details of the growing regions apples were sourced from in relation to closest weather stations.

Apple Growing Region	State	Coordinates	Weather Station	Elevation
Huon Valley	Tasmania	43.0295° S, 147.0580° E	Grove (Research Station)	65 m
Spreyton	Tasmania	41.2167° S, 146.3500° E	Aberdeen (Melrose Road)	85 m
Narre Warren	Victoria	38.0262° S, 145.3067° E	Cranbourne Botanic Gardens	85 m
Shepparton *	Victoria	36.3833° S, 145.4000° E	Shepparton Airport	114 m
Batlow	New South Wales	35.5167° S, 148.1500° E	Tumbarumba Post Office	645 m
Adelaide Hills	South Australia	34.9063° S, 138.8397° E	Mount Lofty	685 m
Manjimup *	Western Australia	34.2492° S, 116.1437° E	Manjimup	287 m
Orange	New South Wales	33.2833° S, 149.1000° E	Orange Agricultural Institute	922 m
Stanthorpe	Queensland	28.6600° S, 151.9376° E	Applethorpe	872 m

\* Apples were collected from Shepparton in 2018 only, and from Manjimup in 2019 only.

### 2.2. Cider Making

Apples were refrigerated for a maximum of one month before being milled and crushed following the method described in Way et al.’s paper [24]. Each 12 kg box of apples was milled to produce juice. Three 50 mL juice samples were taken and frozen for juice analysis before the remaining juice was transferred into four 500 mL Schott bottles fitted with air locks. The pH of each bottle was measured using a handheld WP-81 pH-Cond-Salinity Meter (TPS, Brisbane, Australia) and adjusted with diluted malic acid (400 g/L) (Aldrich, Shanghai, China) to below 3.8 where necessary. Bottles were then treated with potassium metabisulphite (Chem-Supply, Gillman, SA, Australia) at 50 ppm and Novozymes Vinoclear (Winequip, Melbourne, VIC, Australia) at 1 mL/L. After 24 h at  $21 \pm 1$  °C, yeast EC1118 (Lallemand, Edwardstown, SA, Australia) was rehydrated and added at 0.3 g/L to each bottle as well as yeast nutrient Fermaid AT (Lallemand, Edwardstown, SA, Australia) at 0.4 g/L, as in Way et al.’s paper [24]. The weight of each bottle was recorded before being placed in a temperature-controlled room at 14 °C. Bottles were weighed daily to monitor the rate of fermentation. When the weight of the bottles plateaued, it was assumed fermentation had ended. Bottles were then racked into labelled 330 mL crown seal bottles, as well as three 50 mL tubes and one 10 mL centrifuge tube, which were frozen for future laboratory analysis.

### 2.3. Base Cider Quality Analysis

Base ciders were analysed as described in Way et al.’s paper [25]. Samples were centrifuged for 10 min at 4000 rpm and brought to room temperature before a handheld WP-81 pH-Cond-Salinity Meter (TPS, Brisbane, Australia) was used to measure pH, and an automatic titrator (Mettler Toledo g20 Compact Titrator, Greifensee, Switzerland) measured titratable acidity (TA), where results were measured in g/L of malic acid. A handheld digital refractometer (A. Kruss Optronic, Hamburg, Germany) measured total soluble solids

(TSS), with results measured in °Brix. Using a spectrophotometer (SPECTROstar Nano, BMG LABTECH, Windsor, NSW, Australia), TPC was measured using the Somers method by diluting the samples 1:50 in 1M Hydrochloric acid [24]. TPC results are represented as total phenolic index (TPI), with spectrophotometer results, expressed in absorbance units, multiplied by a dilution factor ( $\times 50$ ).

#### 2.4. Site Data

Climate data was provided by the Australian Bureau of Meteorology [26], with observations recorded from the weather stations closest to the apple orchards in each region. Data for monthly rainfall (mm) and maximum and minimum monthly temperature (°C) were collected over the two growing seasons. Growing degree days (GDD), was calculated by adding the daily maximum temperature to the mean daily minimum temperature and dividing the results by two, then subtracting 10 ( $GDD = [(daily\ maximum\ temperature + daily\ minimum\ temperature)/2] - 10\ ^\circ C$ ) (Table 1).

The base installation of R (version 4.1) was used to model all data. Linear regression models were estimated with TPC, TA, pH and TSS outcome measurements selected as the ‘y-variables’ (or dependant variables). The outcome measurements being modelled were the average ( $n = 4$ ) measurements from each orchard’s variety-specific, individually fermented 500 mL bottle of cider.

Two sets of ‘x-variables’ (or explanatory variables) were used, with every set of unique outcome measurements having two models. The first set of models presented is the ‘region model’, defined by using region and variety categorical data as x-variables (including a region/variety interaction term). The second set of models is the ‘climate model’, defined by using region-specific weather data (Table 1). The x-variables in the climate model are the numerical data for GDDs and average rainfall, combined with categorical data for variety. All two-way interactions between these three variables were also included.

The model results were analysed using a conventional ANOVA-style breakdown, which successively tests for the relevance for each of the variables using F-tests. In addition to the  $p$ -values derived from the successive F-tests, we present the marginal increase in adjusted- $R^2$ , which was attributed to adding the respective variable. All references to  $R^2$  in this paper are to its conventional adjusted measure.

To assess the statistical significance of differences between the region and climate models, we compared the regional model with a super-set model that comprised both the climate model along with region and the region/variety interaction x-variables (enabling a conventional F-test for nested models). Regression residuals from all region and climate models were checked; in each case, the residuals were approximately normally and identically distributed.

A principal component analysis was conducted on a comprehensive monthly weather variable matrix, analysing rainfall, solar radiation, mean minimum monthly temperature and mean maximum monthly temperature across all regions for both seasons. A proxy was also calculated for the winter chill measurement in each region, using the average of the daily minimum temperatures for the months of June, July and August (winter).

### 3. Results

#### 3.1. Region Model

Over two growing seasons, the influence of region and variety on base cider chemical parameters was investigated using the region model (Table 2). During both seasons, there was a significant ( $p < 0.005$ ) interaction for TA and TSS, and also for pH in 2018 only. In 2019, there were significant ( $p \leq 0.001$ ) influences of variety and region on pH. There was no interaction for TPC in either season. In 2018, variation in TPC was driven by region only, while in 2019, variation in TPC was influenced by both variety and largely by region.

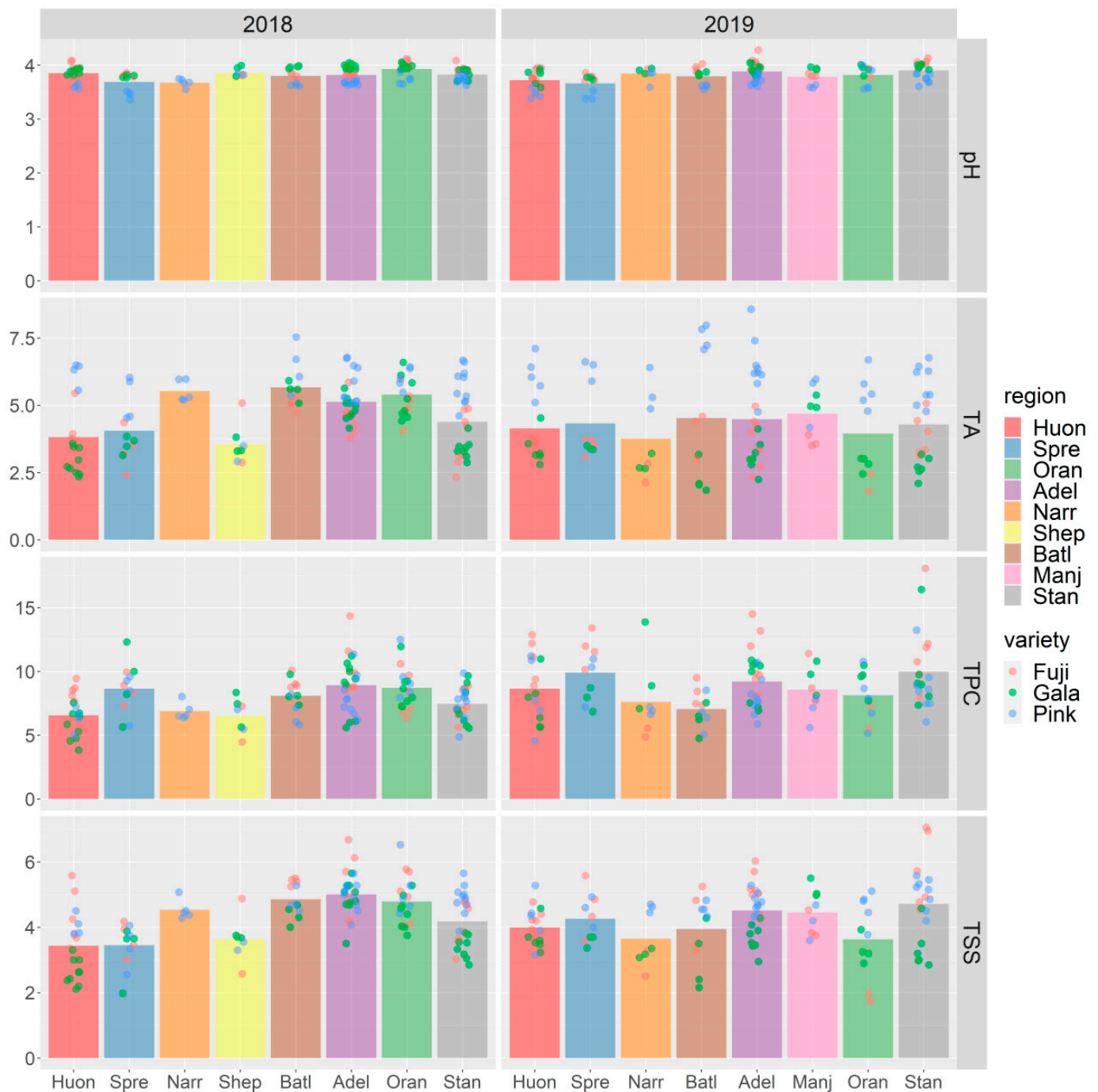
**Table 2.** Key results from the region model, including the  $p$ -values and  $R^2$  adjusted values for base cider chemical parameters (TPC, pH, TSS and TA) of all three dessert varieties, ‘Pink Lady’, ‘Fuji’ and ‘Royal Gala’ combined, over two growing seasons, 2018 and 2019. The  $p$ -values correspond to a series of F-tests within a conventional ANOVA breakdown of variable significance (the order of testing corresponds to the introduction of the ‘variety’, ‘region’ and ‘interaction’ variables, respectively). The  $R^2$  values in the ‘variety’, ‘region’ and ‘interaction’ columns are the marginal increase in  $R^2$  attributable to adding each variable (in sequence). The  $R^2$  in the ‘overall’ column is the total for the region model with all variables included.

		Variety		Region		Interaction		Overall	
		$p$ -Value	$R^2$	$p$ -Value	$R^2$	$p$ -Value	$R^2$	$p$ -Value	$R^2$
2018	TPC	0.095	1.2%	<0.001	21.9%	0.153	3.4%	<0.001	26.5%
	pH	<0.001	55.0%	<0.001	12.4%	<0.001	12.7%	<0.001	80.1%
	TSS	<0.001	12.4%	<0.001	40.2%	0.005	6.4%	<0.001	59.0%
	TA	<0.001	34.6%	<0.001	26.7%	<0.001	9.9%	<0.001	71.2%
2019	TPC	0.001	8.9%	0.004	9.9%	0.071	6.9%	<0.001	25.7%
	pH	<0.001	45.0%	<0.001	24.7%	0.176	1.5%	<0.001	71.2%
	TSS	<0.001	21.7%	<0.001	10.9%	<0.001	28.4%	<0.001	61.0%
	TA	<0.001	71.9%	0.002	2.3%	<0.001	8.2%	<0.001	82.4%

### 3.2. TPC

In 2018, 21.91% of the variation in TPC between all three dessert varieties is explained by the difference in growing region (Table 2). This was the only combination of quality parameter and season where there was no significant influence from variety, either independently or through an interaction. In 2019, both variety and region were found to have a significant ( $p < 0.005$ ) influence on TPC results, but only 8.9% of the variation could be explained by variety, and similarly an additional 9.9% explained by region (Table 2). Across both seasons, the region model was able to explain approximately 25% of the variation in TPC values.

In 2018, ciders made from apples grown in the Huon Valley, the southernmost region, represented the lowest TPC; however, this was not repeated in 2019 (Figure 1). For Stanthorpe, the northernmost region, the TPC of all varieties demonstrated minimal variation in comparison to TPC results from the same region in 2019, which provided the greatest range of TPC values, as well as the greatest individual result. Similarly, across all other regions, no single variety produced consistently higher or lower TPC values (Figure 1).



**Figure 1.** Base cider chemical parameter means as displayed by region and variety over the two growing seasons of 2018 and 2019. Bars are overall mean for region; points are the mean ( $n = 4$ ) measurements from each orchard's variety-specific, individually fermented sample of cider. Regions are displayed as Huon = Huon Valley, Sprey = Spreyton, Narr = Narre Warren, Shep = Shepparton, Batl = Batlow, Adel = Adelaide Hills, Manj = Manjimup, Oran = Orange and Stan = Stanthorpe. Varieties are displayed as Fuji, Gala = Royal Gala and Pink = Pink Lady.

### 3.3. pH

In 2018, 12.71% of the variation between pH results was driven by the interaction between region and variety (Table 2). Specific region and variety combinations influenced pH results. Cider made using 'Pink Lady' apples had significantly lower pH values compared to cider made using 'Fuji' and 'Royal Gala' apples across all regions. An exception

to this trend was Shepparton, where cider made using 'Pink Lady' apples had similar pH values to cider made using 'Fuji' apples (Figure 1).

In 2019, variety and region were separately found to have a statistically significant ( $p \leq 0.001$ ) influence on cider parameters. Variety was found to account for 45% of the variation in pH, while region was found to account for 24.7% (Table 2). In 2019, ciders made using 'Pink Lady' apples were consistently lower in pH as compared to the other two varieties across all regions (Figure 1). Overall, the model accounted for 80.1% of the variation between pH results in 2018, and 71.2% in 2019 (Table 2).

### 3.4. TSS

Cider made from 'Royal Gala' apples grown in the Huon Valley, Batlow and Stanthorpe had significantly ( $p < 0.05$ ) lower TSS readings compared to ciders made using 'Fuji' and 'Pink Lady'. This trend was not observed in the other five regions (Figure 1). In 2019, there was a significant ( $p < 0.001$ ) interaction between variety and region, accounting for 28.43% of the variation in TSS. Across the majority of regions, the TSS results for 'Pink Lady' and 'Fuji' were similar, except for Narre Warren and Orange, where 'Pink Lady' had higher TSS values than both 'Fuji' and 'Royal Gala' (Figure 1). The TSS of ciders made from 'Royal Gala' were always lower than 'Pink Lady' and 'Fuji', except for Narre Warren, where it overlapped with 'Fuji', and also Manjimup, where 'Royal Gala' had higher TSS values than both 'Pink Lady' and 'Fuji' (Figure 1).

### 3.5. TA

In 2018, there was a significant ( $p < 0.001$ ) interaction between variety and region, accounting for 9.86% of the variation in TA results. Cider made using 'Pink Lady' apples had higher TA values than cider made using 'Fuji' and 'Royal Gala' apples across all regions (Figure 1). This excludes Shepparton, where cider made using 'Pink Lady' apples, on average, had the lowest TA values, with no significant difference to ciders made using 'Fuji' or 'Royal Gala' in that region (Figure 1). The model suggests that region and variety accounted for 71.24% of the variation in TA values overall (Table 2).

In 2019, there was a significant ( $p \leq 0.001$ ) interaction between variety and region for TA results, accounting for 8.23% of the variation (Table 2). Overall, the model accounted for 82.4% of the variation, with variety responsible for 71.9% and region for 2.4% (Table 2). In 2019, TA trends were consistent between varieties and regions, with cider made from 'Pink Lady' apples generally having higher TA values than the other varieties. In contrast to this trend, cider made using 'Royal Gala' apples from Manjimup did not have a significantly different TA when compared to cider made from 'Pink Lady' apples sourced from the same location (Figure 1).

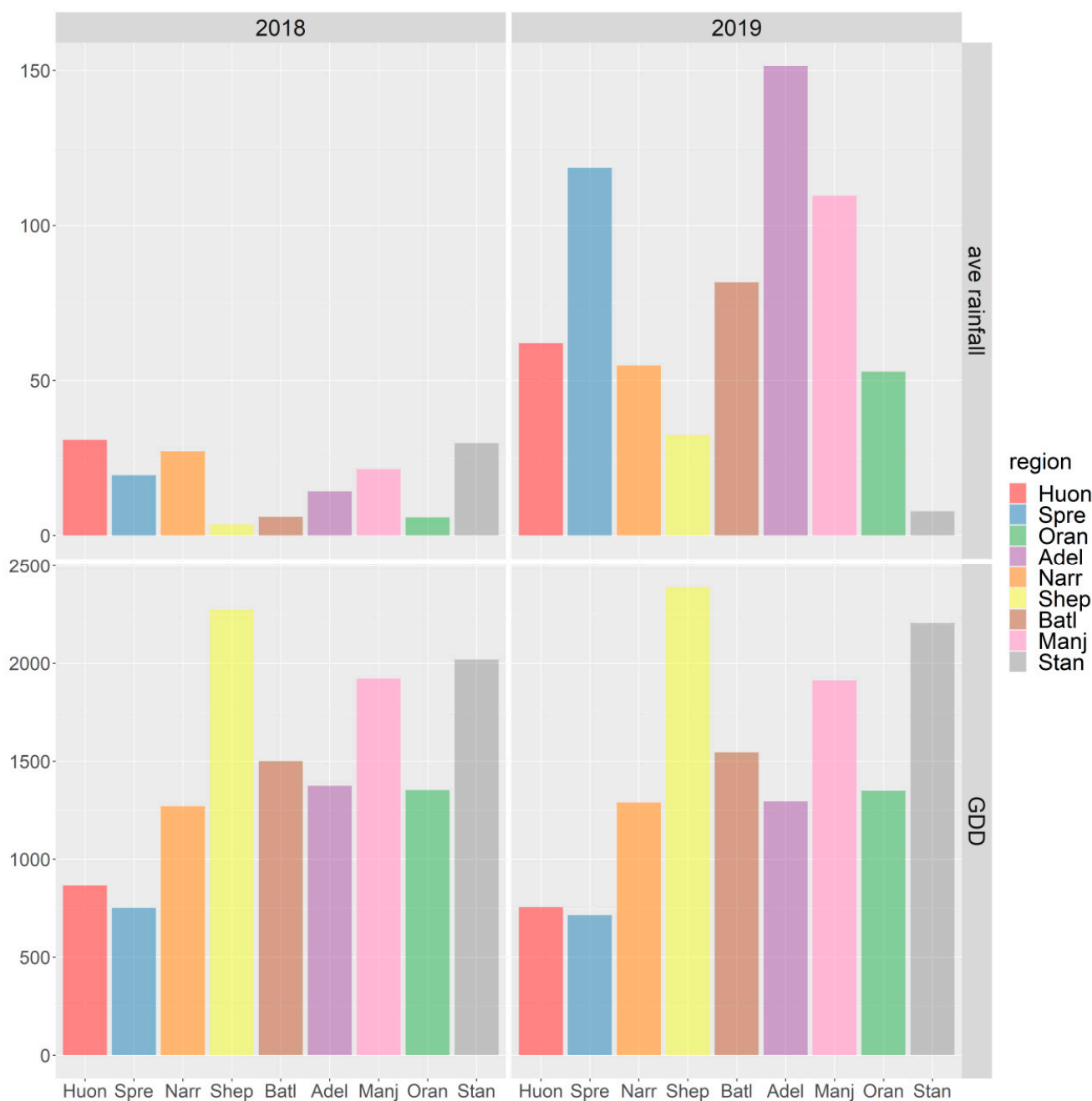
### 3.6. Season

Although there were slight differences between the drivers of variation for each quality parameter, overall,  $R^2$  results were extremely consistent between the seasons. The overall TPC-adjusted  $R^2$  values were 26.5% and 25.7% for 2018 and 2019, respectively (Table 2). This pattern was repeated for pH, with 80.1% and 71.2%, TSS with 59% and 61% and again for TA with 71.2% and 82.4% (Table 2).

### 3.7. Climate Model

The data used as x-variables in the climate model included the numerical measurements of GDDs and average rainfall, the categorical data for variety, and all two-way interactions between these variables over both seasons (Figure 2). Strong regional differences were observed for GDDs. Over both seasons, the regions follow similar patterns in comparison to each other, with little difference between seasons. Significant regional differences were observed for rainfall between seasons. For example, Huon Valley received on average five times more rainfall in the 2019 growing season as compared to 2018

(Figure 2). In contrast, Stanthorpe was the only region to receive less rainfall in 2019 than 2018 (Figure 2).



**Figure 2.** Climate key indicators; mean rainfall and GDDs compared by region over the two growing seasons of 2018 and 2019. Bars are overall mean for region, and points are the mean (n = 4) measurements from each orchard’s variety-specific, individually fermented 500 mL sample of cider. Regions are displayed as Huon = Huon Valley, Sprey = Spreyton, Narr = Narre Warren, Shep = Shepparton, Batl = Batlow, Adel = Adelaide Hills, Manj = Manjimup, Oran = Orange and Stan = Stanthorpe. Varieties are displayed as Fuji, Gala = Royal Gala and Pink = Pink Lady.

The climate model used average rainfall and GDDs as key climate indicators to determine if these variables could explain more variation in cider quality than the region model alone. Overall, the region model without climate data was found to produce higher R<sup>2</sup> values for the data analysed (Table 3). The region model explained more of the variation

in the cider quality data using the variables ‘region’ and ‘variety’, as opposed to the climate model, which used the variables ‘average rainfall’ and ‘GDD’.

**Table 3.** Comparison between overall  $R^2$  values for both the region and climate models.  $p$ -values apply to a standard ANOVA F-test comparing the climate model to the climate model augmented with the region model x-variables. Each  $p$ -value essentially summarises the empirical evidence for the hypothesis that the difference in overall  $R^2$  between the respective region model and climate model is truly zero.

		Overall $R^2$			
		Region Model	Climate Model	Difference	$p$ -Value
2018	TPC	26.5%	21.4%	5.2%	0.170
	pH	80.1%	65.7%	14.4%	<0.001
	TSS	59.0%	41.6%	17.4%	<0.001
	TA	71.2%	62.5%	8.7%	0.004
2019	TPC	25.7%	16.0%	9.7%	0.043
	pH	71.2%	58.2%	13.0%	0.001
	TSS	61.0%	30.9%	30.1%	<0.001
	TA	82.4%	72.0%	10.4%	<0.001

Nonetheless, the  $R^2$  value for the climate model of each quality parameter was still high in an absolute sense (Table 3). For example, for TA in 2019, 82.4% of the variation was explained by the variables ‘region’ and ‘variety’ (Table 2), whilst in comparison, the climate model using ‘average rainfall’ and ‘GDD’ accounted for 72% of the variation (Table 3). This finding suggests that 10.4% of the variation in TA between ‘region’ and ‘variety’ may not be directly related to climate factors, and instead may relate to other variables such as orchard management, growing system, tree age or root stock (Table 3). Results were analogous for the other quality parameters, except for TPC, where the difference in  $R^2$  between the region and climate models was not statistically significant (Table 3).

The data presented in Table 4 demonstrate the increased complexity in the climate model in comparison to the region model (Table 3) due to the additional factors and therefore interactions. Overall, the results presented in Table 4 (consistent with Table 3) show that the climate model does not provide increased explanatory power for understanding variation amongst quality parameters across either season in comparison to the region model.

The results of the PCA on the comprehensive monthly weather variable matrix are summarised in the Supplementary Materials (Figure S1). There was a very high correlation between GDDs and the first principal component (Figure S1). The winter chill proxy is highly correlated with the second principal component, while the average rainfall variable is not highly correlated with any particular principal component.

As a scenario analysis, the winter chill proxy was added as an x-variable to the climate model regressions. The results are not presented in the main text of the paper because the scenario analysis does not change the substantive inferences about the climate model presented in the main text. In particular, the inferences about how the climate model compares to the region model do not change. The scenario analysis is included in the Supplementary Materials (Figure S2) as well as a comparative table to the region model (Table S1).

**Table 4.** Key results from the climate model, including  $p$ -values and  $R^2$  adjusted values for base cider chemical parameters (TPC, pH, TSS and TA) of all three dessert varieties, ‘Pink Lady’, ‘Fuji’ and ‘Royal Gala’ combined, over two growing seasons, 2018 and 2019. The  $p$ -values correspond to a series of F-tests within a conventional ANOVA breakdown of variable significance (the order of testing corresponds to the introduction of the ‘variety’, ‘rain’, ‘GDDs’ and ‘interaction’ variables, respectively). The  $R^2$  values in the ‘variety’, ‘rain’, ‘GDDs’ and ‘interaction’ columns are the marginal increase in  $R^2$  attributable to adding each variable (in sequence, from left to right). The  $R^2$  in the ‘overall’ column is the total for the regional model with all variables included.

	Variety		Rain		GDD		Variety:Rain		Variety:GDDs		Rain:GDDs		Overall	
	$p$ -Value	$R^2$	$p$ -Value	$R^2$	$p$ -Value	$R^2$	$p$ -Value	$R^2$	$p$ -Value	$R^2$	$p$ -Value	$R^2$	$p$ -Value	$R^2$
2018														
TPC	0.11	1.2%	<0.001	7.8%	0.238	0.2%	0.287	0.1%	0.496	−0.6%	<0.001	12.7%	<0.001	21.4%
pH	<0.001	55%	0.067	0.6%	0.004	2%	0.019	1.6%	<0.001	6.8%	0.969	−0.3%	<0.001	65.7%
TSS	<0.001	12.4%	<0.001	14.8%	0.076	0.9%	0.004	4.3%	0.424	−0.3%	<0.001	9.5%	<0.001	41.6%
TA	<0.001	34.6%	<0.001	12.2%	0.499	−0.3%	<0.001	8.8%	0.32	0%	<0.001	7.2%	<0.001	62.5%
2019														
TPC	0.001	8.9%	0.769	−0.8%	0.268	0.1%	0.793	−1.4%	0.317	0.1%	0.001	9%	0.001	16%
pH	<0.001	45%	0.244	0%	<0.001	14.6%	0.861	−0.6%	0.808	−0.6%	0.413	−0.1%	<0.001	58.2%
TSS	<0.001	21.7%	0.524	−0.5%	0.002	5.7%	0.34	0%	0.311	0.2%	0.011	3.8%	<0.001	30.9%
TA	<0.001	71.9%	0.03	1%	0.479	−0.1%	0.533	−0.2%	0.639	−0.3%	0.994	−0.3%	<0.001	72%

#### 4. Discussion

Base cider quality parameters were influenced by region to varying degrees, whilst having some interactions with variety. Overall, the region model was found to explain the greatest amount of variation amongst the outcome measurements when compared to the climate model. Given the specific influence of region on each quality parameter, each is discussed separately in the sections below. Furthermore, the influence of climatic factors was also investigated separately to region in order to determine any climate-specific influence on cider quality parameters.

##### 4.1. TPC

For both seasons, laboratory analysis of the key cider quality parameter TPC was shown to vary substantially between regions, having both variety-related and seasonal influences. Variety did not significantly influence TPC results in 2018 but had a limited influence in 2019. However, studies have shown that dessert apples generally have low TPC values, with limited variation between varieties; hence, any detectable difference in TPC in dessert varieties is likely to be meaningful [6,15,24,25].

As approximately 25% of the variation described in the region model can be attributed to region and variety, there is likely a range of unmeasured factors also contributing to TPC variation. Phenolic content has been found to be influenced by apple maturity at harvest [27], milling and pressing processes [28] and yeast strain used in the fermentation process [29]. These factors were controlled in this study, with apples harvested at the same maturity, as confirmed using the iodine starch test [30]. Furthermore, apples were milled and pressed uniformly, and all ciders were fermented with the same yeast strain. Other factors that may be influencing variation in TPC include rootstock, age of tree and orchard management [3]. Thompson-Witrick et al. [5] showed that apple cultivar, growing location and harvest season create highly variable polyphenol composition and concentration in cider. Thompson-Witrick et al. [5], used a range of apple varieties including cider varieties, which provided a greater diversity of polyphenol content, highlighting the influence of

region [24]. Valois et al.'s [7] findings supported Thompson-Witrick et al.'s [5], noting that phenolic composition may vary between varieties, regions and seasons.

#### 4.2. pH

The region model determined that pH was significantly influenced by region, although individual key drivers and interactions varied between seasons. There was a clear difference between regions for pH across the two seasons studied. The mean pH for ciders made from apples grown in the Huon Valley was the second highest of all regions in 2018; however, in 2019, the same region produced the second lowest mean. This highlights the influence of season, most likely attributed to changes in climatic conditions. These findings are in contrast to Alexander et al.'s [16], where region was not found to significantly impact pH, even though the regions differed in climate factors such as air temperature and GDDs. In wine, seasonal conditions have been shown to influence pH, as has solar exposure [17]. Increased bunch exposure is negatively associated with pH; thus, winemakers now use grape exposure as a tool to manipulate the pH of wine.

Over both seasons, variety was found to influence pH through an interaction with region in 2018, and independently as main effects in 2019. This is consistent with the findings of Kendall et al. [31] which demonstrate a significant difference in pH of juice between varieties, as collected when studying the effects of reduced irrigation on cider cultivars in Washington, USA. Sousa et al. [32] also found variation in pH between varieties used for cider making. Similar to the seasonal variation in this study, Cline et al. [3] found varieties varied in pH from 3.50 to 4.16 between seasons.

The observation of a significant interaction between region and variety in 2018 and not 2019 may mean that the interaction is season-dependent, and therefore relies on a specific combination of climatic conditions. However, cider made from 'Pink Lady' apples grown in Shepperton had similar pH values to cider made using 'Fuji' apples, driving the interaction seen in 2018. For all other regions, cider made using 'Pink Lady' apples had significantly lower pH values compared to both other varieties (Figure 1). The reason this interaction was not observed in 2019 is likely due to the inability to source apples from Shepperton in 2019. Therefore, it remains unknown if this interaction would have occurred in 2019 if the same samples were collected for a second season.

pH plays an important role in microbiological stability for cider fermentation [18]. Of the 1096 ciders produced in this study, the pH of the finished ciders ranged between 3.34 and 4.39. In the early stages of fermentation, before a higher concentration of alcohol increases microbial stability, there is a risk of microbial contamination with high pH juice [18]. A pH greater than 4 exposes juice to spoilage microbes, which can lead to major flavour issues and negatively impacts cider quality [32]. As the main role of acidity in cider is to support fermentation and influence cider flavour, pH balance is a critical component of the process [8] and a pH between 3.3 and 3.7 is ideal for fermentation conditions [16].

#### 4.3. TSS

Sugar in cider primarily acts as a substrate for yeast to convert to ethanol and carbon dioxide via alcoholic fermentation [8]. After fermentation, residual sugar plays a crucial role in cider quality for the sensory perception of sweetness in cider [8]. There were significant interactions between variety and region in both seasons for cider TSS, with 2019 having a lower *p*-value and higher  $R^2$  than 2018. This illustrates the influence of the interaction between region and variety, whilst season drove the degree of intensity. Environmental conditions such as light intensity and canopy exposure have been found to affect TSS, specifically with shading reducing TSS [22].

When studying the influence of fertigation on apple quality, Kuzin et al. [23] determined TSS was largely defined by the weather conditions of the growing season. This is understandable when examining the large seasonal variability between TSS results across all regions in Figure 1 where there is no consistent trend. Others have found seasonal variability in sugar related results; for example, Alexander et al. [16] found variation in specific

gravity between seasons, suggesting the differences may be caused by different storage times at orchards prior to fruit collection. From the results in this study and similar findings across the literature, we can propose that TSS results are influenced by a combination of variety and regional factors, specifically seasonal climate conditions.

#### 4.4. TA

Overall, TA had the highest combined  $R^2$  value over both seasons. Therefore, the region model was best able to explain the variation of TA results using the interaction between region and variety. This is likely a combination of TA being variety-specific and also often influenced by region-specific factors such as climate conditions [16,20,21]. TA has been found to vary between varieties [16], to the extent where TA, alongside polyphenols, has traditionally been used to classify varieties for their flavour category [8]. TA plays an important role in the organoleptic quality of cider [8]. The findings of this study are consistent with Miller et al.'s [20], demonstrating inconsistencies in fruit characteristics across different locations, with TA being particularly higher for many dessert cultivars grown in British Columbia compared to those grown in West Virginia. Differences in TA were attributed to variation in climate conditions between growing locations, including temperature and light intensity [20]. Furthermore, Kuzin et al. [23] concluded that TA content was more dependent on weather conditions rather than by the mineral nutrition treatments tested. In wine production, Sadras et al. [21] found grape TA to be dependent on both variety and season, which was also consistent with the results in this study.

The evidence for varietal and regional influence over TA helps to explain the existence of the interactions found in this study. The interactions indicate that TA varies between some varieties grown in certain conditions and is largely dependent on region. This is highlighted in Figure 1, where there is a clear separation of varieties across regions, as compared to the other quality attributes.

#### 4.5. Season

In this study, a seasonal influence on TPC was observed. In 2018, region was the only significant driving factor, whilst in 2019, variety also played a role. For pH, variation in results was due to an interaction in 2018, whilst in 2019, variety and region influenced results independently. TSS results were influenced by an interaction in both seasons; however, the  $p$ -value and  $R^2$  value varied greatly. These fluctuations in quality, as influenced by season, are likely to impact ciders produced; consequently, seasonal variation can often create inconsistencies for cider makers. TA was the only measure of quality to not differ significantly between seasons according to the region model; however, Figure 1 demonstrates that the variety/region interactions still vary considerably between seasons. Similar studies have found results to be influenced by seasonality. For example, Plotkowski and Cline [8] found seasonal variation in polyphenols and TA across a range of varieties, to the extent they changed classifications from bittersharp to bittersweet across seasons. Additionally, Alexander et al. [16] discovered seasonal variation between tannin results when studying four different varieties.

#### 4.6. Variety

As variety is known to influence cider quality, a greater understanding of variety-specific qualities is required to better equip cider producers and to enhance the quality of their product. Equally important is the ability to source preferred varieties, as cider apples are often in high demand and in short supply, thus resulting in high prices compared to dessert varieties [16]. The influence of variety is evident in every result displayed above, as is often the case in cider studies. Cline et al. [3] found significant differences between varieties for polyphenols, TA, pH and TSS. Alexander et al. [16] studied the effect of region on apple juice quality characteristics in the USA and found no significant differences nor interactions due to region but found that variety affected all quality measurements

significantly. In previous studies, Way et al. [28] found variety to contribute the most to variation in results, regardless of the treatments implemented [24,25].

#### 4.7. Climate Model

The climate model combined variety with average rainfall and GDDs, two major differentiating climatic factors amongst regions. While the climate model predicted similar values to the region model, the region model ultimately accounted for more variation across the seasons and quality characteristics assessed. This indicates that a large percentage of the variation being accounted for in the region model is likely attributed to the underlying climate differences caused by variability in rain fall and GDDs. The moderate differences between the models could be attributed to other region-specific aspects such as soil, tree age, rootstock and orchard management practices, but as seen in Table 3, this influence is minimal.

Although the region model was consistently able to explain a greater percentage of variation in the data, the difference between the two models was not always significant (Table 3). This was evident in 2018 for the TPC results. However, although both models were similar for TPC, the other three quality parameters were more effectively explained by the region model.

Regardless of the model, region-specific conditions such as climate strongly influenced the base cider quality indicators. This new understanding may encourage cider producers to focus on sourcing fruit from specific regions to champion region-specific attributes. It may also provide cider producers with the knowledge that their ciders may be distinctive to other producers using apples from other regions, offering a new marketing strategy, with the knowledge they may have a unique product that cannot be replicated outside of their specific location.

## 5. Conclusions

This research illustrates that apple-growing region influences cider quality, albeit with season and variety dependency. As expected, apple variety has a major influence on cider quality, even when ciders are produced using dessert apple varieties which have minimal variation amongst them. Production region can be used as a proxy for climatic condition variability when comparing growing regions. As the cider industry develops in Australia, further research is warranted to compare the influence of production region on cider quality for cider varieties. This research provides evidence that the concept of terroir is relevant for cider, offering opportunity for cider producers to select apples based on growing location exclusively if seeking consistency, or alternatively over a range of regions in pursuit of enhancing complexity.

**Supplementary Materials:** The following supporting information can be downloaded at: <https://www.mdpi.com/article/10.3390/beverages10040099/s1>, Figure S1: Scatter plots and correlations for the climate model variables (GDD and average rainfall), the chill proxy (the average of the minimum temperatures for the months of June, July and August) and climate data principal components. The "PC" data is for the first four principal components of monthly weather variables (on a monthly weather variable matrix with average rainfall, solar radiation, mean minimum monthly temperature and mean maximum monthly temperature). Data is for all regions and both seasons. The diagonal plots are the smoothed distributions (akin to the top of a histogram). The lower diagonal charts are pairwise scatter plots. The upper diagonal cells contain Pearson correlation coefficients, with standard  $p$ -values for a test on a null hypothesis that the true correlation is zero. 82% of variance in the climate matrix is explained by the first four principal components. (\* represents  $p \leq 0.05$ , \*\* is  $p \leq 0.01$  and \*\*\* is  $p \leq 0.001$ ).; Figure S2: Climate key indicators average rainfall and GDD compared by region over the two growing seasons of 2018 and 2019 with the addition of the winter chill proxy. Regions are displayed as Huon = Huon Valley, Sprey = Spreyton, Narr = Narre Warren, Shep = Shepparton, Batl = Batlow, Adel = Adelaide Hills, Manj = Manjimup, Oran = Orange and Stan = Stanthorpe. Table S1: Comparison between overall  $R^2$  values for both the region and hybrid models. The "hybrid" model combines region, key climate factors (GDD and Average rainfall), as well as the winter chill

proxy. *p*-values apply to a standard ANOVA F-test comparing the climate model to the climate model augmented with the region model *x*-variables. Each *p*-value essentially summarises the empirical evidence for the hypothesis that the difference in overall  $R^2$  between the respective region model and climate model is truly zero.

**Author Contributions:** Conceptualization and methodology of this research were carried out by M.L.W., J.E.J. and N.D.S.; formal analysis, investigation and data curations by M.L.W., I.H. and R.G.D.; writing—original draft preparation by M.L.W.; and writing—review and editing was a joint effort between M.L.W., J.E.J., I.H., R.G.D. and N.D.S. The research was supervised by J.E.J. and N.D.S. All authors have read and agreed to the published version of the manuscript.

**Funding:** This research was funded by a Westpac Future Leaders Scholarship and the Tasmanian Institute of Agriculture at the University of Tasmania. Apples were supplied in kind from the growers around the country.

**Institutional Review Board Statement:** Not applicable.

**Informed Consent Statement:** Not applicable.

**Data Availability Statement:** The original contributions presented in the study are included in the article/Supplementary Materials; further inquiries can be directed to the corresponding author/s.

**Acknowledgments:** The authors wish to acknowledge the apple growers from around the country who generously donated their apples in kind to this study. Thank you John Evans, Andrew Scott, Howard Hansen, Matthew Griggs, Neil Fuller and Justin Heaven for organising applies from Stanthorpe and Kevin Dodds for organising apples from Batlow. The authors also wish to acknowledge Tim Jones from Willie Smith's Cider whose input and support was greatly appreciated.

**Conflicts of Interest:** The authors declare no conflicts of interest.

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