horticulturae

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Effect of Rain Cover on Tree Physiology and Fruit Condition and Quality of 'Rainier', 'Bing' and 'Sweetheart' Sweet Cherry Trees

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Abstract: A study was conducted in a commercial sweet cherry orchard in central Chile. The objective was to evaluate the rain cover effect on changes in the microclimate, vegetative growth, plant physiology and fruit quality of 'Rainier', 'Bing' and 'Sweetheart' sweet cherry trees. The data were compared to a control without a rain cover. The results showed that, under the rain cover, there was a 50–60% reduction in total solar radiation, as well as an increase in air temperature (+0.6 °C) and a decrease in relative humidity (−4.7 percentage points) in the upper canopy zone. Regarding the trees under rain cover, a greater shoot length (28–58%) and leaf area (24–54%) were observed among cultivars compared to the control; the trunk cross-sectional area was only significant in 'Rainier', it being 1.2 times greater under rain cover. CO_2 assimilation showed no differences, but an increase in the leaf transpiration rate was observed. The fruit firmness and sugar content in fruits were negatively affected by the rain cover, those characteristics being of major relevance for the cherry growers. Additionally, the contents of anthocyanins and carotenoids and the antioxidant capacity were significantly lower only in 'Rainier' under rain cover, while the total phenol content decreased in all three cultivars. The rain cover did not negatively affect the tree physiology, but it can be detrimental in bicolor cultivars with a yellow flesh due to a lower color and phenolic compounds development.

Keywords: fruit quality; gas exchange; pigment; protected environment; *Prunus avium*; rain cracking; leaf area

Citation: Pino, S.; Palma, M.; Sepúlveda, Á.; Sánchez-Contreras, J.; Moya, M.; Yuri, J.A. Effect of Rain Cover on Tree Physiology and Fruit Condition and Quality of 'Rainier', 'Bing' and 'Sweetheart' Sweet Cherry Trees. *Horticulturae* **2023**, *9*, 109. https://doi.org/10.3390/horticulturae9010109

Academic Editor: Dong Zhang

Received: 6 December 2022
Revised: 30 December 2022
Accepted: 10 January 2023
Published: 13 January 2023

1. Introduction

Sweet cherry (*Prunus avium* L.) is one of the main fruit trees planted in Chile, with more than 61,000 ha in cultivation and 350,000 t of fresh fruit exported in the 2021/2022 season [1]. One of the main problems affecting cherry production is rainfall-induced cracking in periods close to harvest [2], which causes significant economic losses [3]. In response to this problem, the use of plastic covers has become widespread in Chile to prevent fruit quality deterioration [4]. However, these structures modify light interception and the microclimate around the trees, affecting the physiological performance of the trees.

Depending on the material used and the installation design, the plastic covers reduce incident solar radiation and photosynthetically active radiation at different levels on crops, which could affect flower bud development [5]. In apple trees, it has been observed that 30% full sun light is the threshold value for fruit production [6]. In addition, in sweet cherry trees, the use of covers to protect fruit from rain has shown variable effects on both fruit quality and biochemical composition [7–9]. Regarding vegetative growth, a greater shoot elongation and greater leaf area per tree have been reported generally, causing greater shading inside the canopy, which can be detrimental to fruit production [5]. In sweet cherry trees, summer pruning management and the use of reflective mulch, which allow for a better light distribution in the canopy, have shown a positive effect in the subsequent production season, given by an improved sugar content in the buds [5,10].

Additionally, plastic covers affect air temperature and canopy relative humidity [7,11,12]. In warmer climates, an increase in temperature due to reduced rain cover ventilation could increase crop water demand, causing greater stress on the trees [5]. On the other hand, a reduction in wind together with a higher humidity under rain cover could favor the tree water status due to the lower atmospheric demand [12]. Therefore, modifying the microclimate of the trees during fruit growth, with the implementation of plastic covers, could alter tree physiology as well as fruit quality and composition. This is relevant in cherry trees, since the organoleptic parameters of sweet cherries, such as color, sweetness, firmness and acidity are highly valued by consumers [13]. Likewise, the phenolic content and antioxidant activity of sweet cherries are also appreciated, given the healthy characteristics attributed to them [14,15].

The objective of the present research was to study the effect of rain cover on the microclimate, vegetative growth, plant physiology, fruit quality and biochemical characteristics in three sweet cherry cultivars—'Rainier', 'Bing' and 'Sweetheart'—grown in a commercial orchard in central Chile.

2. Materials and Methods

2.1. Plant Material and Environmental Conditions

The study was conducted in a commercial orchard in Rio Claro, Maule Region, Chile (35°14′ S, 71°14′ W, 342 m a.s.l.) during the 2017/18 growing season. The sweet cherry (*Prunus avium* L.) trees were 'Rainier', 'Bing' and 'Sweetheart' cvs., grafted on 'MaxMa 14' rootstock. Planting was carried out in autumn 2001 at a distance of 4.5 m × 2.5 m (889 trees ha^{-1}) and in an NW–SE orientation. The training system used was central axis, with a 3.5 m height.

The area has a Mediterranean climate with a dry season in the summer, between December and March, and rainfall mostly concentrated between June and August. During 2017, the average annual rainfall was 900 mm, and the average air temperature was 14 °C. The soil is characterized by a clayey texture, Alfisol type. Irrigation was carried out with two dripper lines per row, according to the requirements of each cultivar, without distinguishing between blocks with and without rain cover.

2.2. Treatments

Two treatments were evaluated in a homogeneous area of 2.4 ha. There were trees with rain cover and others without rain cover (control). The rain cover used was a Vöen model of high-density polyethylene (Vóhringer GmbH & Co. Berg, Fronreute, Germany), and it was installed on a structure of wooden posts and wires at a 4 m height. The rain cover was deployed during the entire fruit growth period and removed after fruit harvest. The treatments were distributed in two blocks, and in each block, the cultivars were arranged in seven rows, side by side. All measurements were made on ten trees of the central row of each cultivar and treatment, considering two edge rows per side.

2.3. Environmental Conditions

The total solar radiation (W m^{-2}) was measured with an LI-189SA radiometer (Li-COR Inc., Lincoln, NE, USA), photosynthetically active radiation (PAR; µmol m^{-2} s^{-1}) with an AccuPar LP-80 ceptometer (Decagon Devices, Inc., Pullman, WA, USA) and ultraviolet (UV-B; µW cm^{-2}) with a PMA2100 radiometer (Solar Light Co., Glenside, PA, USA). Evaluations were conducted preharvest (14 December 2017) and postharvest (1 February 2018) at 1.5 m above the ground between rows. On both dates, five readings were taken at 10:00, 14:00 and 18:00 h.

Additionally, the air temperature (T) and relative humidity (RH) were monitored in both treatments using HOBO UX 100 automatic recorders (Onset Computer Co., Bourne, MA, USA). Four sensors were installed in the central rows of both treatments, at 1.5 and

3.0 m above the ground, protected from direct solar radiation, rain and aerosols. Data were recorded every 15 min throughout the season. With that, the mean, maximum and minimum daily T and RH and the thermal accumulation in degrees day (GD, base 10 °C) [16] and growing degree hours (GDH, base 4.5 °C and optimum 25 °C) [17] were calculated. Environmental stress was calculated according to the stress index used in the apple tree study [18].

$$\text{Stress index} = (\text{Tair} - 10)(-0.2\text{RH} + 15)$$

where Tair = air temperature; RH = relative humidity.

2.4. Vegetative Growth

The canopy volume (m^3) and trunk cross-sectional area (TCSA; cm^2) were measured at the end of the season (26 February 2018), prior to rain cover removal, on 10 trees of each cultivar and treatment. Shoot evaluations were performed on four shoots per tree, which were cut, stored in a cooler and transported to the laboratory for immediate measurement. The shoot length (cm), internode length (cm), number of leaves per shoot and shoot leaf area (cm^2) were measured with an LI-3100 leaf area meter (LI-COR Inc., Lincoln, NE, USA). The leaf fresh weight (g) was measured with an analytical balance and dry matter (%) by dehydration in a drying oven (D-6450, Heraeus GmbH & CO., Hanau, Germany) at 60 °C.

2.5. Physiological Measurements

The plant xylem water potential (MPa) was measured with a Scholander 3005 pressure chamber (Soil Moisture, Santa Barbara, CA, USA). Leaves were previously sealed with a plastic bag and aluminized film for at least two hours. Gas exchange was measured with an InfraRed Gas Analyzer LCpro-SD (ADC BioScientific, Herts, UK). Maximum photosystem II efficiency was measured with an OS1-FL fluorometer (Opti-Sciences, Tyngsboro, MA, USA) on leaves acclimated to darkness for at least 20 min. Evaluations were performed on trees with fruit (11 December 2017) and without fruit (11 January 2018) at midday and under clear day conditions, considering five leaves of each treatment and cultivar in each evaluation.

The relative leaf pigment content was measured in a non-destructive form with the Dualex^{TM} (Force-A, Orsay, France) and Spad index with an SPAD 502 Plus (Konica Minolta, Tokyo, Japan), both based on the fluorescence excitation spectrum of chlorophyll. The evaluation was performed pre-harvest (13 December 2017) on the adaxial side of leaves located in the middle third of the current-year shoots. Samples of five leaves from each treatment and cultivar were considered.

2.6. Physicochemical Characteristics of the Fruit

2.6.1. Fruit Quality

Fruit quality evaluations were carried out at commercial harvest on a sample of 25 fruit from the upper and lower canopy zones. The commercial harvest was carried out according to the color scale on 14 December on 'Rainier' and 'Bing' and on 21 December on 'Sweetheart'. The weight (g) was measured with an analytical balance and the diameter (mm) was measured with a caliper on the equatorial zone of the fruit. The color was determined visually by means of a scale (light red = 1, red = 2, mahogany red = 3, dark mahogany = 4 and black = 5; cherry color chart scale, Pontificia Universidad Católica de Chile). In 'Rainier', as it is a bicolor cultivar, the color was determined as the red cover percentage. Firmness (g mm^{-1}) was measured with a FirmTech II (BioWorks, Inc., Wamego, KS, USA). Then, five replicates of five fruits were considered to determine the soluble solids content (°Brix) with a PAL-BX/ACID digital refractometer (Atago, Tokyo, Japan).

2.6.2. Pigment Content

Pigment content evaluations were performed on a random sample of five fruits. The total anthocyanin concentration was quantified according to the method of Fuleki and

Francis [19]. Two discs (0.95 cm^2) of fruit skin were extracted, fragmented and macerated in 500 µL of Ethanol/HCl at 4 °C in darkness for 24 h. After incubation, the extract was centrifuged at 200× *g* for 3 min. After collecting the supernatant, a new extraction was made to the discs using the same procedure. The absorbance was measured at 535 nm using a spectrophotometer (Pharo 300, Merck KGaA, Darmstadt, Germany). The results were expressed as mg per 100 g of fresh weight (mg 100 g^{-1} FW).

Chlorophylls (Chl) and carotenoids were quantified based on the method developed by Lichtenthaler [20]. Two fruit skin discs (0.95 cm^2) were extracted from the fruit, fragmented and macerated in 500 µL of acetone 80% (acetone/water 80:20, *v/v*) for 24 h in the dark at 4 °C. The extract was centrifuged at 200× *g* for 3 min after incubation. Then, it was diluted to 500 µL with acetone 80%. Absorbance was measured at 470, 647 and 663 nm using a spectrophotometer (Pharo 300, Merck KGaA, Darmstadt, Germany). The results were expressed as mg per 100 g of fresh weight (mg 100 g^{-1} FW).

2.6.3. Phenolic Extraction

The total phenols and antioxidant capacity measurements were carried out on an extract of the edible fraction of the fruit (skin plus pulp). A total of 5 g of fruit was pulverized with liquid nitrogen and diluted in 20 mL of 80% ethanol (ethanol: water 80:20, *v/v*). The solution was put in an ultrasonic bath for 15 min and in a thermoregulated bath at 100 °C for 10 min to facilitate the extraction. The extract was filtered under vacuum and diluted to 10 mL with ethanol at 80%. Subsequently, it was stored in the dark at −20 °C until the measurements were performed.

2.6.4. Determination of the Total Phenolic Content

The total phenolic content (TPC) was determined by the Coseteng and Lee [21] method using the Folin–Ciocalteu reagent. Samples were prepared with 10 µL phenolic extract and 490 µL of ethanol 50% (ethanol/water 50:50, *v/v*). They were then mixed with 0.5 mL of Folin–Ciocalteu reagent (Merck, Darmstadt, Germany) and 2.5 mL of deionized water. They were homogenized for 1–3 s and left to stand for 5 min. Then, 0.5 mL of sodium carbonate (Na_2CO_3; in 2.5 mL of deionized water) was added and left to stand for 15 min at room temperature (20 °C). Absorbance was measured at 640 nm with a spectrophotometer (Pharo 300, Merck KGaA, Darmstadt, Germany). The calibration curve was performed with chlorogenic acid, with a difference of 25 µL between each point on the curve, ranging from 0 to 500 µL. The results were expressed as mg chlorogenic acid equivalents per 100 g of fresh weight (mg CAE 100 g^{-1} FW).

2.6.5. Determination of Antioxidant Activity

The Oxygen Radical Absorbance Capacity (ORAC) determination was performed based on the method described by Huang et al. [22] and Prior et al. [23], with some modifications. The radical standard was prepared with 0.1624 g of 2,2′-azo-bis (2-amidino-propane) di-hydrochloride (AAPH) dissolved in 4 mL phosphate buffer 75 mM pH 7.4. The calibration curve was performed using 6-hydroxy-2,5,7,8- tetramethylchromane-2-carboxylic acid (Trolox) with concentrations between 6.5 and 100 µM in phosphate buffer 70 mM pH 7.4. Florescence was measured with a microplate reader (Synergy HT, BioTek Instruments, Winooski, Vermont, USA). The results were expressed as µmol of Trolox equivalents per 100 g of fresh weight (µmol TE 100 g^{-1} FW).

2.7. Statistical Analysis

A one-way analysis of variance (ANOVA) was performed to evaluate if there were significant differences between treatments. Means were compared using the Tukey HSD 95% test ($p \leq 0.05$). Data analysis was performed using Statgraphics Centurion XVI software (Warrenton, VA, USA). Figures were generated using SigmaPlot 10 software (WPcubed GmbH, München, Germany).

3. Results

3.1. Environmental Conditions

The rain cover significantly filtered the solar radiation that impacted the trees in relation to those uncovered. On December 14, the total solar radiation, PAR and UV-B at midday decreased under the rain cover by an average of 50%, 58% and 66%, respectively, while on February 1, the reductions were 60%, 65% and 65%, respectively (Figure 1).

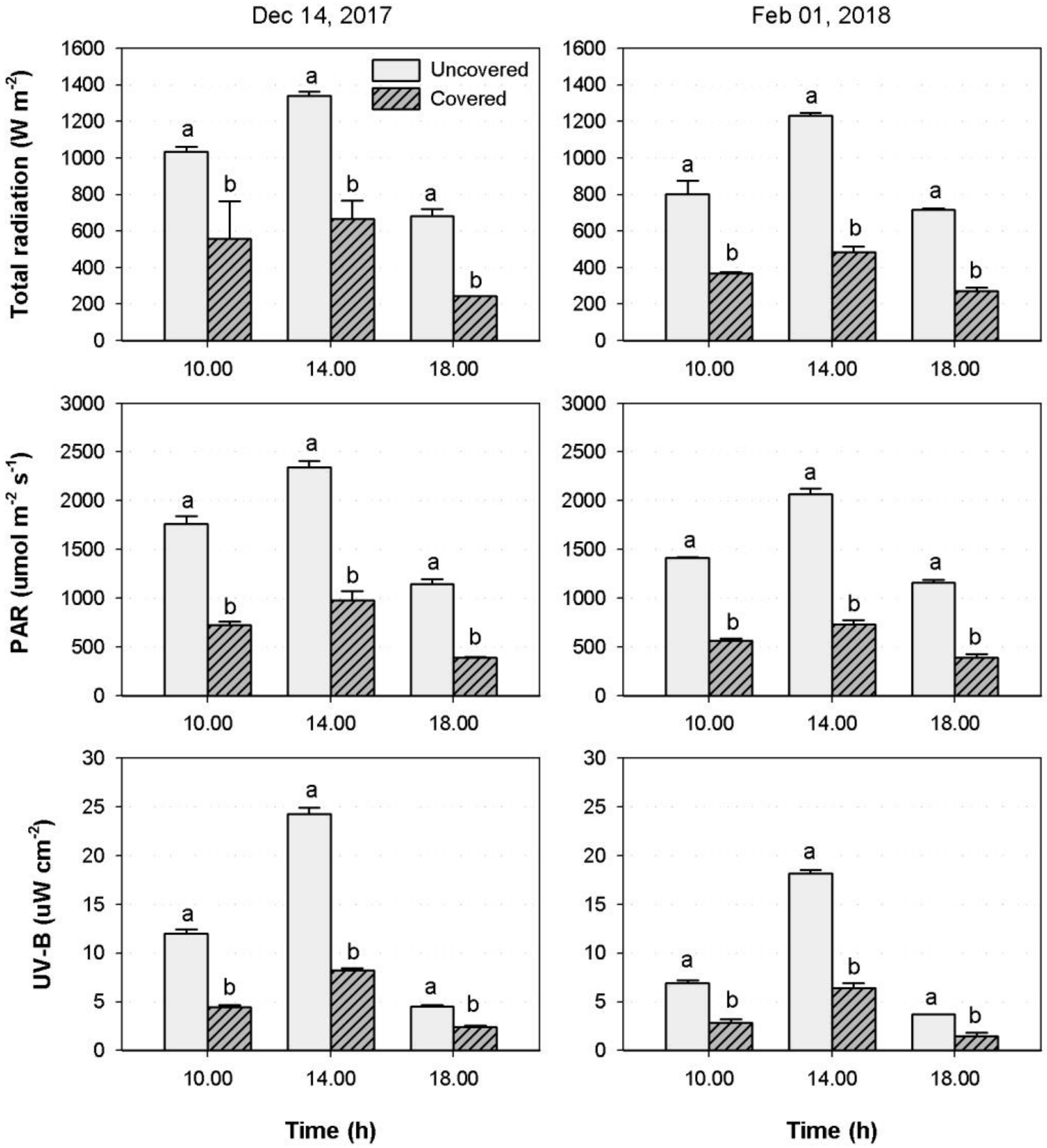

Figure 1. Total solar, photosynthetically active (PAR) and ultraviolet B (UV-B) radiation with and without rain cover at three times of the day on 14 December 2017 and 1 February 2018. Columns followed by the same letters indicate no statistical difference between treatments according to the Tukey test ($p \leq 0.05$). Means + SE (n = 5). Central Valley, Chile.

The rain cover affected the air temperature to a greater extent in the area closest to it, reaching its maximum earlier than in the uncovered treatment and for a longer period of time, while the measurements at 1.5 m did not show major differences, with the minimum temperatures being warmer under the rain cover (Figure 2).

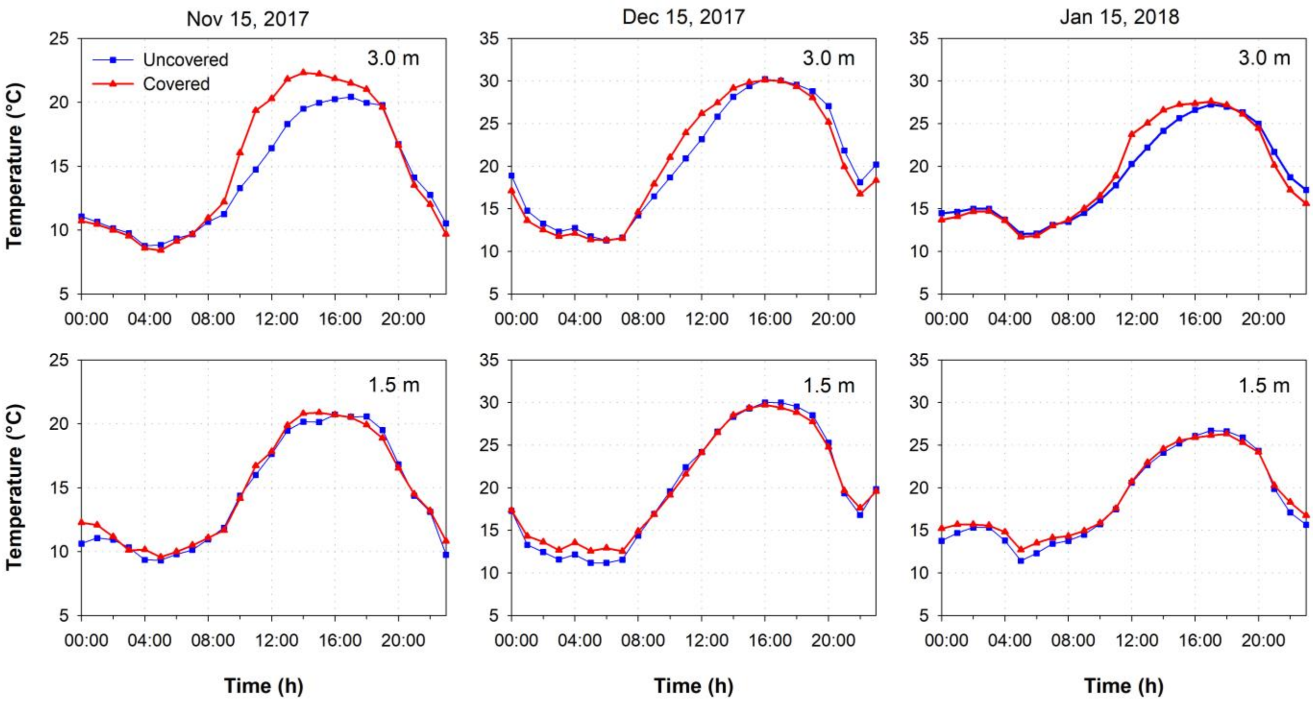

Figure 2. Daily fluctuation of air temperature with and without rain cover at 3 m (**top**) and 1.5 m (**bottom**) above the ground. Days evaluated under cloud-free conditions. Central Valley, Chile.

The relative humidity at a 3 m height was significantly lower during most of the day under rain cover, compared to the control, as a consequence of the higher temperature in the upper zone; at a 1.5 m height, the differences observed between treatments were considerably smaller (Figure 3).

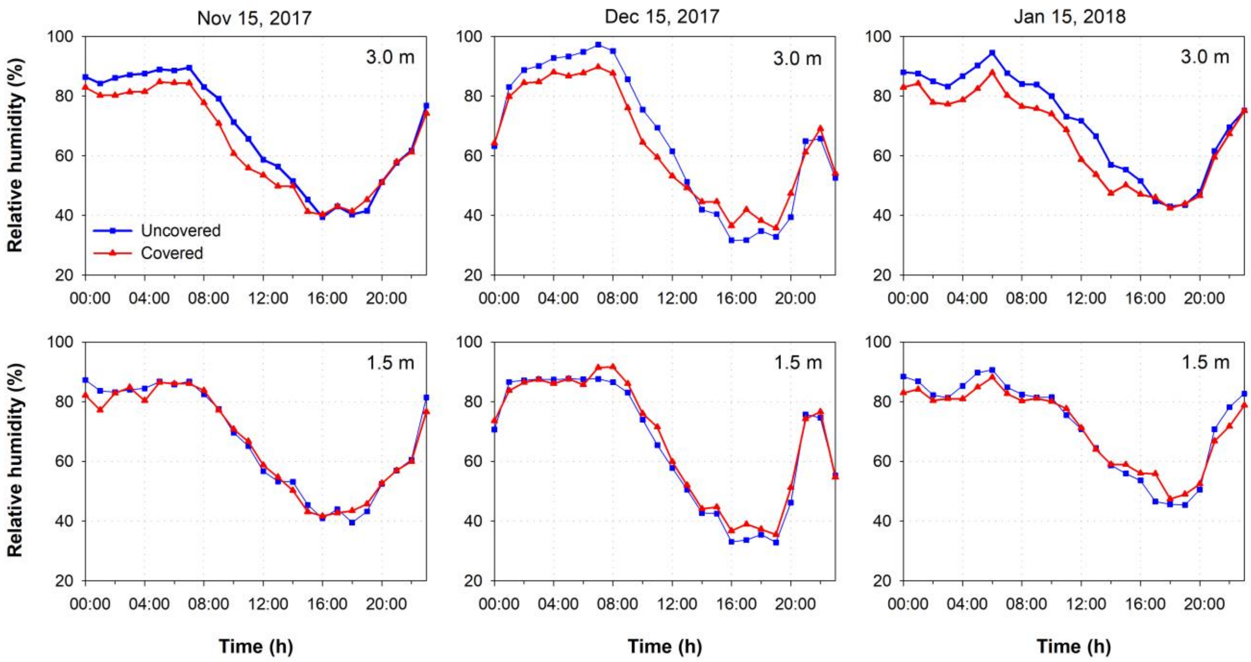

Figure 3. Daily fluctuation of relative humidity with and without rain cover at 3 m (**top**) and 1.5 m (**bottom**) above the ground. Days evaluated under cloudless conditions. Central Valley, Chile.

From 1 November to 15 December 2017, the daily maximum temperatures at a 3 m height were higher under rain cover, but not at 1.5 m. On the other hand, thermal accumulation, expressed in GDH and GD, tended to increase under rain cover at both heights (Table 1). The stress index evaluated at 3 m registered higher values under rain cover; however, the opposite occurred at 1.5 m (Table 1).

Table 1. Air temperature, relative humidity, thermal accumulation and stress index between 1 November 2017 and 15 December 2017 in sweet cherry trees with and without rain cover at 3 and 1.5 m heights. Central Valley, Chile.

Height (m)	Treatment	Air Temperature (°C)			Relative Humidity (%)			Thermal Accumulation		Stress Unit
		Max	Mean	Min	Max	Mean	Min	GDH	GD	
3.0	Uncovered	24.8	16.3	9.1	97.4	75.9	44.8	12,385	296	23,591
	Covered	25.7	16.9	9.2	89.8	71.2	46.4	12,562	323	26,717
1.5	Uncovered	24.5	16.2	9.1	88.9	74.3	46.6	12,436	293	21,804
	Covered	24.2	16.7	10.2	93.2	75.4	49.4	13,238	307	18,741

During the period from 1 November 2017 to 28 February 2018, a similar trend was shown as that before harvest, although with bigger differences in the stress index, which were attributed to the higher temperatures and lower relative humidity due to the advance of the season (Table 2).

Table 2. Air temperature, relative humidity, thermal accumulation and stress index between 1 November 2017 and 28 February 2018 in sweet cherry trees with and without rain cover at 3 and 1.5 m heights. Central Valley, Chile.

Height (m)	Treatment	Air Temperature (°C)			Relative Humidity (%)			Thermal Accumulation		Stress Units
		Max	Mean	Min	Max	Mean	Min	GDH	GD	
3.0	Uncovered	27.4	18.3	10.4	96.1	72.3	40.7	35,850	1011	86,914
	Covered	27.6	18.5	10.2	88.3	67.0	41.3	35,243	1041	102,845
1.5	Uncovered	26.9	18.1	10.1	89.4	71.8	42.2	35,701	985	81,393
	Covered	26.4	18.4	11.2	92.6	72.9	45.4	38,157	1016	69,248

3.2. Vegetative Growth

The canopy volume at the end of the season did not show significant differences for the use of rain cover in any of the cultivars. TCSA tended to be higher in trees under rain cover, being significant only in 'Rainier', with an increase of 15% in relation to the control (Table 3).

Table 3. Vegetative growth of sweet cherry cv. 'Rainier', 'Bing' and 'Sweetheart' with and without late-season rain cover.

Cultivar	Treatment	Canopy Volume (m^3)	TCSA (cm^2)	Shoot				Leaf			Dry Matter (%)
				Length (cm)	Internode Length (cm)	Leaf Area (cm^2)	Fresh Weight (g)	N° Leaves	Area (cm^2)	Fresh Weight (g)	
'Rainier'	Uncovered	16 a	289 b	47 b	2.2 b	1496 b	42 a	21 a	71 b	2.0 a	40 a
	Covered	13 a	354 a	60 a	3.0 a	1854 a	45 a	20 a	91 a	2.2 a	37 b
	p-value	0.14	0.04	0.04	0.01	0.01	0.46	0.57	0.00	0.16	0.00
'Bing'	Uncovered	17 a	343 a	33 b	1.7 b	1094 b	31 a	19 a	55 b	1.6 a	44 a
	Covered	18 a	399 a	49 a	2.3 a	1510 a	37 a	22 a	67 a	1.7 a	38 b
	p-value	0.40	0.12	0.04	0.01	0.03	0.23	0.08	0.03	0.47	0.00
'Sweetheart'	Uncovered	13 a	194 a	33 b	2.2 b	1034 b	28 b	15 b	69 b	1.9 a	43 a
	Covered	15 a	219 a	52 a	2.6 a	1558 a	37 a	20 a	80 a	1.9 a	39 b
	p-value	0.18	0.28	0.00	0.00	0.00	0.00	0.00	0.01	0.98	0.02

Mean values followed by different letters in the same column show significant differences between treatments according to the Tukey's test ($p \leq 0.05$) ($n = 10$).

The shoot growth of trees under rain cover showed increases of 28–58% in length, 18–36% in internode length and 24–54% in area among cultivars. The leaves number per shoot was significant only in 'Sweetheart', with a 25% increase under rain cover. The leaf dry matter was lower under rain cover, while the area was, on average, 22% greater among cultivars (Table 3).

3.3. Plant Physiological Variables

In preharvest measurements, the leaf photosynthetic rate did not show significant differences between treatments, but the transpiration rate did, it being higher in covered trees in all cultivars. The higher transpiration under rain cover resulted in a lower water use efficiency, especially in 'Bing' and 'Sweetheart' (Table 4).

Table 4. Physiological variables in sweet cherry trees cv. 'Rainier', 'Bing' and 'Sweetheart' with and without rain cover on two dates: 11 December 2017 (with fruit); 11 January 2018 (without fruit).

Cultivar	Treatment	Assimilation (µmol CO_2 m^{-2} s^{-1})		Transpiration (mmol H_2O m^{-2} s^{-1})		Water Use Efficiency (µmol CO_2 mmol H_2O^{-1})		Stomatal Conductance (mol H_2O m^{-2} s^{-1})		Stem Water Potential (MPa)		Photon Maximal Efficiency Fv/Fm	
		11-Dec	11-Jan	11-Dec	11-Jan	11-Dec	11-Jan	11-Dec	11-Jan	11-Dec	11-Jan	11-Dec	11-Jan
'Rainier'	Uncovered	9.4 aA	8.4 aA	2.9 bA	1.6 aB	3.2 aB	5.4 aA	0.27 aA	0.11 aB	−0.9 bA	−1.9 aB	0.78 aA	0.81 aA
	Covered	9.9 aA	7.1 aB	3.7 aA	1.3 aB	2.7 aB	5.5 aA	0.25 aA	0.09 aB	−0.6 aA	−1.4 aB	0.82 aA	0.82 aA
'Bing'	Uncovered	9.1 aA	6.8 aB	2.5 bA	2.0 aB	3.7 aA	3.5 aA	0.20 bA	0.13 aB	−1.0 aA	−1.4 bB	0.81 aA	0.83 aA
	Covered	8.5 aA	7.3 aA	3.1 aA	2.1 aB	2.7 bA	3.4 aA	0.26 aB	1.15 aA	−0.9 aA	−0.9 aA	0.81 aA	0.84 aA
'Sweetheart'	Uncovered	9.4 aA	5.7 aB	2.7 bA	1.2 aB	3.4 aB	4.8 aA	0.18 bA	0.07 bB	−0.9 aA	−1.3 aB	0.80 aA	0.81 aA
	Covered	8.7 aA	7.5 aA	3.6 aA	2.0 bB	2.5 bB	3.7 bA	0.27 aA	0.14 aB	−0.6 aA	−1.2 aB	0.78 aA	0.77 bA

Means followed by the same lowercase letter did not differ statistically between treatments according to Tukey's test. Means followed by the same uppercase letter did not differ statistically between sampling dates according to Tukey's test ($p \leq 0.05$) (n = 5).

After the fruit harvest, the leaf gas exchange showed no differences between treatments, except for 'Sweetheart', which showed higher transpiration and stomatal conductance under rain cover (Table 4).

3.4. Leaf Pigments

The relative pigment content in leaves between treatments was different in each cultivar (Table 5). In 'Rainier', this tended to be higher under rain cover, with significant differences in the flavonoid index and SPAD. In contrast, in 'Bing', it tended to be lower, with significant differences in the chlorophyll index, anthocyanin and NBI. No differences were detected in 'Sweetheart' (Table 5).

Table 5. Relative pigment content in leaves of 'Rainier', 'Bing' and 'Sweetheart' sweet cherry trees with and without rain cover.

Cultivar	Treatment	Dualex index				SPAD Index
		Chlorophyll (Chl)	Flavonoid (Flav)	Anthocyanin (Anth)	Nitrogen Balance (NBI)	
'Rainier'	Uncovered	34 a	1.3 b	0.18 a	10 a	37 b
	Covered	37 a	2.0 a	0.19 a	10 a	42 a
	p-value	0.06	0.01	0.32	0.68	0.04
'Bing'	Uncovered	34 a	1.6 a	0.18 a	10 a	41 a
	Covered	29 b	1.7 a	0.15 b	8 b	39 a
	p-value	0.01	0.23	0.01	0.02	0.42
'Sweetheart'	Uncovered	27 a	7.0 a	0.68 a	14 a	33 a
	Covered	27 a	6.5 a	0.68 a	15 a	33 a
	p-value	0.83	0.24	0.85	0.49	0.69

Mean values followed by different letters in the same column show significant differences between treatments according to the Tukey's test ($p \leq 0.05$) (n = 5).

3.5. Physicochemical Characteristics of the Fruit

Although all the physicochemical characteristics were measured considering fruits from the top and bottom of the tree separately, it was decided to use their average because of the complexity in the interpretation of the resulting table, since no substantial differences were found between them.

The weight was 1–2 g greater in fruits under rain cover, with a significant increase in diameter only in 'Sweetheart' (Table 6). The solid soluble concentration tended to be lower in fruit under rain cover, with significant differences only in 'Bing'. Firmness showed significant differences between treatments in the three cultivars, with fruit under rain cover having an 11% lower firmness, on average, compared to the control (Table 6).

Table 6. Quality parameters of 'Rainier', 'Bing' and 'Sweetheart' sweet cherries with and without rain cover.

Cultivar	Treatment	Diameter (mm)	Weight (g)	SSC (°Brix)	Firmness (g mm^{-1})	Color (%*; 1–5)
'Rainier'	Uncovered	27 a	13 b	20 a	260 a	72 * a
	Covered	28 a	14 a	18 a	239 b	42 * b
	p-value	0.06	0.01	0.08	0.01	0.00 (β)
'Bing'	Uncovered	28 a	11 b	23 a	275 a	4.1 a
	Covered	29 a	13 a	20 b	236 b	4.3 a
	p-value	0.53	0.00	0.00	0.00	0.05 (β)
'Sweetheart'	Uncovered	26 b	12 b	21 a	271 a	2.0 a
	Covered	27 a	13 a	20 a	249 b	2.4 a
	p-value	0.00	0.00	0.13	0.00	0.05 (β)

Means followed by the same letter do not differ statistically according to Tukey test ($p \leq 0.05$) ($n = 25$). *: Percentage of red color coverage. β: comparison by Kruskal–Wallis test. SSC: solid soluble concentration.

Regarding color, the rain cover had an influence depending on the cultivar (Table 6). 'Rainier', being a bicolor cultivar, was the most affected by the use of rain cover, with a 71% reduction in the covering percentage. In the case of the red cultivars, 'Bing' and 'Sweetheart', their color intensity did not show significant differences with the control.

The rain cover tended to reduce the anthocyanin and carotenoid contents in the fruit, with a significant decrease in the bicolor cultivar 'Rainier' (Figure 4), which is consistent with the color measurements described previously (Table 6). The chlorophyll content showed no differences between treatments, independent of the cultivar (Figure 4).

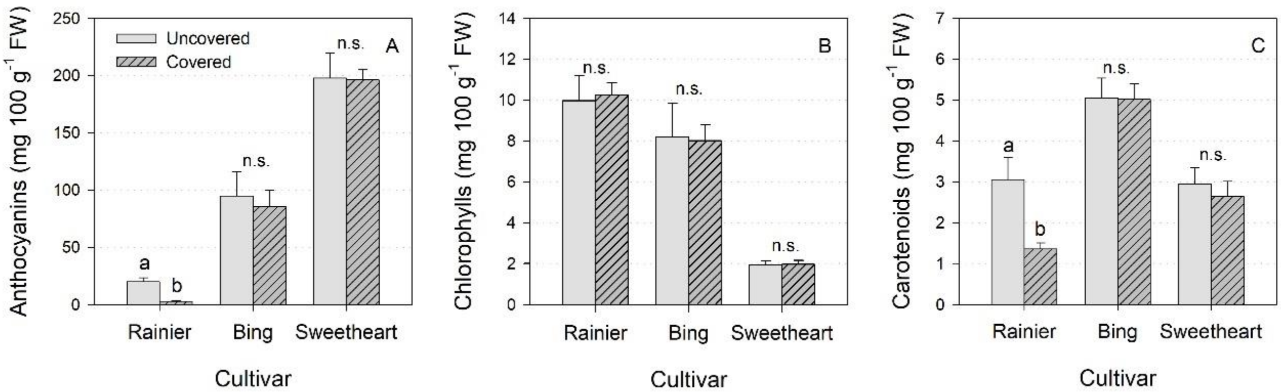

Figure 4. (**A**) Anthocyanin, (**B**) total chlorophyll and (**C**) carotenoid contents of 'Rainier', 'Bing' and 'Sweetheart' sweet cherries with and without rain cover. Columns of a cultivar followed by the same letters indicate no statistical difference between treatments according to Tukey's test ($p \leq 0.05$). Means + SE ($n = 5$).

The total phenolic content of the fruits was significantly lower in the treatments under cover in all cultivars. Likewise, the antioxidant capacity was lower in fruits under rain cover, it being significant only in the bicolor cultivar 'Rainier' (Figure 5).

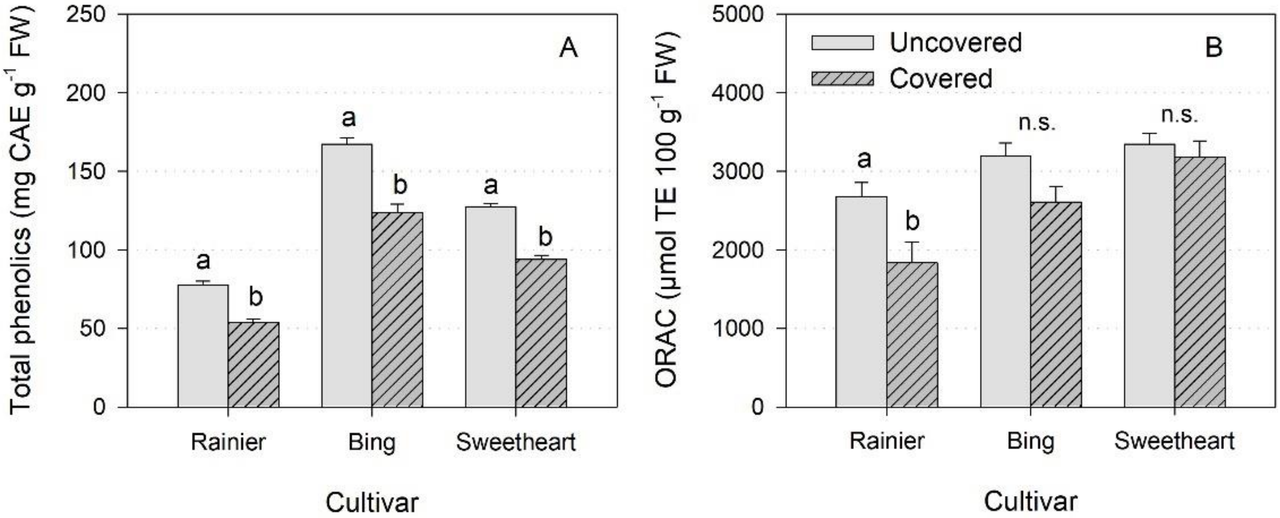

Figure 5. (**A**) Total phenol content and (**B**) ORAC antioxidant capacity of 'Rainier', 'Bing' and 'Sweetheart' sweet cherries with and without rain cover. Columns of a cultivar followed by the same letters indicate no statistical difference between treatments according to Tukey's test ($p \leq 0.05$). Means + SE (n = 5).

4. Discussion

4.1. Environmental Conditions

The rain cover affected the quality and quantity of incident solar radiation (Figure 1). In Chile, previous studies conducted on sweet cherry trees under cover showed PAR reductions of 40% [7]. High PAR levels between 1000 and 1100 µmol m^{-2} s^{-1} are suitable for the leaf in reaching its saturation point [5], which is not limiting under rain cover (Figure 1).

The stress index, related to daylight hours above 29 °C, was higher in the measurements at a 3 m height under rain cover since this section of the trees was exposed to a higher temperature and lower relative humidity for a longer period of time (Figures 2 and 3).

Previous studies showed similar relationships. Mika et al. [9], in evaluations conducted in sweet cherries 'Lapins' in Poland, reported that the mean daily temperature near the ground was lower under rain cover than outdoors, but at a 4.0 m height, the mean daily temperature was 0.4 °C higher. On the other hand, Wallberg and Sagredo [7], in Chilean evaluations also conducted in sweet cherries 'Lapins' at harvest, reported a mean temperature decrease of up to 2 °C under the rain cover, but with higher temperatures also in the near-cover zone. Børve et al. [24], in Norway, evaluated the microclimate generated by different types of plastic covers on different cultivars and training systems of sweet cherries and obtained similar results to those of the present study. These authors proposed a model of the permanent type, similar to the one tested, having temperature difference ranges of at least 1 °C with the control.

In sweet cherry trees under a high tunnel, an increase between 5 and 10 °C in maximum daily air temperature has been reported, given the lower ventilation characteristics of these structures [11], as well as an increase between 10% and 15% in RH, independent of height, reaching the dew point at night [25]. The higher thermal accumulation recorded under rain cover from bloom to harvest (Table 1) could favor an early harvest, an advance that was seen in sweet cherry trees under a high tunnel [26].

In addition to the canopy size and training system, pruning also has a significant effect on the distribution of solar radiation in the canopy [10].

4.2. Vegetative Growth

In 'Lapins' sweet cherry trees grown under rain cover, a greater shoot length and leaf size have been reported [7], while in 'Samba', 'Bellise' and 'Rita' sweet cherry trees with rain cover, Overbeck et al. [27] reported no change in the leaf area. On the other hand, the TCSA tended to be higher in trees under rain cover, it being significant only in 'Rainier', with a 15% increase relative to the control (Table 3). In contrast, Schäfer [28] reported that sweet cherry trees growing under rain cover had a smaller trunk diameter initially; however, over the years, this difference decreased.

Studies of sweet cherry trees under a high tunnel indicate a considerable increase in plant vigor in addition to a greater number and size of leaves, which could also favor greater trunk growth [25,26].

UV-B radiation has been suggested to be involved in the oxidation of cell size-inducing phytohormones, showing a reduction in leaf area in response to increased UV-B radiation [29]. Therefore, the lower UV-B incident radiation under the rain cover could lead to a lower degradation of indole acetic acid (IAA), a precursor of auxin synthesis. Therefore, the hormone would promote the growth of active or annual shoots, elongating internodes and the leaf area.

4.3. Plant Physiological Variables

At harvest, Sotiropoulos et al. [30] observed a slight tendency for lower transpiration in tree leaves under rain cover, suggesting that these would be more "comfortable" than those outdoors, while Zhang et al. [31] showed a lower net photosynthesis, transpiration and stomatal conductance but a higher light use efficiency (A_{net}/PAR) in 'Tieton' and 'Brooks' sweet cherry trees under rain cover.

The maximum photosystem II efficiency (Fv/Fm) obtained among cultivars was close to 0.8 (Table 4). Tartachnyk and Blanke [32] indicate that fruit trees with a ratio of Fv/Fm = 0.8 have an efficient photochemical energy conversion. Between treatments, only the second measurement date, after fruit harvest, showed differences, which was lower under rain cover only for 'Sweetheart', indicating differences among cultivars for acclimatization to light filtration (Table 4). Previous evaluations of sweet cherry trees under rain cover indicated an increase in the minimum blossom yield (Fo) without changes in Fv/Fm [31].

The plant water potential measured before harvest tended to have more negative values in uncovered trees, with significant differences only in 'Rainier', while after harvest, a similar trend was observed, with significant differences only in 'Bing' (Table 4). The differences could be attributed to a decrease in ambient temperature generated by rain cover, which would decrease the transpiration rate of covered trees [12]. Between measurement dates, the water potential values ranged from −0.6 to −1.0 MPa among cultivars, while the postharvest values showed an increase of 44%–133% (Table 4).

Gonçalves et al. [33] evaluated the water potential of sweet cherry trees with different rootstocks, 'MaxMa 14' being the one with the most negative values (−1.42 MPa), with no detrimental effects on plant growth. This was attributed to a deeper root system than those with more dwarfing rootstocks. Carrasco-Benavides et al. [34] reported that, at 100% water replenishment, the stem water potential at midday (Ψs) was −1.02 MPa, while a moderate water deficit in postharvest (Ψs > −1.5 MPa) did not negatively affect the fruit quality or productivity of sweet cherry trees. On the other hand, Shackel et al. [35] reported that, in sweet cherry trees, water potentials between −1.5 and −1.7 MPa result in shoot growth inhibition. In the present study, values exceeded this range, and as shown in Table 3, there was greater vegetative growth in the trees under rain cover.

The rate of CO_2 assimilation, transpiration and leaf stomatal conductance showed a generalized decrease between measurement dates in both treatments (Table 4), which could be explained by the absence of fruits on the last date [36]. An increase in water use efficiency was also observed (Table 4).

4.4. Leaf Pigments

In the present study, the results showed different tendencies among the cultivars, indicating differences in the acclimatization capacity against modifications in light transmission.

SPAD, chlorophyll and nitrogen balance indices are directly related to plant nutritional status [27,37]. Research with Dualex has shown that the chlorophyll index is a good indicator of nitrogen status in grapevine (*Vitis vinifera* L.) [38], showing a high correlation with the SPAD index [39]. A higher chlorophyll index value has coincided with greater fruit production in different cherry varieties, possibly due to the better nitrogen level and photosynthetic capacity of the trees [27].

The flavonoid index has shown a high correlation with the leaf phenol content [40]. In canopies with a high exposure to UV radiation, this index is higher, while in less illuminated canopies, it tends to decrease. Thus, variations in this indicator can reference changes in the synthesis of photoprotective compounds in leaves in the face of light alterations generated by rain cover [27].

4.5. Physicochemical Characteristics of the Fruit

The fruit size was affected by rain cover, showing in all three varieties a greater weight and diameter in fruits grown under cover. According to previous research, the effect of rain cover on fruit weight depends on cultivar. For example, in 'Kordia' and 'Regina', no effect has been found [41], while in 'Lapins', there has been an increase [42].

Soluble solids and firmness tended to be lower in fruit from covered trees. These attributes are of utmost relevance for cherry growers and exporters, as they are determinant in the fruit acceptance process in destination markets. Previous research indicates that plastic covers did not affect the sugar content [41,42] or fruit firmness [43]. However, if the temperature reaches very high levels during the ripening period, fruits tend to have lower firmness and soluble solids [5]. In high tunnel-grown sweet cherries, Schmitz-Eiberger and Blanke [44] detected that early-harvest cultivars presented the same SSC and firmness as the control, while late-harvest cultivars tended to present fruit with a lower sugar content (10–30%) and lower firmness.

Color was not affected in the 'Bing' and 'Sweetheart' varieties, which have red flesh. On the other hand, in 'Rainier', which has yellow flesh and bicolor skin, a significant reduction in color cover was observed, given the filtration of UV radiation generated by the rain cover. Multiple studies have observed a lower color intensity in fruits grown under plastic covers when measured with laboratory instruments. However, these differences are rarely detectable by consumers, especially in cultivars with dark red or dark purple flesh, since pigmentation occurs simultaneously between the flesh and the skin, which would attenuate the effect. [5,7,14] speculate that a low color would be attributed to a reduction in PAR due to the rain cover use. In bicolor varieties such as 'Rainier', the use of plastic covers has significantly hindered color development by filtering UV radiation [45].

Similarly, as with color, the concentration of anthocyanins in cherries skin was only found in the bicolor cultivar 'Rainier'. In sweet cherries, pigment synthesis induction is highly varied and largely cultivar-dependent [46]. In 'Lapins' sweet cherry trees, no significant differences in the anthocyanin content were found between the open air and under rain cover [9]. In 'Hedelfinger' and 'Kordia' sweet cherries, the concentration of cyanidin 3-rutinoside (the main anthocyanin in sweet cherries) tended to be lower under rain cover, while it tended to be higher in 'Regina', although without significant differences [8].

In the present study, the phenol content was significantly lower in fruits grown under cover in the three varieties analyzed. Additionally, the antioxidant capacity was lower in fruits under rain cover, it being significant only in the bicolor cultivar 'Rainier' (Figure 5), which is in agreement with what was observed by Correia et al. [47] and Yuri et al. [48], who reported that higher solar radiation and environmental stress favored the synthesis of these compounds. On the other hand, Schmitz-Eiberger and Blanke [44] showed higher anthocyanin and total phenol contents in fruits under a high tunnel, which were attributed to the heat stress and temperature fluctuation between the day and night; however, the antioxidant capacity only differed in one of the five cultivars studied.

5. Conclusions

The plastic rain cover caused an increase in air temperature and a decrease in relative humidity, especially in the upper canopy zone. These changes did not affect the CO_2 assimilation at harvest; however, the rain cover did affect vegetative growth in the trees, increasing vigor. Additionally, the tree water potential showed fewer negative values in trees under rain cover. The rain cover affected both the fruit quality and condition, with negative consequences regarding the firmness and soluble solids content of the three cultivars, as well as a significant reduction in the rain cover color in 'Rainier'. In addition, the fruits under rain cover had a lower total phenol content and lower antioxidant capacity. The results suggest that the environmental changes observed in the trees under rain cover mainly influenced the fruit quality at harvest, especially in the bicolor cultivar.

Author Contributions: Conceptualization, Á.S., J.S.-C. and J.A.Y.; methodology, Á.S., J.S.-C. and J.A.Y.; validation, Á.S., J.S.-C. and J.A.Y.; formal analysis, S.P. and M.P.; investigation, Á.S. and J.S.-C.; resources, J.A.Y.; data curation, S.P.; writing—original draft preparation, S.P.; writing—review and editing, M.P., Á.S., M.M. and J.A.Y.; visualization, S.P.; supervision, J.A.Y. All authors have read and agreed to the published version of the manuscript.

Funding: This research received no external funding.

Data Availability Statement: Not applicable.

Acknowledgments: The authors would like to acknowledge Agropecuaria Wapri S.A. for their technical support.

Conflicts of Interest: The authors declare no conflict of interest.

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