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EL USO DE CUBIERTAS PLÁSTICAS AL FINAL DE LA TEMPORADA RETRASA LA APARICIÓN DE ESTRÉS HÍDRICO SEVERO Y MEJORA LA EFICIENCIA DEL USO DEL AGUA EN LAS PLANTAS DE KIWI BAJO RIEGO DEFICITARIO

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LATE-SEASON PLASTIC CANOPY DELAYS THE OCCURRENCE OF SEVERE WATER STRESS AND IMPROVES WATER USE EFFICIENCY AND FRUIT QUALITY IN KIWIFRUIT VINES

RESUMEN

Las plantas de kiwi poseen una gran sensibilidad al estrés hídrico. Actualmente, muchas de las áreas productoras se han visto afectadas por la escasez hídrica. El cultivo protegido puede mitigar el estrés hídrico y efectos en la calidad de la fruta, sino también aumentar la eficiencia del uso del agua. Al comienzo de la maduración de la fruta, se evaluaron dos condiciones ambientales (con y sin cubierta de plástico) en plantas adultas sometidas a regímenes de riego deficitarios (RDI) y convencionales (WET) y en San Nicolás, Chile, durante dos temporadas. Las plantas cubiertas bajo RDI requerían el doble de tiempo para exhibir niveles severos de estrés hídrico (~-1,3 MPa) en comparación con las plantas sin cubiertas. El retraso en el inicio del estrés hídrico severo condujo al ahorro de agua y sin afectar el rendimiento ni la calidad de la fruta, lo que aumentó la productividad del agua entre 21% y 71%. Las diferencias en el estado del agua de la planta se asociaron con cambios en la radiación solar, en lugar de diferencias en el déficit de presión de vapor o el contenido de agua del suelo. Los frutos de plantas cubiertas, bajo riego deficitario exhibieron mayor firmeza a mayor madurez (> 7,0 Brix). El aumento de la productividad del agua en las vides severamente estresadas, bajo cubiertas, confirma que el cultivo protegido sería una buena herramienta para reducir el impacto del riego limitado en muchas áreas productoras afectadas por la escasez de agua.

SUMMARY

Kiwifruit is widely recognized as a fruit crop sensitive to water stress due to low stomatal regulation. Unfortunately, many of the most important kiwifruit producing areas have been affected by increasing water scarcity as a consequence of climate change. Protected cultivation may be used in kiwifruit vines not only to mitigate water stress and potential reductions in fruit quality, but also to increase water use efficiency. At the beginning of fruit maturation, two environmental conditions (uncovered and covered with a transparent plastic canopy) were assessed in mature kiwifruit plants (*Actinidia deliciosa* Chev. cv. Hayward) subjected to conventional and deficit irrigation regimes in San Nicolás, Chile, for two consecutive seasons. The results showed that covered plants under deficit irrigation required twice the time to exhibit severe levels of water stress (~-1.3 MPa) compared to plants under open-field conditions. The delay in severe water stress onset led to water savings and caused no reductions in both yield and fruit quality, which increased water productivity between 21% and 71%. Treatment differences in plant water status were associated with changes in solar radiation, rather than differences in vapor pressure deficit or soil water content. Fruits from covered plants subjected to deficit irrigation exhibited higher firmness at greater maturity (>7.0 Brix). The increase in water productivity in severely water-stressed kiwifruit vines, when using late-season plastic canopy cover, confirms that protected cultivation can be a good tool to reduce the impact of limited irrigation in many kiwifruit producing areas affected by water scarcity.

CAPÍTULO 1

INTRODUCCIÓN GENERAL

El kiwi (*Actinidia spp.*) es un cultivo frutal originario de los bosques del Sudeste de China (De la Fuente, 1988), lugar caracterizado por poseer un clima monzónico con promedios de precipitaciones y humedad relativa del aire de 1.183 mm año^{-1} y 80%, respectivamente. Los máximos valores de precipitaciones se presentan en primavera, la época de pleno desarrollo vegetativo del kiwi, pudiendo así mantener al cultivo en condiciones hídricas adecuadas durante toda la temporada. Debido a las condiciones climáticas de su centro de origen, esta especie ha desarrollado características morfológicas propias de especies vegetales adaptadas a una abundancia hídrica y baja disponibilidad de radiación solar, tales como son láminas foliares de gran tamaño (148 a 240 cm^2) (Miller *et al*., 2001), alta densidad estomática (Moncaleán *et al*., 2007, Infante *et al*., 1994), además de un deficiente control estomático en respuesta a condiciones de sequía (Judd *et al*., 1989).

Por lo tanto, los huertos de kiwi tienen grandes demandas de agua para mantener altos rendimientos en comparación con otros cultivos de frutas, lo cual ha llevado a los productores a regar en algunos casos con el 200% de la evapotranspiración de cultivo (ET_c). Por ejemplo, se ha estimado que las necesidades de riego de las plantas de kiwi conducidas en un sistema de pérgola son aproximadamente un 20% más altas (> 10.000 m^3 ha^{-1}) (Holzapfel *et al*., 2000) que las de las vides de mesa que usan el mismo sistema de conducción (± 8.000 y 9.000 m^3 ha^{-1}) (Villagra *et al*., 2014). Sin embargo, más de un 50% de las plantaciones de kiwi en el mundo se encuentran en zonas con clima Mediterráneo, en donde la temporada de crecimiento y desarrollo de las plantas ocurre en presencia de una elevada radiación solar (>1.500 mmol m^{-2}s^{-1}), altas temperaturas del aire (>30°C), y baja pluviometría (<500 mm), siendo estas últimas principalmente en la época de invierno y no en la temporada de desarrollo de las plantas de kiwi. Durante la última década, Chile y otros países productores de kiwi con condiciones climáticas mediterráneas han experimentado incrementos progresivos en la temperatura del aire y reducciones en la precipitación como consecuencia del cambio climático (Boisier *et al*., 2016).

Además, en esta época existe una competencia por el recurso hídrico con otras especies las cuales poseen mayor rentabilidad, lo cual hace que los productores prefieran destinar el suministro hídrico en busca de una mejor producción la siguiente temporada para estas especies en dezmero de las plantas de kiwi. Por lo tanto, la ocurrencia de estrés hídrico severo durante la temporada de crecimiento se ha vuelto inevitable en muchas áreas productoras de fruta, lo cual es particularmente grave para los kiwis, ya que este cultivo es muy sensible al estrés hídrico debido a la baja regulación estomática (Zhang *et al*., 2018, Judd *et al*., 1989) y alta vulnerabilidad a la cavitación a niveles relativamente bajos de estrés hídrico (Clearwater y Clark, 2003).

El uso de distintos tipos de cubiertas protectoras sobre el dosel ha ganado popularidad en muchas áreas productoras de fruta fresca, ya que esta práctica puede minimizar el impacto de las adversidades climáticas sobre la calidad y el rendimiento de la fruta (Olivares-Soto *et al*., 2020, Sotiropoulos *et al*., 2014, Rana *et al*., 2004). Aunque pocos estudios han investigado el papel del cultivo protegido en las relaciones agua-planta de los cultivos frutales, existe evidencia de que su uso puede disminuir la evapotranspiración de las plantas (Albright *et al*., 1989) y aumentar la eficiencia del uso del agua en los cultivos frutales (Da Silva *et al*., 2018). En Citrus, Cohen *et al*. (1997) informaron que el uso de densas redes de sombra bajo clima semiárido redujo las tasas de transpiración en aproximadamente un 25%. En los cultivares de uva de mesa, cubrir los viñedos con una película de plástico en el envero aumentó el potencial hídrico del tallo (Novello y de Palma, 2008). Montanaro *et al.,* (2009) informaron que una disminución del 50% en la radiación solar en las plantas de kiwi con estrés hídrico resultó en un menor nivel de estrés hídrico (~ 10%) que las plantas descubiertas una semana después de que se aplicó el riego y se utilizó una malla de sombra.

Sin embargo, no existe una comprensión integral sobre el papel del cultivo protegido como estrategia de mitigación de ahorro de agua en los kiwis. Se espera que el cultivo protegido no solo disminuya las demandas de agua atmosférica, tal como ha sido reportado anteriormente en la literatura, sino que también afecte la fisiología y el crecimiento y desarrollo de las plantas debido a los cambios en la calidad y cantidad de la radiación solar y en la temperatura del aire. Por consiguiente, en este estudio, el objetivo general es investigar el efecto combinado del déficit hídrico y la cobertura del dosel con una película plástica sobre las variables ambientales vinculadas a las relaciones hídricas de la planta que determinan respuestas fisiológicas frente al estrés hídrico, el rendimiento del huerto y la calidad del fruto.

HIPOTESIS

El uso de cubiertas plásticas sobre el dosel de plantas de kiwi, en condiciones de déficit hídrico, retrasa la aparición de estrés hídrico severo, mejora la eficiencia del uso del agua y mejora la calidad de la fruta.

OBJETIVO GENERAL

Evaluar el efecto del uso de cubiertas plásticas en el dosel de plantas de kiwi, en condiciones de déficit hídrico, sobre la aparición de estrés hídrico severo, eficiencia del uso del agua y calidad de la fruta.

OBJETIVOS ESPECÍFICOS

- Evaluar el efecto del uso de las cubiertas plásticas sobre el estado hídrico de plantas de kiwi bajo riego convencional y riego deficitario controlado
- Evaluar el efecto del uso de cubiertas plásticas en plantas de kiwi bajo riego convencional y riego deficitario controlado
- Evaluar el efecto del uso de cubiertas plásticas sobre la calidad de la fruta, de plantas de kiwi bajo riego convencional y riego deficitario controlado

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CAPÍTULO 2

Late-season plastic canopy delays the occurrence of severe water stress and improves water use efficiency and fruit quality in kiwifruit vines

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Keywords: Protected cultivation, deficit irrigation, *Actinidia deliciosa*, water productivity, leaf water potential.

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Abstract

Kiwifruit is widely recognized as a fruit crop sensitive to water stress due to low stomatal regulation. Unfortunately, many of the most important kiwifruit producing areas have been affected by increasing water scarcity as a consequence of climate change. Protected cultivation may be used in kiwifruit vines not only to mitigate water stress and potential reductions in fruit quality, but also to increase water use efficiency. At the beginning of fruit maturation, two environmental conditions (uncovered and covered with a transparent plastic canopy) were assessed in mature kiwifruit plants (*Actinidia deliciosa* Chev. cv. Hayward) subjected to conventional and deficit irrigation regimes in San Nicolás, Chile, for two consecutive seasons. The results showed that covered plants under deficit irrigation required twice the time to exhibit severe levels of water stress (~-1.3 MPa) compared to plants under open-field conditions. The delay in severe water stress onset led to water savings and caused no reductions in both yield and fruit quality, which increased water productivity between 21% and 71%. Treatment differences in plant water status were associated with changes in solar radiation, rather than differences in vapor pressure deficit or soil water content. Fruits from covered plants subjected to deficit irrigation exhibited higher firmness at greater maturity (>7.0 Brix). The increase in water productivity in severely water-stressed kiwifruit vines, when using late-season plastic canopy cover, confirms that protected cultivation can be a good tool to reduce the impact of limited irrigation in many kiwifruit producing areas affected by water scarcity.

Keywords: Protected cultivation, deficit irrigation, *Actinidia deliciosa*, water productivity, leaf water potential.

1. Introduction

Kiwifruit (*Actinidia* spp.) is a fruit crop originating in southern China (De la Fuente, 1988), where wet and warm conditions due to the East Asian Monsoon prevail during the growing season. These conditions have determined morphological characteristics that are typical of plants adapted to abundant water supply, such as high values of leaf area (Miller et al., 2001) and stomatal conductance (Moncaleán *et al*., 2007, Infante et al., 1994). Therefore, kiwifruit orchards have large water demands to maintain high yields when compared to other fruit crops. For instance, irrigation requirements of kiwifruit vines trained on a pergola trellis have been estimated to be about 20% higher (>10.000 m^3 ha^{-1}) (Holzapfel et al., 2000) than those of table grapevines using the same trellis system (± 8,000 and 9,000 m^3 ha^{-1}) (Villagra *et al*., 2014).

During the last decade, Chile and other kiwifruit producing countries with Mediterranean-climate conditions, have been experiencing progressive increases in air temperature and reductions in precipitation as a consequence of climate change (Boisier et al., 2016). Thus, severe water stress during the growing season has become inevitable in many fruit producing areas, which is particularly serious for kiwifruit, as this crop is very sensitive to water stress due to low stomatal regulation (Zhang et al., 2018, Judd *et al*., 1989) and high vulnerability to cavitation at relatively low levels of water stress (Clearwater and Clark, 2003).

The use of canopy cover as a protection has gained popularity in many fresh fruit producing areas because this practice can minimize the impact of climatic adversities on fruit quality and yield (Olivares-Soto et al., 2020, Sotiropoulos et al., 2014, Rana et al., 2004). Although few studies have investigated the role of protected cultivation in plant water relations of fruit crops, there is evidence that its use can decrease evapotranspiration of plants (Albright et al., 1989) and increase water use efficiency in fruit crops (Da Silva et al., 2018). In Citrus, Cohen et al. (1997) reported that the use of dense shade netting under semi-arid climate reduced transpiration rates by approximately 25%. In table grapes cultivars, covering vineyards with a plastic film at *veraison* slightly increased stem water potential (Novello and de Palma, 2008). Montanaro et al. (2009) reported that a 50% decrease in solar radiation in water-stressed kiwifruit plants resulted in a lower level of water stress (~10%) than uncovered plants one week after irrigation was applied and shade netting was used.

However, there is not a comprehensive understanding regarding the role of protected cultivation as a water saving mitigation strategy in kiwifruit. We expect that protected cultivation will not only decrease atmospheric water demands, as previously reported in the literature, but it will also impact photochemistry and fruit growth and development due to changes in quality and quantity of above canopy radiation and temperature. Therefore, in this study we aim to investigate the combined effect of water deficit and canopy covers on environmental variables linked to soil-plant-atmosphere water relations, as well as physiological responses, fruit yield and quality.

2. Materials and methods

2.1. Description of the study site and weather data

This study was conducted in a commercial kiwifruit orchard (*Actinidia deliciosa* (A. Chev.) C.F. Liang and A.R. Ferguson) cv. Hayward, in San Nicolás (36°32'45.8"S 72°11'18.4"W), Ñuble region, Chile, for two consecutive seasons (2018-2019). The kiwifruit vines were planted in 2006, at 4.0 m x 3.0 m spacing, trained as an overhead pergola system, with four 3.0 m cordons and fruiting canes placed every 0.2 m. Rows were laid out in a north-south direction. The orchard has had an average yield between 35 and 40-ton ha^{-1} during the last five years. Irrigation was applied by micro-sprinklers, using two emitters spaced 0.75 m apart at a flow rate of 30 L h^{-1} per plant. The soil belongs to the Talquipén series, which is classified as a member of the fine-loamy, mixed, mesic family of Ultic Haploxerolls, and is characterized by low porosity, fine textures, and limited internal drainage (CIREN, 1990). Textures in the sites range from loam in the surface to clay loam with depth. Lateral flower buds were thinned manually before bloom to increase fruit size, while fruit thinning was carried out after bloom to remove deformed fruits. In 2019, a severe fruit thinning was applied approximately two weeks after fruit set in order to reduce crop load by 30% and increase fruit size. Forchlorfenuron (CPPU), a synthetic cytokinin, was applied twice during the season (30 and 40 DAF) at a dose of 300 cc hl^{-1} to increase fruit size. Pest and weed management was conducted according to common commercial practices for kiwifruit orchards.

2.2. Experimental design.

The experiment was carried out in a completely randomized block design with four replicates, and repeated measurements over a period of two years. The treatments were arranged in a split plot design, where the main plot was the cover treatment and the sub plot was the irrigation treatment. Two cover treatments (covered and uncovered plants) were applied from the end of stage I of fruit development, corresponding to approximately 80% of final fruit size, to senescence. The uncovered plants remained under open-field conditions throughout the whole season, while a plastic cover was installed above the canopy throughout the above-mentioned period. The cover was installed on January 11th and January 17th in 2018 and 2019, respectively, over a plot of thirty plants, using a whitish low-density polyethylene film of 180 µm thickness (Oroplus HALS 180, Plastik Advanced SRL, Bergamo, Italy). The plastic cover was installed 0.70 m above each of the 3 consecutive rows for each block-treatment combination (10 pl row^{-1}).

Two irrigation treatments were randomly applied within each main plot one week after the plastic cover was installed, aiming to obtain significant differences in plant water status near the end of the growing season when water availability often reaches the minimum value of the growing season. Control (WET) plants were irrigated to satisfy at least 100% ETc throughout the whole season, while in the regulated deficit irrigation (RDI) treatment, water was cut off until plants reached an average midday leaf water potential (Ψ_L) of -1.3 MPa. After harvest, commercial irrigation practices were resumed throughout the entirety of the orchard. Irrigation requirements were estimated based on the evapotranspiration for kiwifruit ($ETc = ETo \times k_c$). The crop coefficients (k_c) for kiwifruit used in this study from budbreak to harvest were obtained from FAO 56 (Allen et al., 1998).

2.3. Environmental conditions

Rainfall and reference evapotranspiration were obtained from the San Nicolás weather station (Agroclima Weather Network) (36°31'4.3"S 72°5'40.9"W), which is located about 10 km from the study site. Temperature and air relative humidity above the plant canopy were recorded in two randomly selected blocks in covered and uncovered plants under WET conditions. An air temperature and humidity probe

(HMP60, Vaisala, Helsinki, Finland) was installed 0.5 m above the plant canopy, with a sampling frequency of 5 minutes. These data were used to estimate vapor pressure deficit.

Spectral radiation measurements were carried out at solar noon using a dual spectrophotometry system (StellarNet INC., Tampa FL, USA), which was configured to measure total irradiance in the wavelength range of the ultraviolet (323-400nm), visible (400-700nm), red (700-800nm) and infra-red (950-1690nm) spectrums. Measurements were made below and above the canopy of covered and uncovered vines, taking three samples per main plot. The ratios of blue light (400-500 nm)/red (600-700 nm) and red light (600-700 nm)/distant red (700-800 nm) were calculated in order to estimate the activity of phytochromes and cryptochromes (Bastías and Corelli-Grappadelli, 2012).

Volumetric soil water content was measured in a single block in the deficit-irrigated plants that were under plastic cover and open canopy conditions. Capacitance sensors (GS1, Decagon Devices, Pullman, WA, USA) were installed between the emitters on the row, at two depths (-0.2 and -0.4 m) and a distance of 0.3 m from the trunk. Data were recorded and stored every 15 minutes in a datalogger (Em5b, Decagon Devices, Pullman, WA, USA).

Photosynthetic photon flux density (PPFD) was measured above and below the plant canopy were weekly performed with a ceptometer (LP-80, Decagon Instruments, Washington, USA). Data collection was carried out at noon, with a sampling frequency of one and three measurements above and below the plant canopy, respectively. Above-canopy PPFD was measured 50 cm above the upper canopy, while below-canopy PAR was measured at the center of the row, right below the plant arm, edge of the ridge (0.5 m from the trunk) and at the center of the inter-row space (2.0 m from the trunk) 1.0 m above the soil surface. Leaf and fruit temperatures were measured with a portable infrared thermometer (Fluke 62 Max, Fluke Corporation, Everett, WA, USA) on the same plants where plant water status was measured. A sample of 5 leaves and 5 fruits randomly selected per plant were measured by distancing the infrared thermometer at 5.0 cm from the sample, which is expected to accurately represent the surface temperature of an area of 10 cm^2.

2.4. Plant water status and growth

Plant water stress levels were determined weekly by measuring midday leaf water potential (Ψ_L) on two leaves per sampled plant in each subplot. Measurements were taken between 12:00 and 15:30 h. using a pressure chamber (PMS 615; PMS Instruments, Washington, USA). Leaf samples for this measurement were obtained from the upper third of the canopy, preferably selecting mature leaves with no visual symptoms of biotic or abiotic stress, according to the methodology described by McCutchan and Shackel (1992). The number of samples used to determine Ψ_L was established based on a statistical power analysis at the beginning of the first season (data not shown). Stomatal conductance and photosystem II efficiency were measured with a steady-state porometer (SC-1, Decagon Instruments, Washington, USA) and a portable fluorescence meter (Pocket PEA, Hansatech Instruments, Norfolk, UK), respectively. Both measurements were conducted at midday, employing the same selection criteria used for Ψ_L determinations. To determine photosystem II efficiency, leaves were adapted to darkness for 30 minutes before the original fluorescence (Fo) and maximum fluorescence (Fm) measurements (Liang et al., 2019). Photosystem II efficiency (*Fv/Fm*) was calculated with the following formula:

$$\frac{Fv}{Fm} = \frac{(Fm - Fo)}{Fm}$$

For shoot growth measurements, 6 shoots per plant were selected at random and marked; shoot length and number of leaves were recorded on a weekly basis starting from fruit set. During the second season, defoliation at harvest (%) was evaluated. For this, 6 fruiting canes per plant were marked at the beginning of the season and their leaves were counted on a weekly basis. Defoliation percentage (DP) was obtained using the following formula:

$$DP = 100\frac{NLF}{NLH}$$

where, NLF is the number of leaves at fruit set and NLH is the number of leaves at harvest.

2.5. *Water productivity and intrinsic water use efficiency*

At harvest, a 10-fruit sample was randomly taken from the upper canopy of two vines from each experimental unit to estimate intrinsic water use efficiency, measured as carbon isotope discrimination ($\Delta^{13}C$), as proposed by Bchir et al., (2016). Fruit samples were dried in an oven at 70°C until there was no reduction in sample weight. Dried samples were ground and sieved to obtain a homogeneous fine powder. The stable carbon isotope composition ($\delta^{13}C$) for each sample was determined with the MAT253 mass analyzer (Finningan Mat Copr. USA), using the following equation (Farquhar et al., 1989):

$$\delta^{13}C\ (‰) = \left[\frac{\left(\frac{^{13}C}{^{12}C}\right)_{sample} - \left(\frac{^{13}C}{^{12}C}\right)_{standard}}{\left(\frac{^{13}C}{^{12}C}\right)_{standard}}\right] x\ 1000$$

where $^{13}C/^{12}C$ sample and $^{13}C/^{12}C$ standard are the measured $^{13}C/^{12}C$ ratios for the leaf sample and the PDB standard (Pee Dee Belmnite), respectively. Carbon isotope discrimination was calculated when the error of previous analyses was ≤ 0.2 ‰:

$$\Delta\ ^{13}C\,(‰) = \left[\frac{\delta_{air} - \delta^{13}C_{leaf}}{(1 + \delta^{13}C_{leaf})}\right] x\ 1000$$

where δ_{air} is the value of CO^2 in the air (-8%) (Brugnoly and Farqhuar, 2000) and $\delta^{13}C_{leaf}$ is the stable carbon isotope of the plant.

Water productivity was estimated as the product of the ratio between water applied (irrigation + precipitation) from budbreak to harvest ($m^3\ ha^{-1}$) and yield per hectare (kg ha^{-1}).

2.6. *Fruit maturity, quality, and yield estimates*

Soluble solids concentration and pulp firmness were measured once a week throughout the last stage III of fruit development to determine an appropriate harvest date. Fruit samples consisted of 15 fruits per plant, following the “poke and grab” sampling method (Rankine et al., 1962). Soluble solids concentration and pulp firmness were measured using an optical refractometer (Atago Hand Refractometer, Atago, Tokyo, Japan) and a digital penetrometer with an 8 mm plunger (FM200, PCE Instruments, Southampton, UK), respectively. Pulp firmness was measured at the midpoint of the longest fruit side, on opposite fruit sides, before manually removing a skin section. Once harvest maturity was achieved, 45 fruits per plant were sampled and divided into 2 subsamples. The first subsample consisted of 30 fruits and was used to determine fruit size, pulp firmness, and soluble solid concentration. The second subsample of 15 fruits was used to determine the percentage of dry matter (DM). For this, a pulp slice of 2 to 3 mm thick was extracted manually from each fruit in its equatorial zone, and its fresh and dry weight (after drying the sample for 8 hours at 60°C) was recorded with an analytical balance (± 0.1 g) (UX6200H, Shimadzu, Japan). The DM value was calculated as the ratio between the dry and fresh weight of each fruit slice. The percentage of fruit affected by sunburn was estimated in six randomly selected fruiting canes per plant. All the fruits of each fruiting cane were collected at harvest and the presence of sunburn was visually evaluated, following the commercial standards of the Chilean kiwifruit industry (Farias, 2010). In this study, the area affected by sunburn was used to classify fruits in the following four categories: 1=no damage (no visual symptom of sunburn); 2=slight damage (sunburn area is lower than 2 cm^2); 3= moderate damage (sunburn area is higher than 2 cm^2); and 4=severe damage (necrotic tissue area is higher than 2 cm^2).

2.7. Statistical analysis

The data were subjected to an analysis of variance (ANOVA), after testing for normality distribution (Shapiro-Wilk), homogeneity of error variances (Levene's test), and additivity (Tukey). Differences between means were determined using the LSD test (alpha = 0.05). All statistical analyses were performed using the statistical software SAS 9.4 (SAS Studio, University Edition, SAS Institute, NC, USA).

3. Results

3.1. Environmental conditions

Very similar VPD and PPFD were observed until the plastic cover was installed in early January (Figures 1.A and 1.B). Once the plastic cover was installed, there was a slight increase in VPD (approximately 1%) and a considerable decrease in PPFD values (approximately 30%). In the first season, the VPD remained at values close to 1.5 kPa between November and January. In the second season, VPD showed a progressive increase that ranged between 1.4 and 1.8 kPa for the same period. The maximum VPD values were found in February for both seasons, but there was a minor increase in VPD values in 2019 due to cooler conditions. The PPFD values remained close to 1,500 umol m^{-2} s^{-1} between November and January in both seasons. Thereafter, a decrease in PPFD values was observed in both cover treatments, ranging between 900 and 1,000 umol m^{-2} s^{-1}, and between 1,200 and 1,500 umol m^{-2} s^{-1} for the open canopy treatment. The use of the plastic cover reduced the total solar energy (323 - 1690 nm) on average by 32% (Figure 2). However, a greater reduction was observed for wavelengths at the longer spectrum range. Transmitted radiation through the plastic cover in the UV (320 - 400 nm), the blue (400 - 500 nm), the red (600 - 700 nm), and the infrared light spectrums were 92%, 50%, 25%, and 15%, respectively, of that measured in uncovered vines.

Irrigation for the WET plants was around 9,800 m^3 ha^{-1} in the first season, while irrigation for the same treatment was reduced by approximately 30% during the second season (Table 1). Cumulative ETc in the first season was approximately 4% higher than in the second season, reaching values that were close to 8,000 m^3 ha^{-1}. Regardless of the cover treatment, the amount of applied water was lower in the plants under RDI. However, under RDI conditions, covered plants received less irrigation water than those uncovered (26% and 12%, respectively). Rainfall was higher in the first season and averaged 2,697 m^3 ha^{-1}. The accumulated rainfall in covered plants was calculated up until the date in which the plastic cover was opened, reaching 1,530 and 1,773 m^3 ha^{-1} for the first and second season, respectively. In the second season, irrigation water for the WET treatment decreased by approximately 25%. In general, there was no clear impact of net cover or deficit irrigation on water productivity, yet covered plants under RDI recorded the highest values in both seasons (6.53 and 4.19 kg m^{-3} in the first season and second season, respectively).

3.2. Physiological responses

Despite the cover treatment, Ψ_L ranged between -0.8 and -0.6 MPa for the WET plants throughout the season in 2018, whereas there was a wider Ψ_L range, between -1.0 to -0.6 MPa for the WET plants in 2019 (Figures 3.A, 3.B, 3.C, and 3.D). Covered plants under RDI required six weeks to reach about -1.3 MPa (Figures 3.B and 3.D), while uncovered plants, under the same irrigation treatment, required four weeks to reach the same water stress level (Figures 3.A and 3.C). In the first season, uncovered plants under RDI exhibited two periods of water stress (Figure 3.A), while, in the second season, only one period of water stress was observed (Figure 3.C). The decrease in the average value of the volumetric soil water content was not different between cover treatments once irrigation was cutoff in RDI plants in January (Figures 4.A and 4.B). Regardless of the cover treatment, soils reached similar minimum values of volumetric water content, ~0.27 m^3 m^{-3} in both seasons. The largest treatment differences in soil water content was observed when irrigation was resumed in uncovered plants, which exhibited a raise between 8 and 10 m^3 m^{-3}.

Stomatal conductance, leaf and fruit temperature, and the percentages of transmitted PPFD and defoliation were similar for both cover treatments in 2018 and 2019 (Table 2). Regarding irrigation treatments, no significant differences were found in stomatal conductance, leaf temperature and transmitted PPFD. In 2019, fruit temperature and the percentage of defoliation were higher by 0.7 °C and 24.1%, respectively, in RDI plants (Table 3). Higher *Fv/Fm* values were generally observed in covered compared to uncovered plants, but significant differences were only detected in two out of seven measurements (Figure 5.A). On the other hand, no differences were found in Fv/Fm between irrigation treatments (Figure 5.B). While the ^{13}C discrimination analysis showed less negative discrimination values in covered plants in 2019 (Figure 6.A), similar ^{13}C discrimination values were found between irrigation treatments (Figure 6.B).

3.3. Yield estimates and fruit quality

The analysis of the yield estimates showed no differences between cover and irrigation treatments in both seasons (Table 4). The number of fruits per plant ranged between 497 and 573, and between 274 and 437 for the first and second season, respectively. The average yield during the first season was 481-ton ha^{-1}, while in the second season was 29.3 ton ha^{-1}. Fruit number, fruit weight, and yield showed a reduction of 56%, 10%, and 64%, respectively, during the second season.

The use of the plastic cover did not result in significant or consistent differences for the majority of the evaluated fruit quality parameters at harvest, such as Brix, pulp firmness, and polar and equatorial fruit diameters (Table 5). In 2019, the percentage of dry matter was slightly higher (0.5%) in covered plants. The percentage of moderate sunburn damage was the only consistent effect of cover on fruit quality, as covered plants showed less fruit damaged by sunburn than uncovered plants, 14% and 23% in 2018 and 2019, respectively.

Similarly, irrigation showed almost no impact on fruit quality, as similar values of Brix, firmness, and polar diameter were found in both irrigation treatments (Table 6). The fruits of plants under RDI exhibited about 1.0% more dry matter content than WET plants during the first season, but this was not observed during the second season as values were similar in both treatments. The only consistent effects of irrigation were found on the equatorial diameter and the percentage of fruit with slight levels of sunburn damage. The equatorial fruit diameter was 5% higher in WET than RDI treatments. The difference in the percentage of mild sunburn damage was 14% and 24% between irrigation treatments for the first and second seasons, respectively, where RDI reached a maximum value of 58% of damaged fruit by sunburn in 2019. Despite the lack of effects of cover and irrigation treatments on pulp firmness at harvest, the relationship between Brix and firmness was not the same for each cover-irrigation combination (Figure 7). The decrease in fruit firmness associated with an increase in soluble solids concentration was similar in all the treatments up to 5.5 Brix. Above this value, fruits from covered plants subjected to RDI maintained their firmness, while the other treatments continued the decrease in fruit firmness.

4. Discussion

The results of this study provide evidence to sustain that protected cultivation, as a water conservation strategy, is effective. The use of late-season plastic cover modified key environmental variables affecting the atmospheric water demand.

Nevertheless, covered plants had a different water status response, in magnitude and consistency, to both irrigation treatments. For instance, under WET conditions, Ψ_L from covered and uncovered plants varied from mild water stress to optimum water status (-0.8 to -0.5 MPa) (Judd et al.,1989) even in months when the orchard reached the highest evaporative demand of the season (January and February). Low sensitivity of Ψ_L of well-irrigated vines to larger atmospheric water demand is consistent with high whole-plant hydraulic conductance of kiwifruit at optimum water conditions (Dichio et al., 2012). Conversely, under RDI conditions, covered plants went for a period of time twice as long as uncovered plants to reach severe levels of water stress (<-1.2 MPa) (35 v/s 14 days, respectively). Despite the faster decrease in Ψ_L of uncovered plants under RDI, both covered and uncovered kiwifruit vines exhibited similar reductions in soil water content as soon as irrigation was cut off, reaching the minimum value of soil water content of 0.3 m^3 m^{-3} in three weeks (Figure 4). Thus, it seems unlikely that the observed effect of cover on Ψ_L at different irrigation treatments is linked to water availability. Other factors, such as the high vulnerability of kiwifruit vines to cavitation at relatively low water stress and the absence of an efficient embolism repair mechanism (Clearwater and Clark, 2003) may have accelerated leaf dehydration and Ψ_L reduction in uncovered deficit-irrigated plants. Besides the abovementioned explanation, PPFD reductions in the cover treatment may have also reduced the vulnerability of leaves to cavitation, and hence, delayed the drop in Ψ_L (Guyot et al., 2012). These results confirm the high susceptibility of *Actinidia* plants to water stress-related hydraulic impairments under Mediterranean environmental conditions, as proposed by Dichio et al., (2013). Consequently, we suggest that the use of plastic covers to manage water stress may not only decrease atmospheric water demand, but also moderate the vulnerability of leaf hydraulic conductivity thanks to reduced PPFD.

The plastic cover had no impact on Red/Far Red and Blue/Red light ratios, which may explain why stomatal conductance, as well as other phytochrome and cryptochrome mediated-responses, such as the rates of leaf appearance and shoot elongation (Bastías and Corelli-Grapadelli, 2012) were similar for both cover treatments. However, the efficiency of photosystem II, estimated as *Fv/Fm*, tended to be higher in covered plants (Figure 5), probably reflecting the effect of a 44% reduction in UV transmission (320–400 nm) on the photoinhibition of leaves (Verdaguer et al., 2017; Pfündel, 2003). The analysis of ^{13}C discrimination indicated that the intrinsic water use efficiency was slightly higher in covered plants, regardless of the irrigation strategy, as ^{13}C discrimination was 0.7‰ less negative in mature fruits from covered plants. Since, in the present study, stomatal conductance was not affected by cover treatments, the slight increase in the intrinsic water use efficiency of covered plants could be linked to higher *Fv/Fm*. Conversely, no significant increase in photosystem II efficiency and intrinsic water use efficiency was observed at the RDI treatment, as fruits from plants under both irrigation treatments showed the same *Fv/Fm* and ^{13}C discrimination values at harvest, respectively. The low sensitivity of stomata to RDI is most likely linked to the lack of irrigation effects on intrinsic water use efficiency, as isotope discrimination has been found to be reduced when stomata close in response to water stress (Farquhar et al., 1989).

Cover and irrigation treatments showed no effect on the number of fruits per plant, despite both treatments induced microclimatic changes at least two weeks before the flower bud induction process was completed, which occurred when fruits reached 90% of the final size (1st week of February) (Pratt and Reid, 1974). The 40% reduction in yield observed during the second season was caused by the application of severe fruit thinning a couple of weeks after fruit set. Although the objective of that cultural practice was to increase fruit size, there was no improvement in fresh fruit weight in 2019. This confirms that, in kiwifruit, fruit thinning after fruit set has a limited impact on fruit size (Xiloyannis et al., 1997). Water supply reduction for several weeks had no impact on yield, confirming the low sensitivity of reproductive growth of fruit lianas to late water deficit, as reported by Calderón-Orellana et al., (2019) for table grapes. Therefore, our experiment showed that RDI improved water productivity, which was between 21% and 71% higher than that found under conventional crop management (i.e., Uncovered WET plants). Despite the expected high seasonal variability in water productivity, an improvement of this parameter is key for kiwifruit production, as this widely recognized high-water demanding fruit crop is currently grown in areas severely affected by water scarcity.

The use of the plastic cover reduced the transmission of near infra-red radiation (750 - 1100 nm) from the canopy surface by 33%, which explained the increase in air temperature above the plant canopy of covered vines (<2 °C). However, the absence of differences in canopy and fruit temperatures between covered and uncovered plants may explain why maturity parameters that have been reported to be sensitive to air temperature in kiwifruit, such as the concentration of soluble solids and the percentage of dry matter (Bouldon et al., 2007) were similar for both cover treatments. About a 20% reduction in fruit moderate sunburn was observed in covered plants, which is linked to a 32% reduction in incoming solar radiation. A reduction in sunburn incidence is of paramount importance in kiwifruit, as sunburn not only reduces the cosmetic fruit value, but also has been associated with greater colonization rates of *Alternaria* spp. (Sommer et al., 1983). Although covered plants showed no impact on fruit firmness at harvest, in the second season, the relationship between the concentration of soluble solids and firmness was not the same for each cover-irrigation treatment combination. While covered plants under RDI maintained high values of fruit firmness (19 lb) at high Brix (7 Brix), the remaining treatments showed a linear decrease in firmness as Brix increased above 6.0. The fact that only covered deficit-irrigated vines had less softening at higher maturity illustrates that the role of water stress on fruit firmness is still unclear, regardless of previous findings reporting higher firmness in fruits from deficit-irrigated kiwifruit plants during cold storage (Currie et al., 2008). Furthermore, these results suggest that the combined use of protected cultivation and RDI may allow harvesting at higher maturity with no reduction in firmness.

Similar to the results observed for the cover treatments, irrigation showed no consistent effects on fruit maturity, which disagrees with several studies that found higher Brix and dry mater in deficit-irrigated kiwifruit vines (Currie et al., 2008). However, plants under deficit irrigation had smaller fruit equatorial diameter in both seasons. This was probably associated to severe water stress during stage III of fruit development when the rate of lateral fruit growth was the highest of the season. Water stress-induced reduction in fruit equatorial diameter may be beneficial for growers, since long fruits are more valuable than round fruits in the kiwifruit market. Nevertheless, the greater percentage of defoliation observed in 2019 and the higher number of fruits damaged by sunburn in deficit-irrigated vines may be a clear indicator that levels of water stress reached in this study were too severe for kiwifruit.

5. Conclusions

Water scarcity due to climate trends and projections in many fruit-producing areas with Mediterranean climate may induce considerable losses in yield and quality of kiwifruit. This study shows evidence that the late installation of a plastic cover delays the occurrence of severe water stress in a mature kiwifruit orchard. The lack of agreement on the patterns of plant water status and soil water content between cover treatments of deficit-irrigated vines suggests that the use of protected cultivation in kiwifruit decreased not only atmospheric water demand, but also the vulnerability of leaves to cavitation. Although regulated deficit irrigation seems to have improved water productivity, the higher number of fruits damaged by sunburn in deficit-irrigated plants suggests that a water stress severity of -1.3 MPa may be harmful to kiwifruit. The combined use of protected cultivation and deficit irrigation seems promising since fruit softening at higher maturity was reduced while water productivity was increased.

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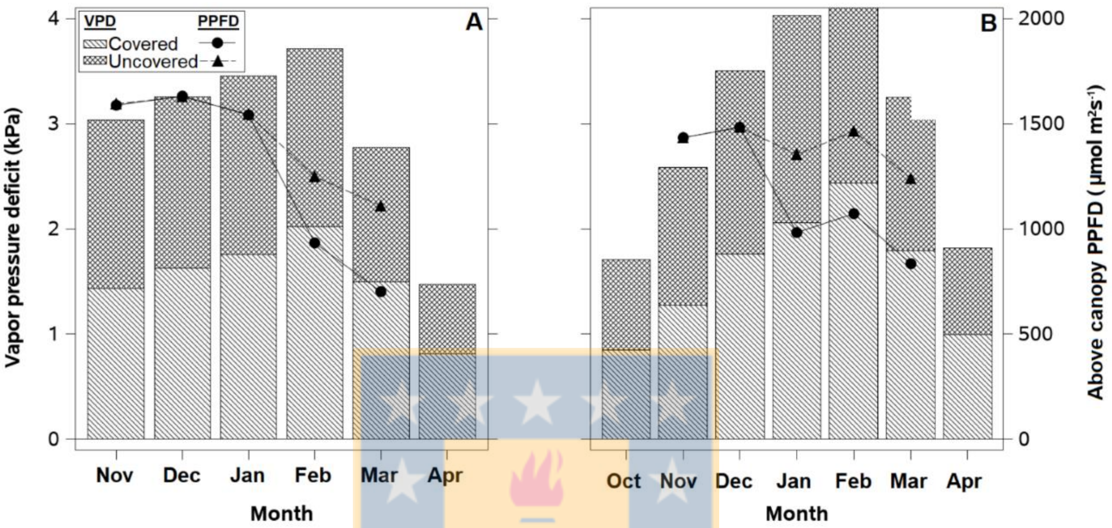
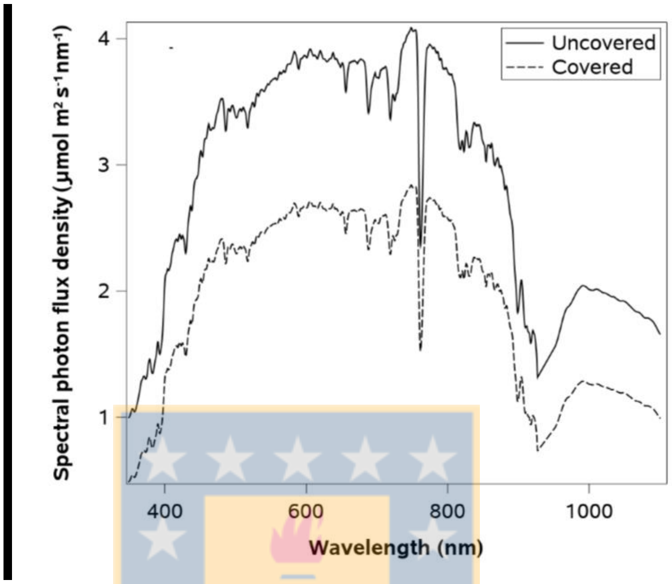
Figure 1. Monthly average of vapor pressure deficit and above canopy photosynthetic photon flux density (PPFD) measured at 0.5 m above the plant canopy of Hayward kiwifruit vines on an overhead “pergola” trellis system in San Nicolás, Ñuble, Chile, subjected to two cover treatments (Covered: low-density polyethylene canopy and Uncovered: open field conditions) from anthesis to harvest (November to April) in (A) 2018 and (B) 2019.



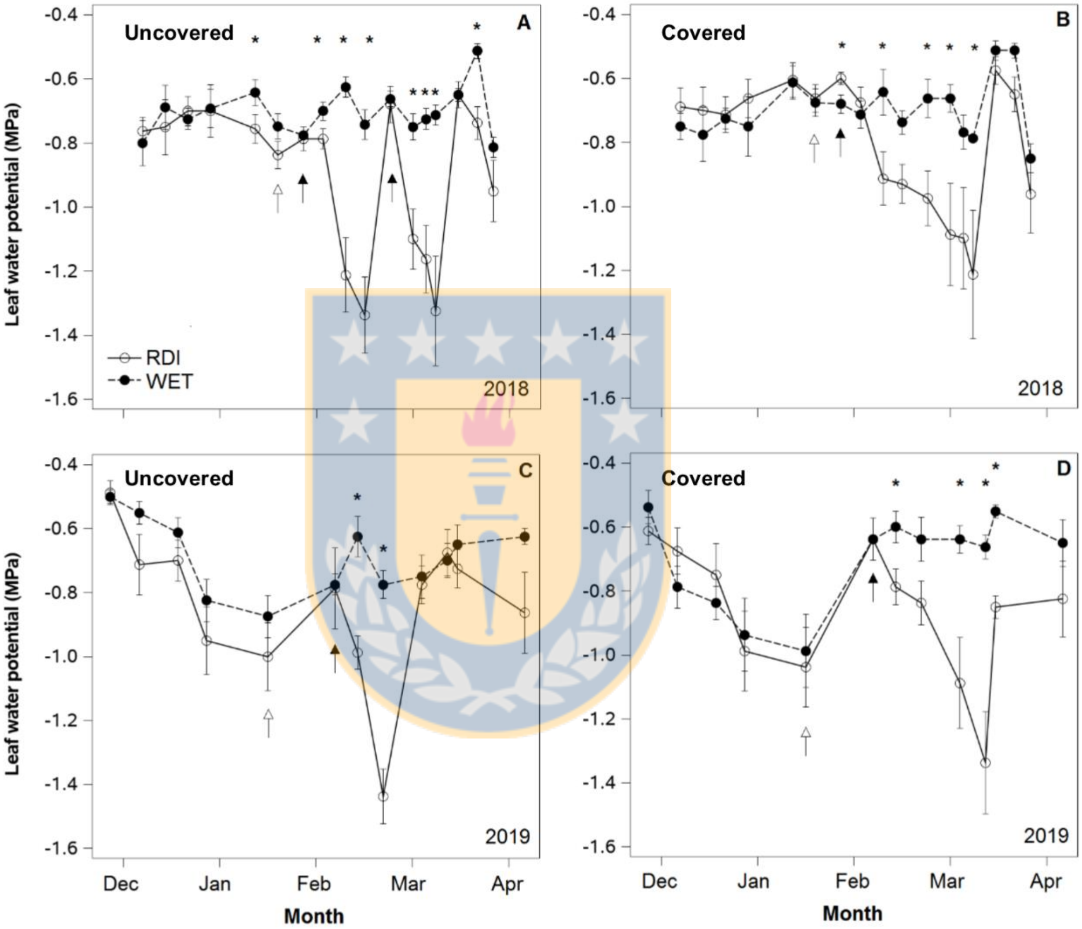
Fuente: Elaboración propia

Figure 2. Spectral photon flux density from 350 to 1,100 nm measured at the soil level (z=0) in Hayward kiwifruit vines on an overhead “pergola” trellis system in San Nicolás, Ñuble, Chile, subjected to two cover treatments (Covered: low-density polyethylene canopy and Uncovered: open field conditions) in January 15th, 2019.



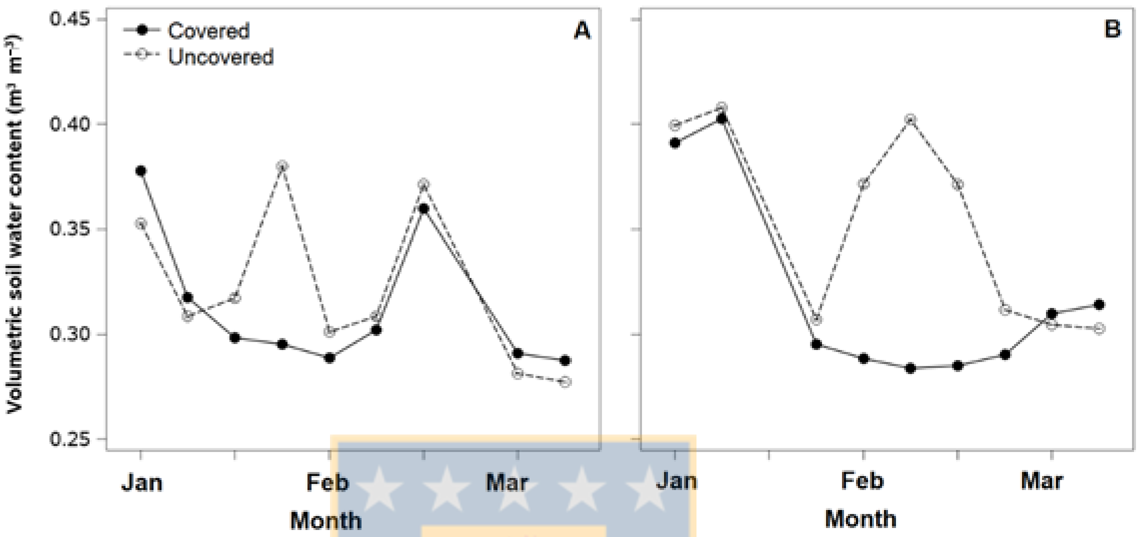
Fuente: Elaboración propia

Figure 3. Average values of midday leaf water potential from fruit set to harvest (November to April) of Hayward kiwifruit vines subjected two irrigation treatments (WET: 100% ET_c and RDI: 0% ET_c) under (A) Uncovered (open-field) conditions in 2018, (B) Covered (polyethylene canopy) conditions in 2018, (C) Uncovered (open-field) conditions in 2019, and (D) and Covered (polyethylene canopy) conditions in 2019. Asterisks indicate significant differences (P≤ 0.05, n=4). White and black arrows indicate the application date of cover and irrigation treatments, respectively. Error bars represent ±1 se.



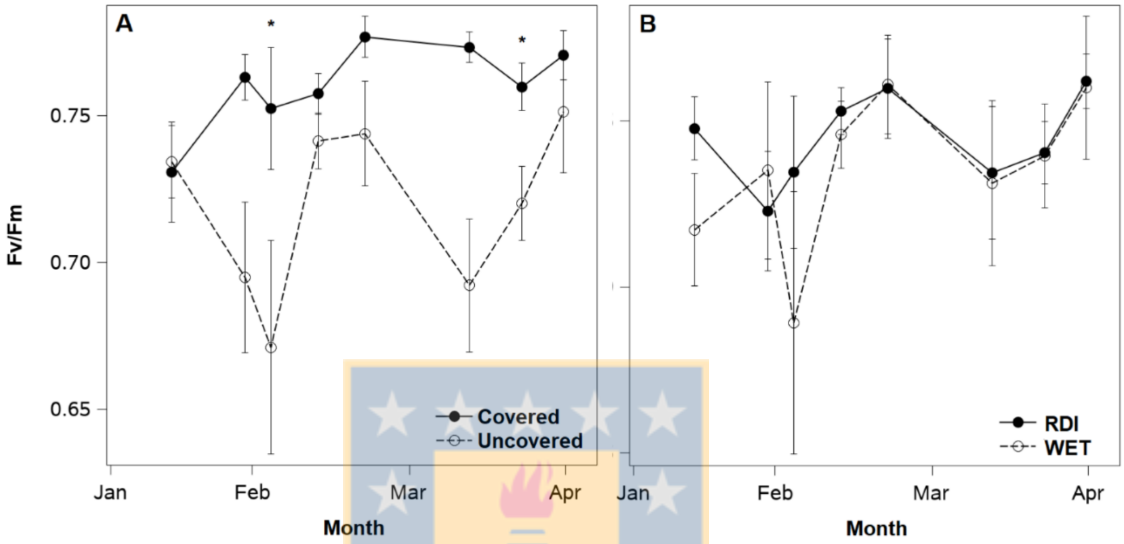
Fuente: Elaboración propia

Figure 4. Weekly values of volumetric soil water content averaged over two depths (-20 and -40 cm) during fruit maturation (January-April) in deficit-irrigated Hayward kiwifruit vines subjected to two cover treatments (Covered: low-density polyethylene canopy and Uncovered: open field conditions) in (A) 2018 and (B) 2019.



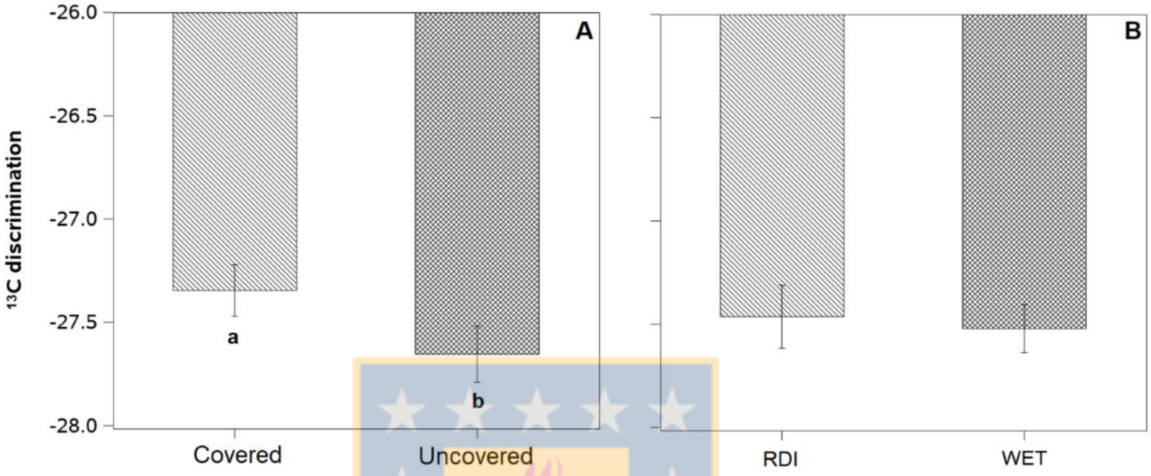
Fuente: Elaboración propia

Figure 5. Average values of *Fv/Fm* during fruit maturation (January-April) in Hayward kiwifruit vines subjected to (A) two cover treatments (Covered: low-density polyethylene canopy and Uncovered: open field conditions) and (B) two irrigation treatments (WET: 100% ET_c and RDI: 0% ET_c) in 2019. Asterisks indicate significant differences ($P \leq 0.05$, n=4). Error bars represent ±1 se.



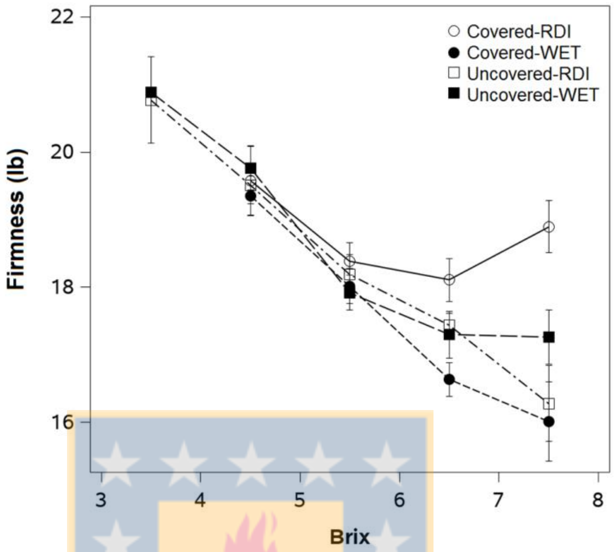
Fuente: Elaboración propia

Figure 6. ^{13}C discrimination measured at harvest time (1st week of March) in mature fruits from Hayward kiwifruit vines subjected to (A) two cover treatments (Covered: low-density polyethylene canopy and Uncovered: open field conditions) and (B) two irrigation treatments (WET: 100% ET_c and RDI: 0% ET_c) in 2019. Different letters indicate significant differences ($P \leq 0.05$, n=4). Error bars represent ±1 se.



Fuente: Elaboración propia

Figure 7. Firmness at various classes of maturity (midpoints of Brix) in fruits from Hayward kiwifruit vines subjected to two cover (Covered: low-density polyethylene canopy and Uncovered: open field conditions) and two irrigation treatments (WET: 100% ET_c and RDI: 0% ET_c) at harvest in 2019. Error bars represent ±1 se.



Fuente: Elaboración propia

Table 1. Cumulative values for irrigation, precipitation, applied water (Irrigation+precipitation), ET_c, and water productivity in Hayward kiwifruit vines subjected to two cover treatments (Covered: low-density polyethylene canopy and Uncovered: open field conditions) and two irrigation treatments (WET: 100% ET_c and RDI: 0% ET_c) from October 1st to April 30th in 2018 and 2019.

	Covered		Uncovered		Covered		Uncovered	
Cumulative values	WET	RDI	WET	RDI	WET	RDI	WET	RDI
	2018				2019			
Irrigation (m^3 ha^{-1})	9,831	7,359	9,831	8,680	7,060	5,106	7,060	6,166
Precipitation (m^3 ha^{-1})	1,530		2,697		1,773		2,098	
Applied water (m^3 ha^{-1})	11,361	8,889	12,528	11,377	8,833	6,879	9,158	8,264
ET_c (m^3 ha^{-1})*	8,163				7,853			
Water productivity (kg m^{-3})	4.25	6.53	3.77	3.39	2.69	4.19	3.44	4.02

* Estimated cumulative crop evapotranspiration for uncovered conditions.

Fuente: Elaboración propia

Table 2. Stomatal conductance, canopy and fruit temperatures, and percentage of defoliation in Hayward kiwifruit vines subjected to two cover treatments (Covered: low-density polyethylene canopy and Uncovered: open field conditions) at the onset of the stage III of fruit development (February) and Harvest (April) in 2018 and 2019.

	2018				2019			
	Stage III		Harvest		Stage III		Harvest	
	COV	UCOV	COV	UCOV	COV	UCOV	COV	UCOV
Stomatal conductance (mmol $m^{-2}s^{-1}$)	653.1	746.8	335.1	371.7	479.7	501.7	413.5	435.9
Canopy temperature (°C)	27.6	26.9	18.7	18.5	23.6	22.9	21.9	21.8
Fruit temperature (°C)	27.4	27.3	19.47	20.04	21.5	22.1	21.9	22.2
Defoliation (%)	-	-	-	-	1.26	0	23.8	33.12

Within-row means followed by different letters are significantly different, n = 4, $P \leq 0.05$.

Fuente: Elaboración propia

Table 3. Stomatal conductance, canopy and fruit temperatures, and percentage of defoliation in Hayward kiwifruit vines subjected to two irrigation treatments (WET: 100% ET$_c$ and RDI: 0% ET$_c$) in 2018 and 2019.at the onset of the stage III of fruit development (February) and Harvest (April) in 2018 and 2019.

	2018				2019			
	Stage III		Harvest		Stage III		Harvest	
	WET	RDI	WET	RDI	WET	RDI	WET	RDI
Stomatal conductance (mmol m^{-2}s^{-1})	701.5	698.4	352.6	354.2	490.1	491.4	429.7	419.8
Canopy temperature (°C)	27.1	27.3	18.45	18.71	23.3	23.0	21.6	22.1
Fruit temperature (°C)	27.3	27.4	19.24	20.26	21.6	22.0	21.7 b	22.4 a
Defoliation (%)	-	-	-	-	0.83	0.43	16.41 b	40.51 a

Within-row means followed by different letters are significantly different, n = 4, $P \leq 0.05$.

Fuente: Elaboración propia

Table 4. Yield estimates in in Hayward kiwifruit vines subjected to two cover treatments (Covered: low-density polyethylene canopy and Uncovered: open field conditions) and two irrigation treatments (WET: 100% ET$_c$ and RDI: 0% ET$_c$) at harvest in 2018 and 2019..

	Covered		Uncovered		Covered		Uncovered	
	RDI	WET	RDI	WET	RDI	WET	RDI	WET
	2018				2019			
Fuits per plant	497	690	573	535	274	369	400	437
Yield per plant (kg)	65	78	64	52	32	39	42	45
Fruit weight (g)	131	113	111	97	117	105	106	102
Yield (ton ha^{-1})	48.3	58,1	47.3	38.6	23.7	28.8	31.5	33.2

Within-row means followed by different letters are significantly different, n = 4, $P \leq 0.05$.

Fuente: Elaboración propia

Table 5. Harvest date, brix, percentage of dry matter, pulp firmness, polar and equatorial diameters, and sunburn severity in Hayward kiwifruit vines subjected to two cover treatments (Covered: low-density polyethylene canopy and Uncovered: open field conditions) at harvest in 2018 and 2019.

	Covered	Uncovered	Covered	Uncovered
	2018		2019	
Harvest date	April 20th		April 10th	
Brix	7.57	7.48	6.80	6.45
Dry matter (%)	17.2	16.6	17.4 a	16.9 b
Firmness (lbf)	14.3	14.8	16.1	16.2
Polar diameter (mm)	52.0	52.3	61.1	60.14
Equatorial diameter (mm)	51.9	50.8	55.5	53.2
Sunburn				
Null (%)	77.5	53.6	47.8	15.6
Mild (%)	16.8	26.3	42.1	50.2
Moderate (%)	5.59 b	19.88 a	10.06 b	33.06 a
Severe (%)	0.12	9.23	0.0	1.11

Within-row means followed by different letters are significantly different, n = 4, $P \leq 0.05$.

Fuente: Elaboración propia

Table 6. Harvest date, brix, percentage of dry matter, pulp firmness, polar and equatorial diameters, and sunburn severity in Hayward kiwifruit vines subjected to two irrigation treatments (WET: 100% ET_c and RDI: 0% ET_c) at harvest in 2018 and 2019.

	RDI	WET	RDI	WET
	2018		2019	
Harvest date	April 20th		April 10th	
Brix	7.57	7.48	6.71	6.53
Dry matter (%)	17.4 a	16.4 b	17.4	17.0
Firmness (lbf)	14.7	14.4	16.3	16.0
Polar diameter (mm)	51.1	53.2	60.3	60.9
Equatorial diameter (mm)	49.9 b	52.7 a	52.9 b	55.7 a
Sunburn				
Null (%)	53.4 b	77.8 a	19.2	14.3
Mild (%)	28.7 a	14.3 b	58.1 a	34.2 b
Moderate (%)	17.5	7.93	21.9	21.2
Severe (%)	0.35	0.0	0.78	0.33

Within-row means followed by different letters are significantly different, n = 4, $P \leq 0.05$.

Fuente: Elaboración propia

CAPÍTULO 3

CONCLUSIONES GENERALES

Este estudio muestra evidencia de que la instalación tardía de una cubierta de plástico retrasa la aparición de estrés hídrico severo en un huerto maduro de kiwi bajo riego deficitario controlado, pero no genera diferencias de importancia en el estado hídrico de plantas regadas abundantemente.

Las claras diferencias de las curvas de desecación de suelo y de potencial hídrico del tallo en plantas bajo riego deficitario con y sin cobertura sugieren que el uso de cultivos protegidos en kiwi disminuyó la demanda de agua atmosférica y así como aumentó la vulnerabilidad de las hojas a la cavitación.

Si bien las plantas bajo estrés hídrico mejoraron la productividad del agua, el mayor número de frutos dañados por “golpe de sol” sugiere que una severidad de estrés hídrico de -1,3 MPa de potencial hídrico de la hoja al mediodía puede ser excesiva para la producción comercial de kiwi. El uso combinado de cultivos protegidos y riego deficitario parece prometedor, ya que se redujo el ablandamiento de la fruta a una mayor madurez y se mejoró la productividad del agua.