Evaluation of the Crop Water Stress Index as an Indicator for the Diagnosis of Grapevine Water Deficiency in Greenhouses

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Abstract: Precise irrigation management of grapevines in greenhouses requires a reliable method to easily quantify and monitor the grapevine water status to enable effective manipulation of the water stress of the plants. This study evaluated the applicability of crop water stress index (CWSI) based on the leaf temperature for diagnosing the grapevine water status. The experiment was conducted at Yuhe Farm (northwest China), with drip-irrigated grapevines under three irrigation treatments. Meteorological factors, soil moisture contents, leaf temperature, growth indicators including canopy coverage and fruit diameter, and physiological indicators including SPAD (relative chlorophyll content), stem water potential (\(\phi_s\)), stomatal conductance (\(g_s\)), and transpiration rate (\(E\)) were studied during the growing season. The results show that the relationship between the leaf-air temperature difference (\(T_c-T_a\)) and the plant water status indicators (\(\phi_s, g_s, E\)) were significant (\(P < 0.05\)), and the relationship between \(g_s, E\) and \(T_c-T_a\) was the closest, with \(R^2\) values ranging from 0.530–0.604 and from 0.545–0.623, respectively. CWSI values are more easily observed on sunny days, and it was determined that 14:00 BJS is the best observation time for the CWSI value under different non-water-stressed baselines. There is a reliable linear correlation between the CWSI value and the soil moisture at 0–40 cm (\(P < 0.05\)), which could provide a reference when using the CWSI to diagnose the water status of plants. Compared with the \(T_c-T_a\) value, the CWSI could more accurately monitor the plant water status, and above the considered indictors, \(g_s\) has the greatest correlation with the CWSI.

Keywords: crop water stress index; deficit irrigation; soil moisture; grapevine; plant water status index; growth index

1. Introduction

Grapevine is one of the important economic fruit trees in China. In recent years, with the adjustment of agricultural industrial structure, the cultivation area of grapevine has increased greatly year by year in China. By the end of 2015, the viticulture area in China had expanded to 800,000 ha, making China the world’s largest grape producer [1]. In recent years, with the improvement of grapevine protected cultivation technology, grapevines are the fruit tree species with the largest protected cultivation area in China, with an output that is 3.5 times in greenhouse higher than that of open planting. This method has significant advantages in ensuring the freshness and supply of fruit during the off season. The development of protected cultivation is an effective way to promote the development of rural economies.
The changes of grapevine water status can affect plant growth, canopy microclimate and fruit metabolism, which will further affect grapevine fruit composition, yield and quality [2,3]. If water stress occurs in the key growth stage of grapevine, it will have adverse effects on the growth, development, and productivity of grapevine. To maximize the economic benefits of grapevines, it is necessary to observe plant water deficit accurately and timely. There are usually two methods to reflect crop water status, which are based on soil measurement and plant measurement [4–6]. However, it is more reliable to use the plant water status as the basis for irrigation than the soil moisture, because the water deficiency of plants is not only determined by the soil moisture but also related to changes in atmospheric evaporation, the distribution of crop roots, and the water transport capacity [7]. Among the methods based on the plant water status, the pressure chamber method is usually used to monitor the stem water potential and leaf water potential of plants [8,9], but there are some disadvantages in these methods, such as complex operation, destructive, time-consuming and laborious characteristics, which are not suitable for long-term monitoring of plant water status.

A portion of plant energy is consumed through transpiration of grapevine leaves to achieve temperature regulation. When soil moisture affects crop growth and development, the change of stomatal behavior will be directly reflected in some physiological indexes, such as stomatal conductance and transpiration rate, which will cause a series of changes of crop water status, such as free water content and relative content, Therefore, stomata tend to close in order to maintain water balance in plants, and the change of transpiration intensity usually determines the degree of heat loss on the surface of leaves, and then leads to the change of leaf temperature [8], leaf temperature is a basic parameter in the study of the physiological characteristics and ecology of plants. Canopy temperature can accurately reflect the water and energy exchange between crops and the atmosphere; Therefore, canopy temperature can be used to predict crop water status. The use of infrared thermometers to measure the leaf temperature provides an important detection method for the crop water deficit. Great progress was made in recent years because the leaf temperature in crops is sensitive to water stress, and the observation method is simple. Research shows that it is feasible to regulate the greenhouse environment based on the leaf temperature, and the mechanism of the leaf temperature model was established [9,10]. When there is enough water in the root zone, the transpiration of leaves will produce a cooling effect, which makes the leaf temperature lower than the ambient temperature. However, when the soil moisture content is lower than a certain value, to prevent the excessive loss of crop water, the decrease in transpiration results in an increase in the leaf temperature [11]. In fact, part of the solar radiation absorbed by leaves is used for transpiration, and the other part is used to raise the leaf temperature.

The CWSI is the most commonly used indicator to diagnose crop water deficits based on the leaf temperature. CWSI was developed as a standardized indicator to diagnose stress and overcome the effects of other environmental factors that affect the relationship between stress and plant temperature [11,12]. The CWSI is usually determined by empirical methods based on relating the leaf-air temperature difference \((T_c - T_a)\) to the air vapor pressure deficit of a non-water-stressed baseline. The CWSI was successfully applied to develop irrigation plans for different crops in different regions over the past 20–25 years [13–22]. Nielsen et al. observed the leaf temperatures of wheat, soybean, bermudagrass, and cabbage under different irrigation treatments and established a CWSI model that could monitor the crop water status in real time [23–28]; the key to building the CWSI model was to determine the lower limit baseline of the model [29]. Veyisi et al. [21] found that the lower limit equation is different at different stages of the growing season. Changes in the baseline are caused by weather differences during the entire growth period and may be the response to the growth stage. The NWSB (non-water-stressed baseline) of CWSI were not consistent during the growth period, but the variation of NWSB during the growth period can be partly explained by the change of solar angle [7]. In recent years, many studies used the soil moisture, leaf relative water content, leaf water potential, and stem water potential to characterize the plant water status. However, few studies
have been conducted on diagnosing the plant water status using the CWSI based on the leaf stomatal conductance and transpiration rate.

The main aims of this study were (1) to analyze the relationship between \( T_c - T_a \) and plant water status indicators, (2) to determine the NWSB (non-water-stressed baseline) for grapevine in greenhouse and as well as CWSI diurnal time courses under different stages, (3) to evaluate the feasibility of the CWSI in predicting the canopy coverage, fruit diameter, and SPAD of grapevines, and (4) to clarify the relationship between the CWSI and plant water status indicators. The above objectives are aimed at evaluating the applicability of the CWSI in monitoring grapevine growth and diagnosing the water deficiency status.

2. Materials and Methods

2.1. Study Area

The experiment was carried out in greenhouse of Yuhe Farm, Shaanxi Province, from March to July 2018 (108.58° E, 37.49° N). The annual average rainfall in this area is 365.7 mm, the annual average temperature is 8.3 °C, the annual relative humidity is 69.37%, and the annual average duration of sunshine is 2893.5 h, which is representative of the typical continental marginal monsoon climate of the area. Table 1 shows the meteorological data (cultivation stage averages) recorded over the experimental year. The test soil was an aeolian sandy soil. The chemical properties of the soil were as follows: the soil ammonium nitrogen was 7.48 mg/kg, the nitrate nitrogen was 22.91 mg/kg, the available phosphorus was 4.07 mg/kg, and the available potassium was 163.47 mg/kg. The physical properties of the soil are shown in Table 2.

Table 1. Meteorological data of different cultivation stages during the growing season.

| Cultivation Stage          | \( T_a \) (°C) | \( R_a \) (w/m²) | RH (%) | VPD (kpa) |
|----------------------------|----------------|-----------------|--------|-----------|
| vegetative stage           | 18.8           | 301.0           | 53.8   | 0.33      |
| flowering stage            | 19.7           | 327.9           | 50.4   | 0.35      |
| fruit expansion stage      | 22.3           | 421.1           | 51.2   | 0.34      |
| coloring mature stage      | 25.1           | 362.3           | 55.7   | 0.32      |

Note: \( T_a \) (°C): air temperature; \( R_a \) (w/m²): solar radiation; RH (%): relative humidity; VPD (kpa): vapor pressure deficit.

Table 2. Physical properties of the soil.

| Depth (cm) | Textural Analysis | FC (g g⁻¹) | PWP (g g⁻¹) | Bulk Density (g cm⁻³) |
|------------|-------------------|------------|-------------|-----------------------|
| 0–40       | 87.54 5.27 7.19   | 13.18      | 2.31        | 1.64                  |
| 40–80      | 70.23 19.53 10.24 | 17.45      | 6.38        | 1.46                  |

Note: FC: field capacity; PWP: permanently wilting point.

2.2. Experimental Design

Six year-old grapevine (early maturing variety 6–12) were planted in greenhouse, and grapevines with good growth and similar shapes were selected for the experiments. The entire growth period of grapevines can be divided into four main growth stages: the vegetative stage, the flowering stage, the fruit expansion stage, and the coloring mature stage, the cultivation period was 121 days during the growth season. The greenhouse was oriented east-west and was 70 m long and 9 m wide. The grapevine row spacing was 0.8 m, and the plant spacing was 0.6 m, with 14 grapevines per row. Artificial warming was carried out in greenhouse to ensure the growth temperature of grapevines on 11 March 2018. Drip irrigation was used in the experiment. A single-wing labyrinth drip irrigation belt (produced by Xinjiang Dayu Water Saving Company, Xinjiang, China) was adopted. The inner diameter and wall thickness of drip irrigation belt was 20 mm and 0.18 mm, respectively. The distance
between the drippers was 300 mm, the design flow of the dripper was 4.0 L/h, and the laying mode of the drip belt was one row of two pipes.

The experiment was conducted with drip-irrigated grapevines under three irrigation treatments: a full irrigation treatment (T₁: 100% M) and two regulated deficit irrigation treatment (T₂: 80% M; T₃: 60% M), M represents the irrigation quota. There were three treatments in total and three plots per treatment (each plot had a length of 8 m, a width of 4.5 m, and an area of 36 m²), with a random block arrangement. The irrigation dates and irrigation amount is shown in Table 3, the grapevines were irrigated 12 times during the entire growth period. The irrigation quota was calculated by Equation (1). The irrigation time was determined according to whether or not T₁ reached the lower limit of the water quantity, which was 65% of β₁ at the vegetative and coloring mature stages and 70% of β₁ at the flowering and fruit expansion stage. The predicted wet layer depth of the soil was 80 cm. The total amount of fertilization during the entire growth period was 0.84 t/ha, and the proportion of N:P:K was 1.0:0.6:1.2. Fertilization was carried out over three periods: the germination stage accounted for 20% of the total amount of fertilization, the flowering and fruit expansion stages accounted for 60%, and the coloring mature stage accounted for 20%. The drip irrigation and fertilization were controlled by integrated irrigation and fertilization equipment. The total irrigation amount during the entire growth period of each treatment was 3810 m³/ha, 3045 m³/ha, and 2280 m³/ha, respectively.

\[
M = 0.1γ_sHP(β_1 - β_2)
\]

where \(M\) represents the irrigation quota, mm; \(γ_s\) represents the soil dry bulk density, 1.64 g/cm³ in 0–40 cm soil depth, 1.46 g/cm³ in 40–80 cm soil depth; \(H\) is the predicted wet layer depth of soil, 0.8 m; \(P\) is the designed wet soil ratio, 0.8; \(β_1\) is the field water holding capacity, 13.18% in 0–40 cm soil depth, 17.45% in 40–80 cm soil depth; and \(β_2\) is the lower limit of the soil moisture content, 65% of \(β_1\) at the vegetative and coloring mature stages and 70% of \(β_1\) at the flowering and fruit expansion stage.

| Cultivation Stage    | Irrigation Date | Irrigation Amount (m³/ha) |
|----------------------|-----------------|--------------------------|
|                      | T₁              | T₂                        | T₃                        |
| vegetative stage     | 3/11            | 293.61                    | 234.80                    | 176.02                    |
|                      | 3/19            | 293.61                    | 234.65                    | 175.55                    |
|                      | 3/27            | 293.60                    | 234.60                    | 175.34                    |
|                      | 4/06            | 293.62                    | 234.65                    | 175.62                    |
| Flowering stage      | 4/15            | 341.40                    | 273.05                    | 204.60                    |
|                      | 4/25            | 341.39                    | 273.04                    | 204.15                    |
|                      | 5/05            | 341.39                    | 273.00                    | 204.20                    |
| Fruit expansion stage| 5/13            | 341.37                    | 272.58                    | 204.14                    |
|                      | 5/20            | 341.42                    | 272.69                    | 204.47                    |
|                      | 5/27            | 341.41                    | 273.10                    | 204.34                    |
| Coloring mature stage| 6/05            | 293.60                    | 234.50                    | 175.59                    |
|                      | 6/15            | 293.61                    | 234.55                    | 176.30                    |

| Total irrigation amount | 3810 | 3045 | 2280 |

2.3. Observation Indicators

2.3.1. Meteorological Factors

The air temperature (\(T_a\)), relative humidity (RH), and solar radiation (\(R_a\)) were recorded automatically every 30 min using a Watchdog micro series (Spectrum Technologies Inc., Chicago, IL, USA) meteorological station in the greenhouse. The vapor pressure deficit (VPD) was estimated by the
relative humidity (RH) and air temperature (Ta) and was calculated by the modified Penman formula. The formula [30] is shown in Equation (2):

\[ VPD = 0.6108 \times \exp\left(\frac{17.27 \times Ta}{Ta + 237.3}\right) \times \left(1 - \frac{RH}{100}\right) \]  

(2)

2.3.2. Soil Moisture Content

The soil moisture automatic monitoring system consisted of an EM50 data recorder (Environmental Logging System, Decision Devices, Inc., Salt Lake City, UT, USA) and four ECH2O5 TE sensors (Decision Devices, Pullman, WA, USA). The soil moisture automatic monitoring system was installed 30 cm from the base of the grapevines and perpendicular to the planting row. Three representative grapevines were selected, and three monitoring systems were installed for each treatment. A sensor was installed every 20 cm, the buried depth was 80 cm, the soil volume moisture content was recorded every 30 min. Before the beginning of the growing season, the ECH2O5 TE sensor was calibrated every 20 cm using the drying method, and then the sensor was calibrated every 10 days using the drying method.

2.3.3. Leaf Temperature and Leaf-Air Temperature Difference

The leaf temperature was continuously monitored and measured every 30 min by an ELV-15A infrared thermometer (Optris CSMTL10, Germany, ± 1.5 °C) that was part of the plant physiological and ecological monitoring system. The probe of the infrared thermometer faced the upper part of the grapevine canopy, and the distance between the detection window and the fully expanded leaves of the grapevine canopy was kept at 5–10 cm. The measured data were recorded by a computer, and the mean of three repetitions was used as the result. The leaf-air temperature difference (Tc - Ta) is defined as the difference between leaf temperature (Tc) and air temperature (Ta) and can reflect the self-regulation ability of plant leaves that are under water stress to a certain degree [31].

2.3.4. Canopy Cover and Fruit Diameter

From the vegetative stage to the fruit expansion stage, the canopy cover was measured once every 5–7 days. Three representative grapevines with good growth conditions were selected for each treatment. Canopeo is an ACT image analysis tool (developed by the University of Oklahoma) using color values in the red-green-blue (RGB) system to measure the canopy cover. Nadir (i.e., downward-facing) images were taken from areas of experimental plots using “point and shoot” type digital cameras. The camera was kept at about 1.5 m from the top of the canopy using a 1.5 m monopod. Maintain adequate distance from the camera to the top of the canopy.

The fruit diameter was measured in the early stage of fruit growth, three grapevines were selected for each treatment, and a grape bunch was marked for each grapevine. The fruit diameter (transverse and longitudinal) in the middle of the grapevine bunch was measured with a digital vernier caliper (Mitutogo, Japan; the measurement range is 0–150 mm and precision is 0.01 mm) once every 3–5 days until the grapevines matured; the measurements were averaged for each replication.

2.3.5. Determination of Plant Physiological Indicators

Three grapevines were selected for each treatment. From the vegetative stage to the mature stage, a chlorophyll SPAD meter ‘SPAD-502’ (Konica Minolta, Tokyo, Japan) was used for the precise, rapid, and nondestructive estimation of SPAD value approximately every 15 days. The same healthy leaf was monitored before the fruit expansion stage, and the fifth healthy leaf was monitored at the internode of fruiting branch in the upper part of the plant after the fruit expansion stage. Five values were measured for each leaf, the measurements were averaged for each replication. The pressure chamber (TP-PW-II, Zhejiang Top Cloud-agri Technology Co., Ltd., Hangzhou, Zhejiang, China) was used to measure the stem water potential (ϕs) every 5–7 days, and the ϕs is measured at 9:00 to 10:00 BJS. Three grapevines were selected for each treatment, and one branch under good growth conditions
was selected as the sample on the sunny side outside the crown. The sample was put into a plastic bag containing moist gauze and quickly brought into the laboratory. The sample was clamped in a pressure chamber and pressurized by gas (compressed nitrogen), the pressure used for exudation of tissue fluid was observed. At this time, the pressure value was the stem water potential. At the same time, stomatal conductance \( g_s \) was measured on the same day, but between 09:00–10:00 BJS. An open gas-exchange system Li-6400 XT (Li-Cor Biosciences, Lincoln, NE, USA) with a 2 cm × 3 cm standard chamber was used to measure the \( g_s \) and leaf transpiration rate \( E \) under the ambient light (1500 \( \mu \)mol m\(^{-2} \) s\(^{-1} \)) and the ambient CO\(_2\) concentration (400 ± 5 \( \mu \)mol CO\(_2\) mol\(^{-1} \)) conditions.

### 2.4. Concept of CWSI and Its Calculation Based on the Idso Method

The empirical form of the CWSI is based on the linear relationship between \( T_c-T_a \) and the VPD of the air for a well-watered crop \[11\] during the day and under homogeneous conditions and a clear sky. NWSB represents non-water-stressed baseline, which is VPD-dependent. According to the experimental design, the soil moisture content of \( T_1 \) was always above 65% of the field water capacity. It could be considered that grapevines under \( T_1 \) were in the potential evaporation state during the entire growth period. Therefore, the lower limit baseline of the CWSI was established by using the \( T_c-T_a \) of \( T_1 \). Based on the lower limit baseline and the temperature and relative humidity \( (RH) \) measured on that day, the upper limit baseline could be calculated by the formula proposed by Idso et al. \[11\] and Jackson et al. \[12\]. The CWSI is calculated as follows:

\[
CWSI = \frac{(T_c - T_a) - (T_c - T_a)_{\text{ll}}}{(T_c - T_a)_{\text{ul}} - (T_c - T_a)_{\text{ll}}} \tag{3}
\]

where \( T_c \) is the leaf temperature (°C); \( T_a \) is the air temperature (°C); \( T_c-T_a \) is the leaf-air temperature difference; \((T_c-T_a)_{\text{ll}}\) is the NWSB; and \((T_c-T_a)_{\text{ul}}\) is the non-transpiring baseline (upper limit equation). \((T_c-T_a)_{\text{ll}}\) and \((T_c-T_a)_{\text{ul}}\) can be estimated as linear functions of the atmospheric VPD and the vapor pressure gradient \( (VPG) \), respectively, using Equations (4)–(9) \[11\].

\[
(T_c - T_a)_{\text{ll}} = b + a(\text{VPD}) \tag{4}
\]

\[
(T_c - T_a)_{\text{ul}} = b - a|\text{VPG}| \tag{5}
\]

\[
\text{VPD} = e_s(T_a) - e_a \tag{6}
\]

\[
\text{VPG} = e_s(a + T_a) - e_s(T_a) \tag{7}
\]

\[
e_s(T_a) = (0.6108 \times \text{EXP}(\frac{17.27 \times T_a}{T_a + 237.3})) \tag{8}
\]

\[
e_a = e_s(T_a) \times \left(\frac{\text{RH}}{100}\right) \tag{9}
\]

where \( a \) and \( b \) are the slopes and constants of the fitting equation, \( e_s(T_a) \) is the saturated vapor pressure at air temperature under specific conditions, and \( e_a \) is the actual vapor pressure. These formulas follow the following fact that transpiration decreases the leaf temperature relative to the air temperature; the decrease in temperature is greater at relatively high VPD values than at low VPD values. The variation range of the index is from 0 to 1. The closer the CWSI value is to 1, the more serious the water stress is. On the contrary, 0 indicates no water stress.

### 2.5. Data Analysis

The correlation analysis and regression analysis were carried out using SPSS 21.0 software (SPSS Inc., Chicago, IL, USA). Multiple comparisons were performed by least significant difference tests, with a significance level of 0.05. Microsoft Excel 2010 Software was used for processing data. The graphs were created by using Origin 9.1. Correlation analysis was conducted between \( T_c-T_a \) and
meteorological and leaf temperature datas every 30 min, and 7 days were selected for each growth stage. The relationships between $T_{c} - T_{a}$ and VPD as well as between $T_{c} - T_{a}$, CWSI and physiological measurements were analyzed through regression analyses. Linear regression were conducted between CWSI and growth index of grapevine, soil moisture content. In all cases, the coefficient of determination ($R^2$) was used to assess the goodness of fit of the associations among variables.

3. Results and Discussions

3.1. Diagnosis of Grapevine Water Deficiency Based on the Leaf-Air Temperature Difference

3.1.1. Daily Changes in the Leaf-Air Temperature Difference before and after Irrigation

The daily changes in $T_{c} - T_{a}$ before and after irrigation under different stages were V-shaped curves (Figure 1). It could be observed that the $T_{c} - T_{a}$ value of each treatment was basically negative the day before and after irrigation, which was consistent with the results of Sepaskhah [32]. At the flowering stage, the $T_{c} - T_{a}$ value of each treatment began to decrease after 13:00 and remained stable at approximately 20:00 (Figure 1c). $T_{c} - T_{a}$ reached a peak under the three treatments during the mature stage, which was $-5.45 - 7.01 \degree$C (Figure 1g). $T_{c} - T_{a}$ decreased rapidly between 16:00–19:00. On the one hand, the leaf temperature was affected by $R_{a}$ and ground radiation; on the other hand, the specific heat capacity of water vapor is large, which made the leaf temperature decrease slowly. Thus, $T_{c} - T_{a}$ appeared to decrease.

The peak of $T_{c} - T_{a}$ after irrigation is higher than that before irrigation (Figure 1). Yeşim [33] found that the range of $T_{c} - T_{a}$ during the entire growth period was $-4 \degree$C–$3 \degree$C, which was slightly smaller than the current experimental findings because $T_{c} - T_{a}$ is affected by the crop variety, canopy resistance, leaf size, and canopy density [34]. $T_{c} - T_{a}$ showed a “double-peak” curve under the three treatments during the fruit expansion stage. The first peak was slightly higher than the second because the leaf stomata were actively closed due to the high temperature (Figure 1f). When the environmental conditions cooled, the leaf stomata could not quickly recover to their original state. There was a certain lag, and the leaf temperature was still high, resulting in the peak of $T_{c} - T_{a}$ being lower than the first peak. The peak of $T_{c} - T_{a}$ at the mature stage developed at approximately 13:00 (Figure 1g,h). We concluded that the $T_{c} - T_{a}$ of each treatment decreased as the water deficit increased because the leaf stomata tended to close under water stress, and the decrease in transpiration intensity would lead to a small amount of heat loss on the leaf surface; therefore, the leaf temperature was high, and $T_{c} - T_{a}$ decreased [35]. Similar results were obtained by Růžička Stričević [36], who found that $T_{c} - T_{a}$ between poplar and sorghum leaves increased as the water deficit increased.

3.1.2. Relationship between $T_{c} - T_{a}$ and Meteorological Factors

The correlation analysis of $T_{c} - T_{a}$ with meteorological factors ($T_{a}$, $R_{a}$, RH and VPD) and leaf temperature are shown in Table 4. The positive correlation between $T_{c} - T_{a}$ and $T_{a}$, $T_{a}$ and VPD was significant ($P < 0.05$), and the negative correlation with RH was significant ($P < 0.05$) (Table 4). The results showed that meteorological factors had a significant effect on $T_{c} - T_{a}$. King and Shellie [37] obtained similar research conclusions. Moreover, Jensen et al. [38] believed that the main meteorological factor affecting $T_{c}$ was $R_{a}$. This study indicated that $R_{a}$ had the least correlation with $T_{c} - T_{a}$, which was mainly because the plastic roofs of greenhouses would block part of the $R_{a}$. Furthermore, $R_{a}$ at night was zero; thus, the change in $T_{c}$ was no longer affected by $R_{a}$. The correlation coefficients of $T_{c} - T_{a}$ and $R_{a}$, $T_{c} - T_{a}$ and RH decreased with the development of growth period, from 0.651–0.734 and 0.711–0.861 to 0.453–0.607 and 0.525–0.621, respectively (Table 4).
Figure 1. Daily change in the leaf-air temperature difference before and after irrigation at different stages. (a,b) represent the vegetative stage; (c,d) represent the flowering stage; (e,f) represent the fruit expansion stage; and (g,h) represent the mature stage. (a,c,e), and (g) represent $T_c - T_a$ before irrigation, while (b,d,f,h) represent $T_c - T_a$ after irrigation. $T_1$ represents 100% M, $T_2$ represents 80% M, $T_3$ represents 60%.

3.1.3. Relationship between $T_c - T_a$ and Plant Water Status Indicators

The fitting diagrams of $T_c - T_a$ with plant water status indicators ($\varphi_s$, $g_s$, and $E$) under the three treatments are shown in Figure 2, and the coefficient of the fitting equation between $T_c - T_a$ and plant water status index ($\varphi_s$, $g_s$, and $E$) is shown in Table 5. The relationship between $T_c - T_a$ and the plant water status indicators showed a correlation when dates were pooled together, and the correlation was significant among all treatments ($P < 0.05$), which indicates that $T_c - T_a$ can reflect the water status of grapevines to a certain extent. The $\varphi_s$, $g_s$, and $E$ values decreased with the decrease of $T_c - T_a$. It was easy to find that the relationship between $\varphi_s$ and $T_c - T_a$ was a quadratic function curve (Figure 2a).
Gonzalez-Dugo et al. [39] also believed that the relationship between the stem water potential and the CWSI was a quadratic function curve, consistent with the results of this paper. The \( g_s \) and \( E \) values were linearly correlated with \( T_c-T_a \) (Figure 2b,c). Khorsandi et al. [35] reported that there is a significant and linear relationship between \( T_c-T_a \) and \( g_s \). In general, the relationships between \( T_c-T_a \) and \( g_s \) and between \( T_c-T_a \) and \( E \) were higher, with \( R^2 \) values ranging from 0.530–0.604 and from 0.545–0.623, respectively (Table 5).

Table 4. The correlation of leaf-air temperature difference with meteorological factors and leaf temperature.

| Cultivation Stage | Treatment | \( T_a \) | \( R_a \) | RH | VPD | Leaf Temperature |
|-------------------|-----------|-----------|---------|----|-----|------------------|
| Vegetative stage  | \( T_1 \) | 0.843 **  | 0.651 ** | −0.711 ** | 0.947 ** | 0.824 ** |
|                   | \( T_2 \) | 0.802 **  | 0.708 ** | −0.861 ** | 0.954 ** | 0.764 ** |
|                   | \( T_3 \) | 0.783 **  | 0.734 ** | −0.716 ** | 0.956 ** | 0.870 ** |
|                   | \( T_1 \) | 0.826 **  | 0.641 ** | −0.741 ** | 0.831 ** | 0.453 *  |
| Flowering stage   | \( T_2 \) | 0.879 **  | 0.675 ** | −0.775 ** | 0.839 ** | 0.615 ** |
|                   | \( T_3 \) | 0.605 **  | 0.531 *  | −0.631 *  | 0.644 ** | 0.579 ** |
| Fruit expansion   | \( T_1 \) | 0.865 **  | 0.607 ** | −0.661 *  | 0.864 ** | 0.675 ** |
| stage             | \( T_2 \) | 0.885 **  | 0.570 ** | −0.584 ** | 0.862 ** | 0.702 ** |
|                   | \( T_3 \) | 0.764 **  | 0.537 ** | −0.563 ** | 0.801 ** | 0.636 ** |
| Mature stage      | \( T_2 \) | 0.760 **  | 0.599 ** | −0.525 *  | 0.665 ** | 0.525 *  |
|                   | \( T_3 \) | 0.627 **  | 0.453 *  | −0.545 ** | 0.709 ** | 0.665 ** |

Note: *Correlation is significant at the 0.05 level. **Correlation is significant at the 0.01 level. \( T_a \) (°C): air temperature; \( R_a \) (w/m²): solar radiation; RH (%): relative humidity; VPD (kpa): vapor pressure deficit.

Figure 2. Relationships between \( T_c-T_a \) and plant-based water status indicators for three treatments. (a) stem water potential \( (g_s) \), (b) stomatal conductance \( (g_s) \) and (c) leaf transpiration rate \( (E) \) for \( T_1 \), \( T_2 \) and \( T_3 \) treatment. The different lines represent the fitting regression lines to the date. \( T_1 \) represents 100% M, \( T_2 \) represents 80% M, \( T_3 \) represents 60%.

Table 5. Regression coefficient and accuracy evaluation of fitting equation between \( T_c-T_a \) and plant water status index.

| Treatment | Indicator | a     | b     | c     | \( R^2 \) | P    |
|-----------|-----------|-------|-------|-------|----------|------|
| \( T_1 \) | \( \psi_s \) | −58.497 | −37.298 | −10.437 | 0.587    | 0.005|
|           | \( g_s \)  | −12.979 | −2.914 | 0.604  | 0.001    |      |
|           | \( E \)    | −0.898 | −0.821 | 0.581  | 0.001    |      |
| \( T_2 \) | \( \psi_s \) | −19.704 | −22.245 | −9.913  | 0.611    | 0.003|
|           | \( g_s \)  | −21.694 | −2.225 | 0.600  | 0.001    |      |
|           | \( E \)    | −0.838 | −1.547 | 0.545  | 0.002    |      |
| \( T_3 \) | \( \psi_s \) | −0.670  | −4.340 | −8.596  | 0.511    | 0.014|
|           | \( g_s \)  | −23.023 | −2.389 | 0.530  | 0.008    |      |
|           | \( E \)    | −0.628 | −2.450 | 0.623  | < 0.001  |      |

Note: “a” represents the coefficient of the quadratic term; “b” represents the coefficient of the first-order term; “c” represents the constant.
3.2. Diagnosis of Grapevine Water Deficiency Based on the CWSI

3.2.1. Daily Changes in the CWSI under Different Weather Conditions

The daily changes in the CWSI under the two weather conditions are shown in Figure 3. The fluctuation of the CWSI under $T_1$ on sunny days was relatively stable (0.078–0.403) (Figure 3). Moreover, Jones [40] found that the leaf temperature of non-transpiration and full-transpiration could be used as the upper and lower limits of the canopy temperature, respectively. The change rule of the CWSI on cloudy days was similar to that on sunny days, but cloudy days showed a significantly larger range in magnitude than sunny days (−0.280–0.556). Gardner et al. [41] concluded that the stable CWSI values could be used to diagnose plant water deficiency, which is consistent with the results of this study. We concluded that the CWSI was more suitable for sunny weather, and the CWSI would be unstable on rainy days, which is also the reason why the CWSI model is limited as a suitable irrigation index [42,43].

![Figure 3. Daily change in the CWSI under different weather conditions.](image)

3.2.2. NWSB and CWSI

There was a significant negative correlation between $T_c-T_a$ and VPD during different stages (Figure 4). Similar results were obtained by Khorsandi et al., who found that the negative correlation between $T_c-T_a$ and VPD was significant [35]. The $R^2$ value was the highest (0.878) during the fruit expansion stage (Figure 4c), which might be related to the changes in transpiration and photosynthesis during the reproductive stage of grapevines. Furthermore, the absolute value of the slope of the equation during the flowering stage was larger than that during the other stages (Figure 4b), which indicates that the transpiration ability of grapevines changes due to different growth conditions under different stages, thus resulting in different ranges of $T_c$ decreases under the same VPD. Bellvert [44] established the upper ($ul$) and lower limit baselines ($ll$) of Pinot-noir grapevines as follows: $ll = -1.709 VPD + 2.534$ and $ul = 0.465 VPD + 6.125 (R^2 = 0.83)$. Bellvert [45] also developed the $ll$ equation of grapevines as follows: $ll = -1.326 VPD + 3.111 (R^2 = 0.424)$. However, differences in the crop varieties, management methods, soil moisture, and climate conditions will affect the upper and lower limit equations.

The daily changes in the CWSI under the three treatments during the different stages were inverted “V” curves that increased first and then decreased (Figure 5). The CWSI of the three treatments reached the maximum at approximately 14:00 during the different stages, and the peaks of the CWSI under the three treatments were the largest during the fruit expansion stage, ranging from 0.51 to 0.81 (Figure 5c). The CWSI under $T_1$ was significantly lower than that under $T_2$ and $T_3$, indicating that the CWSI increased as the irrigation amount decreased (Figure 5). The results of this study confirmed the views of Sezen et al. [46] and of Colak and Yazar [47]. There were two peaks of the CWSI during the flowering and mature stages (Figure 5b,d), and the second peak was larger than the first peak. However, some studies [26,48] reported that the CWSI might exceed the range of 0–1.0. This might be caused by...
inconsistent baseline calculation methods. We concluded that the order of the CWSI for the three treatments during the entire growth period was $T_3 > T_2 > T_1$, and 14:00 was the best observation time for the CWSI to reflect the water status of grapevines.

![Figure 4](image_url)

**Figure 4.** NWSB of the CWSI at different stages: (a) represents the vegetative stage, (b) represents the flowering stage, (c) represents the fruit expansion stage, and (d) represents the mature stage.

![Figure 5](image_url)

**Figure 5.** Daily changes of the CWSI under the three treatments during different stages. (a) represents the vegetative stage, (b) represents the flowering stage, (c) represents the fruit expansion stage, and (d) represents the coloring mature stage. Data are means ± SE of three replicates. $T_1$ represents 100% M, $T_2$ represents 80% M, $T_3$ represents 60%.
The CWSI value of the T₃ treatment showed a zigzag upward trend, while the CWSI of the T₁ and T₂ treatments increased first and then decreased during the growing season (Figure 6 and Table 6). The CWSI values of the T₁, T₂, and T₃ treatments were 0.07–0.30, 0.20–0.51, and 0.40–0.89 during the growing season (Figure 6). Bellvert [44] reported that the CWSI was occasionally negative under full irrigation treatment, which was not found in the results of this study. As can be seen in Figure 6, the CWSI was different in adjacent time periods, indicating that many external factors, including meteorological factors such as the wind speed, wind direction, clouds, VPD, Tₑ, and Rₛ, affect it [49]. The CWSI values of each treatment decreased after irrigation, and there were different degrees of lag (Figure 6). The reason is that stomatal conductance does not reach the maximum rapidly as the soil moisture changes after rewatering; rather, it recovers slowly under the influence of early water stress, so the leaf temperature is still high [50]. The seasonal mean CWSI values of the three treatments were 0.181, 0.350, and 0.677, respectively (Table 6). Bellvert and Girona [51] reported that the CWSI values under a full irrigation treatment were lower than 0.5, while those under a severe water deficit treatment were close to 1.0; the findings of this study agree with their results. Yazar et al. [52] evaluated the CWSI values of ergot seedless and flame seedless and suggested that crops should be irrigated when the CWSI values reached 0.30–0.35 in the Mediterranean area.

![Figure 6](image_url)

**Figure 6.** Time evolution of the CWSI during the growing season. T₁ represents 100% M, T₂ represents 80% M, T₃ represents 60%.

| Months  | Mean CWSI |
|---------|-----------|
|         | T₁        | T₂        | T₃        |
| March   | 0.159     | 0.275     | 0.511     |
| April   | 0.183     | 0.358     | 0.668     |
| May     | 0.217     | 0.386     | 0.761     |
| June    | 0.166     | 0.380     | 0.766     |
| Seasonal mean CWSI | 0.181 | 0.350 | 0.677 |

### 3.2.3. Relations between the CWSI and Canopy Cover, Fruit Diameter and SPAD

Many studies showed that the CWSI values are closely related to the leaf area index [53], photosynthetic index [54], which can be used as an indirect index to identify the drought tolerance of crops. Based on the above analysis, the correlations of the CWSI values with the canopy cover (CC), fruit diameter (FD), and SPAD of grapevines is shown in Figure 7. On May 3, the peaks of CC under the three treatments were 95.32% for T₁, 89.65% for T₂ and 85.61% for T₃ (Figure 7a). The FD of the grapevines maintained a rapid growth trend before May 20, and there was no significant difference among the
treatments (Figure 7c). After May 20, the FD under the three treatments increased with increasing irrigation amount and reached 22.45–24.28 mm at the end of the mature stage. In addition, regarding SPAD, different irrigation amounts had a more significant influence on SPAD from 8 May to 27 June. The maximum SPAD values under T₁, T₂, and T₃ were 43.55, 46.01 and 49.50, respectively (Figure 7e).

As Figure 7b shows, the correlation of the CWSI with CC reached a very significant level (P < 0.01). The current experimental finding agreed with the results of other crops [55,56]. There was a very significant negative correlation between the CWSI and FD (Figure 7d). However, Orta et al. [57] reported that crop morphological size was limited owing to water deficit, and the decrease in crop assimilation was the main reason for the decrease in fruit quantity and diameter at the fruit development stage. There was a very significant negative correlation of the CWSI with SPAD (Figure 7f), and the determination coefficient reached 0.717. By analyzing the relationship of the CWSI and the growth and physiological indexes, we concluded that the CWSI could effectively reflect the water status of grapevines.
3.2.4. Relationship between the CWSI and the Plant Water Status Indicators

Soil moisture is the direct source of plant water. Drought stress will lead to decreases in the soil moisture absorption capacity of the root system and then cause the plant water status to decrease, which is an indirect and adaptive feature of plants to cope with the decrease of soil moisture [58]. To explore the relationship between the CWSI and soil moisture, the correlation between the CWSI and soil moisture at 0–40 cm and 40–80 cm was analyzed. The results showed that the fitting effect of the CWSI and soil moisture (0–40 cm) was sound, and the correlation was significant (\( P < 0.05 \)) (Figure 8a). However, the \( R^2 \) of the CWSI with soil moisture for the deep soil (40–80 cm) was relatively low (Figure 8b) and between 0.123 and 0.403. This indicates that the CWSI more suitably reflects the water status of shallow soil, which is beneficial to understanding the effect of drought stress on the plant water status to a certain extent.

![Figure 8](image)

**Figure 8.** The relationship between the CWSI and the soil moisture of different layers under different treatments. (a) represents the relationship between soil moisture content of 0–40 cm and CWSI; (b) represents the relationship between soil moisture content of 0–80 cm and CWSI. \( T_1 \) represents 100% M, \( T_2 \) represents 80% M, \( T_3 \) represents 60%.

The relationship between plant water status indicators (\( \psi_s, g_s \) and \( E \)) and CWSI were close (Figure 9), and the \( R^2 \) were high (Table 7). Moreover, the relationships between CWSI and \( \psi_s, g_s \), and \( E \) were significant at the level of \( P < 0.01 \), which indicated that CWSI is a suitable indicator for the diagnosis of plant water status. The relationship between \( g_s \) and CWSI was the closest, the \( R^2 \) was between 0.708 and 0.847, whereas the relationship between \( \psi_s \) and CWSI was 0.565–0.730 and 0.569–0.711, respectively (Table 7). When the CWSI is compared to \( T_c - T_a \) as a proxy of the water status (by comparing the \( R^2 \) and \( P \) values of the relationship of both indices to \( \psi_s, g_s, \) and \( E \), Tables 5 and 7), it can be observed that the CWSI more accurately represents the crop water status. Compared to \( T_c - T_a \), the performance of the CWSI is improved by considering the evaporative demand and the species-specific response of the relationship to VPD. These results are similar to the theoretical foundation of CWSI, demonstrating that the change of leaf temperature is mainly determined by the change of \( g_s \) under water stress [59]. Under water stress, the stem and leaf water potential reached the lowest through stomatal regulation, which also explained that the correlation between CWSI and leaf, stem water potential and transpiration rate was not high. Berni et al. [60] reported that for olive trees, there is a linear relationship between CWSI and \( g_s \), the same results were also confirmed in nectarines [61]. The relationship between \( E \) and CWSI was weaker than that with \( g_s \) (Table 7), although the transpiration rate of leaves depended largely on \( g_s \), the reason may be that transpiration rate is not only affected by stomatal conductance but also on the conductivity of the boundary layer [62].
Figure 9. The relationship between CWSI and (a) stem water potential ($\psi_s$), (b) stomatal conductance ($g_s$) and (c) leaf transpiration rate ($E$) for T$_1$, T$_2$ and T$_3$ treatment. The different lines represent the fitting regression lines to the data. T$_1$ represents 100% M, T$_2$ represents 80% M, T$_3$ represents 60%.

Table 7. Regression coefficient and accuracy evaluation of fitting equation between CWSI and plant water status index.

| Treatment | Indicator | $a$    | $b$    | $c$    | $R^2$ | $P$  |
|-----------|-----------|--------|--------|--------|-------|------|
| T$_1$     | $\psi_s$  | -2.801 | -2.110 | -0.045 | 0.730 | $<0.001$ |
|           | $g_s$     | -1.406 | 0.519  | 0.847  | $<0.001$ |
|           | $E$       | -0.087 | 0.688  | 0.663  | $<0.001$ |
|           | $\phi_s$  | 0.069  | -0.560 | 0.298  | 0.702 | 0.001 |
| T$_2$     | $\psi_s$  | -1.612 | 0.696  | 0.708  | $<0.001$ |
|           | $g_s$     | -0.099 | 0.853  | 0.569  | 0.001 |
|           | $E$       | -0.099 | -0.096 | 0.565  | 0.007 |
|           | $\phi_s$  | -0.095 | -0.099 | -0.069 | 0.565 |
| T$_3$     | $\psi_s$  | -3.481 | 0.960  | 0.733  | $<0.001$ |
|           | $g_s$     | -0.103 | 0.945  | 0.711  | $<0.001$ |

Note: “a” represents the coefficient of the quadratic term; “b” represents the coefficient of the first-order term; “c” represents the constant.

In many previous studies to evaluate the applicability of CWSI, $\psi_s$ [39,63] or $\psi_l$ [61] is usually the most suitable indicator to determine the applicability of CWSI as an indicator of water status. However, many studies showed that the relationships between CWSI and plant water status indicators were not always linear, which is consistent with the results of this study (Figure 9a,b). For instance, linear and curvilinear relationships were reported for both grapevine [44,45] and peach [63].

4. Conclusions

It can be concluded that according to different NWSBs, the diurnal change in the CWSI among the three treatments was obviously different, and the best observation time of the CWSI value was 14:00 BJS. The CWSI value can be used to predict the canopy coverage, fruit diameter, and SPAD. Moreover, there was a reliable relationship between T$_c$-T$_b$ and the plant water status indicators; however, the CWSI could more accurately monitor the water stress and assess the water status variability in grapevines in greenhouses. Stomatal conductance had the closest relationship with the CWSI, outperforming other widely used plant water status indicators such as the leaf water potential, stem water potential, and leaf transpiration rate.

Plant water status can be diagnosed by monitoring leaf temperature data. Meanwhile, CWSI is closely related to soil moisture content, it can be combined with plant water status and soil moisture content to guide crop irrigation based on CWSI. The response of plants to CWSI is affected by many factors under water stress, to carry out a successful irrigation schedule based on CWSI, the future research should also consider the influence of cultivation methods, climate conditions and other factors on CWSI, and the threshold of CWSI is further obtained when plants need irrigation under water stress. At present, CWSI is widely used in satellite remote sensing technology. The study of CWSI measured in the field can provide reference for CWSI based on satellite thermal infrared, the combination of
CWSI by the two methods can diagnose plant water deficiency more accurately and predict irrigation schedule in future.

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