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Research Article

ESTIMATING WATER REQUIREMENTS AND REFERENCE EVAPOTRANSPIRATION OF STEVIA IN NORTH-WESTERN MOROCCO

Benhmimou, A[1, 2]*., Ibriz, M[1]., Al Faïz,C[2]., Douaik, A[2]., Zouahri, A[2]., Moussadek, R[2] and Lage, M[2]

[1]Genetic and Biometry Lab, Ibn Tofail University, P.O. Box 133 and Ragional Center of Agricultural Research

[2]Regional Agricultural Research Center, P.O. Box 6356, Avenue Mohamed Belarbi Alaoui, 10101 Rabat, Morocco

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ABSTRACT

The aim of this study was to estimate water requirements for stevia (*Stevia rebaudiana* Bertoni) crop. This research was conducted in the Regional Center of Agronomic Research of Rabat in Morocco (INRA). The data were collected daily at 8 am, from 10 June to 28 August 2014. The crop maximum evapotranspiration (ETm) was obtained from the water balance method, the reference evapotranspiration (ET0) is estimated using a valid FAO-56 Penman-Monteith equation and the pan evapotranspiration (ETp) is calculated by class 'A' pan evaporation. The average maximum evapotranspiration of stevia along the entire cycle of 80 days is 4.36 mm/day with a maximum value of 6.50 mm/day. The FAO-56 Penman-Monteith and Pan evaporation methods showed high correlation with the water balance data with a coefficient of determination of R^2= 0.5548 (slope = 0.432) for ETp and R^2=0.4943 (slope = 0.3001) for ET0. The values of the crop coefficients of three stages (first-stage, second-stage and third-stage) for KCp and KC0 are 0.86, 1.00, 1.15, and 0.83, 0.94, 1.10, respectively. The plant height, stem diameter and dry leaf yield of stevia were 75.33±4.44 cm, 12.33±0.79 and 2.03± 0.38t/ha, respectively. The values obtained during this study allow having better irrigation scheduling and water management.

INTRODUCTION

Stevia is a perennial shrub of Asteraceae (Compositae) family, native to the Amambay region in the north east of Paraguay (Puri *et al.*, 2012). It also occurs in the neighbouring parts of Brazil and Argentina (Madan *et al.*, 2010; Soejarto, 2002). Today its cultivation has spread to other regions of the world, including Canada and some parts of Asia and Europe (Amzad-Hossain *et al.*, 2010). It was introduced in Morocco in 2008 (Aboudrare, 2009). The leaves are the economic part of stevia (Ramesh *et al.*, 2006), with a high concentration of steviol glycosides, possible substitutes of synthetic sweeteners (Ahmed *et al.*, 2007; Ramesh *et al.*, 2006; Santos *et al.*, 2000) which gives stevia a great importance as a natural food sweetener supplier crop (Jarma *et al.*, 2006). The major sweet component present in the leaves of stevia, stevioside, tastes about 300 times sweeter than sucrose (Grammer and Ikan, 2003) that finds applications in food, pharmaceutical industries, and agriculture (Xiangyang *et al.*, 2010). The leaves are found to contain a complex mixture of eight sweet diterpene glycosides, including stevioside, steviolbioside, rebaudioside (A, B, C, D, and E) and dulcoside A (Esmat Abou Arab *et al.*, 2010).

Stevia can, apparently, be successfully grown under different conditions regarding climate and soils (Hajar *et al.*, 2014). The plant is adapted to poor soils, with low nutrient requirements, but for an economic production, crop irrigation is required (Ramesh *et al.*, 2006). However, the amount of active principles depends on total biomass, which further depends of water management (Donalisio *et al.*, 1983).

Stevia can survive in areas of continuous moisture but not withstand the prolonged water logging conditions (Donalisio *et al.*, 1983). Plant growth was optimal at soil water content of 43- 47.6% (Goenadi, 1983). Studies carried out by Shock (1982) revealed that stevia does not require frequent irrigation, but found that it is susceptible to moisture stress and poor quality water.

Goenadi (1983) found that the average water requirement per day was 2.33 mm/plant and experiments carried out in Brazil revealed that the average ETc was measured as 5.75 mm/day,

Corresponding author:* **Benhmimou, A
Genetic and Biometry Lab, Ibn Tofail University, P.O. Box 133 and Ragional Center of Agricultural Research

and water consumption was high during the entire cycle (Goenadi, 1983). The KC plays an essential role in various agricultural practices and it has been widely used to estimate the actual ETc in irrigation scheduling (Pereira *et al*., 1999).

Research at Pisa University (Italy) to estimate the water consumption of stevia using two constant water table micro lysimeters with irrigation levels of 64, 80, 100, and 117 % of evapotranspiration revealed that the ETc for 80 days cycle is 464 mm. The results also indicated that irrigation at 117 % of ETc (538 mm) resulted in 13% higher stevia biomass yield than 100 % of ETc (464 mm) and these values are 113 and 105 % higher than the treatment without irrigation. The corresponding KC values are 1.45, 1.14, and 1.16 (Fronza and Folegatti, 2003). Those values are higher than KC values reported by Gonzalez (2000) in Paraguay, which are respectively of 0.25, 0.56, and 0.85 and by Lavini (2008) based on an experiment using water balance method.

Irrigation scheduled at 1.0 IW/CPE i.e., irrigation at 60 mm cumulative pan evaporation (CPE) resulted in higher fresh biomass; fresh leaf yield and dry leaf yield (Aladakatti *et al*., 2012).

In Moroccan conditions, stevia's yield is still low mainly because no information is available on water consumption on different growing phases. Therefore, the present study was undertaken to estimate stevia ETm for a crop cycle by using the water balance method. In addition, the stevia ETm for irrigation planning, the references ET were determined. Moreover, the FAO-56 Penman-Monteith equation calculated for a grass crop at the weather station and the pan evaporation method were tested against stevia ETm, and the crop coefficient in a sub humid region.

MATERIALS AND METHODS

Experimental site

The study was conducted from 10 June to 28 August 2014, in the Regional Centre of Agronomic Research of Rabat in Morocco (INRA) (34.21 N, 6.40 E, 10.5 m above mean sea-level). The climate in the experimental site is Mediterranean sub humid. The mean annual rainfall is about 523 mm. The mean annual maximum and minimum daily air temperatures are 22 and 11 °C, respectively (Fadil *et al*., 2009).

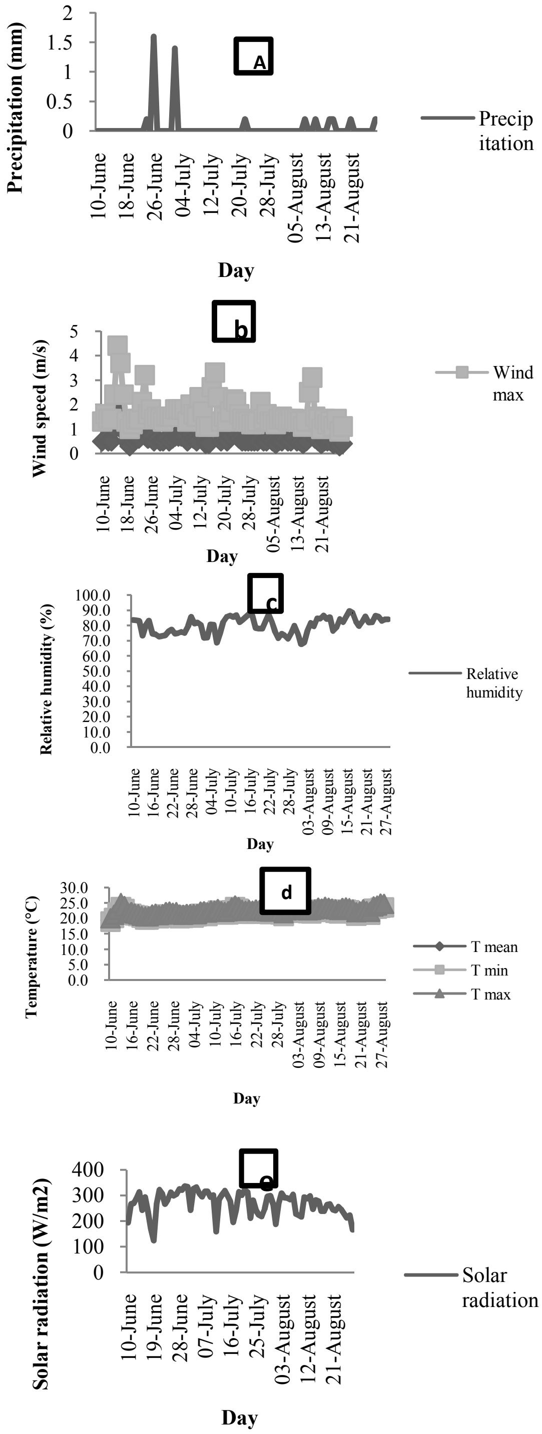
The soil of the experimental site was collected from 0 to 30 cm soil depth before commencement of the experiment and was analysed in laboratory of research of environment and conservation of natural resources INRA, RCAR-Rabat. The soil contained 5. 1% clay, 11.4% silt, and 83.5% sand. The organic matter content was 2.2%, the pH was 7.8 and the N, P and K contents were 120, 106.6 and 154 ppm, respectively. Soil moisture at field capacity was 13.41 % and soil moisture at permanent wilting point was 4.32%. The soil moistures are based on volume.

Plant variety

The variety, INRA, is tested. The sowing was performed on April 1, 2014 on plug trays filled with land and commercial substrate. Plants aged 70 days were transplanted on June 10, 2014 into nine plastic pots, with one plant per pot. Each pot is placed inside a plastic container to collect water drained. Equipments were installed inside the plot measuring 7m×4m (28 m²).

Climatic data

Daily measurements of precipitation, wind speed, relative humidity, air temperature, and solar radiation at a height of 2m during the stevia-growing period are shown in Graph1. These values were measured by an automatic weather station (iMETOS, Pessl Instruments, Austria), located near the experimental site. This data are required for the FAO-56 Penman-Monteith method.



Graph 1 The mean daily climatic data for 80 days of cultivation

Estimation of stevia maximum evapotranspiration

The ETm was determined during growing cycle by water balance method used by Tahi (2008) with a small change (using a plastic container to collect the drained water and a transparent plastic to ovoid water evaporation).

After plot preparation and cover the black plastic mulch, nine plastic pots were installed inside the plot measuring 7m×4m (28 m^2), refilled the same soil and are planted manually; each one is placed inside a plastic container to collect the drained water. Also the containers were covered with transparent plastic to ovoid water evaporation.

The dimensions of the pots are 0.35m in length with an area of 0.141 m^2. Each pot is surrounded by four plants with a spacing of 0.50 m 40000 plant/ha. The pots were irrigated manually using graduated cylinder 250 ml and beaker 1L based. Irrigation was supplied to meet the estimated maximum evapotranspiration of the crop (fully irrigated treatment). Also, the plants surrounding the pots were irrigated similarly. The data were recorded by water volume. After that, a ratio between the consumed water volume and the pots area (0.141 m^2) is estimated to obtain a daily ETm (mm/day).

Daily stevia ETm values for 80 days were generated by weighing the pots and measuring the drained water according to the equation (1):

$$ETm = (P1 - P2) - D \tag{1}$$

where:

$ETm =$ crop maximum evapotranspiration (mm/day),

$P1 =$ pot weight at day's beginning,

$P2 =$ pot weight at day's end,

$D =$ drain water collected.

FAO-56 Penman–Monteith method

Daily values of reference evapotranspiration (ET0) by the FAO-56 Penman–Monteith method (Allen *et al*., 1998) were computed through equation 2:

$$ET0 = \frac{0 \cdot 408\Delta(Rn - G) + \gamma \dfrac{900}{T_{mean} + 273} U_2 (e_s - e_a)}{\Delta + \gamma(1 + 0 \cdot 34U_2)} \tag{2}$$

where Δ is the slope of the saturation vapor pressure (kPa/°C), Rn the net radiation (MJ/m^2), G the soil heat flux (MJ/m^2), T_{mean} the mean temperature (°C), U_2 the wind speed at the 2 m height (m/s),

γ the psychometric constant (kPa/°C) and (e_s-e_a) represents the vapor pressure deficit (kPa).

The net radiation (Rn) was estimated as the difference between net short-wave radiation and the net long-wave radiation. The meteorological data desired, i. e. wind speed, maximum and minimum daily temperature, relative humidity and solar radiation.

Class A evaporation pan method

Reference evapotranspiration by Pan Method was estimated according to the following equation (3):

$$ETp = Kp \times Ep \tag{3}$$

where ETp is the daily reference evapotranspiration (mm), Kp the pan coefficient, and Ep is the pan evaporation (mm) estimated from a class A evaporation pan. The Kp is dependent on the type of pan involved, the pan environment in relation to near by surfaces and the climate. In this environment the value of Kp is 0.8 (Chati, 1991).

The daily values of ETm, ET0, and ETp were measured at 8 am during the stevia-growing period.

Plant data record

Nine plants are harvested at the 80th day after planting, just prior to flowering when steviol glycoside content in the leaves is maximum (Madan *et al*., 2010). The growth and yield parameters are recorded and their mean values are computed. The height of the plant is measured from ground to the base of the fully opened leaf. The diameter of the stem was measured with slide calipers up to 0.01 mm accuracy. Fresh and dry leaf yield was recorded as total biomass yield and expressed in t/ha. Leaves were dried under shade and kept at 50°C in oven for one hour. Dry leaf yield is the economic yield of stevia used for extraction of stevioside (Grammer and Ikan, 2003).

Statistical analysis

Statistical analyses were performed to determine the Coefficient of Determination (R^2) and root mean square error (RMSE) using Excel 2007 and SAS, version 9.1software (SAS Institute Inc., Cary, NC, USA), respectively.

RESULTS AND DISCUSSION

Measured stevia maximum evapotranspiration

The daily values of ETm for stevia during 80 days after planting (from 10 june to 28 august) are showen in Graph 2. This Fig shows the ETm ranges from 3 mm/day to 6.5 mm/day in the first days of growth and in the last days just prior to flowering, respectively. The nine pots had similar measure during the total period. The average total water consumption of stevia during the growing period is 349.30±0.76 mm with a daily average value of 4.37± 0.76 mm/day. The average ETc given by (Fronza and Folegatti, 2003; Gonzalez, 2000), which varies between 5.75 and 6.73 mm/day because of dry season and days hours long during the study, represented 76% and 65% mean daily required in this study, respectively.

The daily ETm was transformed into three developmental stages of stevia: first stage: from 0 to 25 days after planting (DAP); second stage: from 26 to 50 DAP and third stage: from 51 to 80 DAP.

ETm for the first-stage, second-stage, and third-stage are 3.56, 4.32, and 5.07 mm/day, respectively (Graph 3).

In the first stage of growth, water consumption was lower, compared to the second and third stages of high evapotranspiration. This small consumption in the first stage occurred because of the small leaf area of plants (low number of stomata). However, the air temperature, solar radiation and relative humidity assumed values between, 21.17°C, 279.26 W/m^2, and 77.50%, respectively (Graph 1). Vicente (2012) found low initial ET sugarcane by using water balance method in tropical conditions.

The study reported by Fronza and Folegatti (2003) for a Mediterranean region in Italy showed higher results which indicate this study. In this stage, days are 11 hours long with high solar radiation are lower than study reported by (Fronza and Folegatti, 2003); days are 15.3 hours long.

In the second stage of growth, ETm increased to 4.32 mm/day, that increases due to temperature increase up to 22.16°C (Graph 1d). In addition to increase in the leaf area. However, the relative humidity and solar radiation reaching to 80.07% and 274.95 W/m^2, respectively (Graph 1c, e).

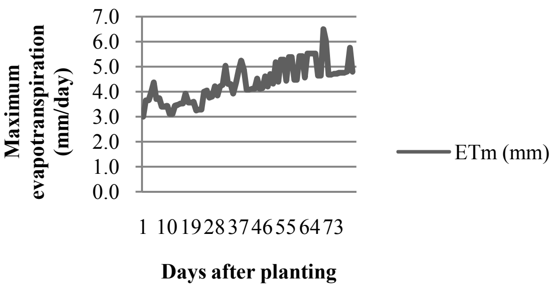
In the third stage of growth, a higher ETm in this stage resulted from lower period rainfall (Graph 5), combined effects of T= 22.59 °C, RH=81.78% and Sr= 255.69 W/m^2 (Graph 1 c, d, e), and development of leaf area, except the last days of this stage (Graph 2) are marked by a step decline in ETm, while leaf area reduce only gradually. Generally, during this stage, the transpiration is driven by an augmentation in stomata vapour conductance.

This tendency was also described by Oliveira *et al* (1993) found similar tendency, detecting good relationships between the crop evapotranspiration and leaf area.

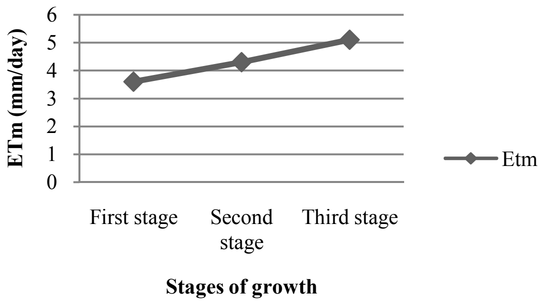
During this study the soil moisture had no limitation effect on the stevia growth, because the water content in the soil was almost at the field capacity.

These values are higher than those obtained in Paraguay (Gonzalez, 2000), and Southern Italy (Lavini, 2008), and lower than those obtained in Italy (Fronza and Folegatti, 2003). This is because the results of these studies were obtained either for other climate conditions or stevia variety.

There are about 90 varieties of *Stevia rebaudiana* developed all around the world depending upon the different climatic requirements (Ibrahim *et al*., 2008; Singh and Rao, 2005).



Graph 2 Variation of daily ETm along the entire cycle of stevia



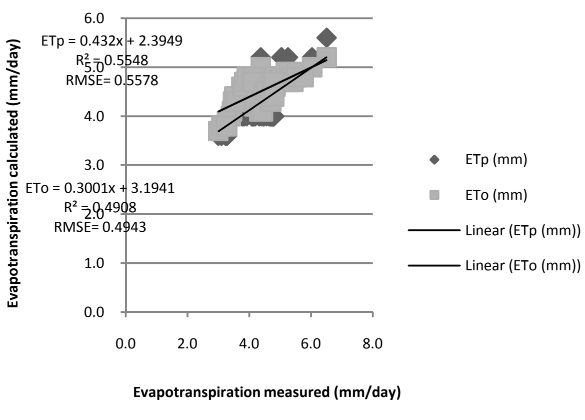
Graph 3 Stevia ETm during the three stages of growth

Statistical comparison of ETm values with ET_0 and ETp values

The Coefficient of Determination (R^2) and root mean square error (RMSE) statistical indicators were used to compare the daily values of ETm using water balance with the values of ETp estimated from Class A pan evaporation and ET0 calculated by Penman–Monteith method.

The results of these comparisons of the above parameters are illustrated in Graph 4. The Both methods gave a good relationship with the water balance ETm values, with significantly high Coefficient of Determination of R^2= 0.5548 (slope = 0.432) for ETp and R^2=0.4943 (slope = 0.3001) for ET0. The values of RMSE of 0.5578 for ETp and 0.4943 also represent a high accuracy. The above results were confirmed by many authors (Pereira and Pruitt, 2004; Sentelhas and Folegatti, 2003) who found a good correlations between daily ETc and those calculated by PM and Pan in Piracicaba (Brazilian state of São Paulo), but their observations were conducted in weighing lysimeters, which are more accurate for daily estimates of ET0 (Allen *et al*., 1998).

The evapotranspiration values over the first, second and third developmental stages are 3.56, 4.32, and 5.07 for the ETm, 4.10, 4.30 and 4.41 for the ETp and 4.27, 4.60 and 4.61 for the ET0, respectively. These last results showed that the values of ETp and ET0 are higher than ETm in the first stage. In this stage, for most of the plants, the ET0 and ETp are higher than ETm because of the initial growing phase of crops with small leaf area and the air temperature was lowest. After 25 days of cultivation (during the second and third stages), ET0 and ETp data presented better correlation with ETm data.



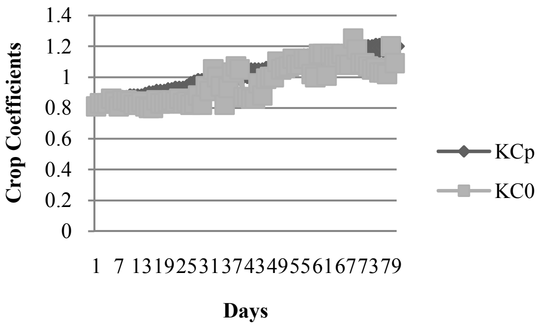
Graph 4 Comparison between the Measured Evapotranspiration and Calculated Evapotranspiration in the axes derived from the Penman-Montheith (ET0) and Pan evaporation (ETp) methods

Measured crop coefficient

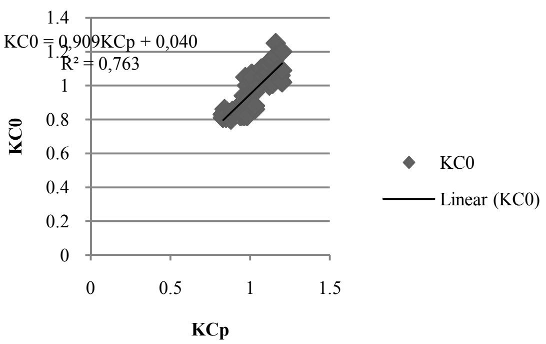
The KC, for a specific crop, was defined by Doorenbos and Pruitt (1974) as the ratio between the evapotranspiration of this crop grown under optimum conditions (ETm) against a reference crop evapotranspiration (ET0). In the present study the daily values of ETm were split by the values of ETp and ET0 to obtain the daily values of KCp and KC0, respectively. The daily values of KCp and KC0 during the growing cycle are given in Graph 5. This graph shows two highest values for KCp and KC0, which were 1.16 and 1.25, respectively. These values were obtained at the 69 day after planting, just prior to flowering when the development of leaf area and augmentation in stomata vapour conductance of stevia, after that a step decline in KCp and KC0 (Graph 5), while leaf area was reduced only gradually. The daily data have been transformed into three values (the first-stage, second-stage and third-stage). These three stages were defined by Gonzalez (2000). The values of three stages for KCp and KC0 are 0.86, 1.00, 1.15, and 0.83, 0.94, 1.10, respectively. These values are higher than the stevia KC found in Paraguay which are respectively for the three stages 0.25, 0.56, and 0.85 Gonzalez (2000) and lower than that obtained by Fronza and Folegatti (2003) which are 1.45, 1.14, and 1.16. During the first stage of stevia growth, KCp and KC0 values were low because the leaf area was small and the transpiration rate was low. During the second and third stages of stevia growth, the highest values of KCp and KC0 were observed. Because, during these stages, the leaf area, the air temperature and the root length, were highest.

This study also shows the relationship between KCp and KC0 (Graph 6). The coefficient of determination of 0.763 (slope= 0.909) confirmed a highly correlated between the KCp and KC0 data during the period of this study.

The variability in KC observed of this study is dependent to variability in factors such as plant age, variety, management system and micrometeorological conditions (Allen *et al*., 1998). Several authors have demonstrated that KC is highly correlated with leaf area (Williams and Ayars, 2005) and ground cover (López-Urrea *et al*., 2009).



Graph 5 Variation of daily KCp and KC0 values



Graph 6 Relationship between the KC0 and KCp

Growth and yield parameters

The plant height and dry leaf yield of stevia were 75.33±4.44 cm and 2.03± 0.38 t/ha, respectively. These values are higher than those find by (Gonzalez, 2000) which are 71 cm and 2.00 t/ha respectively and lower than 86 cm and 4.37 t/ha obtained by Fronza and Folegatti, (2003). Stevia grows up to 1 m tall (Mishra *et al*., 2010). The higher height and yield are justified by long days (15.9 h), solar radiation, and location of the experiment area (43ºN), close to the maximum incoming radiation for the study realized by (Fronza and Folegatti, 2003). The stem diameter was 12.33±0.79 mm and is lower that find by Lidia *et al* (2015) which was 45 mm. The variability in these parameters is dependent to variability in micrometeorological conditions, soil type and variety.

CONCLUSIONS

This study estimated crop coefficient values for stevia in a Mediterranean region, Morocco, using ETm measured by water balance method, ETp measured from Class A pan and ET0 measured by Penman–Monteith method. The results obtained in this study show that stevia crop needs an average amount of water during the three stages of growth, especially during the second stage (from 26 to 50 DAP) (4.32 mm/day). The quantity needed during that period increased slightly during the next stage. The average stevia KC; KCp and KC0 for this growth cycle corresponded to 0.86, 1.00, 1.15, and 0.83, 0.94, 1.10, respectively. Deferent results than the stevia KC found in Paraguay (Gonzalez, 2000), and Italy (Fronza and Folegatti, 2003) which are 0.25, 0.56, and, 0.85 and1.45, 1.14, and 1.16, respectively. This is because the results of those studies were obtained either for other climate conditions or stevia varieties. The values obtained during this study allow having better irrigation scheduling and water management.

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