




## Article

# Water Use, Growth, and Yield of Ratooned Guayule under Subsurface Drip and Furrow Irrigation in the US Southwest Desert

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**Abstract:** Guayule (*Parthenium argentatum*, A. Gray) is a perennial desert shrub with ratoon-cropping potential for multiple harvests of its natural rubber, resin, and bagasse byproducts. However, yield expectations, water use requirements, and irrigation scheduling information for ratooned guayule are extremely limited. The objectives of this study were to evaluate dry biomass (DB), contents of rubber (R) and resin (Re) and yields of rubber (RY) and resin (ReY) responses to irrigation treatments, and develop irrigation management criteria for ratooned guayule. The water productivity (WP) of the yield components were also evaluated. Guayule plants that were direct-seeded in April 2018 were ratooned and regrown starting in April 2020, after an initial 2-year harvest at two locations in Arizona: Maricopa and Eloy on sandy loam and clay soils, respectively. Plots were irrigated with subsurface drip irrigation (SDI) at 50, 75, and 100% replacement of crop evapotranspiration (ET<sub>c</sub>), respectively, and furrow irrigation at 100% ET<sub>c</sub> replacement, as determined by soil water balance measurements. The Eloy location did not include the 100% irrigation treatment under SDI due to unsuccessful regrowth for this specific treatment. The irrigation treatments at the locations were replicated three times in a randomized complete block design. After 21–22 months of regrowth, the guayule plants were harvested in plots. The results showed that DB increased with the amount of total water applied (TWA, irrigation plus precipitation), while R and Re were reduced at the highest TWA received at both locations. Ultimately, the SDI treatments with 75% ET<sub>c</sub> replacement resulted in the best irrigation management in terms of maximizing RY and ReY, and WP for both locations and soil types. Compared to the initial 2-year direct-seeded guayule crop, ratooned guayule required less TWA and attained higher DB, RY, and ReY, as well as higher WP, with average increases of 25% in dry biomass, 33% in rubber yield, and 32% in resin yield. A grower's costs for planting the initial direct-seeded guayule crop would be offset by the additional yield revenue of the ratooned crop, which would have comparatively small startup costs.

**Keywords:** irrigation management; evapotranspiration; soil water depletion; rubber yield



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## 1. Introduction

Guayule (*Parthenium argentatum*, A. Gray) is a native perennial shrub in the desert of northcentral Mexico and southwestern Texas and can be exploited for its high-quality

natural rubber [1], hypoallergenic latex [2], resin [3,4], and other valuable byproducts. According to Wang et al. [5], rubber can be derived from different parts of the plant, with the plant stem comprising 55–65% of the dry biomass, while the remaining portion of the dry biomass is attributed to roots and branches. Methods to increase guayule production have been developed, including selective breeding [6], appropriate rubber and latex extraction techniques [7], procedures to generate valuable byproducts from non-rubber material [8,9], and improvement of physiological aspects to increase yield [10].

Guayule is a xeric shrub with roots capable of extracting moisture from deep soil profiles. It withstands drought and dehydration by entering into a semi-dormant state [11,12]. Based on initial research conducted during World War II, the National Academy of Science [13] reported that total water applications (TWA, irrigation plus precipitation) in excess of 640 mm yr<sup>-1</sup> could cause excessive vegetation growth and hinder rubber formation. Subsequent research has shown that substantial amounts of water are needed for crop establishment and that guayule production benefits from greater application amounts than reported by [13]. Studies conducted in the Arizona desert have shown that between 470 and 635 mm of water are needed for crop establishment if using primarily surface irrigation [11,14]. Working with transplanted guayule, [11,15,16] found that dry biomass, rubber, and resin yield were significantly reduced after two years of growth in a loam soil when TWA was less than ≈1400 mm yr<sup>-1</sup>. In contrast, yield components were maximized when TWA was 1550 mm and 2300 mm during the first year and the second year of growth, respectively. Very similar water requirements (1600 and 2300 mm for the first and second year, respectively) were determined more recently on studies conducted with transplanted guayule on sandy clay loam soil [14,17] and direct seeded guayule on sandy loam and clay soils [18]. Altogether, studies show a high linear correlation between water application and dry biomass and resin yield [16,17,19–22]. However, since shrub rubber content generally increased in water stressed treatments, rubber yield was less correlated with TWA [17,22]. Thus, water application must be optimized to achieve higher rubber and resin yields.

Irrigated crop water productivity (WP) is defined as the ratio of crop yield to the total water use [23]. Currently available data show that WP for guayule rubber yield varies with irrigation method, TWA, total crop evapotranspiration (ET<sub>c</sub>), and soil type [14,16–18,20]. A 2.5-year study conducted on sandy clay loam soils [14,17] compared water productivity of rubber yield using five irrigation treatments, with total water application varying from 2080 to 4910 mm, and two irrigation methods, subsurface drip and level furrow. Water productivity was 25% greater at low water application and 94% greater at high water application with subsurface drip irrigation (SDI) than with level furrows. In contrast, water productivity was the same for the two irrigation methods when the crop was irrigated at 100% ET<sub>c</sub> replacement but only for the tests conducted on a sandy loam soil. Productivity was significantly greater with furrow irrigation than with SDI on a predominately clay soil [18]. Furthermore, in the sandy loam soil, RY-WP was not increased with less TWA compared to the 100% ET<sub>c</sub> treatment, while it was significantly increased when TWA was less than that of the 100% ET<sub>c</sub> treatment in the clay soil [18]. This result was similar in a later study on the same clay soil that showed RY-WP was significantly greater when TWA was reduced by 33% compared to the fully irrigated treatment.

A limiting factor for guayule commercialization is the high cost of planting guayule seeds in greenhouses and transplanting seedlings to the field [24–28]. In the early 2000s, the estimated cost to transplant one hectare of guayule was 1600 USD [29], or about 2400 United States Dollars (USD) in present prices. The cost of guayule establishment can be reduced by using the direct-seeding method [25]. Therefore, direct-seeded guayule combined with appropriate irrigation methods could be a potential alternative to transplanting seedling supplies from nurseries, as successfully demonstrated by Bridgestone Americas, Inc. in the southwest US [5,18]. Although direct seeding is possible, the process of conditioning guayule seeds and the choice of appropriate planting depths [18,30,31], as well as the application of frequent light irrigations [24,31,32] are critical for successful germination and growth establishment.

Ratooning, the practice of allowing guayule shrubs to regrow after an initial harvest, has been recommended for commercial guayule production [27,29]. Studies have shown that guayule rubber yield is the same or better with a ratooned crop than with a non-ratooned crop over the same time period [13,27]. Ratooning after an initial two-year growth period and harvest provides an earlier return on the crop establishment startup costs, as those costs are eliminated after the first year, as well as additional rubber yield. For direct-seeded guayule, initial investments include costs for land and seedbed preparation, precision planting operations, and, possibly, costs associated with installing a portable sprinkler irrigation system to provide the high-frequency soil wetting required to promote germination, prevent soil crusting, and facilitate crop emergence [29]. Also, initial irrigation amounts needed to reestablish the growth of ratooned guayule are less than those needed for the reseeded field. Furthermore, pre-plant irrigation may be needed when reseeding to leach salts below the seedbed depth. Current recommendations for the optimal timing of the initial guayule harvest are after  $\approx$ two years of growth, and shrubs are typically cut at 0.10 m above the soil surface [8,27]. A literature review did not reveal data on yield response to irrigation and corresponding irrigation management strategies for ratooned guayule crops, whether by transplants or direct seeding.

The present paper is a follow-up analysis of the direct-seeded guayule irrigation studies initiated in Apr. 2018 at two locations in Arizona [18]. Those guayule plots were harvested after approximately two years and allowed to regrow. Since the initial study revealed no yield and water productivity benefit for irrigation treatments beyond 75–100%  $ET_c$ , the follow-up study used the same treatment strategy as the original study but excluded the higher  $ET_c$  replacement rates. Thus, the objective of the present paper is to evaluate growth and yield responses to irrigation scheduling and total water applied for the ratooned direct-seeded guayule. The specific objectives are to (1) determine the optimal irrigation management rates for the sandy loam and clay soils, (2) assess the differences in  $ET_c$ , TWA, yield, and WP between the initial and ratooned crop, and (3) determine whether the initial and ratooned guayule crops should be managed differently.

## 2. Materials and Methods

### 2.1. Experimental Layout

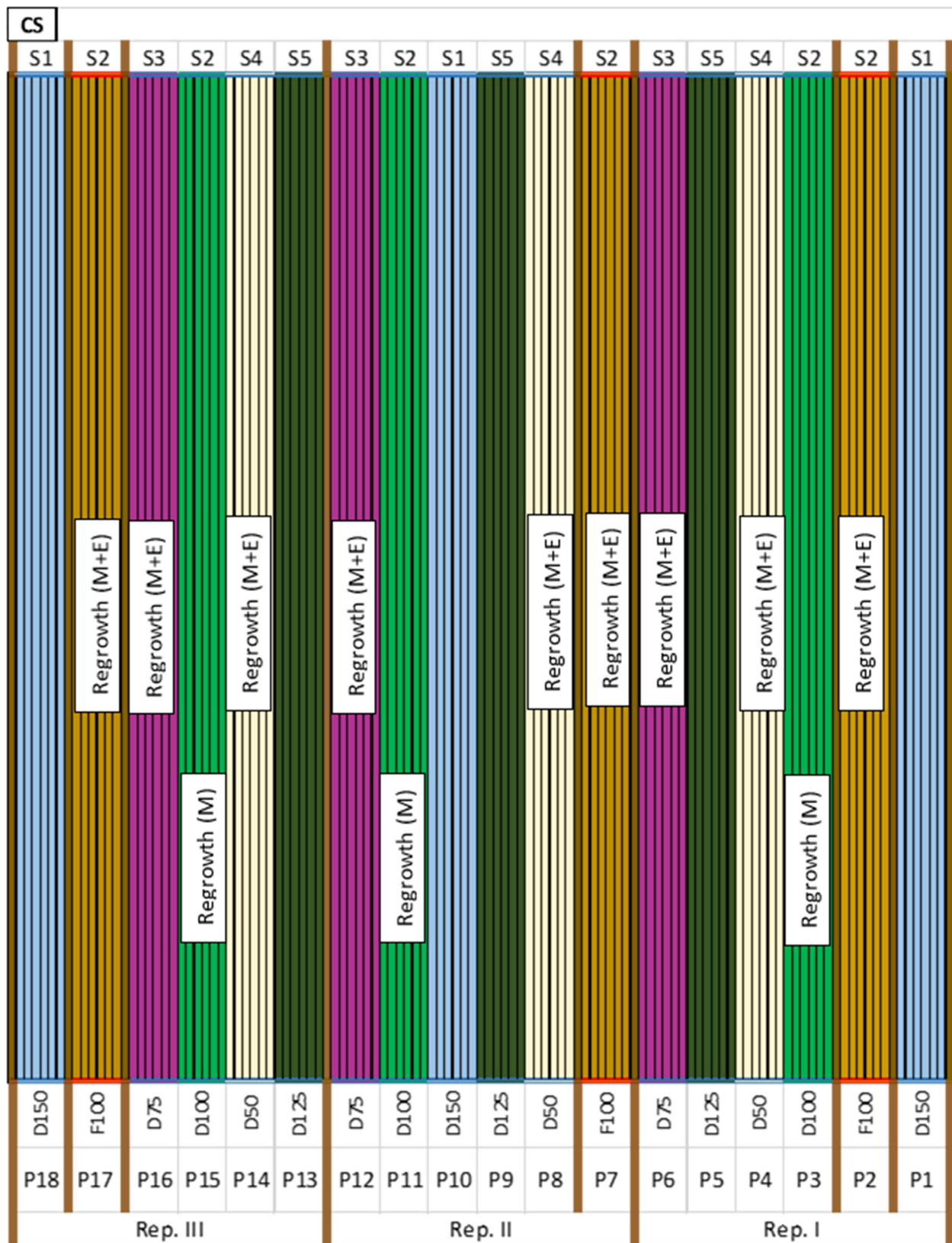
The follow-up study with ratooned guayule was conducted starting in April 2020 and ended in February 2022. The experimental setup is similar to that of the initial direct-seeded study. Details about planting and crop establishment for the initial study are reported in [18]. The experiments were conducted in fields at: (1) The University of Arizona, Maricopa Agricultural Center (MAC) farm, in Maricopa, Arizona (33.07° N lat; 111.97° W long; 361 m a.s.l.), and (2) Bridgestone Americas, Inc., Guayule Research Farm in Eloy, Arizona, USA (32.67° N lat; 111.63° W long; 482 m a.s.l.). Both fields included 18, 75 m long plots that were 6.1 m wide (six rows) at Maricopa and 8.1 m wide (eight rows) at Eloy. The soil at the Maricopa field belongs to a Casa Grande series [33] that includes various sandy textures, with high to moderate water permeability and moderate water holding capacity. The Eloy field-site is mapped as a Gadsden series [34], which has predominantly clay or silty clay loam, and relatively high water holding capacity but low permeability through the deep soil profile. Measured soil texture fractions and estimated soil water properties of the profiles at the two field sites are presented in [18]. Following the harvest of the initial direct-seeded study, plants in those plots were harvested to  $\approx$ 0.10 m above the soil surface in early April 2020 at Maricopa and early May in Eloy.

The split-plot design for the initial 2018–2020 experiments included location as the main plot and irrigation treatments as the split-plots [18]. Within each location, the irrigation treatments were randomized in a complete block design, with three blocks and six irrigation treatments (Figure 1). Five of the irrigation treatments, identified as D50, D75, D100, D125, and D150, were subsurface drip irrigated and received 50–150% of the calculated  $ET_c$ . The sixth treatment was furrow irrigated (F100) and received 100%  $ET_c$  replacement. A similar split-plot design was also adopted for the regrowth experiments

(Figure 1). As previously noted, in the initial study the higher  $ET_c$  replacement treatments produced significantly lower rubber yield and productivity. Hence, the higher replacement treatments were not included in the regrowth evaluation, but the plots were maintained with irrigation to provide adjacent canopy cover for the experimental treatment plots. Thus, treatments reported herein at Maricopa included three SDI treatments (D50, D75, and D100) that received three respective levels of irrigation (50, 75, and 100% replacement of  $ET_c$ ) and one furrow irrigated treatment (F100) at 100%  $ET_c$ . The same treatments were established at Eloy with the exception of D100, which not only had poor regrowth but also had significantly lower water productivity in the harvest of the initial crop than the D50 and D75 treatments in the heavy clay soil. Guayule plants in the D100 treatment (as well as D125 and D150) in the initial study were overgrown with high biomass yield, which likely reduced regeneration rate and resulted in lower ratooned stand. The initial direct-seeded and regrowth experimental layouts are both shown in Figure 1. The guayule was grown from USDA germplasm line AZ-2 seeds (PI 599675) [35], planted in rows spaced 1.02 m apart using a precision vacuum planter, as described by [18]. Irrigation was applied to all treatment plots to activate plant growth, starting on 16 April 2020 at Maricopa and 8 May 2020 at Eloy. The studies ended at both locations in mid-February 2022, when plots were harvested, as described later in Section 2.5.

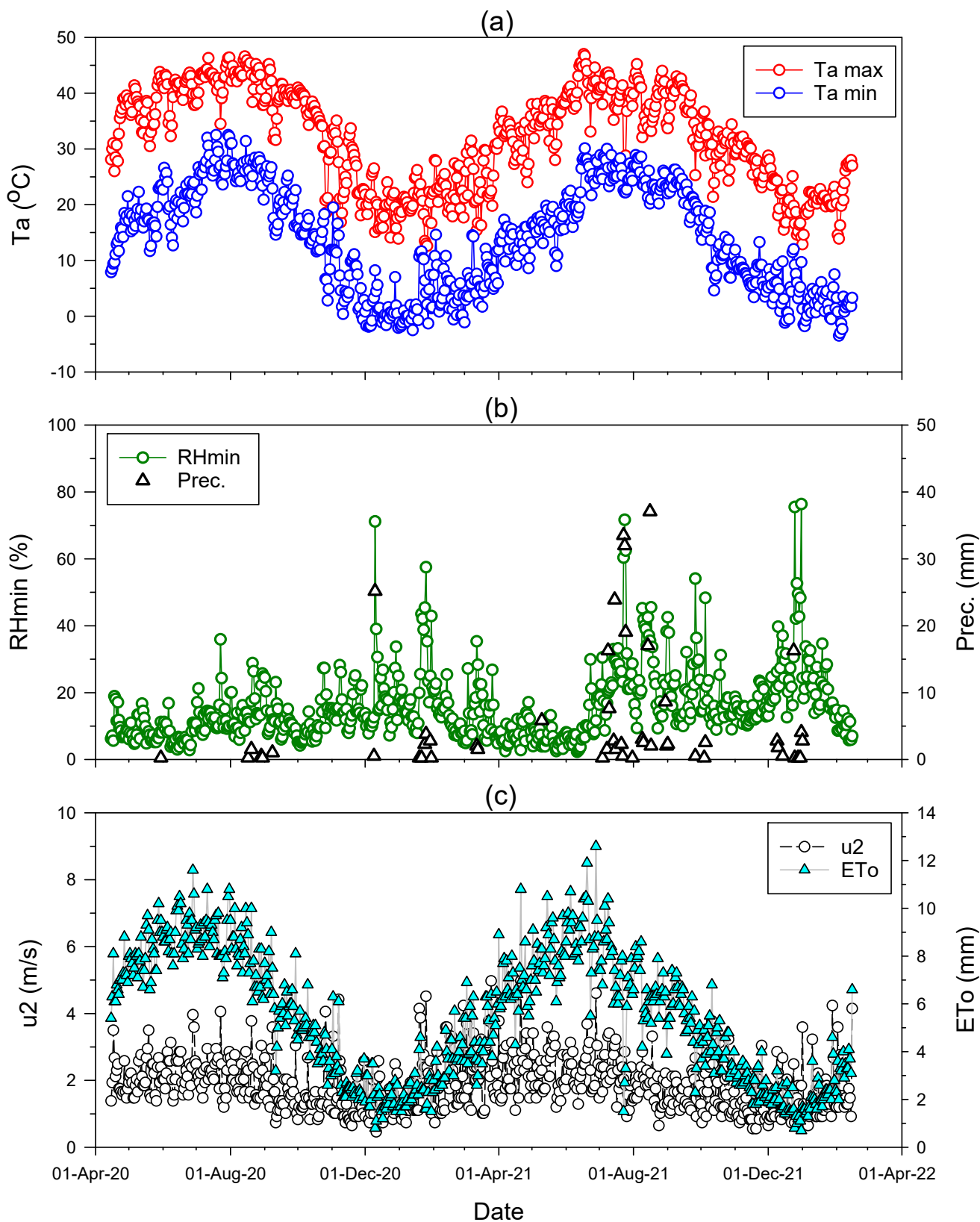
## 2.2. Meteorological Data

Meteorological data for the study years were collected from the Arizona Meteorological Network (AZMET; <https://cals.arizona.edu/AZMET/index.html> [accessed on 16 April 2020]). The MAC AZMET station is located 200 m from the Maricopa field site, while for Eloy the closest station is Coolidge, Arizona (32.98° N lat; 111.61° W long; 423 m a.s.l.), located about 30 km north of the site. Elshikha et al. [22] presented mean daily historical data (2003 to 2021) recorded at the Coolidge AZMET, including maximum and minimum air temperatures ( $T_a$  max and  $T_a$  min, respectively), minimum relative humidity (RHmin), precipitation, windspeed at 2 m height ( $u_2$ ), and the standardized Penman–Monteith grass-reference evapotranspiration ( $ET_o$ ) [36]. The daily  $T_a$  max and  $T_a$  min (Figure 2a), RHmin and precipitation (Figure 2b), and  $u_2$  and  $ET_o$  (Figure 2c) are shown for the ratooned studies at Maricopa (15 April 2020 through 15 February 2022) and at Eloy (1 May 2020 through 15 February 2022) in Figure 3a–c, respectively. The 2020 summer (21 June 21 to 20 September) following ratooning was extremely hot and dry, where daily  $T_a$  max averaged 42.0 °C at Maricopa and 41.0 °C at the higher elevation in Coolidge/Eloy. During that period,  $T_a$  max was above 43.3 °C on 38 and 20 days and the RHmin averaged 12.9 and 13.0%, respectively, at Maricopa and Eloy. In contrast, the summer of 2021 was somewhat cooler and much wetter. In that summer, daily  $T_a$  max averaged 39.2 °C at Maricopa and 38.1 °C at Eloy, where  $T_a$  max above 43.3 °C only occurred on only 15 and 3 days, respectively. While recorded precipitation was 3.6 and 25.9 mm at Maricopa and Eloy, respectively, in summer 2020, it was 216 and 153 mm at the locations in summer 2021, respectively. Historical AZMET data for summer at the Maricopa and Coolidge/Eloy stations indicate that  $T_a$  max averages about 39.6 and 39.1 °C, respectively [22,37], while summer precipitation totals on average 59 and 57 mm, respectively. Thus, the primary growth-producing periods for the ratooned guayule were hotter and drier than normal during the first summer and slightly cooler but much wetter than normal during the second summer. The cumulative precipitation and cumulative  $ET_o$  for the regrown guayule crops were 297 and 3661 mm at Maricopa, respectively, and 268 and 3348 mm at Eloy, respectively.

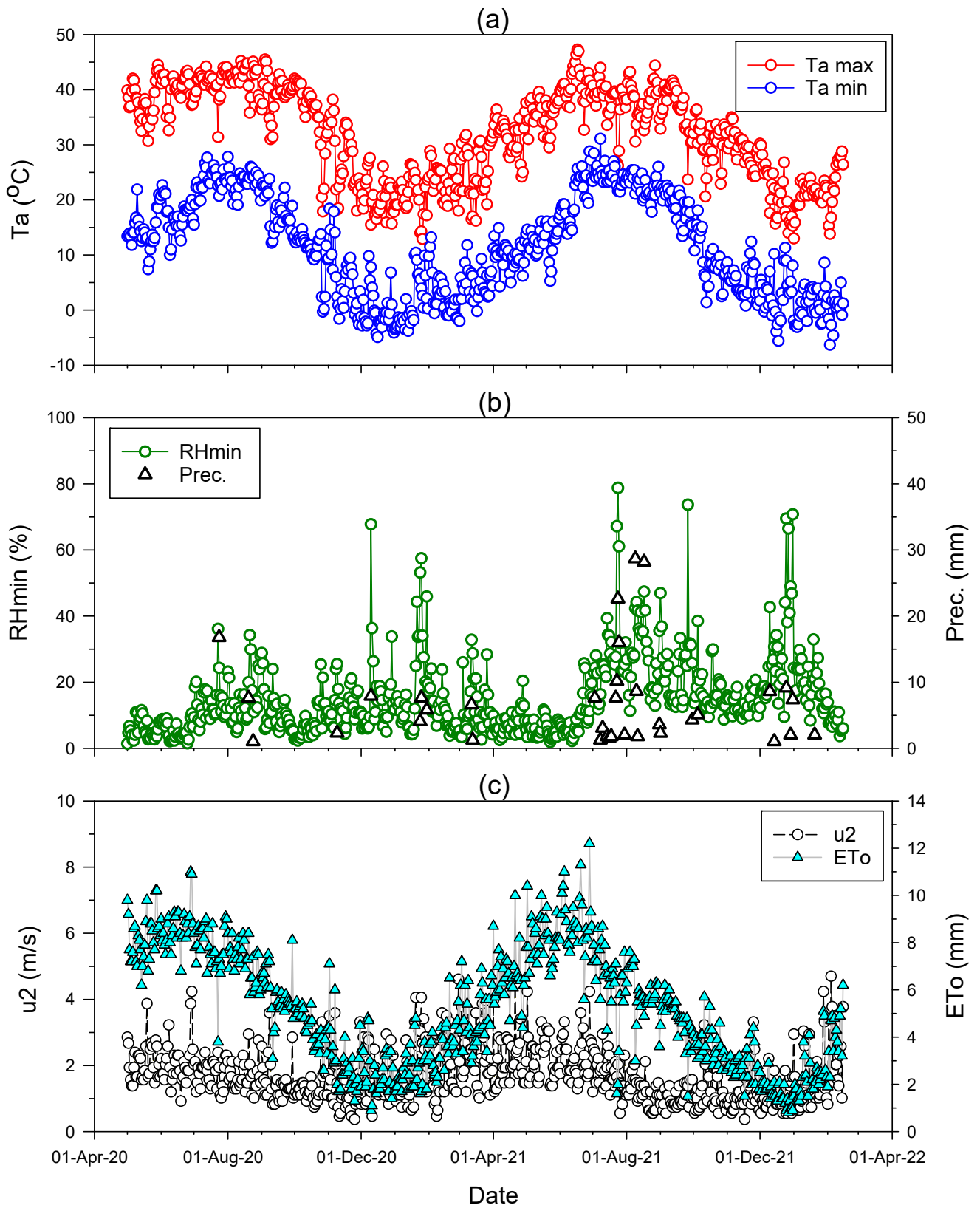


**Figure 1.** Field layout of irrigation treatments in initial direct-seeded studies (D50–D150 and F100) at Maricopa (M) and Eloy (E) and treatments in regrowth studies (D50–D100 and F100 at M and D50–D75 and F100 at E). Field blocks are denoted as (Reps. I–III). The control stations (CSs) for the subsurface drip systems were located at the NE corner of the fields.





**Figure 2.** Daily values of maximum and minimum air temperatures (Ta max and Ta min) (a), minimum relative humidity (RHmin), precipitation (b), windspeed at 2.0 m height (u2), and grass-reference evapotranspiration (ET<sub>o</sub>) (c) from the AZMET station in Maricopa for the guayule regrowth study period from 2020 to 2022.



**Figure 3.** Daily values of maximum and minimum air temperatures (Ta max and Ta min) (a), minimum relative humidity (RHmin), precipitation (b), windspeed at 2.0 m height (u2), and grass-reference evapotranspiration (ET<sub>0</sub>) (c) from the AZMET station in Coolidge, near Eloy, for the guayule regrowth study period from 2020 to 2022.

### 2.3. Irrigation Scheduling and Crop Evapotranspiration

Irrigation scheduling for the regrowth treatments was based on  $ET_c$  replacement and targeted soil water depletion (SWD) within the guayule root zone ( $Z_r$ ). The procedures described in [18] (Section 2.5) for computing a soil water balance (SWB) based on soil water content and irrigation/precipitation measurements were used. For the regrowth study,  $Z_r$  was assumed as 2.0 m from the start to the end of the regrowth period. The soil water parameters used in computing the SWB for the regrowth treatments were those previously measured at the two locations by [18], where field capacity (FC) and permanent wilting point (PWP) volumetric soil water contents were 27.1 and 14.4% at Maricopa, respectively, and 39.6 and 24.2% at Eloy, respectively. The total available water (TAW) of the guayule root zone was computed as:

$$TAW = 10 Z_r (FC - PWP) \quad (1)$$

where:

TAW is in mm,  $Z_r$  is 2.0 m, and FC and PWP were the water content values (% basis) for the two sites, shown above.

At the two locations, a neutron moisture meter (NMM) was used to measure the volumetric soil water content. Readings were obtained from 0.15 m to 1.95 m below the soil surface in 0.30 m increments in each treatment replicate [18]. Soil water content was measured approximately at 1–2-week intervals during spring through fall, and about every 3 weeks during the crop's winter dormancy period when irrigation was not applied. Soil water was last measured on 31 January 2022, at both locations, about two weeks prior to plant harvest sampling. To compute the guayule crop ET by the SWB, the incremental water content depth readings on each date were averaged over  $Z_r$  and then converted to depletion ( $D_r$ ) in units of mm:

$$D_r = 10 Z_r (FC - \text{avg soil water content}) \quad (2)$$

where:

$D_r$  is in mm,  $Z_r$  is 2.0 m, and FC and average soil water content values are on a % basis. Thus,  $D_r$  is equal to zero when soil water content is at FC and is equal to TAW when soil water content is at PWP.

For all treatments, the  $ET_c$  was calculated for periods measured at two adjacent NMM dates by the SWB (Equation (3)):

$$ET_c = (D_{r,2} - D_{r,1}) + I + P - DP \quad (3)$$

where:

$ET_c$  is the total ET (mm) that occurred in the period from the first (1) to second (2) measurement date,  $D_{r,1}$  and  $D_{r,2}$  are the average treatment measured depletion (mm) on the first and second date, respectively,  $I$  (mm) is average total treatment irrigation,  $P$  (mm) is the total precipitation (excluding days when  $P < 1.5$  mm), and  $DP$  is the deep percolation during the period. For all treatments,  $DP$  in Equation (3) was assumed as zero because measured  $D_r$  never exceeded FC on any measurement date.

The irrigation treatments pursued different soil water depletion targets depending on site and water application method, which were based on measured SWD expressed in terms of TAW:

$$SWD = 100 \times [1 - (TAW - D_r)/TAW] \quad (4)$$

where:

SWD is in percent, TAW is in mm, and  $D_r$  (mm) is measured depletion in mm.

At Maricopa, the allowable SWD for the D100 subsurface irrigation treatment was limited to 35% of TAW and each irrigation replaced 100% of the calculated  $ET_c$ . The D50 and D75 treatment irrigations replaced, respectively, 50 and 75% of the D100  $ET_c$ . The furrow treatment (F100) at Maricopa limited depletion to 55% of the TAW and replaced



100%  $ET_c$  with irrigation. At Eloy, both the D75 and F100 irrigation treatments targeted a 55% allowable SWD and irrigations replaced 75% and 100% of  $ET_c$  for the two treatments, respectively. Irrigation applied to the D50 at Eloy was 67% of that applied to the D75. The irrigation treatments were applied in both years during the period April–November and no irrigation was applied during the winter and early spring months, when guayule plants were semi-dormant.

At Maricopa, the three SDI and furrow treatments received differential irrigation applications starting on 16 April 2020. However, at Eloy, the two SDI treatments received equal irrigation from May 8 through 31 May 2020, which was applied to the plots by three furrow irrigations. Furrow irrigation was used because the SDI system was not operable at that time. Differential irrigation applications for the D75 and D50 at Eloy, thereafter, commenced using SDI on 17 June 2020. Irrigation applied to the F100 treatment at Eloy was applied according to the F100 SWB.

#### 2.4. Irrigation System and Fertilization

The SDI and level furrow irrigation systems were designed and set up at both experimental sites in 2018 [18]. The SDI drip tape was buried 0.20 m below the soil surface in the center of each bed. At Maricopa, a 51 mm diameter main line buried 0.75 m below the surface supplied water to all SDI treatment replicates. At Eloy, each treatment replicate had a separate station and main line. Each irrigation station was equipped with a solenoid valve, a flow meter, a pressure regulator and gauge, and an air vent. Water was delivered to each level furrow plot with a riser valve, with the applied volume measured with an in-line flow meter. An end-berm prevented any runoff from these plots.

Fertilizer was applied each year in the form of urea–ammonium–nitrate (32% N) at a rate of 65 kg N ha<sup>-1</sup>. The fertilizer was injected in the irrigation water using a single head hydraulic diaphragm chemigation injection pump. The pump was connected, through injection ports, to the mainline of the SDI system and to an aluminum pipe delivering the water to the furrow plots. All treatment plots were fertigated in mid-September 2020 and early July 2021 at both locations.

#### 2.5. Plant Growth Measurements and Yield Assessment

Crop measurements included crop height, canopy cover, and final biomass. Crop height and crop width needed to determine canopy cover were sampled about once per month using three 1.0 m<sup>2</sup> sections selected along the inner four crop rows within each experimental plot. The canopy cover fraction ( $f_c$ ) was estimated from measured canopy width using the formula presented in [18]. Plant biomass was sampled in mid-February 2022 by hand-harvesting the plants from the ground level, from three, 3.0 m<sup>2</sup> sections within the inner rows of each plot. Thus, the ratooned guayule was allowed to regrow for 22 and 21 months at Maricopa and Eloy, respectively. The moisture content of the harvested biomass was measured by air-drying, grinding, and oven-drying the plant samples [8,19]. After complete drying, the rubber (R) and resin (Re) contents were determined using a Soxhlet-based near-infrared spectroscopy (NIR) method [10,18]. Rubber and resin yield were determined from the biomass yield and rubber and resin content. The water productivity for DB, RY, and ReY were then calculated using Equation (5):

$$WP \text{ (kg m}^{-3}\text{)} = \text{yield (kg)}/\text{total water applied (m}^3\text{)} \quad (5)$$

where total water applied (TWA) is the sum of irrigation and precipitation received by the plants during the two years of regrowth.

#### 2.6. Statistical Analysis

The statistical procedures described by [18] were adopted for the present analysis. A split-plot model was used, considering the experiment locations as main plot treatment with 2 levels, and irrigation as the split-plot treatment with 3 (Eloy) and 4 (Maricopa) levels. Using values from the replicated samples, statistical analyses of R, Re, DB, RY, ReY,

as well as the WP for DB (DB–WP), RY (RY–WP), and ReY (WP–ReY), were performed with the Proc Mixed procedure (SAS v.9.4, Institute Inc., 2016, Cary, NC, USA). Both location and irrigation treatments, as well as their interaction, were considered as fixed effects, while block and block  $\times$  location were random effects. Proc Mixed estimated the random components and the residual by the residual maximum likelihood (REML). When location, treatment, or interaction F tests were significant ( $p < 0.05$ ), least square means were separated using the *P* diff option in SAS (with  $p < 0.05$ ).

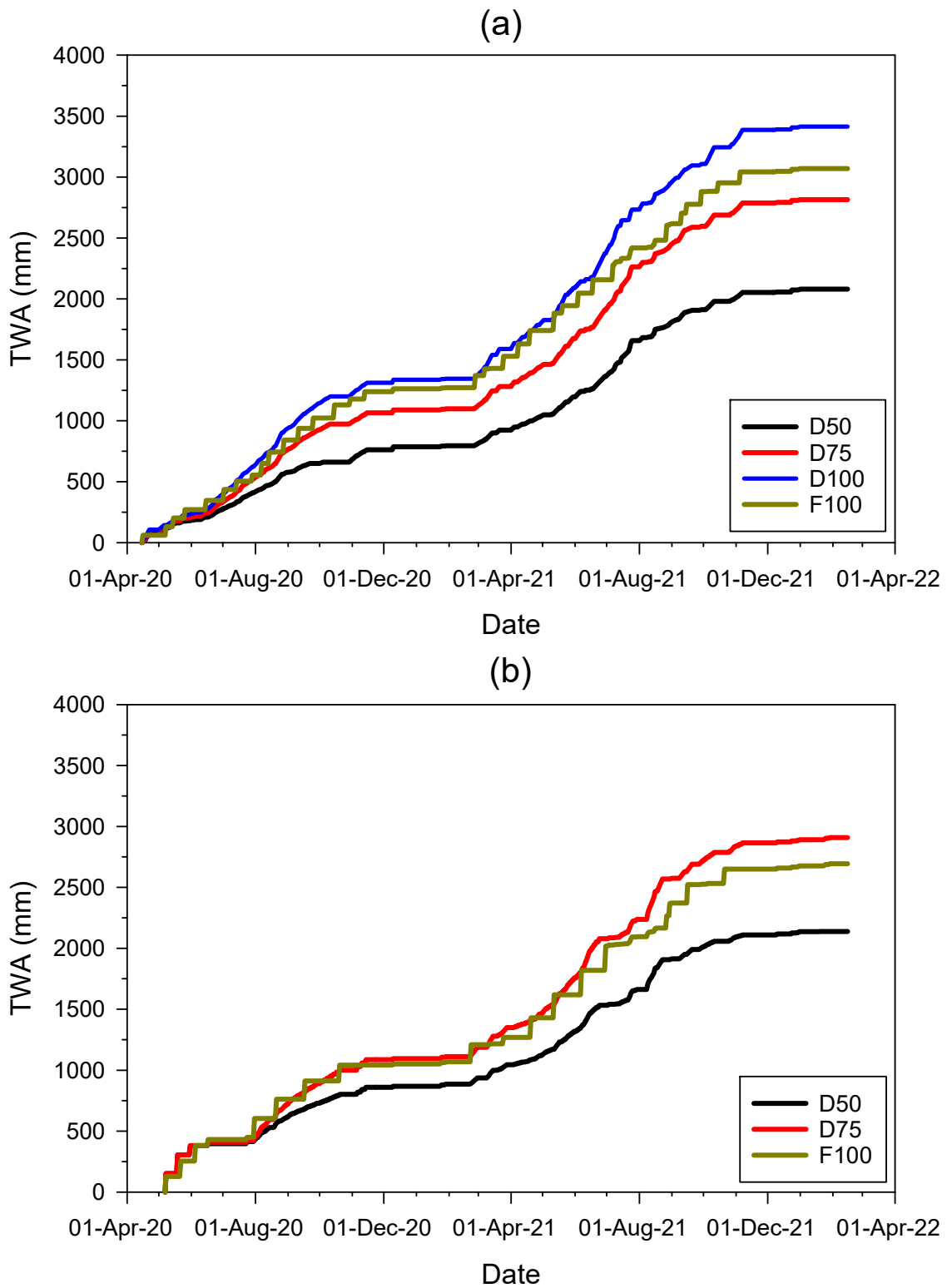
### 3. Results

#### 3.1. Irrigation and Crop Growth

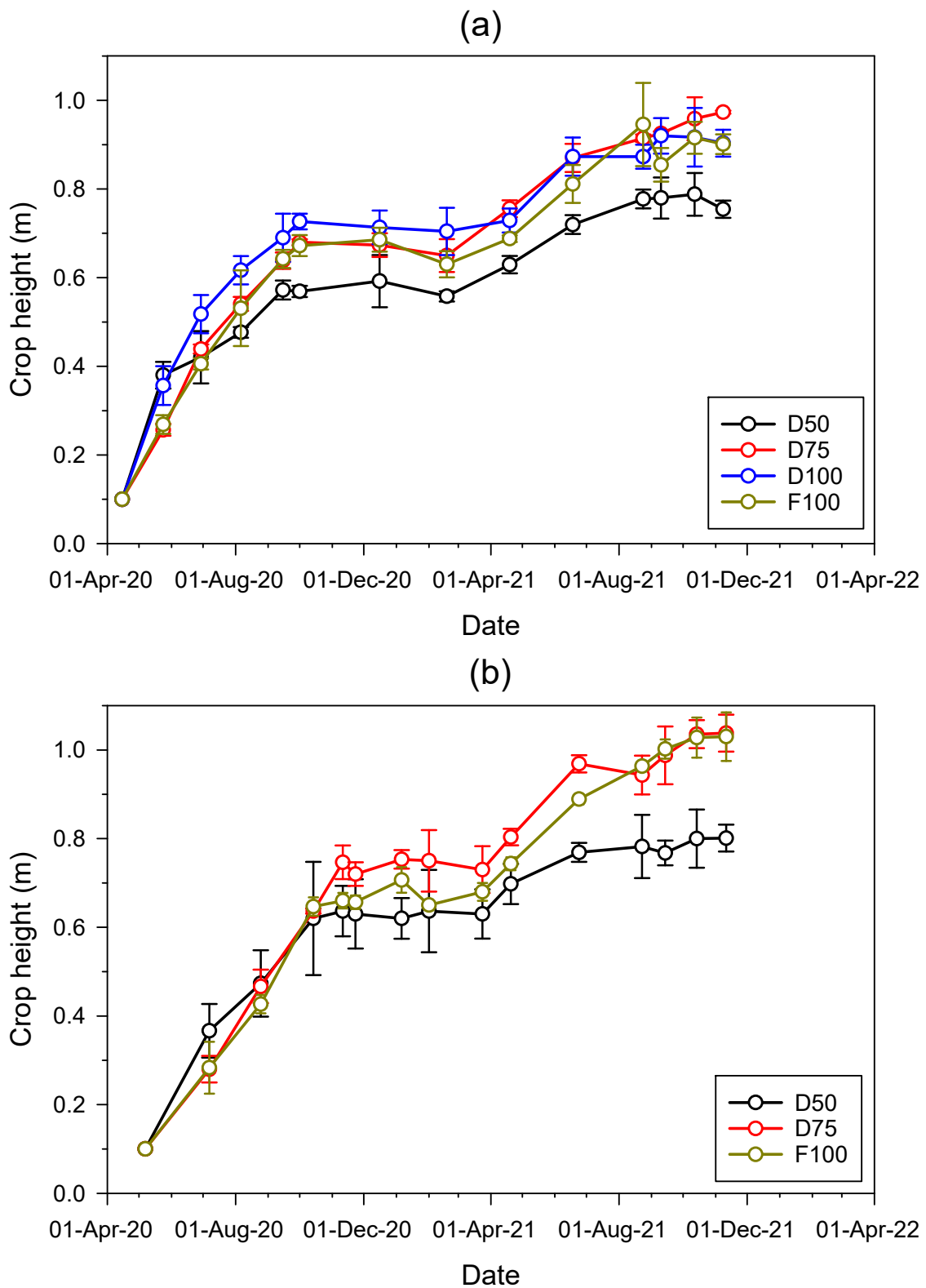
The cumulative TWA, which includes irrigation and precipitation, is shown with date for each treatment at Maricopa (Figure 4a) and Eloy (Figure 4b). No irrigations were applied at any location after early November 2021. The cumulative effective precipitation ( $>1.5$  mm per day) was 290 and 264 mm at Maricopa and Eloy, respectively. The treatment differences in TWA were more pronounced at Maricopa than at Eloy during the first year of regrowth (Figure 4a,b). At Eloy, guayule regrowth for all treatments during summer 2020 lagged Maricopa, as illustrated by crop height and  $f_c$  data for treatments at the two locations (Figures 5 and 6, respectively). For example, measured crop heights for Maricopa treatments averaged 0.33 m on 24 May 2020 (Figure 5a), about five weeks after initial regrowth irrigations, while it took about 8 weeks for Eloy treatments to attain that height (Figure 5b). With the well-watered treatments  $f_c$  reached 50% in early August at Maricopa (Figure 6a), which was about three weeks earlier than at Eloy (Figure 6b). On 1 October 2020, the D75, D100, and F100 treatments were near or above 90%  $f_c$  while crop heights were near 0.70 m. Comparable  $f_c$  and crop heights for the D75 and F100 treatments at Eloy occurred in about mid-November 2020. However, by early spring in 2021, both the D75 and F100 treatments at Eloy had 100% cover, as did the D100 treatment at Maricopa, whereas the D75 and F100 at Maricopa reached full cover in around late spring (Figure 6). The  $f_c$  for the D50 treatments lagged behind the other treatments and D50 was significantly lower from fall 2020 through spring 2021 at both locations (Figure 6). However, at each location, the D50 treatment  $f_c$  reached 100% by summer in 2021. The D50 treatment produced shorter plants (final average heights of about 0.75–0.80 m as measured in November 2021) than other treatments (0.90–1.0 m) at both locations. At each location, the D50 treatment final plant heights were significantly lower than that for all other treatments. At Maricopa, final plant height was significantly greater for the D75 treatment than the D100 and F100 treatments. However, there was no difference in final plant height between the D75 and F100 treatments at Eloy.

Maricopa experienced favorable weather conditions for guayule regrowth in April, a month earlier than at Eloy. Consequently, considerably less irrigation was applied to the treatments at Eloy than at Maricopa during summer 2020 (Table 1). The later initiation of irrigation treatments at Eloy also accounts for smaller differences in cumulative TWA between the D50 and other treatments at Eloy in comparison with Maricopa (Figure 4). However, cumulative TWA differences for D50 were more extensive during the second year than the first at both locations. Starting with the initial irrigations at Maricopa (16 April 2020), the D100 treatment received a total of 278 irrigations and a total of 3125 mm of applied irrigation water applied through early fall 2021 (Table 1). In contrast, the F100 treatment at Maricopa received 32 furrow irrigations and 2781 mm of irrigation water during the same time period. At Eloy, the F100 treatment received 18 irrigations and 2430 mm of irrigation water through fall 2021 (Table 1). The smaller number of irrigation events and applied water is explained by the  $\approx 25\%$  higher TAW for the Eloy clay soil (Table 1), less robust first summer growth, and somewhat lower  $ET_o$  demand than at Maricopa, particularly in the fall of 2021 (Figures 2 and 3). The D75 treatment at Eloy, which generally received only 75%  $ET_c$  replacement by irrigation, got 184 irrigations and a total of 2644 mm of irrigation water through fall 2021 (Table 1). Total irrigation for The D75 treatment at Maricopa was less, 2526 mm, even though it was irrigated more frequently

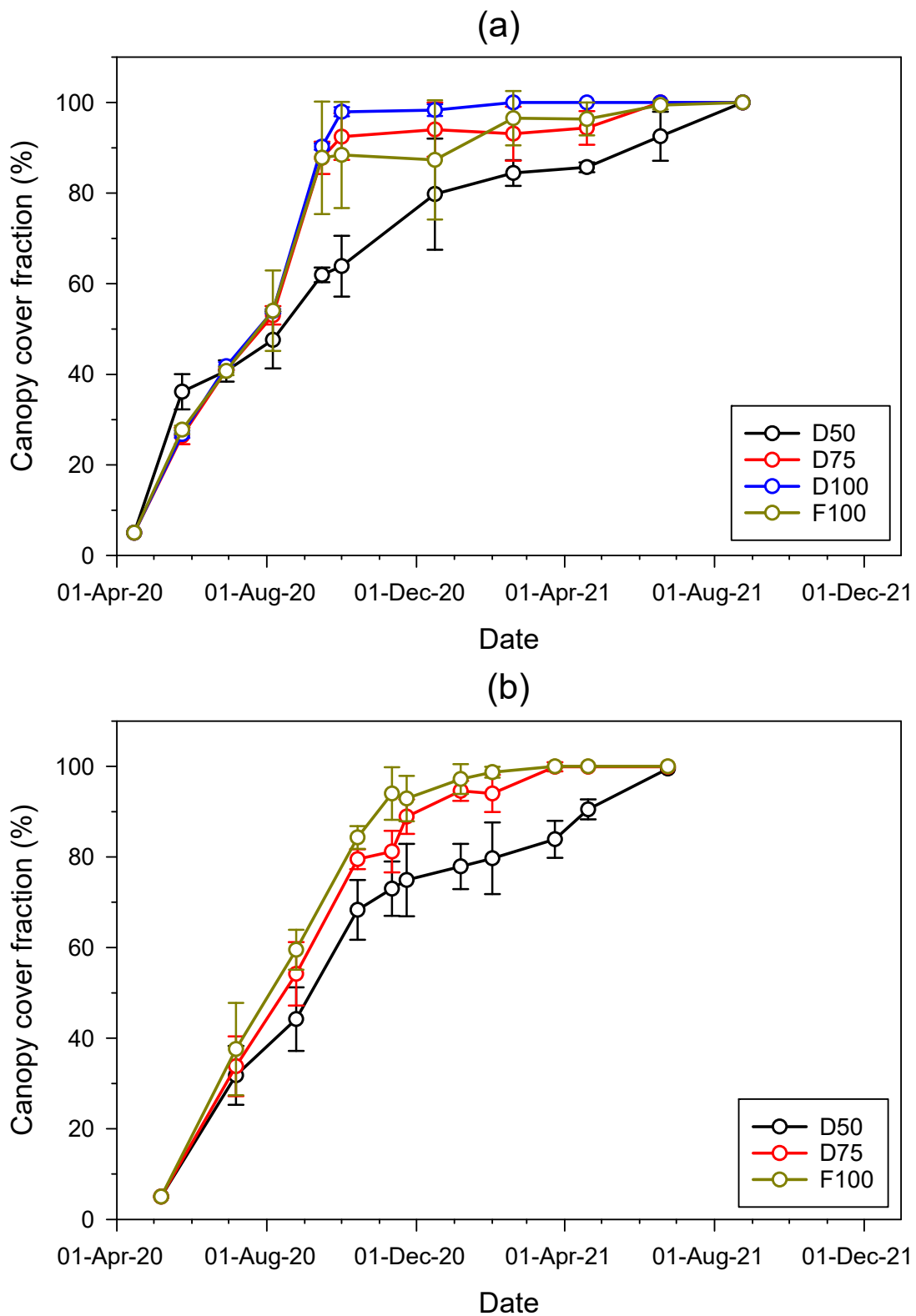
than at Eloy. At the low end, the D50 treatments received irrigation totals of 1791 and 1890 mm at Maricopa and Eloy, respectively.



**Figure 4.** Cumulative total water applied (TWA) with time for subsurface drip irrigation (D50-D100) and furrow (F100) treatments at Maricopa (a) and Eloy (b). The numbers following the letters D (for SDI) and F (furrow) are irrigation treatment rates (i.e., percent replacement of crop evapotranspiration,  $ET_c$ ).



**Figure 5.** Crop height for SDI (D) and furrow (F) treatments with time: (a) Maricopa and (b) Eloy. Error bars are one standard deviation of replicate treatment average. Treatments in legends are as described in Figure 4.



**Figure 6.** Canopy cover fraction for SDI (D) and furrow (F) treatments with time: (a) Maricopa and (b) Eloy. Error bars are one standard deviation of replicate treatment average. Treatments in legends are as described in Figure 4.



**Table 1.** Irrigation amounts and number of events for ratooned guayule for the D100 (SDI) and F100 (furrow) treatments at Maricopa and the D75 and F100 treatments at Eloy. Data were summed for each season (Spring [21 March–20 June], Summer [21 June–20 September], Fall [21 September–20 December] and Winter [21 December–20 March]). Water applied by irrigation and total water applied including irrigation and precipitation over the seasons are summed at the bottom of each treatment column for the two locations.

Maricopa	SDI		Furrow	
	Amount (mm)	Events	Amount (mm)	Events
Spring-20	313	32	346	5
Summer-20	771	79	591	7
Fall-20	227	32	300	4
Winter-20–21	224	18	154	2
Spring-21	652	45	721	7
Summer-21	649	51	406	4
Fall-21	289	21	263	3
Winter-21–22	0	0	0	0
Irrigation	3125	278	2781	32
Irrigation + prec.	3415		3071	
Eloy	Amount (mm)	Events	Amount (mm)	Events
Spring-20 <sup>a</sup>	406	4	432	4
Summer-20	419	31	455	3
Fall-20	236	41	130	1
Winter-20–21	171	14	140	1
Spring-21	766	49	605	4
Summer-21	481	31	550	4
Fall-21	166	14	118	1
Winter-21–22	0	0	0	0
Irrigation	2644	184	2430	18
Irrigation + prec.	2908		2693	

Note(s): <sup>a</sup> The SDI treatments at Eloy received three furrow irrigations and one by SDI in Spring-20.

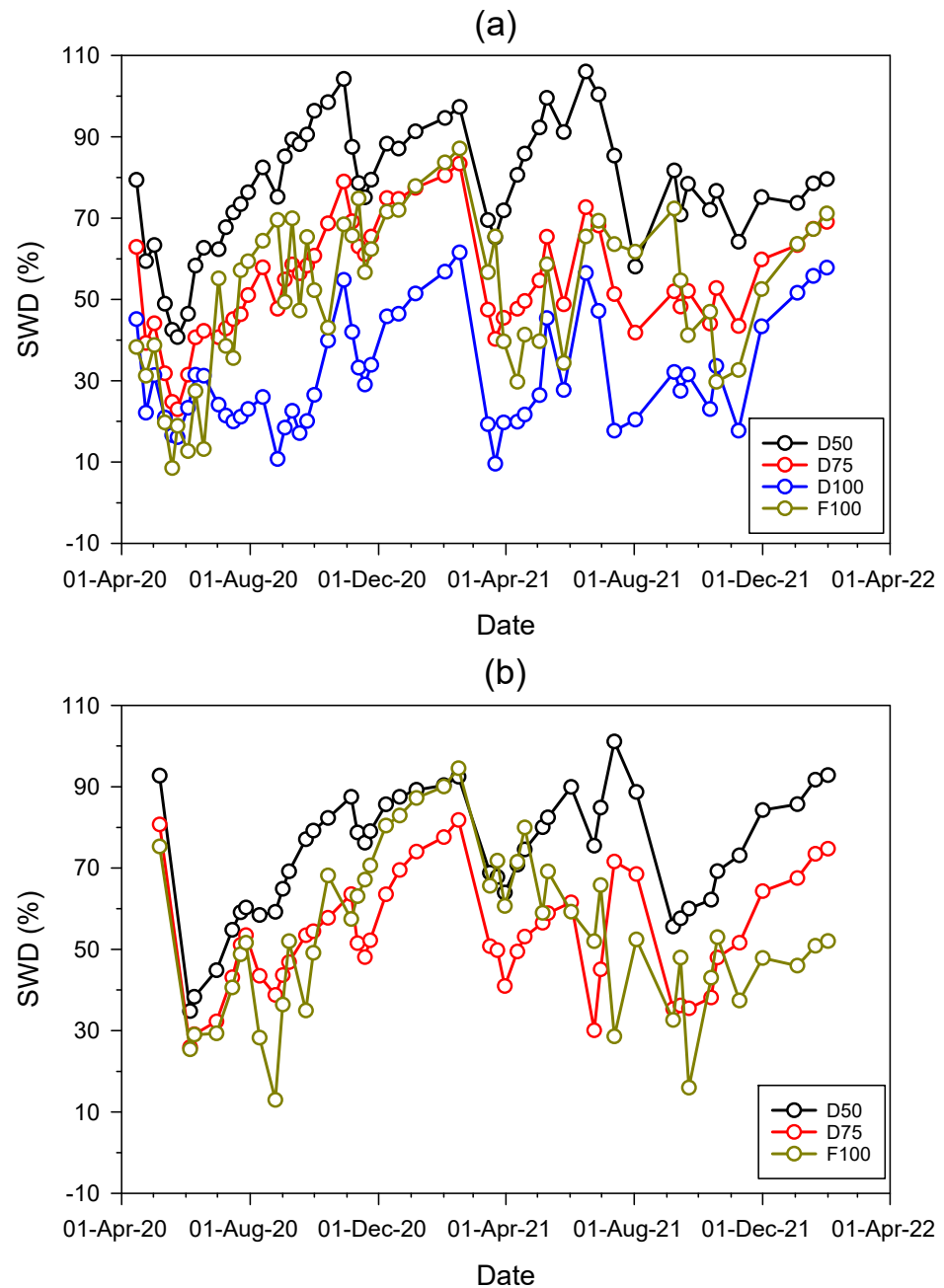
### 3.2. Soil Water Depletion and Crop Evapotranspiration

Figure 7a (Maricopa) and Figure 7b (Eloy) show the differences in soil water depletion (SWD) with time for all treatments. Larger depletion levels were experienced prior to the initial irrigations at Eloy (on 7 May 2020) than at Maricopa (on 15 April 2020). Consequently, about 90 mm more irrigation was applied to Eloy treatments during spring in 2020 than to the Maricopa D100 and F100 treatments (Table 1) to reduce the SWD for the Eloy D75 and F100 treatments to target levels.

During the 2020 summer months at Eloy, the measured SWD varied from about 30 to 55% for the D75 and F100 treatments (Figure 7b). Thereafter and through mid-November, SWD varied from 48% to 64% for D75 and from 35% to 68% for F100 (Figure 7b), when irrigation ceased until late February 2021. The SWD for the D50 treatment at Eloy increased from 45% in early summer to nearly 88% in early November.

At Maricopa, the SWD for the F100 treatment approached 70% on several occasions in summer to mid-November 2020, while the SWD for the D100 treatment remained at less than 40% except in late October, when it reached 55% (Figure 7a). For the D75 treatment the SWD varied from 42% in early summer to 79% in late October 2020, while for the D50 treatment it varied from 62% to 100% during the same period. The SWD for all treatments gradually increased from late November 2020 to mid-February 2021 (Figure 7a,b), as no irrigation was applied until late February 2021 and winter precipitation was relatively light. Measurements made in mid-February 2021 indicated SWD was at or above 90% for D50 and F100 treatments at both locations, above 80% for D75 treatments at both locations, and 62% for the D100 treatment at Maricopa. In 2021, the measured SWD for the D100 at Maricopa was less than 45%, except in early June (55% SWD), whereas the SWD for the F100 treatment was above 65% for measurement dates in spring, early summer, and early fall, 2021 (Figure 7a). While measured SWD for the D75 at Maricopa was similar to that in F100 in 2021, SWD for D50 at Maricopa was extreme, near 100% during spring to early summer in 2021. During summer in 2021 at Eloy, the measured SWD for the D75 and F100

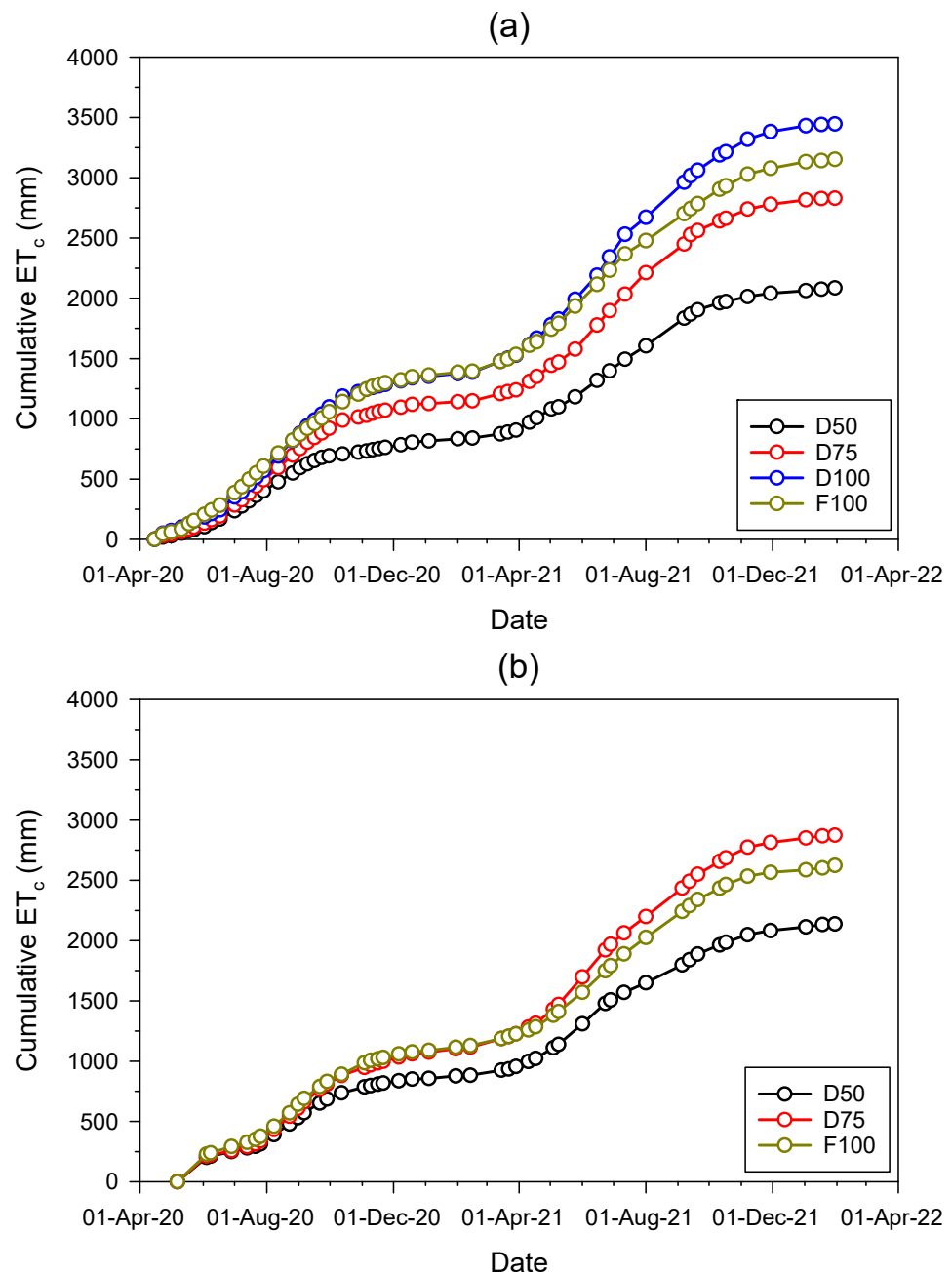
treatments was within a comparable range ( $\approx 30\text{--}70\%$ ), though SWD often varied between the treatments on different dates due to the irrigation method (Figure 7b). The D50 at Eloy had measured SWD near 100% in July 2021 but was generally lower than that for the D50 in Maricopa, particularly after July 2021. Excluding SWD data prior to the initial irrigations of the ratooned crops, the average measured SWD was 78, 54, 31, and 52% for the D50, D75, D100, and F100 treatments at Maricopa, respectively, and 73, 53, and 54% for the D50, D75, and F100 treatments at Eloy, respectively.



**Figure 7.** Soil water depletion (SWD, %) with time for SDI (D) and furrow (F) treatments: (a) Maricopa and (b) Eloy. Treatments in legends are as described in Figure 4.

The calculated cumulative  $ET_c$  for treatments at Maricopa (Figure 8a) and Eloy (Figure 8b) grew similarly as cumulative TWA (Figure 4a,b). At Maricopa, where irrigation treatments began immediately for the ratooned crop,  $ET_c$  for the D75 and D50 treatments began to drop relative to the D100 and F100 in the latter-half of May 2020). At Eloy, where all treatments were furrow irrigated and received approximately equal

amounts of water for the first month, cumulative  $ET_c$  was the same until late June, when differential irrigation started. Cumulative  $ET_c$  was essentially the same for the F100 treatments at both locations ( $\approx 240$  mm) through 8–9 June 2020, albeit with irrigations starting 3 weeks later at Eloy. Differences in cumulative  $ET_c$  among treatments increased during the summer to mid-fall 2020 at Maricopa (Figure 8a). By the end of the first-year irrigation cycle in mid-November, cumulative  $ET_c$  was about 1280 mm for the two 100% treatments, 1060 mm for D75, and 750 mm for D50 at Maricopa. In contrast, at Eloy, the cumulative  $ET_c$  for the F100 treatment (1020 mm) was lower than for F100 at Maricopa. Additionally, the  $ET_c$  for the D50 treatment was only 810 mm, and the D75  $ET_c$  was (990 mm), which was slightly less than measured for the F100 treatment. During winter months of 2020–2021 (i.e., mid-November 2020 to mid-February 2021), cumulative  $ET_c$  was increased by about 75–130 mm, where the increase was generally less for the D50 treatments.



**Figure 8.** Cumulative crop evapotranspiration ( $ET_c$ ) with time for SDI (D) and furrow (F) treatments: (a) Maricopa and (b) Eloy. Treatments in legends are as described in Figure 4.

Differences in cumulative  $ET_c$  among treatments were more prominent during the second-year irrigation cycle (mid-March to mid-November 2021). Notably, cumulative  $ET_c$  for the D100 treatment began to exceed the F100 treatment at Maricopa around late May 2021 (Figure 8a). Around the same time, cumulative  $ET_c$  for the D75 treatment began to exceed the F100 treatment at Eloy (Figure 8b). In the second year, cumulative  $ET_c$  for the D100 at Maricopa was about 1930 mm, which ultimately had 3346 mm of  $ET_c$  through the end of January 2022. This compares to the F100 treatment at Maricopa that had 1635 mm of  $ET_c$  during the irrigation cycle and a total  $ET_c$  of 3152 mm. At Eloy, cumulative  $ET_c$  during the second year was 1660 mm and total  $ET_c$  was 2875 mm for D75 compared to 1406 mm and 2623 mm for the F100, respectively. For the D75 at Maricopa, cumulative  $ET_c$  in the second-year cycle was 1590 and total  $ET_c$  was 2870 mm, results similar to the D75 at Eloy. The  $ET_c$  for the D50 treatments were 1174 mm and 1165 mm for the second irrigation cycle and totals were 2081 and 2138 mm at Maricopa and Eloy, respectively.

### 3.3. Guayule Yield and Water Productivity

Final dry biomass DB for the ratooned guayule varied from 20.6 to 34.5  $Mg\ ha^{-1}$  for treatments at Maricopa and from 16.6 to 29.8  $Mg\ ha^{-1}$  at Eloy (Table 2). For the SDI treatments, DB increased significantly from the lowest to highest irrigation rates at both locations. Statistical comparisons also indicated that DB of the D100 treatment was greater than the DB for F100 at Maricopa, whereas at Eloy, the DB for D75 was greater than that for the F100 treatment. The location effects for DB and other yield parameters were not provided in the statistical model output due to the absence of the D100 treatment at Eloy. However, alike treatments at the locations can be compared, such as the D50 and F100 treatments, which were significantly greater at Maricopa than Eloy (Table 2). The DB for the D75 treatments at the two locations were numerically similar and not significantly different. Linear regression of treatment DB against treatment TWA was significant,  $p < 0.013$ , with a coefficient of determination ( $r^2$ ) of 0.74 and standard error of 3.8  $Mg\ ha^{-1}$ . Regression was somewhat better for DB against cumulative  $ET_c$ ,  $p < 0.01$ ,  $r^2 = 0.78$ , and SE of 3.5  $ha^{-1}$ .

**Table 2.** Irrigation treatment means of rubber content (R), resin content (Re), dry biomass (DB), rubber yield (RY), and resin yield (ReY) for ratooned guayule harvested after 22 and 21 months of growth at Maricopa and Eloy, respectively. Subsurface drip treatments were D50, D75, and D100 with 50, 75, and 100%  $ET_c$  replacement, respectively, and furrow treatments (F100) had 100%  $ET_c$  replacement.

Location	Treatment	R (%)	Re (%)	DB ( $Mg\ ha^{-1}$ )	RY ( $kg\ ha^{-1}$ )	ReY ( $kg\ ha^{-1}$ )
Maricopa	D50	5.1a	10.9a	20.6c	1053b	2239b
	D75	4.9a	10.8a	30.4b	1475a	3256a
	D100	4.0b	9.4b	34.5a	1384a	3255a
	F100	5.1a	10.6a	28.8b	1461a	3047a
	Average	4.8	10.4	28.6	1343	2949
Eloy	D50	4.9a	9.3b	16.6d	811c	1540c
	D75	3.9b	8.4c	29.8b	1126b	2441b
	F100	4.2b	9.3b	18.7cd	782c	1738c
	Average	4.3	9.0	21.7	906	1906

Note(s): Different letters following parameter means indicate a significant difference ( $p < 0.05$ ).

Rubber content decreased from the lowest to the highest SDI treatments at both locations (Table 2). Although the R for D75 at Maricopa (4.9%) was lower than that for D50 (5.1%), the difference was not significant, whereas both treatments had significantly higher R than D100 (4.0%). Notably, the R for the F100 treatment at Maricopa was the same as the R for the D50 treatment. However, the R for the D50 treatment at Eloy (4.9%) was significantly greater than either the D75 or the F100 treatment at Eloy, which were also lower than those for the D75 and F100 treatments at Maricopa. The resin content did not show a trend with irrigation treatment at Maricopa, where the Re for D50, D75, and F100

were similar and each greater than the Re for the D100 treatment. Similarly, Re for the D50 and F100 were greater than that for the D75 treatment at Eloy. For the alike treatments at the two locations, the Re was greater at Maricopa.

The rubber yield, a function of DB and R, varied from 811 kg ha<sup>-1</sup> (D50 at Eloy) to 1475 kg ha<sup>-1</sup> (D75 at Maricopa). At Maricopa, the RY was significantly lower for the D50 treatment than the RY for the other three treatments, which were not significantly different from each other (Table 2). At Eloy, the RY was greater for D75 than D50 and F100. The RY for Maricopa treatments were all significantly greater than those for Eloy treatments with the exception of D50 at Maricopa and D75 at Eloy, which were the same statistically. Linear regression of treatment RY versus TWA and versus cumulative ET<sub>c</sub> were not significant at the  $p = 0.05$  level.

The ReY varied from 1544 kg ha<sup>-1</sup> (D50 at Eloy) to 3266 kg ha<sup>-1</sup> (D75 at Maricopa). Treatment differences for ReY were the same as those for RY, i.e., D50 ReY was lower among Maricopa treatments and D75 was greater among Eloy treatments (Table 2). The ReY was significantly greater for all Maricopa treatments in comparison to Eloy. Linear regression of treatment ReY against treatment TWA was not significant as also found for R against TWA. However, unlike R regression results, ReY against cumulative ET<sub>c</sub> regression results were significant,  $p < 0.037$ ,  $r^2$  of 0.62, and SE of 480 kg ha<sup>-1</sup>.

Water productivity of DB, based on TWA, was not related to irrigation rate at Maricopa, where DB–WP was found to be only significantly greater for D75 (1.08 kg m<sup>-3</sup>) than F100 (Table 3). Similarly, DB–WP for the D75 treatment at Eloy (1.03 kg m<sup>-3</sup>) was significantly greater than the DB–WP for both the D50 and F100 treatments. All Maricopa treatments had higher DB–WP than treatments at Eloy except for the D75 treatment. For RY, WP was significantly lower for the D100 treatment (0.041 kg m<sup>-3</sup>) than for all other treatments at Maricopa (average of 0.048 kg m<sup>-3</sup>), while the RY–WP for the F100 treatment was significantly lower than that for the other Eloy treatments. Overall, RY–WP was greater for Maricopa treatments than those at Eloy. The ReY–WP varied from 0.065 to 0.116 kg m<sup>-3</sup> (Table 3), where the highest and significantly greater ReY–WP were achieved for the D50 and D75 treatments at Maricopa. The ReY–WP for Eloy treatments were much lower than those at Maricopa, where all Maricopa treatments had significantly greater ReY–WP than those at Eloy.

**Table 3.** Irrigation treatment means of water productivity for dry biomass (DB–WP), rubber yield (RY–WP), and resin yield (ReY–WP) for ratooned guayule harvested after 22 and 21 months of growth at Maricopa and Eloy, respectively. Subsurface drip treatments were D50, D75, and D100 with 50, 75, and 100% ET<sub>c</sub> replacement, respectively, and furrow treatments (F100) had 100% ET<sub>c</sub> replacement.

Location	Treatment	DB–WP <sup>1</sup> (kg m <sup>-3</sup> )	RY–WP <sup>1</sup> (kg m <sup>-3</sup> )	ReY–WP <sup>1</sup> (kg m <sup>-3</sup> )
Maricopa	D50	0.99ab	0.051a	0.107a
	D75	1.08a	0.053a	0.116a
	D100	1.01ab	0.041b	0.095b
	F100	0.94b	0.048a	0.099b
	Average	1.00	0.048	0.104
Eloy	D50	0.77c	0.038b	0.072d
	D75	1.03ab	0.038b	0.084c
	F100	0.70c	0.029c	0.065d
	Average	0.83	0.035	0.074

Note(s): <sup>1</sup> Water productivities are based on total water applied (TWA). Different letters following parameter means indicate significant differences ( $p < 0.05$ ).

#### 4. Discussion

The irrigation water applied to activate growth for the ratooned crops in the spring of 2020 at Maricopa and Eloy, ≈330 and 420 mm, respectively, was about 50 and 33% less than that applied to germinate and establish the direct-seeded guayule crops in spring 2018



in the initial studies [18]. Bucks et al. [25], using sprinkler irrigation, recorded as much as 560 mm of irrigation to establish a direct-seeded guayule crop during spring. Moreover, all other reported direct-seeded guayule studies used sprinkler irrigation to germinate and establish the crops (e.g., [30,31]). Our results indicate that a ratooned guayule crop will need significantly less irrigation water to promote regrowth than that needed to establish the direct-seeded crop.

In addition, the costs associated with installing a sprinkler system for the initial establishment would be eliminated since the existing (drip or furrow) irrigation method would be adequate for regrowth. In the pre-ratooned, direct-seeded studies (2018–2020), the D100 and F100 at Maricopa received irrigation totals of 3682 and 3506 mm, respectively, and 436 mm of precipitation [18]. The TWA was 17 and 22% less for the D100 and F100 treatments, respectively, in the regrowth study at Maricopa. The TWA for the F100 treatment in the regrowth study at Eloy was 27% less, or 970 mm, than for the F100 in pre-ratooned study. Besides the lower irrigation requirements needed for regrowth establishment versus direct-seeded establishment, comparatively less irrigation was needed in the second year of the regrowth study because of the higher precipitation in summer 2021 versus summer 2019. Nevertheless, considering the  $\approx$ 1-month shorter season and less TWA for the regrowth study at Maricopa, the cumulative  $ET_c$  was similar to the cumulative  $ET_c$  in the pre-ratooned studies at the location. For the D100 and F100 treatments at Maricopa, cumulative  $ET_c$  was only 6% and 10% less than that in the pre-ratooned studies, respectively. The regrowth study at Eloy was about 2 months shorter than the pre-ratooned study at that location. For regrowth at Eloy, cumulative  $ET_c$  for the D75 treatment was about the same as in the pre-ratooned study for that treatment, whereas the cumulative  $ET_c$  for the F100 treatment was about 20% lower, corresponding to 27% less TWA.

Final DB, RY, and ReY were higher for the ratooned than pre-ratooned guayule [18] for corresponding treatments at Maricopa, where the treatments averaged 25% more DB, 33% more RY, and 32% more ReY. At Maricopa, the highest increases in the three yield parameters for the ratooned guayule were obtained for D50 and D75 treatments, though all treatments had positive increases. As expected, the WP for the ratooned treatments were greatly improved at Maricopa, where the increase in WP for corresponding treatments over pre-ratooned treatments averaged 36, 43, and 43% for DB–WP, RY–WP, and ReY–WP, respectively. In addition to the pre-ratooned study, the WP (based on TWA) for the Maricopa ratooned guayule (provided in Table 3) were notably higher than those in other studies, e.g., [17,22]. The highest WP in literature was reported by [14] for transplanted guayule using SDI.

The final DB for the D50 and F100 treatments for the ratooned guayule at Eloy averaged 13 and 41% less than that for the pre-ratooned D50 and F100 treatments, respectively, while DB for the ratooned D75 treatment was 3% higher than the pre-ratooned D75. However, the DB–WP for the D50 and F100 treatments at Eloy was about the same for the ratooned and pre-ratooned studies, as TWA was also lower for those treatments in the ratooned studies. On the other hand, the RY of the F100, D50, and D75 Eloy treatments in the ratooned study at Eloy exceeded the RY of those in the pre-ratooned study by 3, 9, and 26%, respectively, while RY–WP for the ratooned were increased 24, 12, and 25%, respectively. The ReY–WP for the ratooned studies also were increased: 18, 4, and 12% for the F100, D50, and D75 treatments, respectively.

The ratooned guayule regrowth showed expected differences in crop height and canopy cover as differences in cumulative irrigation increased for treatments. Differential irrigation treatments started later at Eloy than Maricopa where differences in the growth parameters were unclear until late summer 2020 versus October 2020 at Eloy. While the regrowth of the treatments at Eloy lagged treatments at Maricopa in 2020, crop height and cover measurements indicated the parameters were similar at the two locations by about mid-November 2020. However, at either location, the ratooned regrowth developed more rapidly during the first year than did growth following direct seeding in the pre-ratooned studies at the locations [18]. A benchmark canopy cover of 90% was reached by the D100

at Maricopa and the F100 treatment at Eloy about 1.5–2.0 months earlier than for those treatments in the pre-ratooned studies at the locations. Similarly, well-watered treatments in a separate direct-seeded guayule study planted in April 2018 at Eloy also reached 90% cover about 2.0 months later than that in the regrowth at Eloy [22]. The more rapid regrowth for the F100 treatment at Eloy occurred with about 17% less irrigation from Spring-Fall in the first year than that applied to the direct-seeded crop. A higher rate of crop height regrowth during the first year for the ratooned crops compared to the pre-ratooned crops at the Eloy location is also noted. The quicker regrowth of ratooned compared to pre-ratooned guayule may be due to the fact that the ratooned plants already have strong, established roots from its previous growth. This gives it a head start in growing taller during the first year after regrowth, while pre-ratooned guayule has to develop its roots, slowing down its initial growth. The final measured crop heights in November 2021 for the ratooned crops were about 0.90 m at Maricopa and 1.0 m at Eloy excluding the D50 treatments, which were lower. Comparable final crop heights were attained for the well-watered treatments in direct-seeded studies after two years [18,22].

Final guayule DB is typically a linear function of TWA or cumulative  $ET_c$  [14,22,24] as in the present ratooned guayule studies. For this study, the slope of the linear regression of DB vs. TWA was  $0.0123 \text{ Mg ha}^{-1} / \text{TWA}$ , which suggests about  $5.0 \text{ Mg ha}^{-1}$  of DB gained for every 400 mm of TWA. The slope is consistent with the relationship of DB vs. TWA (0.015), where only SDI treatments were used [14]. In studies in which only furrow irrigation was used, the regression slope of DB vs. TWA was found to be much flatter, i.e., 0.05–0.075 [16,17,22]. Thus, guayule DB response to increasing TWA appears to be much lower in furrow irrigation than SDI. The differences in low versus high frequency irrigation between furrow and SDI change the way soil water fluctuates for guayule during the season. The RY and ReY were not as well correlated to TWA or cumulative  $ET_c$  as DB because the R and Re were significantly lower for the wettest irrigation treatments at the locations, which also had the highest DB.

Prior studies using furrow irrigation have indicated that guayule biomass at harvest may not differ among treatments when the average SWD throughout the season is maintained at less than 70% [11,15–17,22]. However, this generality is not appropriate when SDI is used because SWD for guayule can be maintained at much lower levels during the growing season than that for furrow irrigation as shown by [17,18]. This was confirmed in the present regrowth studies on the sandy loam soil at Maricopa, in which DB for SDI treatments decreased significantly as average SWD increased from 31% for the D100 to 54 and 78% for the D75 and D50 treatments, respectively. The F100 at Maricopa had an average SWD similar to the D75 and produced the same DB as the D75. However, on the higher soil water retention soil at Eloy, the pre-ratooned SDI study reported that DB was not significantly different between D75 and a D100 treatment, where average SWD was 47 and 26%, respectively. The 47% SWD at Eloy for D75 represents significantly higher TAW than that for the D75 at Maricopa. Consequently, for the present ratooned study a D100 treatment at Eloy was omitted and the D75 treatment, maintained at less than 55% SWD, was deemed the optimum SDI treatment. At Eloy, the DB for the D50 treatment with an average SWD of 74% was significantly lower than that for the D75 in the ratooned study. While average seasonal SWD was about the same for the D75 and F100 treatments at Eloy, SWD was much higher for the F100 than D75 for extended periods in late fall 2020 and spring 2021. This likely reduced the production of biomass for the F100 treatment during these periods, which was suggested by the lower gain in crop height compared to the D75.

The effects of prolonged water stress due to limited irrigation and higher SWD often produce higher rubber content compared to treatments under wetter soil moisture conditions [17,22,27]. This was found to be the case in the ratooned studies at Eloy, where R was significantly greater for the D50 treatment. Differently, R was the same for all treatments except the D100 at Maricopa. For alike treatments, the R was the same for D50 at both locations but higher for D75 and F100 at Maricopa. These results do agree with treatment differences for R at the two locations in the pre-ratooned study [18] and may suggest an

effect on R due to the different soil types. It is also worth noting that the R was higher about 0.6% higher in the ratooned than pre-ratooned for alike treatments at both locations. Previous studies have shown unclear trends for differences in Re due to irrigation or stress [18]. In the present studies, irrigation treatment trends were essentially the same as those for R at each location, where Re was the same for all treatments at Maricopa except for the D100, which was lower, and the same for the D50 and F100 treatments at Eloy and lower for the D75. As for R, the Re for alike treatments was also significantly greater at Maricopa than at Eloy. Also, as for R, the Re was greater for the ratooned than pre-ratooned treatments at both locations. The increase in ratooned Re averaged 1.1% at Maricopa and 1.4% at Eloy.

## 5. Conclusions

Regrowth of ratooned guayule (*Parthenium argentatum* A. Gray) was conducted for 21–22 months under well-watered and reduced irrigation treatments comparative to subsurface drip and furrow irrigation treatments imposed during a first two-year growth cycle of direct-seeded guayule at Maricopa and Eloy in Arizona. The overall assessment showed that ratooning guayule is a promising agronomic practice with high yield potential and efficient water productivity. The ratooned guayule crop results were generally more favorable under a sandy loam soil at Maricopa than under the heavier clay soil at Eloy, particularly when considering the furrow irrigation method. However, when using frequent subsurface irrigation on either soil type, maximum rubber and resin yields for ratooned treatments were obtained by replacing 75% of crop evapotranspiration, while maintaining soil water depletion below 55%. This irrigation management also achieved the highest water productivities for rubber yield. When irrigation is applied by less frequent furrow irrigation, it is recommended to replace 100% of crop evapotranspiration and maintain soil water depletion at less than 55% for the ratooned guayule.

The irrigation water use for ratooned guayule should be considerably less than that needed to establish and grow a direct-seeded guayule crop, perhaps on the order of 17–27% less total water applied. Much of the water savings can be gained for the ratooned crop during initial stages when less irrigation water is needed to stimulate regrowth in the ratooned crop than that needed to emerge and establish a direct-seeded crop. Additionally, rubber and resin contents and rubber and resin yields for the ratooned guayule crop could be considerably higher compared to the yields of the pre-ratooned crop after two years. Thus, ratooning the direct-seeded guayule can provide the grower with significant additional yield revenue that can be attained with less water input and negligible startup costs. These factors would help offset the initial investment costs related to growing and establishing the guayule crop from seed.

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## References

1. Eranki, P.L.; El-Shikha, D.; Hunsaker, J.; Bronson, K.F.; Landis, A.E. A comparative life cycle assessment of flood and drip irrigation for guayule rubber production using experimental field data. *Ind. Crops Prod.* **2018**, *99*, 97–108. [[CrossRef](#)]
2. Rasutis, D.; Soratana, K.; McMahan, C.; Landis, A.E. A sustainability review of domestic rubber from the guayule plant. *Ind. Crops Prod.* **2015**, *70*, 383–394. [[CrossRef](#)]
3. Nakayama, F.S.; Vinyard, S.H.; Chow, P.; Bajwa, D.S.; Youngquist, J.A.; Muehl, J.H.; Krzysik, A.M. Guayule as a wood preservative. *Ind. Crops Prod.* **2001**, *14*, 105–111. [[CrossRef](#)]
4. Thames, S.F.; Kaleem, K. Guayule resin in amine-epoxy strippable coatings. *Bioresour. Technol.* **1991**, *35*, 185–190. [[CrossRef](#)]
5. Wang, G.; Elshikha, D.M.; Katterman, M.E.; Sullivan, T.; Dittmar, S.; Cruz, M.V.; Hunsaker, D.J.; Waller, P.M.; Ray, D.T.; Dierig, D.A. Irrigation effects on seasonal growth and rubber production of direct-seeded guayule. *Ind. Crops Prod.* **2022**, *177*, 114442. [[CrossRef](#)]
6. Ray, D.T.; Coffelt, T.A.; Dierig, D.A. Breeding guayule for commercial production. *Ind. Crops Prod.* **2005**, *22*, 15–25. [[CrossRef](#)]
7. Cornish, K.; Pearson, C.H.; Rath, D.J. Accurate quantification of guayule resin and rubber requires sample drying below a critical temperature threshold. *Ind. Crops Prod.* **2013**, *41*, 158–164. [[CrossRef](#)]
8. Coffelt, T.A.; Ray, D.T. Cutting height effects on guayule latex, rubber, and resin yields. *Ind. Crops Prod.* **2010**, *32*, 264–268. [[CrossRef](#)]
9. Boateng, A.A.; Elkasabi, Y.; Mullen, C.A. Guayule (*Parthenium argentatum*) pyrolysis biorefining: Fuels and chemicals contributed from guayule leaves via tail gas reactive pyrolysis. *Fuel* **2016**, *163*, 240–247. [[CrossRef](#)]
10. Placido, D.F.; Dierig, D.A.; Cruz, V.M.V.; Ponciano, G.; Dong, C.; Dong, N.; Huynh, T.; Williams, T.; Cahoon, R.E.; Wall, G.W.; et al. Downregulation of an allene oxide synthase gene improves photosynthetic rate and alters phytohormone homeostasis in field-grown guayule. *Ind. Crops Prod.* **2020**, *153*, 112341. [[CrossRef](#)]
11. Bucks, D.B.; Nakayama, F.S.; French, O.F.; Legard, W.W.; Alexander, W.L. Irrigated guayule—Evapotranspiration and plant water stress. *Agric. Water Manag.* **1985**, *10*, 61–79. [[CrossRef](#)]
12. Hammond, B.L.; Polhamus, L.G. *Research on Guayule (Parthenium argentatum): 1942–1959*; USDA Technical Bulletin No. 1327; U.S. Government Printing Office: Washington, DC, USA, 1965; pp. 1–157.
13. National Academy of Science (NAS). *Guayule: An Alternative Source of Natural Rubber*; NAS: Washington, DC, USA, 1977.
14. Hunsaker, D.J.; Elshikha, D.M.; Bronson, K.F. High guayule rubber production with subsurface drip irrigation in the US desert Southwest. *Agric. Water Manag.* **2019**, *220*, 1–12. [[CrossRef](#)]
15. Bucks, D.B.; Nakayama, F.S.; French, O.F.; Rasnick, B.A.; Alexander, W.L. Irrigated guayule—Plant growth and production. *Agric. Water Manag.* **1985**, *10*, 81–93. [[CrossRef](#)]
16. Bucks, D.B.; Nakayama, F.S.; French, O.F.; Legard, W.W.; Alexander, W.L. Irrigated guayule—Production and water use relationships. *Agric. Water Manag.* **1985**, *10*, 95–102. [[CrossRef](#)]
17. Hunsaker, D.J.; Elshikha, D.M. Surface irrigation management for guayule rubber production in the US desert Southwest. *Agric. Water Manag.* **2017**, *185*, 43–57. [[CrossRef](#)]
18. Elshikha, D.M.; Waller, P.M.; Hunsaker, D.J.; Dierig, D.; Wang, G.; Cruz, M.V.; Thorp, K.R.; Katterman, M.E.; Bronson, K.F.; Wall, G.W. Growth, water use, and crop coefficients of direct-seeded guayule with furrow and subsurface drip irrigation in Arizona. *Ind. Crops Prod.* **2021**, *170*, 113819. [[CrossRef](#)]
19. Miyamoto, S.; Davis, J.; Piela, K. Water use, growth, and rubber yields of four guayule selections as related to irrigation regimes. *Irr. Sci.* **1984**, *5*, 95–103. [[CrossRef](#)]
20. Bucks, D.A.; Roth, R.L.; Nakayama, F.S.; Gardner, B.R. Irrigation water, nitrogen, and bioregulation for guayule production. *Trans. ASAE* **1985**, *28*, 1196–1205. [[CrossRef](#)]
21. Nakayama, F.S.; Bucks, D.A.; Roth, R.L.; Gardner, B.R. Guayule biomass production under irrigation. *Biores. Technol.* **1991**, *35*, 173–178. [[CrossRef](#)]
22. Elshikha, D.M.; Wang, G.; Waller, P.M.; Hunsaker, D.J.; Dierig, D.; Thorp, K.R.; Cruz, M.V.; Thompson, A.; Katterman, M.E.; Herritt, M.T.; et al. Guayule growth and responses to deficit irrigation strategies in the U.S. desert. *Agric. Water Manag.* **2023**, *277*, 108093. [[CrossRef](#)]
23. Pereira, L.S.; Cordery, I.; Iacovides, I. Improved indicators of water use performance and productivity for sustainable water conservation and saving. *Agric. Water Manag.* **2012**, *108*, 39–51. [[CrossRef](#)]
24. Miyamoto, S.; Bucks, D.A. Water quantity and quality requirements of guayule: Current assessment. *Agric. Water Manag.* **1985**, *10*, 205–219. [[CrossRef](#)]
25. Bucks, D.A.; Roth, R.L.; Powers, D.E.; Chandra, G.R. 1986 Direct seeding for economical guayule field establishment. In *Guayule, a Natural Rubber Source, Proceedings of the Fourth International Conference on Guayule Research and Development Conference, Tucson, AZ, USA, 16–19 October 1985*; Fangmeier, D.D., Alcorn, S.M., Eds.; Guayule Rubber Soc., Inc.: College Station, TX, USA, 1986; pp. 71–87.
26. Estilai, A.; Waines, J.G. Variation in regrowth and its implication for multiple harvest of guayule. *Crop Sci.* **1987**, *27*, 100–103. [[CrossRef](#)]
27. Ray, D.T.; Foster, M.A.; Coffelt, T.A.; McMahan, C. Guayule: Culture, breeding and rubber production. In *Industrial Crops and Uses*; CABI: Wallingford, UK, 2010; pp. 384–410.

28. Wang, G.; Lynch, A.L.; Cruz, V.M.V.; Heinitz, C.C. Temperature requirements for guayule seed germination. *Ind. Crops Prod.* **2020**, *157*, 112934. [[CrossRef](#)]
29. Foster, M.A.; Coffelt, T.A. Guayule agronomics: Establishment, irrigated production, and weed control. *Ind. Crops Prod.* **2005**, *22*, 27–40. [[CrossRef](#)]
30. Dissanayake, P.; George, D.L.; Gupta, M.L. Direct seeding as an alternative to transplanting for guayule in southeast Queensland. *Ind. Crops Prod.* **2008**, *27*, 393–399. [[CrossRef](#)]
31. Foster, M.A.; Fowler, J.L.; Kleine, L.G.; Grote, M.M.; Puppala, N. Performance of direct-seeded and transplanted guayule. *Ind. Crops Prod.* **2002**, *15*, 23–31. [[CrossRef](#)]
32. Foster, M.A.; Fowler, J.L.; Kleine, L.G.; Puppala, N. Establishing new guayule lines by direct seeding. *Ind. Crops Prod.* **1999**, *9*, 93–100. [[CrossRef](#)]
33. Post, D.F.; Mack, C.; Camp, P.D.; Sulliman, A.S. Mapping and characterization of the soils on the University of Arizona Maricopa Agricultural Center. In Proceedings of the 1988 Meetings of the Arizona Section—American Water Resources Association and the Hydrology Section—Arizona-Nevada Academy of Science, Tucson, AZ, USA, 1988; pp. 49–60.
34. US Department of Agriculture. *Soil Conservation Service (USDA-SCS)*; U.S. Department of Agriculture, Soil Conservation Service: Part 650: Washington, DC, USA, 1993. Available online: <https://www.nrcs.usda.gov/sites/default/files/2022-08/Engineering-In-The-Soil-Conservation-Service.pdf> (accessed on 30 August 2023).
35. Ray, D.T.; Dierig, D.A.; Thompson, A.E.; Coffelt, T.A. Registration of six guayule germplasm with high yield ability. *Crop Sci.* **1999**, *39*, 300. [[CrossRef](#)]
36. Allen, R.G.; Pereira, L.S.; Raes, D.; Smith, M. *Crop Evapotranspiration*; FAO Irrigation and Drainage Paper 56; Food and Agriculture Organization of the United Nations: Rome, Italy, 1998.
37. Hunsaker, D.J.; French, A.N.; Waller, P.M.; Bautista, E.; Thorp, K.R.; Bronson, K.F.; Andrade-Sanchez, P. Comparison of traditional and ET-based irrigation scheduling of surface-irrigated cotton in the arid southwestern USA. *Agric. Water Manag.* **2015**, *159*, 209–224. [[CrossRef](#)]

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