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Approach to Yield Response of Young Almond Trees to Deficit Irrigation and Biostimulant Applications

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Abstract: Water is the most limiting resource in many semi-arid areas of Mediterranean countries. Among the strategies to improve water productivity, the implementation of deficit irrigation (DI) strategies and the introduction of drought-tolerant crops in irrigated areas (such as almond) are being widely studied. Recently, the use of biostimulants to enhance crop tolerance to drought under water-scarcity scenarios is increasing. This work examines the response of three almond cultivars ('Guara', 'Marta', and 'Lauranne') in terms of yield and associated physiological responses in the main phenological stages to biostimulants (HYT® A and HYT® B plus) applied to young trees subjected to different irrigation levels: (i) a full irrigation treatment (FI), irrigated at 100% of crop evapotranspiration (ET_C); and (ii) sustained-deficit irrigation (SDI_{75}), irrigated at 75% of ET_C. Significantly higher yields were obtained with HYT applications in 2 of 3 cultivars; these differences were most evident in the SDI_{75} treatment. In particular, 'Guara' registered the most significant improvements in nut yield when the HYT product was applied (15–20% higher). With regard to crop physiological responses, higher values of leaf water potential and stomatal conductance were noted with the HYT application in some cultivars and phenological stages. These results indicated that the use of biostimulants can be a feasible strategy for almond cultivation, especially when SDI is used.

Keywords: almond; deficit-irrigation; irrigation water productivity; kernel yield; biostimulant

1. Introduction

Almond (*Prunus dulcis* Mill.) represents the third largest crop in terms of surface area of cultivation in Spain and comprises 84% of the total European production (380,341 t in 2017). However, this performance represents only 5% of worldwide production, the USA being the largest international producer with 80% of the world market [1]. In Spain, almond production is relatively low because this crop has been grown in marginal areas where it is traditionally cultivated under limiting conditions, with production around 150 kg·ha^{-1}. In contrast, when almond is cultivated under optimum conditions and with maximum irrigation doses, higher production is achieved, in many cases up to 3000 kg·ha^{-1}.

In terms of yield and quality, irrigation is considered the main limiting factor for almond [2]. Goldhamer and Girona [3] estimated that the water requirements of this crop for the climatic conditions of California, similar to those existing in southern Spain, oscillate between 9000 and 13,500 m^3·ha^{-1}, depending on the plant density, rootstock, crop management or pruning system, among other factors.

Recently, López-López et al. [4] defined the maximum irrigation volumes for almond under the climatic conditions of southern Spain close to 8000 $m^3 \cdot ha^{-1}$.

Taking into account the maximum irrigation rates required by this crop, its introduction as an alternative in arid and semi-arid areas would be justified only if its productivity is improved by means of deficit irrigation (DI) strategies [5]. In this sense, almond is considered a drought-tolerant crop and many authors have already studied its response under DI strategies, with the aim of improving its development under water stress conditions, minimizing production losses, and keeping or even improving nut quality [6–8]. Egea et al. [8] in a three-year experiment demonstrated that the agronomic response of almond trees to DI strategies exhibited direct and strong links between the intensity of the water restriction and the response of several parameters related to tree growth, yield, and water productivity.

However, the improvement of almond production by using DI requires knowing the agronomic and physiological responses at different phenological stages when water withholding is applied [5]. Moreover, the application of a DI strategy not only implies water restriction with respect to the crop water demand, but also a decrease in the mineral nutrient contribution and in the volume of wet soil. These constraints can cause alterations in vegetative development and consequently affect final production. To solve these problem,, in the last few years, the use of biostimulants have been introduced [9].

Biostimulants are among the oldest products used in agriculture, although this term has been recently introduced. In this sense, the first definition that appears in a web journal in 1997 defines biostimulants as "*materials that, in minute quantities, promote plant growth*" [10]. Thus, plant biostimulants are substances that have the capacity of modifying physiological processes in plants, providing potential benefits to growth, development, or stress responses [11]. In 2010, Spann et al. [12] studied the effects of the Stimplex® biostimulant in sweet orange (*Citrus x sinensis*) and demonstrated that this biostimulant was an optimum product to maintain the growth of citrus trees grown in greenhouse conditions under non-uniform irrigation systems. In the same vein, Zaghloul et al. [13] in a study with orange trees demonstrated the improvements achieved when these kinds of products were applied under water limiting scenarios. Recently, Koleška et al. [11] in an experiment with tomato (*Solanum lycopersicum* L.) proved that biostimulant applications prevented yield losses and reduced oxidative damage. In spite of the efficacy of biostimulants having been demonstrated, to date there are few studies of the benefits of these kinds of products when DI strategies are being applied.

Taking into account all of the above-mentioned considerations, the main objective of this study was to examine the effects on the physiological and yield responses to the biostimulants HYT A® VitaComplex and HYT B® AminoVita plús applied to young almond trees subjected to full irrigation and a sustained-deficit-irrigation strategy.

2. Materials and Methods

2.1. Experimental Location

The trial was conducted during the irrigation season (late March to September) of 2018, in a commercial almond (*P. dulcis* Mill. cvs. Guara, Marta, and Lauranne) orchard, grafted onto GN15 rootstocks, and located in the Guadalquivir river basin (Figure 1) (SW Spain, 37°30′27.4′′, 5°55′48.7′′ W). Trees were planted in 2013, spaced 8 × 6 m, and drip irrigated using two pipe lines with emitters of 2.3 $L \cdot h^{-1}$, spaced at 0.75 m intervals.

Figure 1. Commercial orchard "Cartuja" in Seville (SW Spain) and the experimental area used for the experiment with 1.8 ha (red area).

Taking into account that, before this experiment, no irrigation restriction had been applied in the previous seasons, and that, all trees had received the same fertilizer applications, the canopy volumes were very similar within each cultivar. For 'Marta', canopy volumes ranged between 64.5 and 65.7 m^3; 'Lauranne' trees ranged between 72.9 and 74.3 m^3, and 'Guara' trees showed canopy volumes' ranging between 65.6 and 66. 6 m^3.

The soil is a silty loam typical Fluvisol, more than 2 m deep, with organic matter < 15.0 $g \cdot kg^{-1}$. Roots were located predominately in the first 50 cm of soil, corresponding to the intended wetting depth. Soil water content values at field capacity (−0.033 MPa) and permanent wilting point (−1.5 MPa) were close to 0.40 and 0.15 $m^3 \cdot m^{-3}$, respectively.

The climatology of the study area is meso-Mediterranean, with an annual ET_0 rate of 1400 mm and accumulated rainfall of 540 mm, distributed mainly from October to April. During the experimental period, the rainfall and ET_0 were close to 70 and 580 mm, respectively. Additional information regarding climatic conditions and irrigation volumes applied can be found at Table 1.

Table 1. Climatic conditions, water requirements, and irrigation doses applied during the season.

DOY [z]	T_{air} (°C)	RH (%)	Rainfall (mm)	ET_0	K_C	ET_C	FI (m^3 ha^{-1})	SDI_{75} (m^3 ha^{-1})
115–123	24.25	59.9	20	26.57	0.6	23.91	63.52	41.13
124–127	21.6	54	12	26.12	0.9	23.51	122.31	86.73
128–134	23.9	43.63	2.3	27.6	0.9	24.84	272.58	174.98
135–148	24.6	39.8	5.6	23.71	0.9	21.34	330.07	240.52
149–162	25.5	44.1	4	27.32	1.1	33.81	702.64	468.72
163–166	27.8	53.95	0	38.21	1.1	47.28	824.18	566.53
167–183	26.6	48.1	1	32.52	1.2	43.9	1427.18	952.71
184–190	33.5	34.9	0	43.79	1.2	59.12	1717.42	1184.72
191–197	26.25	53.45	0	35.51	1.2	42.61	2055.53	1401.9
198–206	28.45	49.5	0	47.14	1.2	49.5	2472.89	1647.32
207–211	26.05	52.25	0	39.61	1.1	49.02	2718.33	1858.22
212–218	33.2	41.2	0	55.36	1.1	68.51	3037.43	2089.4
219–225	32.5	42.6	0	38.16	1.1	47.22	3277.26	2392.38
226–232	32.6	36.3	0	36.65	1.1	45.35	3512.22	2687.63
233–240	20.18	42.6	0	45.97	0.8	36.78	3772.32	2744.94
241–283	25.16	58.1	2.3	39.37	0.8	29.52	4848.31	3626.59
284–293	18.3	82.56	21.9	15.05	0.7	9.22	5073.89	3787.92

[z] DOY, day of the year; T_{air}, average air temperature; RH, average relative humidity; ET_o, reference evapotranspiration; K_C, crop coefficient; ET_C, crop evapotranspiration; FI, full-irrigated treatment at 100 ET_C; SDI_{75}, sustained-deficit irrigation at 75% ET_C.

2.2. Irrigation Strategies and Fertilization

Two irrigation treatments were applied: (i) a full-irrigated treatment (FI), which received 100% of the crop evapotranspiration (ET_C) during the irrigation period, and (ii) a sustained-deficit-irrigation (SDI_{75}) treatment, which received 75% ETc during the irrigation period.

Two different fertilization strategies were differentiated. One was defined as a treatment characterized by HYT product application. This treatment was based on the application of two HYT products: HYT A®, composed by an aerobic and anaerobic bacteria community; and HYT B® AminoVita plús, a biostimulant and nutrition platform composed by carbohydrates, nitrogen, true protein, amino acids, and essential metabolic micronutrients.

HYT A® is a soil applied microbial biostimulant produced by a co-fermentation process utilizing Agrinos' proprietary consortium of microbes including *Azotobacter vinelandii* and *Clostridium pasteurianum*. HYT B® is a biostimulant extracted from shrimp meal through microbial fermentation. The product contains 6% free L-amino acids, 1% nitrogen, and trace levels of other nutrients, and provides a source of time-released nitrogen that is highly bioavailable and other nutritional complements.

By contrast, the second treatment was no application of HYT products, but with the same irrigation strategies and the same cultivars as the HYT treatment.

HYT A® and HYT B® were periodically applied through the irrigation system (Table 2), following the date and doses proposed by Agrinos (Agrinos AS, Oslo, Norway; www.agrinos.com), the manufacturer of these products.

Table 2. Applications of HYT products along the irrigation period.

DOY [z]	HYT A® ($L \cdot ha^{-1}$)	HYT B® ($L \cdot ha^{-1}$)	Phenological Stage
72	4	4	Fruit setting
93	2	2	Fruit growth
114	2	2	Fruit growth
142	1	1	Fruit growth
173	1	1	Kernel filling
199	1	1	Kernel filling
296	1	1	Post-harvest

[z] DOY = Julian day of year.

Moreover, all trees of both treatments received a traditional fertilization according to 155-60-177 units of NPK in the case of FI treatment and 116-45-135 units of NPK for the SDI_{75} treatment. Figure 2 summarizes the irrigation and fertilization strategies considered in this work.

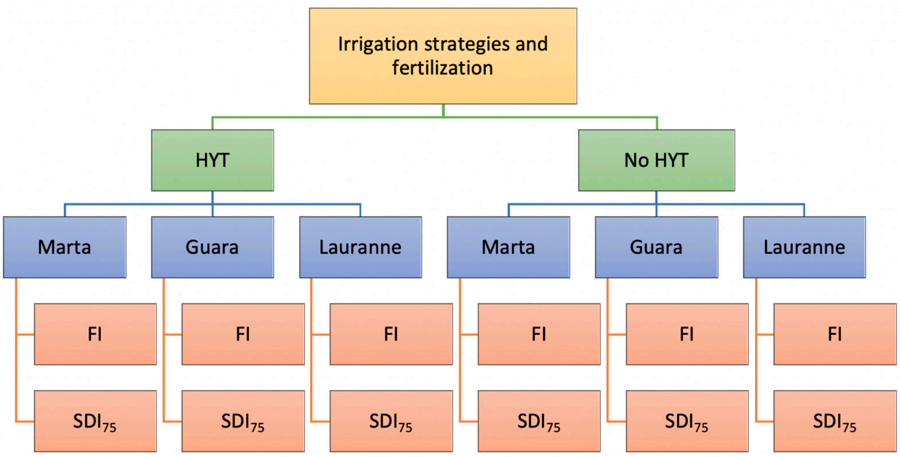

Figure 2. Description of the irrigation treatment and fertilization applied. FI, full irrigation at 100% ET_C; SDI_{75}, sustained-deficit irrigation at 75% ET_C.

Irrigation strategies were performed simulating two different scenarios—a full irrigation treatment (FI), which was irrigated to replace 100% of the crop evapotranspiration (ET_C) during the irrigation period; and a sustained-deficit irrigation (SDI_{75}), which was irrigated at 75% of ET_C during the season. Irrigation volumes were calculated according to the methodology proposed by Allen et al. [3], obtaining the values of reference evapotranspiration according to Penman–Monteith equation with a weather station located in the same experimental orchard; and using the crop coefficients obtained by García-Tejero [14], which ranged between 0.6 and 1.2. According to this strategy, irrigation doses applied to FI and SDI_{75} were 5073 and 3787 $m^3 \cdot ha^{-1}$, respectively.

2.3. Plant Measurements

Measurements of leaf water potential (Ψ_{leaf}) were performed using a pressure chamber (Soil Moisture Equipment Crop., Sta. Barbara, CA, USA), monitoring six trees per irrigation treatment (two shaded leaves per tree, located in the north side of the tree and being totally mature, fresh and at 1.5 m of height, approximately). This measurement offers relevant information about the crop water status, depending on the soil water content and the atmospheric conditions [15]. In addition, in these same trees, the stomatal conductance (g_s) was measured using a porometer SC-1 (Decagon Devices, INC, WA, USA). These measurements were done using two sunny leaves per monitored tree, at 1.5 m height, and following the methodology proposed by the manufacturer (Decagon Devices, 2016). From April to September, crop-water was monitored throughout the measurements of Ψ_{leaf} and g_s between 10:00 and 11:00 (solar time), and with a periodicity of 7–10 days.

Moreover, a foliar sampling was done at the end of June, just a week before the kernel filling stage. The analyses of these samples were done with a Bran–Luebbe autoanalyzer to determine N-Kjeldahl [16] and a ICP-OES Varian ICP 720-ES to analyze P, K, Na, Ca, Mg, S, Fe, Cu, Mn, Zn, Ni, B, and inorganic contaminant [17].

2.4. Experimental Details

The experimental design was randomized blocks, with three replications per irrigation treatment, fertilization system, and cultivar. Each replication had 12 trees (3 rows and 4 trees per row), the two central trees for each replication were monitored. Thus, six trees per treatment (irrigation by fertilization strategy) were used. For each measurement day, an exploratory and descriptive analysis of the data (Ψ_{leaf} and g_s) was developed, applying a Levene's test to check the variance homogeneity of the variables studied. Significant differences between irrigation treatments ($p \leq 0.05$) and fertilization system ($p \leq 0.05$) in the variables studied were identified by applying a one-way ANOVA, with the SPSS statistical software (SPSS Inc., 15.0 Statistical packages; Chicago, IL, USA).

At the end of each season, kernel yield was obtained, measuring the total yield of each test tree for each irrigation treatment by fertilization system (n = 6), obtaining the irrigation water productivity (IWP) for each irrigation and fertilization treatment. To study the differences promoted by the irrigation doses and biostimulant applications, a Tukey's test was applied ($p < 0.05$) to seek significant differences between the interaction (irrigation × fertilization).

3. Results and Discussion

3.1. Effects of DI and HYT Application Products in the Crop's Physiological Status

Table 3 shows the main values of Ψ_{leaf} in shaded leaves, obtained for each cultivar, irrigation and fertilization treatment, and phenological state. In general, trees that received the application of HYT products showed higher Ψ_{leaf} values. This indicated that those trees that were treated with the biostimulant were less affected by drought conditions, and with a better hydration level than the untreated trees. As Table 3 shows, HYT biostimulant had some positive effects in all phenological states, these being most significant for 'Guara'. These results showed that those trees that had been fertilized with the HYT products had a greater capacity to capture the water from the soil, and therefore to keep higher levels of hydration than those that not had received this product. This hypothesis has been corroborated by other authors. Thus, Marulanda et al. [16] demonstrated that inoculation under drought conditions increased shoot and root biomass and water content. Moreover, Sharp et al. [17] found an increase in root elongation and root biomass in drought-tolerant species when these were subjected to water stress conditions.

Table 3. Mean values of Ψ_{leaf} (MPa) in cultivars, irrigation and fertilization treatments in each phenological stage.

Phenological Stage	FI (HYT)	SDI_{75} (HYT)	FI	SDI_{75}
		'Guara'		
Vegetative growth	−0.68 ± 0.15 ns ^{z}	−0.64 ± 0.18	−0.72 ± 0.13	−0.75 ± 0.15
Kernel filling	−0.66 ± 0.10 a	−0.74 ± 0.1 a	−1.01 ± 0.11 b	−0.95 ± 0.16 b
Harvest	−1.59 ± 0.11 a	−1.75 ± 0.16 b	−1.76 ± 0.13 b	−2.28 ± 0.21 c
		'Marta'		
Vegetative growth	−0.49 ± 0.1 a	−0.6 ± 0.12 b	−0.43 ± 0.11 a	−0.55 ± 0.13 ab
Kernel filling	−0.86 ± 0.16 ns	0.83 ± 0.18	−0.92 ± 0.16	−0.94 ± 0.13
Harvest	−1.65 ± 0.15 ns	−1.7 ± 0.14	−1.74 ± 0.16	−1.78 ± 0.16
		'Lauranne'		
Vegetative growth	−0.66 ± 0.11 ns	−0.63 ± 0.10	−0.66 ± 0.14	−0.76 ± 0.12
Kernel filling	−0.88 ± 0.14 ns	−0.89 ± 0.12	−0.88 ± 0.12	−0.92 ± 0.13
Harvest	−1.71 ± 0.14 a	−1.8 ± 0.16 a	−1.88 ± 0.14 a	−2.1 ± 0.13 b

^{z} Different letters indicate significant differences (within each row) among treatments ($p < 0.05$); ns indicates no significant differences among treatments within each cultivar; FI, full irrigation at 100% ET_c; SDI_{75}, sustainable deficit irrigation at 75% ET_C; HYT, treatments that received additional fertilization by using the biostimulant HYT.

Similar results were observed with stomatal conductance (Table 4). In the case of g_s for 'Lauranne', in the kernel filling and vegetative growth period, the application of HYT biostimulant increased the values of g_s. In 'Guara', neither application of HYT products or the irrigation treatments had effects.

Table 4. Mean values of g_s (mmol m^{-2} s^{-1}) in each of the cultivars, irrigation and fertilization treatments during phenological stages.

Phenological Stage	FI (HYT)	SDI_{75} (HYT)	FI	SDI_{75}
		'Guara'		
Vegetative growth	122.8 ± 11.6 ns ^{z}	116.1 ± 14.9	106.3 ± 11.7	103.5 ± 12.1
Kernel filling	219.8 ± 13.8 ns	211.3 ± 13.4	214.8 ± 19.1	217.7 ± 18.3
Harvest	198.3 ± 21.5 ns	188.5 ± 21.1	204.5 ± 21.9	197.4 ± 16.6
		'Marta'		
Vegetative growth	159.8 ± 16.3 a	148.7 ± 14.9 a	113.1 ± 11.2 b	112.3 ± 16.8 b
Kernel filling	204.5 ± 17.3 ns	198.4 ± 20.1	204.4 ± 19.6	199.3 ± 18.8
Harvest	215.7 ± 16.3 ns	213.6 ± 11.7	198.5 ± 19.9	204.5 ± 14.5
		'Lauranne'		
Vegetative growth	185.8 ± 16.7 a	198.1 ± 19.2 a	137.7 ± 19.3 b	137.2 ± 14.5 b
Kernel filling	257.9 ± 34.3 a	263.1 ± 27.9 a	207.7 ± 21.6 b	225.5 ± 22.2 b
Harvest	173.6 ± 21.4 ns	167.4 ± 22.3	169.1 ± 37.9	173.2 ± 31.2

^{z} Different letters indicate significant differences (within each row) among treatments ($p < 0.05$); ns indicates no significant differences among treatments within each cultivar; FI, full irrigation at 100% ET_c; SDI_{75}, sustainable deficit irrigation 75% at ET_C; HYT, treatments that received additional fertilization by using the biostimulant HYT.

3.2. Effects of HYT Products on the Nutritional Status of the Crop

In order to test the effects of biostimulant application on mineral nutrition, a foliar sampling was done at the end of June, just a week before the kernel filling stage. The results of these analyses are shown at Table 5. Overall, the results were not clear enough to conclude improvements in relation to the HYT product or negative effects in relation to the water stress application. According to the findings, it should be noted that in the case of Ca, Fe, and Mn, the treatments with HYT products showed higher values. By contrast, no effects were observed for the remaining elements in the trees without HYT application. These data are in line with the results obtained by other authors. Creus et al. [18] reported that the grains harvested from plants with *Azospirillum* and subjected to water stress had a higher concentration of Mg, K, and Ca compared to plants that were not treated. Other bacterial species, such as *Pseudomonas spp*. and *Bacillus spp*., have also been reported to stimulate plant growth under dry conditions [19]. Moreover, Parađiković et al. [20], in a study with begonia (*Begonia* spp.) transplants, found that the plants treated with biostimulant had higher levels of K in the root, and significantly higher concentrations of total N, K, Ca, and Mg were recorded in above-ground parts of biostimulant-treated plant.

Table 5. Average foliar mineral content for almond cultivars subjected to irrigation and fertilization treatments.

Nutrients	'Guara'				'Marta'				'Lauranne'			
	FI [z] (HYT)	SDI_{75} (HYT)	FI	SDI_{75}	FI (HYT)	SDI_{75} (HYT)	FI	SDI_{75}	FI (HYT)	SDI_{75} (HYT)	FI	SDI_{75}
Ca (%)	4.19 a [y]	3.49 a	3.03 b	4.19 a	4.24	4.67	3.76	3.78	4.19 a	2.81 b	3.16b	4.12 a
P (%)	0.154	0.163	0.158	0.148	0.15	0.135	0.16	0.144	0.167 a	0.187 a	0.181 a	0.15 b
Mg (%)	0.542	0.559	0.523	0.608	0.548	0.635	0.541	0.524	0.586	0.46	0.536	0.609
K (%)	1.94	1.79	1.9	2.04	1.93	1.84	1.97	2.01	2.0	1.88	1.93	1.88
N-Kjeldhal (%)	2.66	2.55	2.67	2.61	2.57	2.4	2.64	2.49	2.72	2.77	2.8	2.79
Cu ($mg \cdot kg^{-1}$)	5.66	4.88	5.21	5.73	5.75	5.16	5.73	6.03	6.66	5.79	6.1	6.34
Fe ($mg \cdot kg^{-1}$)	70.3 a	50.8 b	46.5 b	68.9 a	58.9 b	76.9 a	57.1 b	78.7 a	90.4 a	51.1 b	57.4 b	76.1 a
Mn ($mg \cdot kg^{-1}$)	77.6 a	54.5 b	42.5 b	83 a	68.6 a	72.1 a	48.2 b	90 a	93.1 a	47.5 b	54.4 b	87.2 a
Zn ($mg \cdot kg^{-1}$)	27.6	24.6	23.9	26.4	25.8	20.3	28.1	23.7	27.7	23.2	22.0	22.5

[z] FI, full-irrigated at 100% ET_C; SDI_{75}, sustained-deficit irrigation at 75% ET_C; HYT, treated with biostimulant.
[y] Different letters indicate significant differences among treatments within each cultivar ($p < 0.05$). The absence of letters indicates no significant differences among treatments within each cultivar.

3.3. Effects of DI and the Application of HYT Products in the Crop Yield Response

The effects of the deficit-irrigation and fertilization strategies were studied in the final values of yield and the irrigation water productivity (IWP) for the cultivars (Table 6). In the case of 'Guara', yield values close to 1900 $kg \cdot ha^{-1}$ of kernel yield for FI without HYT applications and reductions of 14% for SDI_{75} were obtained under the same fertilization program. The kernel yield values obtained for this cultivar and with the application of the biostimulant were 2221 $kg \cdot ha^{-1}$ for FI treatment and 1980 $kg \cdot ha^{-1}$ for SDI_{75} (HYT). The improvements obtained in the yields of those trees that had received the HYT products were 15% and 19% for FI (HYT) and SDI_{75} (HYT), respectively. Even, the SDI_{75} (HYT) treatment overcame the effect of the FI treatment without HYT application, to produce a comparable yield. These improvements were linked to higher values in almond unit weight, with only significant differences between FI (HYT) and SDI_{75} (HYT) in comparison to FI and SDI_{75}.

Table 6. Mean values of kernel yield, almond unit weight, and almonds number per tree for each cultivar subjected to irrigation and fertilization treatments.

Yields Parameters	FI (HYT) [z]	SDI_{75} (HYT)	FI	SDI_{75}
'Guara'				
Kernel yield ($kg{\cdot}ha^{-1}$)	2221 a [y]	1980 ab	1928 b	1659 c
Unit weight (g)	1.57 a	1.56 a	1.40 b	1.40 b
Almonds number per tree	6790 a	6092 a	6610 a	5688 b
'Marta'				
Kernel yield ($kg{\cdot}ha^{-1}$)	1947 a	1962 a	1933 a	1677 b
Unit weight (g)	1.33 ns	1.40	1.33	1.39
Almonds number per tree	7026 a	6727 a	6976 a	5791 b
'Lauranne'				
Kernel yield ($kg{\cdot}ha^{-1}$)	2486 ns	2578	2349	2343
Unit weight (g)	1.09 b	1.28 a	1.12 b	1.13 b
Almonds number per tree	10,948 a	9668 b	10,067 a	9953 b

[z] FI, full-irrigated at 100% ET_C; SDI_{75}, sustained-deficit irrigation at 75% ET_C; HYT, treated with biostimulant.
[y] Different letters indicate significant differences among treatments within each cultivar ($p < 0.05$); ns indicates no significant differences among treatments within each cultivar.

In relation to the IWP, the FI NB treatment resulted in a value of 0.35 $kg{\cdot}m^{-3}$, while FI (HYT) exhibited a value of 0.49 $kg{\cdot}m^{-3}$. This result indicated that the application of HYT products coupled with a DI strategy allowed correction for possible yield losses, increasing the IWP in a remarkable way.

With regards to 'Marta', the results were similar to those obtained with 'Guara'. In this sense, a kernel yield value of 1950 $kg{\cdot}ha^{-1}$ for the FI treatment was reached under both fertilization systems. Regarding SDI_{75}, the production values amounted to 1677 $kg{\cdot}ha^{-1}$ without HYT application and 1933 $kg{\cdot}ha^{-1}$ for SDI_{75} (HYT), differences that were significant. For 'Marta', these improvements in final yield were linked to effects on the fruit number per tree, the response being different from that for 'Guara'. Thus, for 'Marta' there were no significant improvements in the final yield for FI (HYT), probably because it might have reached its maximum productive potential. Thus, the biggest yield increments were measured in SDI_{75} (HYT). Yield losses for SDI_{75} without HYT were 13%, whereas in the case of SDI_{75} (HYT), there were no yield losses. Finally, regarding IWP, values of 0.38 and 0.41 $kg{\cdot}m^{-3}$ were noted in the case of FI and SDI_{75}, respectively, whereas for the FI (HYT) and SDI_{75} (HYT) treatments, values of 0.38 and 0.48 $kg{\cdot}m^{-3}$ resulted.

The variety 'Lauranne' exhibited the best yield in comparison to the other two cultivars. In addition, this cultivar showed the best response in terms of water stress. There were no significant differences in yield among the treatments. As occurred in 'Marta', the trees under the FI treatment (with and without HYT application) were close to their maximum productive potential; therefore, there were no improvements with the application of the HYT products. Finally, the IWP values were 0.46 and 0.57 $kg{\cdot}m^{-3}$ for the FI and SDI_{75} treatments, respectively, whereas in the case of trees fertilized with HYT, they were 0.49 and 0.63 $kg{\cdot}m^{-3}$ for FI and SDI_{75}, respectively.

Overall, the results suggested improvements in terms of yield, especially, when a DI is being applied. In this regard, 'Guara' offered the most positive response to biostimulant applications, whereas for 'Lauranne', there were no treatment effects. Finally, 'Marta' did not show differences between FI (HYT) and SDI_{75} (HYT), whereas these differences were more evident and significant between FI and SDI_{75} without HYT application. More interesting were the parameters that explained these effects in terms of kernel yield. In this aspect, for 'Guara', the effects on the final yield were related to the unit weight of the almond kernel and the fruit number per tree. By contrast, 'Marta', the yield response was similar to that observed with fruit number per tree, whereas for 'Lauranne', these relationships were not as clear as in the remaining cultivars.

Casanovas et al. [21] demonstrated that inoculation of maize (*Zea mays* L.) seedlings with *A. brasilense* resulted in the mitigation of many negative effects of drought stress. In addition, Cohen et al. [22] reported that plants inoculated with the PGPB *A. brasilense* Sp245 showed more abscisic acid content than non-inoculated plants, and this resulted in enhanced plant drought tolerance.

4. Conclusions

This work summarizes the most relevant effects in terms of the crop physiological and yield response of three almond cultivars subjected to different irrigation regimes, and its interaction with the biostimulant application. Although this experiment occurred in a single season, similar responses were observed among the three cultivars. Each of the three varieties showed improvements with the application of the HYT product. These improvements were detected in physiological and/or production terms, and these effects were especially remarkable in the case of SDI_{75}.

In the yield response, the effect differed depending on the irrigation strategy imposed. Thus, in the FI treatment the improvement ranged between 2 and 15%, whereas for the SDI_{75} treatment, this enhancement differed between 8 and 19%, depending on the cultivar.

In relation to physiological variables, the application of biostimulants improved the response to water stress of the three varieties, showing higher Ψ_{leaf} values. In this regard, HYT biostimulant had a positive effect in all the crop phenological states, these being more significant for 'Guara'. On the other hand, with respect to the stomatal conductance behavior, the improvement was not as evident as it was for Ψ_{leaf}.

Overall, taking into account all of the results, we can conclude that the use of biostimulants under drought conditions can improve the almond yield response. However, some variation was observed relating to the cultivar. It is essential to continue this type of work with the aim of defining the best DI strategies and increasing the experimental surface to corroborate these conclusions.

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