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Vegetative and Reproductive Responses Induced by Organo-Mineral Fertilizers on Young Trees of Almond cv. Tuono Grown in a Medium-High Density Plantation

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Abstract: Field experiments were conducted in three successive seasons (2019–2021) to evaluate the effects of four commercial organo-mineral fertilizers with biostimulating action (Hendophyt®, Ergostim®, and Radicon®) on the vegetative and productive performance of young almond trees (*Prunus dulcis*, cv. Tuono) grown in a semiarid climate in Southern Italy. Foliar treatments were applied three times during each season (at the swollen bud, beginning of flowering, and fruit set-beginning of fruit growth stages). Both 2020 and 2021 were adversely affected by late frosts, resulting in damage to the flowers and small fruits without any positive effect of the biostimulant applications. In contrast, the results obtained during the normal climate year (2019) indicated that the growth of trunk diameter and shoot length of trees tended to increase in biostimulant treatments compared to those of the control. The number of buds and flowers per unit length of the branch revealed no significant differences among years and all compared treatments. However, in 2019, the fruit set percentage, number, and weight of kernels per tree were significantly higher in the biostimulant treatments compared to those of the control. To this regard, the use of biofertilizers is suitable for maintaining soil fertility and improving crop productivity This information holds significance for almond tree growers.

Keywords: almonds; foliar spray; humic and fulvic acids; carboxylic acids; polyglucosamine; flowering; yield

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1. Introduction

The almond tree (*Prunus dulcis* [Mill.] D.A. Webb) is cultivated in the Northern hemisphere between 30° and 44° latitude and in the Southern hemisphere between 20° and 44° latitude. This cultivation spans over 40 countries, covering a total area of 2,283,414 hectares and yielding a production of 3,993,998 tons of almonds in their shells [1]. Italy holds the seventh position among the world's largest almond producers, with 54,939 Ha cultivated and a total production of 77,677 tons in 2023 [2,3]. The majority of almond cultivation in Italy is concentrated in the Southern regions, particularly in Sicily (52,185 tons on 32,905 hectares) and Puglia (18,445 tons on 18,891 hectares) [3]. There is currently a growing interest in this cultivation even in emerging regions with climates and temperatures favorable to the development and fruiting of the almond tree [4,5].

Over the past 10 years, new almond cropping systems, inspired by the Californian model, have been emerging worldwide in irrigated areas. These systems, including medium-high density (MHD, about 300 to 1000 trees ha^{-1}) and super-high density (SHD, resulting more than 2000 trees ha^{-1}) of trees, prioritize mechanization and sustainability to enhance efficiency and productivity [6,7]. In Italy, a significant portion of the new almond acreage is dedicated to the cultivation of both Lauranne® Avijor (constituting 48% of the total) and Guara-Tuono (comprising 39% of the total). The choice of Rootpac-20® as rootstock is crucial for controlling tree vigor, promoting early production, and improving adaptation to specific soil conditions [7]. A new cultivation system requires the definition of efficient agronomic techniques, with fertilization as a crucial management tool to enhance both growth and quantitative–qualitative yield parameters. In this context, the goal of modern agriculture is to employ a sustainable fertilization strategy that complements chemical fertilizers. In this regard, the use of biofertilizers is suitable to improve nutrient use efficiency and ensure the stability of crop yields under both optimal and suboptimal conditions [8,9]. In this context, agricultural biostimulants (ABs), composed of organic and inorganic materials of various origins, many of which are still unknown, constitute an important category of agricultural inputs with multiple functions [10–12]. These products are utilized to support crop growth, enhance yield, and improve the final quality of produce [13–17]. Specifically, they aim to mitigate nutritional stresses arising from abiotic factors such as drought, soil salinity, and various climatic parameters to which crops may be exposed [18,19]. In almonds, buds are regarded as a crucial yield component [20], and their development is significantly influenced by environmental and management factors [21]. Furthermore, flowering is a fundamental phase in the plant's life cycle, as the overall yield is contingent upon both the number of flowers on a tree and the percentage of flowers that ultimately result in fruit formation [22]. For this reason, agronomic practices should strive to maintain the highest and consistent number of flowers throughout the growing season of the orchard. Regarding climatic conditions, the almond tree's early flowering (in February in Italy) compared to that of other fruit trees makes it susceptible to damage from frosts, occurring at temperatures as low as −2 °C during this period. Late frosts, in particular, constitute the primary limiting factor in the cultivation of this species in the Mediterranean basin [5,23,24]. Furthermore, during flowering, leaves are either absent or still too small to provide the necessary nutrients at the time of setting and in the immediate post-setting moments. As a result, plants can benefit from the foliar absorption of nutrients, such as the small organic molecules contained in biostimulants. Although the effect of foliar-applied biostimulant substances on plant growth, yield, and fruit quality has been studied in various fruit tree species [16], the availability of information on this effect on almonds is relatively limited. Existing studies refer to research conducted in pots [25] or in fields using traditional low-density systems [26–30]. Notably, no research has been published on almond trees in the medium-high density (MHD; about 300 to 1000 trees/ha) and super-high density (SHD; resulting in more than 2000 trees/ha) systems. The results of research on potted plants [25] indicate that foliar applications of two biostimulants derived from microbial fermentation and algae extraction, respectively, demonstrate a substantial positive effect on the total leaf shoot area. There was also a significant increase in shoot length and biomass. Regarding the nutritional content of almond fruit, various types of plant biostimulants, in general, led to elevated levels of important bioactive compounds, particularly concerning γ-tocopherol and β-tocopherol [27]. The use of biostimulants under drought conditions improves the almond yield response of three varieties (Guara, Marta, and Lauranne), demonstrating higher leaf water potential values [28]. In another study [29], the results indicate that certain treatments involving foliar fertilization with urea and humic acid at different concentrations lead to a significant increase in components of vegetative growth, including the length and diameter of the stem, leaf area, fresh weight, and dry weight. In an experimental test conducted in Egypt [30], the foliar application of humic acid and milagro enhanced the vegetative growth of the seedlings. This improvement was evident in the length of the stem, diameter, number of branches and leaves, leaf area, fresh and dry leaf weight, and specific weight of dry leaf. Additionally, there was an increase in chlorophyll and leaf mineral content compared to those of untreated young trees.

Taking into account all the considerations mentioned above, the objective of this study was to assess, in the semiarid environment of the Apulia region in Italy, the effects of foliar applications of four commercial organo-mineral fertilizers (Hendophyt® PS, Iko-Hydro, Rutigliano BA, Italy; Ergostim® XL, Isagro SpA, Sumitolo Chemical, Italy; and Radicon®, Fertek, Cavizzano NA, Italy) on the vegetative growth (shoot and trunk growth), bud production, flowering, fruit set, and yield of almond cv. Tuono in the SHD system.

2. Materials and Methods

2.1. Trials Site and Bioastimulant Treatments

A three-year study was conducted in an irrigated almond orchard using the medium-high density (MHD) system from 2019 to 2021, corresponding to the 3rd, 4th, and 5th years after planting (YAP). The commercial orchard is situated in the Foggia countryside, located in the Apulia region of southern Italy at coordinates 41°27′08″ N, 15°31′56″ E and an elevation of 54 m above sea level. The orchard consists of Tuono variety almond trees (synonymous with Guara) [6,31], grafted on a hybrid Rootpak 20® of *Prunus besseyi* × *Prunus cerasifera* L-H. Bailey and Ehrh. The trees are spaced 4 × 1.5 m^2 apart (1666 trees ha^{-1}) and are grown in a vase shape with three production axes, oriented in rows from North to South. Tuono is a native variety from Apulia and is currently cultivated in the primary Italian almond cropping areas and other European regions due to its self-fertility and favorable fruit characteristics [32].

The soil texture is a silty-clay vertisol of alluvial origin (1.20 m depth) (Typic Chromoxerert, fine, thermic, according to the Soil Taxonomy-USDA-NRCS 1999 [33]). The soil composition includes sand (36.8%), silt (32.7%), and clay (30.5%), with various essential parameters: total N (Kjeldahl) = 1.5‰, assimilable P_2O_5 (Olsen) = 56 mg kg^{-1}, exchangeable K_2O (Schollemberger) = 1390 mg kg^{-1}, exchangeable Ca = 3128 mg kg^{-1}, electrical conductivity (ECe) = 0.68 dS cm^{-1}, pH (soil: water 1:2.5) = 8.0, and organic matter = 1.6%. In this study, four water-soluble commercial organo-mineral fertilizers with biostimulant action—Hendophyt® PS (Iko-Hydro), Ergostim® XL (Isagro), and Radicon® (Fertek)—were applied through foliar spraying and compared to a control (sprayed with water). Table 1 presents the composition, including the main active compounds and the dosage of different products used in the trials. Specifically, these formulations include polysaccharide biopolymers (polyglucosamine), carboxylic acids (N-acetylthiazolidin-4-carboxylic acid—AATC and triazolidinecarboxylic acid—ATC), as well as humic and fulvic acids. These substances contribute to the biostimulating action in plants [34]. The products were applied three times during each growing season, specifically at the swollen bud, beginning of flowering, and fruit set–beginning of fruit growth stages. The application dates were 5 March, 12 April, and 8 May in 2019; 3 March, 10 April, and 4 May in 2020; and 26 February, 16 March, and 20 April in 2021. All treatments were administered between 10:00 and 11:30 am, with a total volume of 550 L ha^{-1}. Each tree was sprayed using a pulled sprayer under favorable weather forecasts, ensuring no rainfall was expected in the following 24 h. The experimental setup followed a completely randomized block design, with three replications per treatment and five trees per plot. The trial was inserted in an orchard with a surface area of approximately 2 hectares. One buffer row was located between replicates and blocks, and two or more buffer rows were around the perimeter of the experimental field. Each replicate had 15 plants, and three centrally located plants per plot were used to collect vegetative and reproductive parameters.

Table 1. Formulations and doses of foliar application of agricultural biostimulant (AB) commercial products used in the experiment.

ABsTreatment
HENDOPHYT PS (Iko-Hydro): a fully water-soluble powder comprising biopolymers of polysaccharides (polyglucosamine), 60%; carbon, 35%; organic nitrogen, 4%; boron, 0.25%; applied at a dose of 150 g 100 L^{-1} of water.
ERGOSTIM XL (Isagro): a concentrated water-soluble liquid N-acetiltiazolidin-4-carboxylic acid (AATC), 2.5%; and triazolidine-carboxylic acid (ATC) 2%; applied at a dose of 200 mL 100 L^{-1} of water.
RADICON (Fertek): a suspension–solution of humic and fulvic acids, obtained from worm compost (night crawled). Dry composition: total organic matter, 60%; extractable organic substance of organic matter, 4%; humified organic substance extractable organic matter, 90%; organic substance of extractable organic nitrogen, 1.0%; C/N ratio = 4; applied at a dose of 500 g 100 L^{-1} of water.

To prevent contamination between treatments, a buffer row was positioned between replicates and blocks, and two or more buffer rows were established around the perimeter of the experimental field. In each replicate, three centrally located plants per plot were selected for the collection of vegetative and reproductive parameters. Trees were chosen to be healthy and as uniform as possible. The same set of trees was consistently selected for the experiment across the three growing seasons under consideration.

2.2. The Climate

The research site was situated in a typical semi-arid zone, characterized by a Mediterranean climate classified as an accentuated thermomediterranean climate [35]. The temperatures in this region may fall below 0 °C in the winter and exceed 40 °C in the summer. Rainfall is unevenly distributed throughout the year, with the majority concentrated in the winter months, resulting in a long-term annual average of 559 mm [36]. Daily climatic parameters, including maximum and minimum temperatures, air humidity, wind speed, and total precipitation, during the three growing seasons were recorded by the meteorological station nearest to the experimental area, supplied by Syngenta [37]. The weather conditions varied significantly among the three years, particularly in terms of air temperature and rainfall (Table 2). A notable difference in air temperatures was observed during the flowering and fruit set period in the frost-heavy seasons of 2020 and 2021. During these seasons, trees and flowering plants were affected by actual ice stalactites (Figure 1), in contrast to the more favorable temperature trend for almond growth observed in 2019.

Figure 1. Ice stalactites on almond trees.

Specifically, in 2020, frosts were recorded on 24 and 25 March (−0.24 and −1.43 °C, respectively), occurring after the first biostimulant treatments. In 2021, the frosts occurred very late, on 8, 9, and 10 April (−0.6, −2.6, and −0.9 °C, respectively), after both the first and the second biostimulant treatments had taken place (Table 3). Consequently, the average maximum and minimum temperatures in March 2020 were colder, with averages of 15.6 °C and 2.1 °C, respectively, compared to those in 2019, which had averages of 18.6 °C and 8.2 °C, respectively. Similarly, in April 2021, the average maximum and minimum temperatures (15.4 °C and 3.4 °C, respectively) were lower than those recorded in April 2019. Furthermore, the annual precipitation was higher in 2021, reaching 627.8 mm, compared to 527.1 mm in 2020 and 461.7 mm in 2019.

The orchard was managed using common practices prevalent in the area. Drip lines with 2 L h^{-1} drippers spaced 40 cm apart were positioned 50 cm from the ground along the tree rows. Controlled irrigation was implemented, with a mean seasonal irrigation volume of 3500 m^3 ha^{-1}. Fertilization was conducted annually through the fertigation system, involving 100 kg ha^{-1} of N, 60 kg ha^{-1} of P, and 80 kg ha^{-1} of K. Protection against fungal diseases primarily occurred in the autumn–winter period using copper-based products compliant with phytosanitary regulations outlined in the Integrated Production Regulations of the Puglia Region [38].

Table 2. Monthly mean maximum and minimum temperatures (T_{max}, T_{min}) and relative air humidity (RH_{max} and RH_{min}), wind speed (W_s), and total precipitation (P) in 2019, 2020, and 2021.

Month	**T_{max} (°C)**	**T_{min} (°C)**	**RH_{max} (%)**	**RH_{min} (%)**	**W_s (m s^{-1})**	**P (mm)**
2019						
Jan	10.6	1.6	99.2	63.3	3.4	61.0
Feb	14.6	2.6	95.1	51.2	4.3	21.2
Mar	18.6.	4.5	98.8	44.2	4.4	32.0
April	20.6	8.2	94.4	51.0	3.7	40.3
May	21.3	10.2	95.3	56.3	4.0	86.7
June	33.2	17.5	85.9	35.1	3.7	9.2
July	33.7	19.5	84.0	33.9	3.7	30.0
Aug	34.8	20.3	79.9	33.9	3.6	5.7
Sept	29.5	16.8	88.7	42.6	3.6	3.8
Oct	25.5	11.5	93.2	43.9	2.6	29.2
Nov	19.3	9.4	98.5	62.2	5.2	112.6
Dec	14.7	5.0	99.0	65.2	6.5	30.0
Mean	23.4	10.6	92.7	48.6	4.1	
Total						461.7
2020						
Jan	10.5	1.6	98.3	55.1	4.8	3.6
Feb	14.6	2.9	94.8	42.6	5.1	51.0
Mar	15.6.	2.1	96.4	60.8	3.3	83.0
April	18.8	6.1	94.1	53.2	3.4	48.9
May	27.5	14.7	90.8	43.1	3.8	25.8
June	28.8	17.7	80.5	48.3	4.0	19.7
July	31.0	21.2	79.7	40.6	3.9	20.4
Aug	31.5	21.8	83.7	44.3	3.9	40.0
Sept	22.2	17.4	72.8	58.4	4.0	38.5
Oct	25.5	9.7	97.1	47.6	3.9	44.6
Nov	19.3	7.7	99.5	72.8	4.2	68.6
Dec	14.7	5.2	99.6	71.9	4.3	83.0
Mean	21.7	10.9	90.6	53.2	4.1	
Total						527.1
2021						
Jan	12.2	2.4	99.5	63.3	5.8	58.2
Feb	15.5	3.4	99.6	56.0	5.1	35.2
Mar	15.4	3.4	98.9	52.3	4.7	57.8
April	19.9	4.7	99.5	44.7	4.3	40.4
May	26.5	10.8	95.7	30.3.	3.5	26.0
June	33.2	15.9	85.1	24.7	3.3	8.6
July	35.4	19.3	83.8	26.1	3.7	100.8
Aug	34.9	19.4	92.3	28.3	3.8	29.2
Sept	29.5	15.4	94.8	35.6	3.5	19.4
Oct	21.2	10.9	98.5	54.9	3.5	70.2
Nov	17.2	10.8	99.6	80.0	3.1	135.4
Dec	13.7	4.8	99.0	64.9	4.7	46.6
Mean	22.9	10.1	95.5	48.3	4.1	
Total						627.8

Table 3. Daily mean maximum and minimum temperatures (T_{max}, T_{min}), relative air humidity (RH_{max} and RH_{min}), wind speed (W_s), and total precipitation (P) for the frosty days: 24 and 25 March, 2020 and 8, 9, and 10 April, 2021.

Date	T_{max} (°C)	T_{min} (°C)	RH_{max} (%)	RH_{min} (%)	W_s (m s^{-1})	P (mm)
				2020		
24 March	5.8	−0.3	99.5	67.0	5.0	10.8
25 March	6.5	−1.4	99.6	81.2	1.1	14.4
Mean	6.1	−0.8	99.5	74.1	3.05	
Total						24.4
				2021		
8 April	13.9	−0.6	99.4	23.2	1.7	1.0
9 April	18.1	−2.6	99.3	23.4	2.7	0
10 April	21.1	−0.9	99.4	15.9	3.7	0
Mean	17.7	−1.4	99.4	20.8	2.7	
Total						1.0

2.3. Plant Measurements

2.3.1. Vegetative Growth

The shoot length (SL) and trunk diameter (TD) were determined at the beginning (in February) and the end (in September) of each season on three central plants in each treated plot. SL was determined on two well-lit one-year-old shoots (subsamples) randomly selected from opposite sides (east and west) of the outer canopy of each plant. The selected shoots were marked and measured using a tape, with the measurements expressed in centimeters. TD was measured at a marked point 50 cm above the ground level using a Vernier digital caliper, and the measurements were expressed in millimeters. Annual shoot growth (ASG) and annual trunk growth (ATG) were calculated based on the difference between the measurements taken in February and those in September for each year.

2.3.2. Bud, Flower, and Fruit Counting

Throughout each year, on the previously mentioned three central trees, four branches per tree were randomly selected for measurements, including 1-year-old shoots and spurs. All selected branches were chosen as homogeneously as possible, originating from opposite sides of the canopy and being of the same order of branching, with an approximate length of 1 m and positioned ≈1.7 m above the ground. Approximately 150–200 buds (both flower and vegetative) were counted and recorded at the pre-blossom phase on each of these selected branches. Thus, 600–800 buds were found on each tree. Subsequently, the length of all the branches was measured, and the count of all buds was conducted on each of the four branches [39]. Measurements were taken when flower buds were just before bloom, at phenological stage B [40] (on 1 March 2019, 24 February 2020, and 20 February 2021) and when the flowers were completely open at stage F [40] (on 11 March 2019, 4 March 2020, and 10 March 2021). The parameters considered for analysis were bud density (buds cm^{-1}) and flower density (flowers cm^{-1}). Finally, the final fruit set, expressed as a percentage of fruit per total open flowers, was evaluated at a later date (on 10 July 2019, 17 July 2020, and 15 July 2021).

2.4. Harvesting, Fruit Collection and Yield

In each year, almond fruits for each treatment were hand-harvested at the commercial maturity stage (on 20 September 2019, 24 September 2020, and 30 September 2021), and the number and weight of fresh almond fruits per tree were measured. Samples of 2 kg of almonds with hulls were taken from each replicate, stored in plastic bags, and transported to the laboratory. Each fruit in the samples was separated from the hull, and the nuts were left to dry on the ground in the sun for 5 days, bringing the humidity to about 10% of the weight. The results were expressed as hull per fruit (% of the total fresh weight), kernel dry yield (in % of kernel per nut), and double seeds (%). Furthermore, 10 fruits were randomly collected from each replication and subjected to the following morphological analyses: weight, length, width, thickness of the nuts, and kernels.

2.5. Statistical Analysis

The results were assessed using one-way ANOVA with JMP® software version 8 (SAS Institute Inc., Cary, NC, USA), and average values were compared using Tukey's test. Standard deviations (SD) were calculated using Excel from the Office 2007® suite (Microsoft Corporation, Redmond, WA, USA). Percentage values were transformed to arcsine before conducting the analysis of variance.

3. Results

3.1. Trunk and Shoot Development

Annual trunk growth (Figure 2) exhibited no significant differences among treatments over the years. However, overall, it tended to increase in the biostimulant treatments (average 20.5 mm) compared to that of the control (18.9 mm). Additionally, there was a decreasing trend from the first to the third year, with average values ranging from 22.4 to 20.8 and 17.0 mm, respectively.

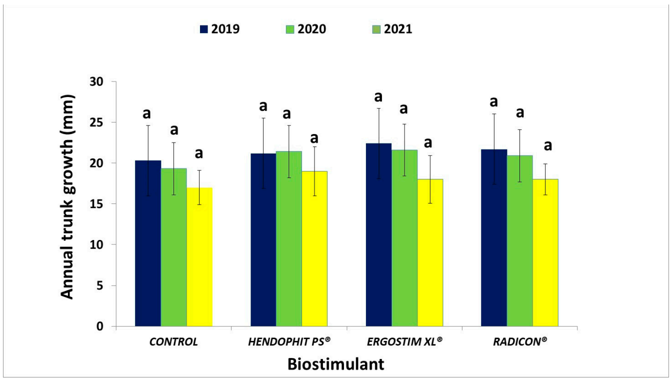

Figure 2. Annual trunk growth in different biostimulant and control treatments. The data are average values ± SD in three subsequent years (2019–2021). Similar letters per year and treatment indicate no significant differences according to Tukey's test ($p < 0.05$).

Additionally, shoot development (Figure 3) exhibited no significant differences among treatments and the control. However, the average shoot length each year tended to be higher under the biostimulant treatments (59.4 cm in 2019, 24.2 cm in 2020, and 23.3 cm in 2021) compared to that of the control (50.1 cm in 2019, 11.5 cm in 2020, and 15.3 cm in 2021).

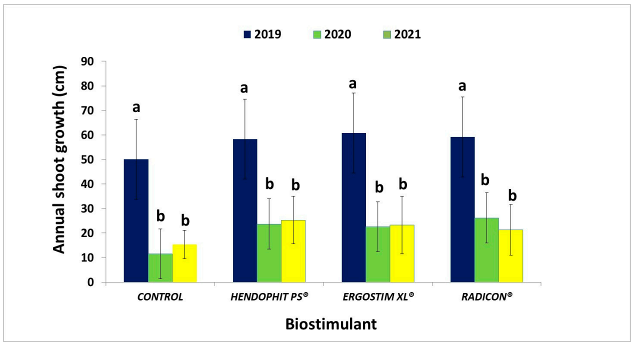

Figure 3. Annual shoot growth in different biostimulant and control treatments. The data are average values ± SD in three subsequent years (2019–2021). Different letters among years indicate significant differences according to Tukey's test ($p < 0.05$).

A significantly higher shoot length was observed in 2019 (ranging from 50.1 to 59.2 mm) compared to that of both 2020 and 2021 (ranging between 21.5 and 26.2 mm, respectively). The lower vegetative growth recorded in 2020 and 2021 could be attributed to the frosts that occurred in the respective months of March during these years, indicating the susceptibility of almond trees to climatic conditions.

3.2. Agronomical Characteristics: Bud, Flower, and Fruit Productivity

In Table 4, bud, flower, and fruit productivity is reported. Regarding total bud density, no statistical differences were found among treatments and years. However, it tended to be higher in both 2019 and 2020 (averaging 1.00 and 1.08 buds cm^{-1}, respectively) than in 2021 (averaging 0.89 buds cm^{-1}). Similarly, flower density showed no significant differences among years and treatments (averaging 0.50 flowers cm^{-1}), corresponding to flowering in 49% of the total bud population.

Table 4. Agronomical characteristics of almond trees in different biostimulant treatments and control in three subsequent years (2019–2021).

Parameter	Year	Treatment				
		Control	Hendophit PS®	Ergostim XL®	Radicon®	Average
Bud density (No cm^{-1})	2019	1.02 ± 0.30	0.99 ± 0.17	1.02 ± 0.17	0.97 ± 0.12	1.00 ± 0.19
	2020	1.10 ± 0.30	0.97 ± 0.28	1.08 ± 0.25	1.15 ± 0.16	1.08 ± 0.22
	2012	0.88 ± 0.18	0.89 ± 0.21	0.85 ± 0.26	0.94 ± 0.16	0.89 ± 0.20
Flower density (No cm^{-1})	2019	0.51 ± 0.10	0.43 ± 0.07	0.54 ± 0.10	0.44 ± 0.08	0.48 ± 0.08
	2020	0.45 ± 0.11	0.44 ± 0.18	0.50 ± 0.11	0.40 ± 0.09	0.45 ± 0.12
	2021	0.53 ± 0.19	0.56 ± 0.11	0.54 ± 0.15	0.65 ± 0.21	0.57 ± 0.20
Final fruit set incidence (%)	2019	21.5 ± 4.5 b	28.3 ± 1.3 a	28.4 ± 4.5 a	34.4 ± 5.4 a	28.4 ± 4.9 A
	2020	5.8 ± 7.8	2.4 ± 4.7	1.6 ± 5.2	2.8 ± 4.3	3.1 ± 5.5 B
	2021	9.8 ± 9.2	6.2 ± 5.1	6.9 ± 6.8	15.9 ± 9.9	9.7 ± 7.7 B
Fruit set per tree (No $tree^{-1}$)	2019	66.3 ± 8.5 b	85.7 ± 4.6 a	96.7 ± 5.3 a	81.6 ± 6.5 a	82.6 ± 6.2 A
	2020	55.3 ± 12.3	65.7 ± 11.4	55.0 ± 9.0	71.9 ± 8.2	65.0 ± 10.2 B
	2021	44.3 ± 8.0	48.5 ± 9.4	46.6 ± 10.0	46.9 ± 11.3	46.6 ± 9.7 B
Fresh kernel yield per tree (g)	2019	359.5 ± 74.5 b	460.2 ± 14.9 a	477.3 ± 40.0 a	405.7 ± 32.8 a	425.7 ± 56.3 A
	2020	298.2 ± 29.6	420.6 ± 57.9	333.6 ± 29.6 a	296.4 ± 65.1	337.2 ± 40.5 A
	2021	221.4 ± 16.1 c	242.0 ± 15.3 c	251.0 ± 18.4 c	235.2 ± 16.1 b	237.4 ± 16.5 A

The data are averages ± sd of different treatments in each year and averages ± SD across all seasons. Different lowercase letters on the lines indicate significant differences among biostimulant treatments, while lines followed by no letter are not significantly different (Tukey's test, $p < 0.05$). Different capital letters among year averages indicate significant differences at $p < 0.05$. The absence of letters indicates no significant differences among the years.

Furthermore, the observations in Table 4 itself indicate that only in 2019 the fruit set percentage was statistically higher in the biostimulant treatments than in the control. This parameter was the highest in the Radicon® treatment (34.4%), although it was not significantly different from that in both Hendophyt® and Ergostim XL® treatments (28.3 and 28.4%, respectively), and was significantly higher than that in the control (22.5%). A remarkably low percentage of fruit set was detected in both 2020 (ranging from 2.6 to 5.8%) and 2021 (ranging from 6.2 to 15.9%), with no discernible differences among the treatments. These results could be explained by the aforementioned adverse weather conditions that occurred in these last years.

The final number and weight of fruits per tree, parameters related to fruit set, indicated significantly higher average values in 2019 (82.5 No $tree^{-1}$ and 425.7 g, respectively) than in both 2020 (65.0 No $tree^{-1}$ and 337.2 g) and 2021 (46.6 No $tree^{-1}$ and 237.4 g). It should be noted that considering the data reported above, the reductions in fruit set percentage that occurred in 2020 and 2021 compared to that in 2019 were higher than the relative reductions detected in the same years in both the total number and weight of fruits per tree. This, of course, was due to the increase in plant canopy that certainly occurred over the years.

The positive effects of biostimulant treatments were noticed only in the 2019 season when the number and weight of fruit per plant were significantly higher (on average 88.0 No tree^{-1} and 447.3 g, respectively) compared to those of the control (66.3 No tree^{-1} and 359.5 g, respectively). On the contrary, no significant differences were found among treatments in both 2020 and 2021 when the late frosts occurred.

3.3. Yield-Related Variables

The fruit quality parameters reported in Table 5 showed no statistical differences both among the years and biostimulant treatments. Mean percentage values for hull per fruit, kernel per nut, and double seeds in the three years ranged from 39.6 to 52.3, from 28.1 to 31.4, and from 6.9 to 10.4%, respectively.

Table 5. Fruit quality parameters of almonds in different biostimulant treatments and control in three subsequent years (2019–2021).

Parameter	Year	Treatment				
		Control	Hendophit PS®	Ergostim XL®	Radicon®	Average
Hull per fruit (% of total fresh weight)	2019	52.3 ± 7.1	48.4 ± 8.9	44.9 ± 0.9	44.8 ± 3.4	47.6 ± 5.1
	2020	44.6 ± 9.4	43.7 ± 2.1	44.3 ± 7.6	39.6 ± 2.7	43.0 ± 5.4
	2021	44.1 ± 3.1	45.0 ± 4.1	43.9 ± 3.7	43.3 ± 5.0	44.1 ± 4.0
Shelling: Kernel per nut dry (%)	2019	28.1 ± 4.5	30.1 ± 4.9	31.4 ± 5.2	29.8 ± 6.1	29.8 ± 5.2
	2020	31.3 ± 0.9	30.3 ± 0.8	30.8 ± 0.8	30.2 ± 2.9	30.6 ± 1.3
	2021	30.2 ± 1.3	30.7 ± 0.9	29.9 ± 1.0	29.5 ± 1.4	30.1 ± 1.1
Double Seeds (%)	2019	6.4 ± 2.4	7.3 ± 3.5	10.4 ± 4.4	9.3 ± 2.5	8.3 ± 3.2
	2020	7.5 ± 1.3	8.1 ± 2.9	7.2 ± 2.4	6.9 ± 1.0	7.4 ± 1.9
	2021	6.9 ± 3.5	7.1 ± 2.4	6.9 ± 1.2	8.0 ± 3.5	7.2 ± 2.6

The data are averages ± sd of different treatments in each year and averages ± SD across all seasons. The absence of letters indicates no significant differences both among treatments and years.

3.4. The Nut and Kernel Morphological Traits

The morphological characteristics of the nuts, such as weight, length, width, and thickness reported in Table 6, showed no statistical differences between the biostimulant treatments and the control in each year, but they were higher in 2019 (on average 6.1 g, 42.7, 32.7, and 23.2 mm, respectively) than in both 2020 (on average 5.2 g, 34.9 mm, 27.1 mm, and 17.4 mm, respectively) and 2021 (5.5 g, 36.3 mm, 28.5 mm, and 18.2 mm, respectively).

Table 6. Morphological characteristics of almond nuts in different biostimulant treatments and control in three subsequent years (2019–2021).

Parameter	Year	Treatment				
		Control	Hendophit PS®	Ergostim XL®	Radicon®	Average
Nut dry weight (g nut^{-1})	2019	6.8 ± 0.8	5.7 ± 0.9	5.8 ± 0.9	6.1 ± 0.8	6.1 ± 0.8
	2020	5.1 ± 0.7	5.1 ± 1.0	5.4 ± 0.8	5.1 ± 0.9	5.2 ± 0.7
	2021	5.8 ± 0.6	5.4 ± 0.7	5.3 ± 0.8	5.6 ± 0.7	5.5 ± 0.7
Nut length (mm)	2019	41.6 ± 2.0	41.9 ± 2.0	44.3 ± 1.9	42.9 ± 2.1	42.7 ± 2.0 A
	2020	33.1 ± 2.3	35.5 ± 3.9	35.4 ± 2.8	35.6 ± 2.1	34.9 ± 2.8 B
	2021	34.0 ± 2.3	36.1 ± 2.7	37.2 ± 2.5	37.9 ± 2.3	36.3 ± 2.8 B
Nut width (mm)	2019	32.0 ± 1.4	32.0 ± 3.7	33.8 ± 1.8	32.9 ± 2.0	32.7 ± 2.2 A
	2020	27.1 ± 1.4	27.0 ± 2.2	27.6 ± 1.6	26.7 ± 2.3	27.1 ± 1.9 B
	2021	28.2 ± 2.3	28.9 ± 2.0	28.3 ± 2.2	28.5 ± 1.8	28.5 ± 2.1 B
Nut thickness (mm)	2019	22.5 ± 0.8	22.5 ± 5.4	24.1 ± 1.5	23.7 ± 0.9	23.2 ± 2.1 A
	2020	17.4 ± 0.5	17.3 ± 1.0	17.7 ± 0.9	17.1 ± 1.0	17.4 ± 0.8 B
	2021	18.0 ± 0.9	17.9 ± 0.8	18.4 ± 0.5	18.5 ± 0.8	18.2 ± 0.7 B

The data are averages ± sd of different treatments in each year and averages ± across all seasons. The absence of letters indicates no significant differences both among treatments and years. Different capital letters among year averages indicate significant differences (Tukey's test, $p < 0.05$). The absence of capital letters indicates no significant differences among years.

Likewise, the weight, length, width, and thickness of the kernels (Table 7) showed no statistical differences both among years and biostimulant treatments, with average values ranging from 1.5 to 1.7 g, from 24.3 to 26.6 mm, from 15.5 to 16.6 mm, and from 7.3 to 8.3 mm, respectively.

Table 7. Metric traits of almond kernel in different biostimulant treatments and control in three subsequent years (2019–2021).

Parameter	Year	Treatment				
		Control	Hendophit PS®	Ergostim XL®	Radicon®	Average
Kernel dry weight (g kernel^{-1})	2019	1.6 ± 0.3	1.6 ± 0.2	1.5 ± 0.2	1.6 ± 0.2	1.6 ± 0.2
	2020	1.7 ± 0.3	1.5 ± 0.2	1.6 ± 0.2	1.7 ± 0.2	1.7 ± 0.2
	2021	1.5 ± 0.2	1.6 ± 0.2	1.5 ± 0.2	1.6 ± 0.2	1.5 ± 0.2
Kernel length (mm)	2019	26.6 ± 2.4	25.4 ± 1.4	25.7 ± 1.3	26.0 ± 1.1	25.9 ± 1.5
	2020	26.3 ± 1.9	25.9 ± 1.0	26.0 ± 1.0	26.3 ± 1.6	26.1 ± 1.4
	2021	24.3 ± 2.3	24.3 ± 2.3	24.3 ± 2.3	26.3 ± 1.98	24.8 ± 2.2
Kernel width (mm)	2019	15.9 ± 1.5	15.8 ± 1.2	16.0 ± 1.0	16.0 ± 1.2	15.9 ± 1.2
	2020	16.6 ± 1.2	15.5 ± 1.4	16.4 ± 1.4	16.6 ± 1.1	16.3 ± 1.3
	2021	15.9 ± 1.1	16.1 ± 1.1	15.7 ± 1.2	16.3 ± 1.3	16.0 ± 1.2
Kernel thickness (mm)	2019	7.3 ± 1.5	7.9 ± 0.5	7.3 ± 0.7	7.6 ± 0.4	7.5 ± 0.8
	2020	8.1 ± 0.6	8.0 ± 0.7	8.1 ± 0.6	7.9 ± 0.8	8.0 ± 0.7
	2021	8.3 ± 0.6	8.0 ± 0.5	7.9 ± 0.7	7.7 ± 0.6	8.0 ± 0.6

The data are averages ± sd of different treatments in each year and averages ± sd across all seasons. The absence of letters indicates no significant differences both among treatments and years.

4. Discussion

This study aimed to evaluate the influence of biostimulant treatments on the vegetative growth and reproductive behavior of young almond trees. The products were applied three times during each growing season—at the swollen bud, beginning of flowering, and fruit set–beginning of fruit growth stages. The impact of the tested biostimulants on the vegetative system primarily focused on the growth of trunk diameter and shoots. Specifically, long shoot growth during the early years of orchard establishment is the main component of vegetative development in almonds [41]. Our results, indicating a slight positive effect of biostimulants on the increase in trunk diameter and shoot length, align with previous studies [25,30]. This increase in vegetative shoot growth can result in more buds that will support future production. Growers should expect the mainstay of vegetative growth to be the production of long vegetative shoots. Regarding the number of buds per unit of branch length, mostly detected before or during the application of the biostimulant products, no statistical differences were found among all treatments. Overall, the average total bud density in each year (ranging from 0.89 to 1.08 buds cm^{-1}) was close to the range (0.46 to 1.02 buds cm^{-1}) reported in other research [42]. However, in the last year of this study (2021), our data tended to be low, likely due to the impact of the spring frost the previous year (2020), which negatively affected tree performance and also the formation of buds, which occurred during the prior season [41]. This dynamic of both the vegetative growth of the shoots and of all the buds (vegetative and floral) are key components for the development of an economically sustainable and productive orchard. Even the density of the flowers (varied between 0.40 and 0.65 flowers cm^{-1}) did not highlight significant differences either between years or between biostimulant treatments and fell within the wide range (from 0.03 to 1.52) detected in different almond genotypes in previous research [43]. The percentage of fruit set in 2019 was significantly higher in the biostimulant treatments than in the control. This phenological stage is delicate for the tree, and the application of external energy sources plays a vital role in ensuring the quality of pollen and nectar in the flowers [44]. Among the three types of organo-mineral fertilizers used, the best result was observed for Radicon®, which contains humic and fulvic acids, and similar positive effects have been detected in another study [45]. Furthermore, overall, our range of relative fruit set values (varying from 22.5% to 34.4%) is in agreement with previous reports on several almond cultivars (ranging between 15% and 40%) [5,22,46]. In both 2020 and 2021, significantly lower fruit set percentages than in the previous season were observed, with no significant differences among biostimulant treatments and the control. The decreases in fruit set percentage in these last years were undoubtedly due to the frosts that occurred during and after the flowering period (almond phenological states from B, "Swollen bud", to I, "Young fruit, Jacket stage", of the Felipe classification), as previously reported in paragraph 2.1. These results align with those of previous studies [23,47], which demonstrated that almond flowers and young fruits are extremely sensitive to frost, suffering damage at temperatures below 0 °C (−1 or −2 °C), depending on the exposure time. In these phenological states, a couple of hours at these temperatures can cause serious damage and even ruin the year's production [47]. To this regard, the foliar application of biostimulants did not produce any effect on crops subject to frost, due to the formation of tiny ice crystals outside and inside the plant cell, which are lethal for them. In general, the ability of crops to defend themselves from frost is determined by the cultivar's ability to escape freezing temperatures over time. However, a possible positive action of biostimulants to alleviate non-excessive thermal stress from cold in plants is to improve the absorption of nutrients, increasing their concentration within the plant tissues, making them more resistant to low temperatures [48].

In consonance with the fruit set incidence, in the 2019 season, the yield, in terms of the number and weight of fruit per tree, was significantly higher in the biostimulant treatments than in the control. On average, an increase in the number of fruits per tree and fruit yield per tree achieved with the application of biostimulants relative to the control was 24.7% and 19.7%, respectively. These results are in accordance with some previous research [49]. The smaller increase in weight per tree compared to the number of fruits in this season could be due to the slightly higher weight values of the fruits recorded in the control (Table 6). The fruit incidence characters, such as the hull, shelling, and double seeds percentage, showed no statistical differences both among the years and biostimulant treatments. Our hull percentage data (on average 44.9%) are consistent with data previously reported in the literature [50], as is the percentage of shelling (on average 30.1%), which fells within the 30–40% range reported in previous research [5,7,24,51–55]. On the contrary, our data regarding the percentages of double seeds (on average 7.6%) are lower than those obtained for the same Tuono cultivar (between 15% and 31%) by other authors [7,53,56,57]. Indeed, as for the incidence of each single part of the fruit, they are primarily determined by genotype but also by environmental factors [49,58–60]. Therefore, in this regard, our data showed distinctive and commercially interesting agronomic characteristics.

Based on the use of almond components, the following information is known: Almond hull is a by-product that can be used as supplemental livestock feed or, due to its beneficial properties (mainly caused by polyphenols and unsaturated fatty acids), in the food, cosmetic, and pharmaceutical industries [61]. The shelling percentage parameter is used to obtain a quantitative measure of shell density and is utilized commercially to calculate kernel yield [56]. Finally, a high presence of double-seeded nuts significantly reduces their commercial value, as having a flat or concave face is undesirable both for the industry (since they present difficulties for confectionery use) and for consumers (because they are less attractive than single-seeded nuts) [62]. Regarding the morphological traits of nuts, such as weight, length, width, and thickness, there were no statistical differences between biostimulant treatments and the control (on average 5.6 g, 38.0 mm, 29.4 mm, and 19.6 mm, respectively), but significantly higher values were observed in 2019 (on average 6.1 g, 42.7 mm, 32.7 mm, and 23.2 mm) compared to both 2020 and 2021 (on average of the two years 5.3 g, 35.6 mm, 27.8 mm, and 17.8 mm, respectively). Regarding the weight, length, width, and thickness of the kernels, there were no statistical differences both among years and biostimulant treatments, with average values of 1.5 g, 25.6 mm, 16.1 mm, and 7.8 mm, respectively. Overall, our data on the characteristics of the nuts and kernel morphological traits were somewhat superior to those of the same Tuono cultivar reported in other research [7,53,56,63,64], in which for the nuts they varied between 3–4 g, 28–34 mm, 21–23 mm, and 15–20 mm, respectively, and for the kernel they varied between 1.2 and 1.4 g, 23.4 and 23.9 mm, 12.2 and 14.9 mm, and 6.3 and 7.2 mm, respectively. Socias i Compañy et al. [65] commented that the general trend in the industry is the preference for large kernels in order to facilitate and cheapen the processes of cracking and blanching. Nonetheless, for some special confectioneries, very small sizes are chosen, as well as those with definite shapes. For sugared almonds (peladillas or dragées) and for chocolate almonds, large kernels are selected, preferably round to reduce the layer of sugar or chocolate covering the kernel

5. Conclusions

In the three years of experimentation (2019, 2020, and 2021), only in the first year, characterized by a normal climate trend, did the biostimulant treatments show a slight positive effect on the growth of the trees and on the percentage of total fruit set. Furthermore, a significantly higher fruit load and weight per plant were observed. Therefore, the use of biostimulants proved to be crucial during the flowering of almond trees. On the contrary, the second and third experimental years were affected by late frosts, causing damage to the flowers and small fruits. This resulted in reduced growth of the trees, a lower percentage of fruit set, and diminished yield. Furthermore, during these years, no significant effect of the biostimulant treatments on tree crops was observed. Additionally, the study's findings highlight that the frequent occurrence of late frosts, likely influenced by climate change, poses a greater risk to almond production than anticipated. Therefore, further research on the use of extra and ultra-late cultivars is needed to address this challenge. Furthermore, characteristics such as fruit, nut, and kernel quality were not significantly affected by the foliar application of biostimulants, probably because they could have reached their maximum quality potential in this growing environment. However, considering the positive results in terms of yield mentioned above, the foliar application the biostimulants Hendophyt®, Ergostim®, and Radicon® could be recommended to enhance the performance of almond tree cv. Tuono under normal climatic conditions in arid and semi-arid areas, similar to those covered by this study, such as Southern Italy. Finally, further research is needed on different almond cultivars and application methods, as well the specific mechanisms of action of the biostimulant treatments.

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