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ROOT-ZONE BIOPOLYMER APPLICATION IMPROVES DROUGHT RESILIENCE, WATER-USE EFFICIENCY AND RECOVERY OF *BOUGAINVILLEA SPECTABILIS*

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Abstract

Water shortage is becoming a challenge for ornamental plants, especially in arid and semi-arid regions where a significant amount of irrigation is required to maintain attractive landscapes. *Bougainvillea spectabilis* is well adapted to warm climates and widely used in landscapes where water is limited, but prolonged drought can limit vegetative growth, photosynthesis, flowering and overall ornamental quality. The present study investigated a root-zone strategy to improve drought tolerance by combining the water-holding capacity of a biodegradable hydrogel with the biological activity of alginate oligosaccharide (AOS) and chitosan. Plants were subjected to three water regimes corresponding to 100%, 70% and 40% field capacity and were subjected to one of four treatments: untreated control, alginate hydrogel, AOS or AOS incorporated into a chitosan-based hydrogel. Growth, flowering, photosynthetic pigments, gas exchange, leaf water relations, osmotic adjustment, oxidative stress, antioxidant enzymes, root architecture, and water use efficiency were measured. A seven-day period of rewatering was then employed to assess the recovery of plants from severe drought. Water deficiency decreased plant height, branching, leaf area, biomass, chlorophyll concentration, photosynthetic rate, stomatal conductance and relative water content in a progressive manner. It also enhanced hydrogen peroxide, malondialdehyde (MDA) and electrolyte leakage. The combined AOS + chitosan treatment was the best protection, especially under severe drought conditions. Leaf water content, photosynthetic activity and root development of the treated plants at 40% field capacity were significantly higher than the untreated plants. The treatment also increased proline, soluble sugars and glycine betaine accumulation and SOD, CAT and APX activities, decreased oxidative membrane damage. After rewatering, AOS +chitosan–alginate hydrogel-treated plants recovered faster, had higher relative water content and photosynthetic activity, and lower MDA and electrolyte leakage than untreated drought-stressed plants. In general, these results indicate that the use of a water-retaining root-zone hydrogel in conjunction with AOS and chitosan can confer physical and physiological protection against drought. This strategy can be useful to improve the performance and water use efficiency of Bougainvillea under intermittent irrigation in dry environments.

Key words: *Bougainvillea spectabilis*; Drought stress; Deficit irrigation; Alginate oligosaccharide; Chitosan; Hydrogel; Water relations; Osmotic adjustment; Antioxidant defense; Drought recovery

Introduction

Maintaining high-quality ornamental plants underwater-limited conditions is a major challenge, particularly in arid and semi-arid regions where high temperatures, low precipitation, and increasing irrigation demand can substantially constrain landscape production. In ornamental crops, drought stress affects not only biomass production but also plant architecture, leaf development, photosynthetic activity, water relations, flowering, and overall ornamental quality. In Mediterranean ornamentals it was shown that drought resistant plants manage the root development, stomatal control, osmotic adjustment and antioxidant defense in a concerted manner to cope with water shortage during growth (Toscano *et al.,* 2019; Giordano *et al.,* 2021).

Bougainvillea is a popular landscaping plant because it grows rapidly, has colorful bracts, blooms for a long period, and can survive in a hot, dry climate. But this does not mean that even if it has a fame of ornamental that can survive dryness there is no impact of water stress on the performance of the plant. In trials with deficit irrigation, reduced water availability may lead to loss of biomass production, leaf number and leaf area, although the degree of response depends on the genotype (Giordano *et al.,* 2021). It has also been shown that the ability of Bougainvillea cultivars to maintain consistent physiological activity under stress circumstances varies widely. Recent studies have shown large variety in cultivars' sensitivity to salt and other environmental circumstances, which further emphasize the amazing physiological flexibility of Bougainvillea (Zhang *et al.,* 2024a). An alternative for improving drought management is to use the inherent drought tolerance of the plant coupled with treatments to strengthen its physiological capacity to cope with lack of water. These improvements provide a chance to improve the management of drought.

Less water available for plants causes drought and closure of their stomata. This decreases transpiration, and hence the amount of carbon dioxide taken in. Chronic stress may cause overproduction of reactive oxygen species (ROS) and decrease of photosynthesis. ROS may damage membranes, proteins and the photosynthetic equipment. Relative water content and chlorophyll were reduced, whereas proline, MDA and antioxidant enzyme activity were raised in *Bougainvillea glabra* during water stress (Wang *et al.,* 2026). Interestingly, these plants may produce a set of defensive mechanisms and, thus, drought tolerance in Bougainvillea is controlled by a cross-talk between water

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saving, osmotic adjustment and ROS detoxification. Thus, the improvement of such activities may be regarded a feasible method for maintaining the quality of attractive plants in situations of restricted watering.

Natural polysaccharides are gaining growing interest as possible biostimulants to improve plant performance under abiotic stress. As shown by Zhang *et al.,* (2020) and Zhang *et al.,* (2024b), alginate oligosaccharides may function as signalling molecules and were correlated with enhanced antioxidant activity, osmotic control, and stress-responsive metabolism. Moreover, chitosan and chitooligosaccharides have attracted great interest for triggering plant defense responses and regulating physiological processes related to drought tolerance (Liu *et al.,* 2022; García-García *et al.,* 2023; El-Araby *et al.,* 2024). They suggested that these biostimulant may be applied to the root zone while increasing the quantity of water accessible in the vicinity of the plant roots. This may allow a reaction over a longer time period particularly in perennial landscaping plants that have been water-stressed more than one occasion. However, these procedures may be combined with the use of hydrogel. They may absorb and hold water and hence may increase to the quantity of moisture in the root zone. Furthermore, co-application of AOS and chitosan may have other biological stimulation effects. For instance, alginate-based hydrogels were shown to enhance water availability and root development under drought stress (Idrissi *et al.,* 2024), while chitosan–alginate systems have showed potential for water retention and impacts on plant growth underwater-limited conditions (Gomaa & Aldaby, 2023; Nordin *et al.,* 2024). While drought responses of Bougainvillea have been studied (Toscano *et al.,* 2019; Giordano *et al.,* 2021; Wang *et al.,* 2026), limited studies have examined the use of root-zone biopolymer treatments to enhance drought tolerance and recovery in this species (Liu *et al.,* 2024). The present work was thus aimed to investigate whether an AOS + chitosan–alginate hydrogelmay help *B. spectabilis* to maintain its growth, hydration status, photosynthesis and antioxidant protection under drought circumstances and also to recover more rapidly after rewatering. The combination treatment was expected to be more efficient than either alginate hydrogel or alginate oligosaccharide (AOS) alone due to the improved water availability in the root zone and the simultaneous stimulation of the plant's defense mechanisms.

Materials and Methods

Plant material and growing conditions: Uniform, healthy *Bougainvillea spectabilis* L. plants of similar age, height, and developmental stage were selected for the experiment to minimize variation among experimental units during September 2024. Plants were selected at around 10–12 months of age to minimize disparities in initial height, stem diameter and number of leaves. Each plant was individually placed in plastic pots, 8 L each, filled with a 2:1:1 (Peat/sand/perlite) combination of peat moss, washed sand and perlite. The plants were grown in a greenhouse with natural illumination at King Saud University. Relative humidity was maintained at 55-70%. The typical air temperature ranged between 25 to 30°C. The plants had 4 weeks of adjustment before the start of the treatment. In addition to having a full fertilizer solution throughout this period each plant was regularly watered to maintain the substrate at its greatest capacity. Bougainvillea has been found to respond to irrigation reductions with changes in its growth, leaf development, stomatal regulation and water-use efficiency (Toscano *et al.,* 2019; Giordano *et al.,* 2021). Based on these results, experimental conditions and deficit irrigation treatments were selected for this investigation.

Biopolymer treatments: Four root-zone treatments were evaluated: Untreated control, alginate hydrogel, alginate oligosaccharide (AOS) and AOS incorporated into a chitosan–alginate hydrogel. AOS was utilized as 200 mg L^{-1}, and the hydrogel was prepared with 0.2% (w/v) chitosan. The concentrations were chosen to get moderate elicitation effect without exceptionally high polymer concentrations (Zhang *et al.,* 2020; Zhang *et al.,* 2024b), and were within the range usual in research on polysaccharide-mediated stress protection. The alginate-based hydrogel was prepared by incorporating the specified concentration of sodium alginate into the hydrogel formulation, followed by cross-linking with calcium chloride. Chitosan was incorporated into the formulation used for the combined AOS + chitosan treatment. The prepared hydrogel was thoroughly mixed to ensure homogeneous distribution of the components before application to the root zone. Calcium chloride was employed as cross linking agent for creating stable hydrogel matrix. The hydrogel was rinsed with distilled water following the cross-linking and adjusted the water absorption capacity. The combined application was the incorporation of AOS into the hydrogel prior to application. Approximately 100 g of freshly prepared hydrogel was applied to each pot. Hydrogel preparations were standardized before application to ensure comparable amounts and physical conditions among experimental units. The AOS treated plants were given the same amount of AOS solution without hydrogel. Control plants received the same volume of distilled water without hydrogel or $CaCl_2$. Hydrogel composition: Sodium alginate: 2% (w/v), Sigma-Aldrich, St. Louis, MO, USA; $CaCl_2$: 0.5 M, contact time 30 min; AOS: 200 mg L^{-1}, degree of polymerization 2–8, Sigma-Aldrich; Chitosan: 0.2% (w/v), molecular weight 100–300 kDa, degree of deacetylation ≥85%, dissolved in 1% acetic acid; pH of chitosan solution adjusted to 5.5 before mixing. The formulation was based on the premise that alginate and chitosan can create biodegradable hydrogel networks that can trap and release water gradually and may function as transporters for physiologically active compounds (Gomaa & Aldaby, 2023; Nordin *et al.,* 2024).

Water-stress treatments: Three irrigation levels were established:100% field capacity (FC): well-watered condition, 70% FC: moderate water deficit, 40% FC: severe water deficit. Field capacity was determined gravimetrically. Representative pots were saturated with water, allowed to drain freely for 24 h and then weighed. The resulting substrate weight was used as the reference for 100% FC. During the stress period, each pot was weighed daily and the required amount of water was added to maintain the assigned moisture level. Field capacity targets were set per treatment by accounting for the additional water-holding capacity of the hydrogel. For hydrogel-treated pots, the hydrogel was added first, and then the substrate was saturated and allowed to drain to determine the 100% FC reference for that

treatment. The experiment followed a 4 × 3 factorial design consisting of four biopolymer treatments and three water regimes. Six plants were used for each treatment combination, resulting in 72 experimental plants.

Growth and flowering measurements: Plant height was measured from the surface of the substrate to the top of the plant. At the conclusion of stress period, number of main branches was recorded. Total leaf area was determined using digital image analysis of leaves after harvest (ImageJ 1.53 (NIH, Bethesda, MD, USA). Shoots and roots were weighed immediately after harvest to assess fresh biomass and then dried at 70 °C to constant weight. Shoot biomass, root biomass and root/shoot ratio were calculated using dry weights. For Root measurements the equipment WinRHIZO Pro 2021 (Regent Instruments, Quebec, Canada) was used. Flowering performance was evaluated by recording days to first flowering, number of flowers/plant, flower diameter and bract diameter. The diameter of the bract cluster (the showy colored structure), measured at its widest point. Flower number is the number of bract clusters per plant. Days to first flowering was recorded from the date of treatment initiation. These were included because the drought tolerance of ornamental plants should be assessed in terms of both physiological performance and preservation of marketable decorative traits (Toscano *et al.,* 2019).

Photosynthetic pigments: Fully expanded leaves were collected from the middle portion of the shoots. Approximately 0.1 g fresh tissue was homogenized and extracted in 80% acetone. Absorbance was measured spectrophotometrically, and chlorophyll a, chlorophyll b, total chlorophyll and carotenoids were calculated using standard spectrophotometric equations at wavelengths 663 nm, 645 nm, 470 nm (Lichtenthaler 1987; Moussa *et al.,* 2026).

Gas exchange: Net photosynthetic rate (Pn), stomatal conductance (gs) and transpiration were measured on fully expanded leaves using a portable infrared gas analyzer. Measurements were conducted during the morning under stable environmental conditions. The equipment used was LI-6400XT (LI-COR, Lincoln, NE, USA); PAR 1000 µmol m^{-2} s^{-1}, CO_2 400 µmol mol^{-1}, leaf temperature 25°C, measurements between 09:00–11:00.

Intrinsic water-use efficiency was calculated as:

$$iWUE = Pn / gs$$

where Pn represents net photosynthesis and gs represents stomatal conductance. WUE (iWUE = Pn/gs)

Gas exchange measurements were particularly important because stomatal regulation is one of the major physiological responses of *Bougainvillea* to limited water availability (Giordano *et al.,* 2021).

Water relations: Relative water content was calculated using fresh, turgid and dry weights:

$$RWC\ (\%) = [(FW - DW)/(TW - DW)] \times 100$$

where FW is fresh weight, TW is fully hydrated weight and DW is dry weight.

Leaf water potential was determined using a pressure chamber (PMS Model 1000 (PMS Instrument Co., Albany, OR, USA). Relative water loss was calculated by recording the decrease in leaf mass during a controlled dehydration period (25°C, 50% RH, 0–6 h) and expressed as the percentage of initial fresh weight lost per hour.

Osmotic adjustment: Free proline was quantified using the ninhydrin-based colorimetric method and expressed as µmol g^{-1} FW (Bates *et al.,* 1973; Al-Yafrsi *et al.,* 2026). Soluble sugars were determined colorimetrically and expressed as mg g^{-1} FW. The concentration of glycine betaine was determined by a periodide method and expressed as µmol g^{-1} FW (Grieve and Grattan, 1983). The compounds were selected because osmotic adjustment is a key component of plant response to water restriction and has been seen in drought-stressed Bougainvillea earlier (Wang *et al.,* 2026).

Oxidative stress: Hydrogen peroxide was determined spectrophotometrically from fresh leaf extracts (Moussa *et al.,* 2026; Al-Yafrsi *et al.,* 2026). Lipid peroxidation was estimated from malondialdehyde (MDA) concentration using the thiobarbituric-acid-reactive substances method. Membrane damage was estimated through electrolyte leakage (Heath & Packer, 1968). Leaf discs were incubated in distilled water and their initial electrical conductivity was recorded. The samples were then autoclaved to release the total cellular electrolytes and final conductivity was measured.

Electrolyte leakage was calculated as:

$$EL\ (\%) = EC_1 / EC_2 \times 100$$

where EC_1 is the conductivity before autoclaving and EC_2 is the conductivity after autoclaving.

Antioxidant enzymes: Fresh leaves were homogenized in chilled extraction buffer and centrifuged under refrigerated conditions. The supernatant was used for enzyme assays. Activities of superoxide dismutase (SOD), catalase (CAT), and ascorbate peroxidase (APX) were determined spectrophotometrically and expressed relative to soluble protein content (Moussa *et al.,* 2026; Al-Yafrsi *et al.,* 2026). These enzymes were selected because the antioxidant system was closely associated with drought adaptation in *Bougainvillea* and related ornamental shrubs (Wang *et al.,* 2026).

Root architecture: At the end of the experiment, roots were gently removed from the substrate and washed under running water. Root samples were scanned using a digital root-analysis system. The following characteristics were determined: total root length, root surface area, root volume, and number of root tips. Root length was measured on a subsample of roots (approximately 10% of total root fresh weight) and scaled to the whole root system based on fresh weight ratios. The relatively low specific root length may reflect the coarse root system of container-grown Bougainvillea and the inclusion of thick structural roots in the subsample. Root measurements were

particularly important for determining whether the hydrogel treatment affected drought tolerance through changes in the root system.

Water-use efficiency: Whole-plant water-use efficiency (WUE, g L^{-1}) was calculated as biomass production relative to the amount of water supplied, whereas intrinsic water-use efficiency was considered in relation to carbon assimilation and stomatal conductance. Whole-plant water-use efficiency was estimated from dry biomass relative to the total amount of water supplied during the experimental period:

$$\text{WUE} = \frac{\text{Total dry biomass}}{\text{Cumulative water supplied}}$$

Instantaneous intrinsic WUE was also calculated from gas-exchange measurements.

Rewatering and recovery: At the end of the severe drought treatment (lasted 30 days from 1 March 2025 to 30 March 2025). The recovery period was 7 days (1–7 April 2025)., plants exposed to 40% FC were rewatered to 100% FC. Plants exposed to 40% FC were rewatered to 100% FC and measured after 7 days. Physiological recovery was assessed seven days later. Relative water content, photosynthetic rate, stomatal conductance, MDA and electrolyte leakage were measured again. Recovery index (%) was calculated as the percentage of the corresponding pre-drought value recovered after rewatering. For positive physiological parameters, the index was calculated as (value after rewatering / corresponding value before drought) × 100. For damage-related parameters, including MDA and electrolyte leakage, the index was calculated inversely so that lower residual damage corresponded to a higher recovery index. Recovery index calculation is the mean of the percentage recovery of RWC, Pn, gs, MDA and electrolyte leakage, with damage parameters inverted. Plants for biomass and root measurements were harvested at the end of the drought period (before rewatering). A separate set of plants (n = 6 per treatment combination) was used for the recovery experiment. This part of the experiment was intended to determine whether the biopolymer treatment simply reduced drought injury or also improved the ability of Bougainvillea to recover after water became available again.

Statistical analysis

Data were analyzed using two-way analysis of variance (ANOVA), with water regime and biopolymer treatment considered as fixed factors, including their interaction. When significant effects were detected, means were separated using Tukey's test at $p \leq 0.05$. Six biological replicates were used for each treatment combination. Results were expressed as mean ± standard deviation. Pearson correlation analysis was performed to examine relationships among relative water content, photosynthesis, MDA, antioxidant enzyme activity, root development and water-use efficiency.

Results

Growth and flowering: Water deficiency clearly reduced the development of *B. spectabilis* (Table 1). The impact was especially strong at 40% field capacity. Plant height (cm) in untreated plants was decreased from 74.7 cm in well-watered circumstances to 51.7 cm in severe drought. Also the shoot dry weight was decreased from 42.7 to23.1 g. Treatments with biopolymers resulted in lower losses. The strongest response was consistently obtained for the AOS + chitosan–alginate hydrogelhydrogel in combination. Plants treated at 40% FC were 69.4cm tall and generated 35.6 g shoot dry weight compared with 51.7cm and 23.1 g in untreated plants. The leaf area showed the similar pattern. Severe dryness decreased leaf area to 1018 cm^2 $plant^{-1}$ in untreated plants while AOS + chitosan–alginate hydrogeltreated plants retained about 1507 cm^2 $plant^{-1}$. In addition, the dry weight of the roots was greater in the treated plants, ranging from 6.7 g in the untreated plants to 10.4 g with AOS + chitosan–alginate hydrogel. Water scarcity slowed flowering. Untreated plants at 40% FC took about 95 days to blossom and produced only 17.4 flower $plant^{-1}$. AOS + chitosan–alginate hydrogelreduced the flowering time to around 80 days and raised flower number to 30.7 flower per plant. The treated plants also had higher retention of flower and bract size.

Photosynthetic pigments and gas exchange: Decrease of photosynthetic pigments was associated with the rise of water stress (Table 2). Total chlorophyll was decreased from 2.54 mg g^{-1} FW in well-watered control plants to 1.51 mg g^{-1} FW in untreated plants subjected to 40% FC. Under severe drought stress, the decline was much less in AOS + chitosan–alginate hydrogel treated plants, which preserved around 2.21 mg g^{-1} FW of total chlorophyll. A large reduction in net photosynthesis was the main cause of water constraint. Untreated plants showed a drop from 15.8 to 6.2 umol CO_2 m^{-2} s^{-1} from 100 to 40% FC. AOS + chitosan–alginate hydrogel maintained Pn at 11.8 µmol CO_2 m^{-2} s^{-1} under the same drought situation. Stomatal conductance was lowered similarly in dryness but yet remained much higher in the treated plants. At 40% FC, gs was 0.071 mol H_2O m^{-2} s^{-1} in the untreated plants and 0.132 mol H_2O m^{-2} s^{-1} in the combined treatment.

Water relations and osmotic adjustment: The shortage of water caused a great degradation of the water status of the plants (Table 3). RWC declined from 88.6% in well-watered control plants to 61.2% in untreated plants at 40% FC. Biopolymer applications partially alleviated this decline with the greatest effect shown for AOS + chitosan–alginate hydrogel. These plants maintained their RWC of 78.9% under severe drought stresses.

The leaf water potential was also more negative with decreased water supply. The water potential of untreated plants at 40% FC was −1.47 MPa while that of AOS + chitosan–alginate hydrogel treated plants was less negative at −1.03 MPa. Proline, soluble sugars and glycine betaine increased with drought stress and the increases were more in biopolymer treated plants. For example, the proline level at 40% FC in AOS + chitosan–alginate hydrogel treated plants was 29.8 µmol g^{-1} FW compared to 18.9 µmol g^{-1} FW in untreated plants.

Table 1. Effects of water regime and biopolymer treatments on growth and flowering of *Bougainvillea spectabilis*.

Water regime	Treatment	Plant height (cm)	Branch number	Leaf area (cm² plant⁻¹)	Shoot DW (g)	Root DW (g)	Root/shoot ratio	Flowering variables			
								Days to flowering	Flower number plant⁻¹	Flower diameter (cm)	Bract diameter (cm)
100% FC	Control	74.7 ± 2.8 ab	12.5 ± 0.6 c	1824 ± 74 ac	42.7 ± 2.2 d	10.9 ± 0.6d	0.252 e	74.2 ± 2.1 d*	38.5 ± 2.4 d	5.82 ± 0.21 b	7.42 ± 0.27 b
100% FC	Alginate hydrogel	77.3 ± 2.4 b	13.3 ± 0.5 b	1936 ± 81 b	45.1 ± 2.3 c	12.0 ± 0.6 c	0.266 d	72.8 ± 1.9 d	41.7 ± 2.1 c	6.01 ± 0.19 ab	7.68 ± 0.24 ab
100% FC	AOS	79.6 ± 2.7 a	14.1 ± 0.9 b	2015 ± 69 a	47.0 ± 2.0 b	12.6 ± 0.8 b	0.268 d	71.4 ± 2.0 e	44.3 ± 2.5 b	6.14 ± 0.22 a	7.83 ± 0.28 a
100% FC	AOS + chitosan–alginate hydrogel	82.1 ± 3.0 a	14.8 ± 0.8 a	2094 ± 76 a	49.2 ± 2.4 a	13.5 ± 0.7 a	0.274 c	69.8 ± 1.8 e	47.6 ± 2.6 a	6.31 ± 0.20 a	8.05 ± 0.25 a
70% FC	Control	66.3 ± 2.7 c	10.8 ± 0.7 b	1498 ± 61 e	34.6 ± 1.8f	9.4 ± 0.5 g	0.272 c	81.5 ± 2.4 c	29.8 ± 1.9 f	5.12 ± 0.18 d	6.61 ± 0.23 d
70% FC	Alginate hydrogel	71.7 ± 2.6 b	12.0 ± 0.6 d	1667 ± 64 d	38.9 ± 1.7 e	10.9 ± 0.6 b	0.280 b	78.3 ± 2.2 c	34.2 ± 2.0 e	5.46 ± 0.20 c	6.98 ± 0.26 c
70% FC	AOS	74.8 ± 2.9 b	12.7 ± 0.8 c	1769 ± 70 c	41.2 ± 2.0 d	11.7 ± 0.7 b	0.284 b	76.4 ± 2.1 d	36.9 ± 2.2 d	5.63 ± 0.19 c	7.21 ± 0.25 c
70% FC	AOS + chitosan–alginate hydrogel	78.3 ± 2.5 ab	13.6 ± 0.7 b	1876 ± 73 b	44.7 ± 2.1 c	12.8 ± 0.6 b	0.286 b	73.1 ± 1.9 d	40.8 ± 2.4 c	5.92 ± 0.21 b	7.57 ± 0.27 b
40% FC	Control	51.7 ± 2.3 f	7.2 ± 0.5 g	1018 ± 48 h	23.1 ± 1.4 i	6.7 ± 0.5 i	0.290 a	94.7 ± 2.8 a	17.4 ± 1.4 i	4.21 ± 0.16 g	5.38 ± 0.21 f
40% FC	Alginate hydrogel	59.8 ± 2.4 e	8.9 ± 0.6 f	1226 ± 52 g	27.8 ± 1.5 h	8.1 ± 0.h	0.291 a	89.1 ± 2.5 b	21.8 ± 1.5 h	4.57 ± 0.17 f	5.81 ± 0.22 e
40% FC	AOS	63.7 ± 2.5 d	9.8 ± 0.7 e	1338 ± 57 f	30.9 ± 1.6 g	8.9 ± 0.6 g	0.288 b	85.6 ± 2.3 b	25.1 ± 1.7 g	4.83 ± 0.18 e	6.12 ± 0.23 e
40% FC	AOS + chitosan–alginate hydrogel	69.4 ± 2.7 c	11.4 ± 0.7 d	1507 ± 63 e	35.6 ± 1.8 f	10.4 ± 0.6 f	0.292 a	80.2 ± 2.2 c	30.7 ± 1.9 f	5.24 ± 0.19 d	6.67 ± 0.24 d

Table 2. Photosynthetic pigments and gas exchange.

Water regime	Treatment	Chl a (mg g⁻¹ FW)	Chl b (mg g⁻¹ FW)	Total Chl (mg g⁻¹ FW)	Carotenoids (mg g⁻¹ FW)	Pn (µmol CO_2 m⁻² s⁻¹)	gs (mol H_2O m⁻² s⁻¹)	E	iWUE
100% FC	Control	1.82 ± 0.08 c*	0.72 ± 0.04 c	2.54 ± 0.10 b	0.42 ± 0.02 d	15.8 ± 0.7 c	0.214 ± 0.012 d	(mmol H_2O m⁻² s⁻¹)	(µmol mol⁻¹)
100% FC	Alginate hydrogel	1.91 ± 0.07 b	0.76 ± 0.04 b	2.67 ± 0.09 ab	0.44 ± 0.02 a	16.7 ± 0.6 b	0.226 ± 0.011 c	2.84 ± 0.14 b	73.8 ± 3.2 a
100% FC	AOS	1.97 ± 0.08 a	0.79 ± 0.03 ab	2.76 ± 0.10 a	0.46 ± 0.02 b	17.4 ± 0.7 ab	0.234 ± 0.012 b	2.98 ± 0.13 b	73.9 ± 3.1 a
100% FC	AOS + chitosan–alginate hydrogel	2.05 ± 0.07 a	0.83 ± 0.04 a	2.88 ± 0.09 a	0.48 ± 0.02 a	18.2 ± 0.8 a	0.243 ± 0.013 a	3.08 ± 0.15 a	74.4 ± 3.3 a
70% FC	Control	1.51 ± 0.06 e	0.60 ± 0.03 e	2.11 ± 0.08 e	0.39 ± 0.02 e	11.7 ± 0.6 b	0.151 ± 0.009 h	3.19 ± 0.16 a	74.9 ± 3.4 a
70% FC	Alginate hydrogel	1.66 ± 0.06 d	0.66 ± 0.03 d	2.32 ± 0.08 d	0.42 ± 0.02 d	13.5 ± 0.6 e	0.170 ± 0.010 g	1.89 ± 0.10 f	77.5 ± 3.5 b
70% FC	AOS	1.73 ± 0.07 c	0.69 ± 0.03 c	2.42 ± 0.09 c	0.44 ± 0.02 c	14.4 ± 0.7 d	0.181 ± 0.010 f	2.12 ± 0.11 e	79.4 ± 3.6 b
70% FC	AOS + chitosan–alginate hydrogel	1.87 ± 0.06 b	0.75 ± 0.03 b	2.62 ± 0.08 b	0.47 ± 0.02 a	15.8 ± 0.6 c	0.198 ± 0.011 e	2.25 ± 0.12 d	79.6 ± 3.7 b
40% FC	Control	1.08 ± 0.05 h	0.43 ± 0.03 h	1.51 ± 0.06 g	0.32 ± 0.02 g	6.2 ± 0.4 i	0.071 ± 0.006 l	2.46 ± 0.13 c	79.8 ± 3.8 b
40% FC	Alginate hydrogel	1.28 ± 0.05 g	0.51 ± 0.03 g	1.79 ± 0.07 f	0.36 ± 0.02 f	8.3 ± 0.5 h	0.091 ± 0.007 k	0.84 ± 0.05 i	87.3 ± 4.1 c
40% FC	AOS	1.38 ± 0.06 f	0.55 ± 0.03 f	1.93 ± 0.08 e	0.38 ± 0.02 e	9.4 ± 0.5 g	0.104 ± 0.008 j	1.08 ± 0.06 h	91.2 ± 4.3 c
40% FC	AOS + chitosan–alginate hydrogel	1.58 ± 0.06 d	0.63 ± 0.03 e	2.21 ± 0.08 d	0.43 ± 0.02 c	11.8 ± 0.6 f	0.132 ± 0.009 i	1.23 ± 0.07 g	90.4 ± 4.2 c

*Values are mean ± SD (n = 6). Different lowercase letters within columns indicate significant differences according to Tukey's test at $p \leq 0.05$. E = transpiration; iWUE = intrinsic water-use efficiency (Pn/gs)

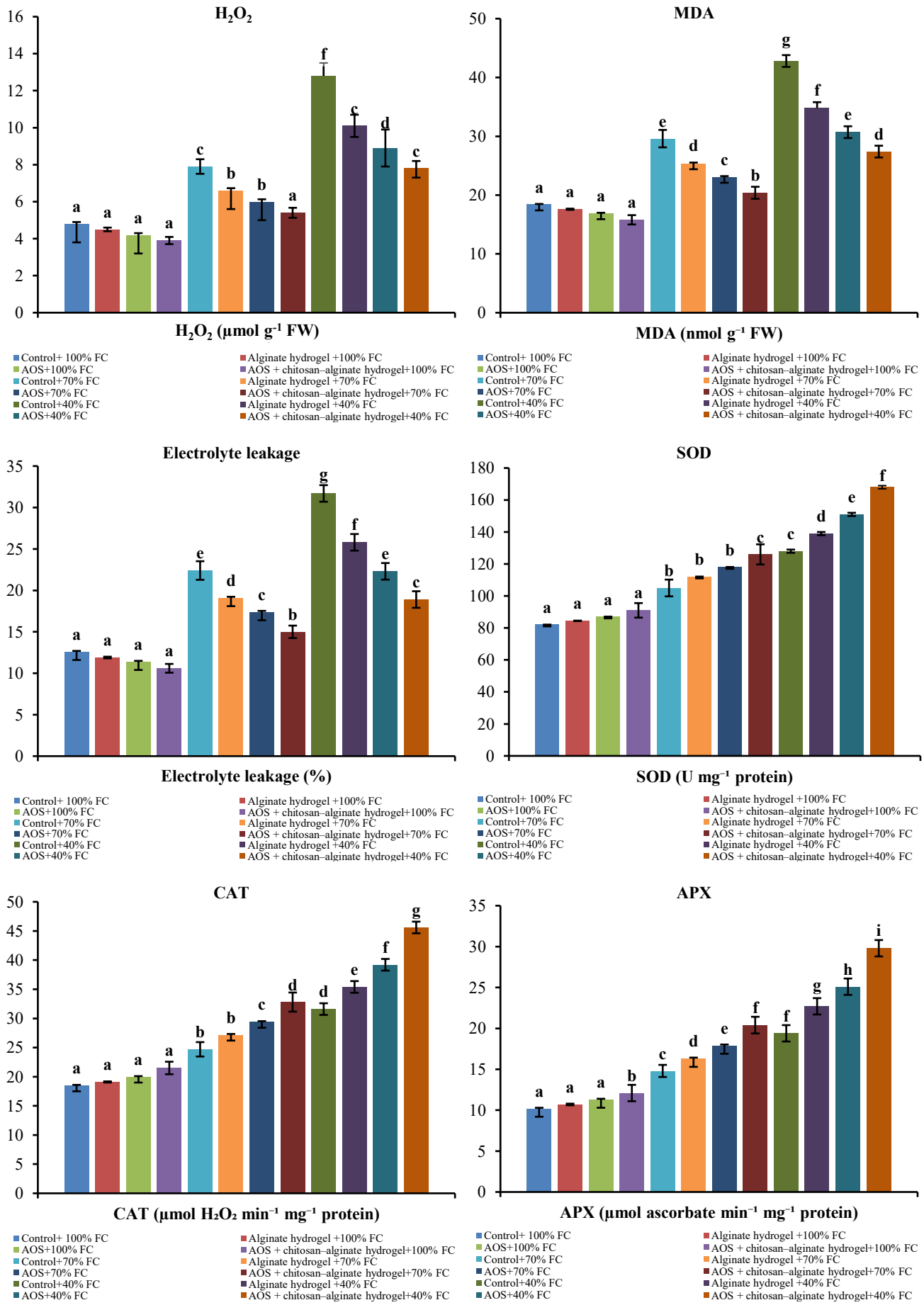


Fig. 1. Oxidative stress and antioxidant defense. Values are mean ± SD (n = 6). Different lowercase letters within columns indicate significant differences according to Tukey's test at $p \leq 0.05$.

Oxidative stress and antioxidant activity: Figure 1 shows that water deficit markedly increased oxidative stress in *Bougainvillea spectabilis*, as indicated by higher H_2O_2 accumulation, MDA concentration and electrolyte leakage. The increase was most pronounced under severe water stress. Application of AOS, alginate hydrogel and particularly AOS + chitosan–alginate hydrogelconsiderably reduced these stress indicators. In parallel, water deficit stimulated the activities of SOD, CAT and APX, with the greatest enzyme activities recorded in plants treated with AOS + chitosan–alginate hydrogel. The lower accumulation of oxidative damage together with stronger antioxidant activity suggests that the combined treatment enhanced ROS detoxification and maintained membrane stability under drought.

Root development and water-use efficiency: Root growth was strongly affected by drought (Table 4). In untreated plants, root length was decreased from 684 cm plant^{-1} under 100% FC to 421 cm plant^{-1} under 40% FC. AOS + chitosan–alginate hydrogel substantially reduced this decline. Root length reached 631 cm plant^{-1} under severe drought, while root surface area and root volume were also considerably higher than in untreated plants. Water-use efficiency was increased with the decrease of irrigation, reflecting greater biomass production per unit of water supplied. The highest WUE was consistently observed in AOS + chitosan–alginate hydrogel-treated plants. Under 40% FC, WUE reached 6.89 µmol CO_2 mmol^{-1} H_2O compared with 5.76 in untreated plants. Table 5 showed the significant correlation among all parameters studied.

Recovery following rewatering: Differences among treatments were obvious after rewatering (Table 6). Untreated drought-stressed plants recovered to an RWC of 76.8% after seven days. In comparison, the AOS + chitosan treated plants recovered up to 90.8% RWC. Photosynthesis also recovered more robustly in the treated plants. In AOS + chitosan–alginate hydrogel-treated plants, Pn was 16.7 µmol CO_2 m^{-2} s^{-1} while in untreated plants it was 10.4 µmol CO_2 m^{-2} s^{-1}. Also, the combined application maintained decreased MDA and electrolyte leakage after recovery. For AOS + chitosan treated plants the calculated recovery index was 86.7% whereas in untreated plants it was 63.4%.

Table 3. Water relations and osmotic adjustment.

Water regime	Treatment	RWC (%)	Leaf water potential (MPa)	Relative water loss (%)	Proline (µmol g^{-1} FW)	Soluble sugars (mg g^{-1} FW)	Glycine betaine (µmol g^{-1} FW)
100% FC	Control	88.6 ± 2.1 b*	−0.62 ± 0.04 d	19.4 ± 1.2 b	3.8 ± 0.3 b	18.6 ± 0.9 h	2.8 ± 0.2 i
100% FC	Alginate hydrogel	90.1 ± 1.9 a	−0.58 ± 0.03 c	18.2 ± 1.0 b	4.2 ± 0.3 ab	19.7 ± 0.8 g	3.0 ± 0.2 h
100% FC	AOS	91.3 ± 2.0 a	−0.55 ± 0.03 b	17.5 ± 1.1 a	4.6 ± 0.3 a	20.4 ± 0.9 g	3.2 ± 0.2 h
100% FC	AOS + chitosan–alginate hydrogel	92.5 ± 1.8 a	−0.51 ± 0.03 a	16.8 ± 0.9 a	5.0 ± 0.4 a	21.3 ± 1.0 g	3.4 ± 0.2 h
70% FC	Control	76.4 ± 1.9 d	−0.93 ± 0.05 h	27.8 ± 1.4 f	8.7 ± 0.5 c	25.9 ± 1.1 f	4.6 ± 0.3 g
70% FC	Alginate hydrogel	80.2 ± 1.8 c	−0.83 ± 0.04 g	25.1 ± 1.3 e	10.4 ± 0.6 d	28.2 ± 1.2 e	5.2 ± 0.3 f
70% FC	AOS	82.1 ± 1.7 bc	−0.78 ± 0.04 f	23.8 ± 1.2 d	11.6 ± 0.6 e	29.4 ± 1.3 d	5.6 ± 0.3 f
70% FC	AOS + chitosan–alginate hydrogel	85.7 ± 1.9 b	−0.69 ± 0.04 e	21.4 ± 1.1 c	13.2 ± 0.7 f	31.6 ± 1.4 d	6.2 ± 0.4 e
40% FC	Control	61.2 ± 1.8 g	−1.47 ± 0.07 l	39.6 ± 1.7 i	18.9 ± 0.9 g	38.4 ± 1.6 c	7.1 ± 0.4 d
40% FC	Alginate hydrogel	67.8 ± 1.7 f	−1.29 ± 0.06 k	35.1 ± 1.5 h	22.7 ± 1.0 h	42.1 ± 1.7 b	8.3 ± 0.5 c
40% FC	AOS	71.4 ± 1.9 e	−1.20 ± 0.06 j	32.7 ± 1.4 g	25.4 ± 1.1 i	44.6 ± 1.8 b	9.1 ± 0.5 b
40% FC	AOS + chitosan–alginate hydrogel	78.9 ± 1.8 c	−1.03 ± 0.05 i	28.4 ± 1.3 f	29.8 ± 1.3 j	48.7 ± 2.0 a	10.6 ± 0.6 a

* Values are mean ± SD (n = 6). Different lowercase letters within columns indicate significant differences according to Tukey's test at p≤0.05

Table 4. Root architecture and water-use efficiency.

Water regime	Treatment	Root length (cm plant^{-1})	Root surface area (cm^2)	Root volume (cm^3)	Root tips	Cumulative water applied (L plant^{-1})	Whole-plant WUE (g L^{-1})
100% FC	Control	683 ± 29 c	214 ± 11 d	38.6 ± 2.3 c	438 ± 23 c	48.1 ± 2.2 a	1.13 ± 0.05 e
100% FC	Alginate hydrogel	731 ± 33 b	228 ± 15 c	41.8 ± 2.3 c	471 ± 25 b	48.5 ± 2.2 a	1.19 ± 0.05 d
100% FC	AOS	757 ± 33 ab	241 ± 15 b	43.7 ± 1.6 b	493 ± 27 a	48.3 ± 2.1 a	1.21 ± 0.06 d
100% FC	AOS + chitosan–alginate hydrogel	793 ± 33 a	256 ± 11 a	46.9 ± 2.5 a	518 ± 29 a	48.6 ± 2.2 a	1.27 ± 0.06 c
70% FC	Control	603 ± 27 d	188 ± 10 e	33.7 ± 2.1 e	377 ± 21 e	38.7 ± 1.8 b	1.15 ± 0.05 d
70% FC	Alginate hydrogel	672 ± 30 c	210 ± 11 d	37.8 ± 2.1 d	426 ± 23 c	38.9 ± 1.8 b	1.26 ± 0.06 c
70% FC	AOS	708 ± 31 b	224 ± 12 c	40.4 ± 2.3 c	451 ± 25 b	38.8 ± 1.8 b	1.32 ± 0.06 c
70% FC	AOS + chitosan–alginate hydrogel	756 ± 33 ab	243 ± 13 b	44.1 ± 2.5 b	489 ± 26 a	39.0 ± 1.8 b	1.41 ± 0.07 b
40% FC	Control	421 ± 22 g	129 ± 8 h	23.2 ± 1.6 h	264 ± 17 g	28.4 ± 1.4 c	1.05 ± 0.05 e
40% FC	Alginate hydrogel	504 ± 23 f	156 ± 12 g	28.2 ± 1.8 g	319 ± 19 f	28.6 ± 1.4 c	1.21 ± 0.06 d
40% FC	AOS	548 ± 25 e	170 ± 10 f	30.8 ± 1.9 f	348 ± 20 e	28.5 ± 1.4 c	1.31 ± 0.06 c
40% FC	AOS + chitosan–alginate hydrogel	631 ± 321 d	198 ± 12 e	35.7 ± 2.1 d	406 ± 23 d	28.7 ± 1.4 c	1.55 ± 0.07 a

*Values are mean ± SD (n = 6). Different lowercase letters within columns indicate significant differences according to Tukey's test at p≤0.05. Whole-plant WUE = total dry biomass / cumulative water supplied

Table 5. Pearson correlation matrix among key physiological and biochemical parameters.

Parameter	RWC	Pn	gs	MDA	EL	SOD	CAT	APX	Proline	Root length	WUE
RWC	1.00										
Pn	0.92**	1.00									
gs	0.89**	0.94**	1.00								
MDA	−0.87**	−0.91**	−0.88**	1.00							
EL	−0.84**	−0.89**	−0.86**	0.93**	1.00						
SOD	0.78**	0.82**	0.79**	−0.76**	−0.74**	1.00					
CAT	0.81**	0.85**	0.83**	−0.79**	−0.77**	0.88**	1.00				
APX	0.79**	0.83**	0.80**	−0.78**	−0.75**	0.86**	0.91**	1.00			
Proline	0.75**	0.79**	0.76**	−0.72**	−0.70**	0.73**	0.76**	0.74**	1.00		
Root length	0.86**	0.88**	0.85**	−0.83**	−0.81**	0.77**	0.80**	0.78**	0.72**	1.00	
WUE	0.73**	0.71**	0.68**	−0.70**	−0.69**	0.65**	0.68**	0.66**	0.62**	0.70**	1.00

*Values are Pearson correlation coefficients (n = 72). **Significant at p≤0.01

Table 6. Recovery after rewatering.

Treatment	Pre-drought RWC (%)	RWC after recovery (%)	Pre-drought Pn (μmol CO_2 m^{-2} s^{-1})	Pn recovery (μmol CO_2 m^{-2} s^{-1})	Pre-drought gs (mol H_2O m^{-2} s^{-1})
Control	88.6 ± 2.1	76.8 ± 2.1 c	15.8 ± 0.7	10.4 ± 0.6 c	0.214 ± 0.012
Alginate hydrogel	90.1 ± 1.9	82.7 ± 2.0 b	16.7 ± 0.6	12.8 ± 0.7 b	0.226 ± 0.011
AOS	91.3 ± 2.0	85.6 ± 2.2 b	17.4 ± 0.7	14.1 ± 0.8 b	0.234 ± 0.012
AOS + chitosan–alginate hydrogel	92.5 ± 1.8	90.8 ± 2.3 a	18.2 ± 0.8	16.7 ± 0.8 a	0.243 ± 0.013

Treatment	gs recovery (mol H_2O m^{-2} s^{-1})	MDA (nmol g^{-1} FW)	EL (%)	Recovery index (%)
Control	0.118 ± 0.008 c	31.6 ± 1.5 c	23.1 ± 1.2 c	63.4 ± 2.8 c
Alginate hydrogel	0.146 ± 0.009 b	26.8 ± 1.3 b	19.4 ± 1.0 b	71.8 ± 3.1 b
AOS	0.161 ± 0.010 b	24.3 ± 1.2 b	17.2 ± 0.9 b	76.4 ± 3.2 b
AOS + chitosan–alginate hydrogel	0.194 ± 0.011 a	19.1 ± 1.0 a	13.5 ± 0.8 a	86.7 ± 3.5 a

*Values are mean ± SD (n = 6). Different lowercase letters within columns indicate significant differences according to Tukey's test at $p \leq 0.05$. Recovery index = mean percentage recovery of RWC, Pn, gs, MDA and EL, with damage parameters inverted

Discussion

Water deficit compromises both growth and ornamental quality: The results indicate that severe water deficiency leads to a considerable decrease in the growth of *B. spectabilis*. This is in agreement with the usual performance of ornamental plants during drought, which is that the availability of less water causes reduction in cell growth and photosynthetic acquisition of carbon (Toscano *et al.,* 2019; Giordano *et al.,* 2021). These effects are associated with declines in height, branching, leaf area and biomass. It has been established that the deficit irrigation may restrict the development of leaves and the production of biomass, although the degree of the response depends on the genotype and level of irrigation (Giordano *et al.,* 2021). The results reported here show that treatment of the roots-zone with biopolymer may reduce greatly these losses. The increase in flowering is especially relevant to our subject. In a commercially appealing shrub, the preservation of flowers and bracts is a commercial consideration of more importance than the mere maintenance of the biomass of the vegetative structures of the plant. Under severe drought circumstances, there was a large drop in the number of flowers and a delay in the time of flowering, both of which are indicators that water stress may directly affect the value of attractive plants. This loss was substantially prevented by AOS and chitosan, showing that the treatment was beneficial in preserving the physiological resources needed for biological development.

Maintaining plant water status appears to be central to the protective response: AOS + chitosan–alginate hydrogel had one of the most significant effects on the preservation of leaf moisture due to the capability of AOS and chitosan. The RWC of untreated plants was 61.2% whereas the RWC of treated plants was roughly 79% under acute drought conditions. The closing of the stomata becomes more and more severe with the significant drop of the RWC and the amount of photosynthesis continues to be more and more restricted. Similarly, recent investigations in *B. glabra* have shown the ability of melatonin to improve plant water status during drought stress (Wang *et al.,* 2026). Declining RWC was also identified as a critical component of drought damage in this study.

The increased RWC seen in the hydrogel treatment is likely due to the physical properties of the material. Alginate and chitosan are hydrophilic networks capable of absorbing enormous volumes of water. Alginate-based hydrogels have been reported as the best for improving water retention of the soil and root growth in drought situations (Gomaa & Aldaby, 2023). Chitosan-based hydrogels have also been studied as possible water-retaining agents for crops grown in water-limited situations (Nordin *et al.,* 2024). Chitosan and AOS may be both work as elicitors of the plants and they may change the physiological responses of the treated plants. Thus, the combination of treatments may have provided Bougainvillea with a bigger volume of available water in the root zone and also a significant improvement in its physiological capacity to absorb soil water.

Osmotic adjustment helped maintain cellular hydration: Bougainvillea plants showed increased quantity of proline, soluble carbohydrates and glycine betaine as response to the drought. These compounds play a vital role during the process of osmotic adjustment and may help cells retain water when the water potential of the environment drops. The higher accumulation of proline, soluble carbohydrates and glycine betaine in the AOS + chitosan–alginate hydrogel treated plants than the control indicates that this application promoted the natural acclimatization response of the plants. Similar alterations in proline and other stress-related metabolites have been found in Bougainvillea under drought stress (Wang *et al.,* 2026). Furthermore, this response is in agreement with the outcomes of previous research on AOS and chitosan in other plant species (Zhang *et al.,* 2020; Zhang *et al.,* 2024a; García-García *et al.,*2023). In addition, AOS treatment has been associated with improved drought tolerance and induced stress-related metabolic pathways. In contrast, the application of chitosan oligosaccharides at appropriate doses may improve the water status of plants in drought conditions (Zhang *et al.,* 2020; El-Araby *et al.,* 2024; Zhang *et al.,* 2024a; García-García *et al.,*2023). The most crucial point is that the combination treatment didn't only protect the plants from drying. Instead, it seems to have allowed plants to maintain osmotic adjustments while only a small amount of cellular damage occurred at the same time.

Photosynthesis was preserved by improving water status and reducing oxidative damage: During extreme droughts we expect declines in chlorophyll and gas exchange. This is because the stomata will shut and therefore the amount of carbon dioxide absorbed will decrease and dehydration may damage photosynthetic

structures. Relationships between water status and photosynthetic performance similar to those reported in Bougainvillea have been found in other ornamental plants (Dahab *et al.,* 2022; Liang *et al.,* 2023; Wang *et al.,* 2026). AOS + chitosan–alginate hydrogel prevented significant drop in chlorophyll. The treated plants maintained about twice the photosynthetic rate compared to the untreated plants growing in the high dryness. Furthermore, the stomatal conductance was higher in the treated plants. In contrast, a too much stomatal closure might lead to a significant decrease in photosynthesis. Closing of stomata is helpful as it prevents water loss in drought conditions. The treated plants seemed to manage to keep RWC at a higher level while at the same time maintaining a sufficient stomatal openness so that they could absorb carbon. Zhang *et al*., (2024a,b) recently demonstrated that the responses of Bougainvillea cultivars to environmental stress were reflected in coordinated changes in water status, leaf structure and photosynthetic performance. These results support the hypothesis that the preservation of photosynthetic activity is contingent on the maintenance of leaf hydration as an important need.

Stronger antioxidant protection reduced oxidative injury: Drought increased H_2O_2, MDA and electrolyte leaks indicating that water deprivation caused considerable oxidative stress in Bougainvillea. The rise in antioxidant enzymes suggested that plants aimed to detoxify the excess ROS. The recent drought study in *B. glabra* also found a similar trend with increased activity of antioxidant enzymes and elevated MDA during protracted water scarcity (Wang *et al.,* 2026). It was observed that the effect of AOS + chitosan was optimum, because it showed an increase in antioxidant enzyme activity and a reduction in oxidative damage. SOD transforms superoxide radicals to H_2O_2 and then H_2O_2 is removed by CAT and APX. Thus, increased activity of these enzymes gives a better mechanism to manage ROS. The decrease in MDA and electrolyte leakage confirmed that the higher antioxidant response resulted in improved protection of the membranes. This provides more information than just the enzyme activity since increased antioxidant activity in heavily damaged plants may be due to a response to excessive oxidative strain. Similar increases in antioxidants have been documented after AOS application in drought conditions (Idrissi *et al.,* 2024). Chitosan-based application have similarly been related with enhanced antioxidant defense (García-García *et al.,* 2023; Al-Yafrsi *et al.,* 2026). In the current investigation, the two polysaccharide-based strategies are combined in a root-zone delivery system extending these results.

Root development provides an additional explanation for improved drought tolerance: One of the most important findings to which the study led was the root response. Roots of untreated plants under high drought conditions were much shorter and had a smaller surface area. By contrast, the roots of AOS- and chitosan-treated plants were much more developed. The greater the root system the more substrate is reached and the more water that would be available and accessed. This is particularly important to consider in circumstances when the water supply to the substrate is not steady. In this procedure the hydrogel that retains water near the developing roots may also be useful. According to Gomaa & Aldaby (2023), hydrogels made of alginate may stimulate root development even when the plant is under drought stress. The statistics shown here indicate that a similar approach might be particularly useful in Bougainvillea, where root development and hydraulic qualities play a role in the ability of the plant to tolerate minimum watering. This provides a crucial mechanical link between the root-zone treatment and the advantages above ground:

Better root environment → Greater root development → Improved water uptake → Higher leaf hydration → Improved photosynthesis and growth.

Improved water-use efficiency has practical significance: The WUE increase was observed for AOS + chitosan–alginate hydrogelis of particular importance to ornamental horticulture in dry conditions. Deficit irrigation is more than just saving water. It's about getting plants to operate to a reasonable level with less water. Thus Bougainvillea has previously been considered as a useful plant for water conservation landscaping. Previous research has shown the role of irrigation management and genotype choice in increasing water efficiency (Giordano *et al.,* 2021). In addition to the selection of tolerant cultivars and to the decrease in frequency of watering, the present method provides another possibility: to adjust the root environment itself, in an attempt to obtain a higher water availability and physiological efficiency. This could be particularly advantageous if watering is inconsistent. A biodegradable hydrogel made from natural elements might reduce watering demands while keeping leaves and flowers alive. The findings suggest that AOS + chitosan–alginate hydrogel root-zone treatment has potential for improving the performance of *Bougainvillea spectabilis* under water-limited conditions. However, further validation under nursery, landscape, and field conditions is required before broad commercial recommendations can be made. The consistent treatment ranking across all measured variables likely reflects the uniform genetic material and controlled greenhouse conditions. Field validation with diverse genotypes is needed to confirm the generality of these findings.

Rapid recovery after drought may be more important than drought survival alone: The rewatering experiment is a significant addition to the research. Plants treated with AOS + chitosan–alginate hydrogel recovered their water status and photosynthetic activity substantially better than untreated plants. It implies that the treatment was sufficiently protective for the plant during drought to preserve functioning tissues that could restart activity when water was available. Conversely, untreated plants acquired more membrane damage that presumably reduced their capacity to recover swiftly. This is a critical difference for landscape plants since drought is never consistent or enduring. Plants may undergo multiple cycles of appropriate irrigation, water restriction and re-watering. Therefore, an application that speeds up recovery may be more effective than one which increases survival just in the near term. The results consequently suggest that AOS + chitosan–alginate hydrogel may operate as a kind of drought preconditioning. The treatment may prime the

plant to react more effectively when water becomes limited and then recover more quickly following replenishment. In an effort to determine whether this is actual physiological stress memory, more trials with repeated drought-rewatering cycles are required. Although the improved recovery observed after rewatering is consistent with enhanced drought preparedness or priming-like physiological responses, the present experiment does not directly establish drought stress memory. Confirmation of a true priming mechanism would require repeated stress–rewatering cycles and direct investigation of the underlying signaling pathways. The observation that AOS + chitosan-treated plants recovered Pn to levels slightly above the well-watered control (16.7 vs. 15.8 μmol CO_2 m^{-2} s^{-1}) suggests a possible hormetic or priming-like effect, though this requires further investigation.

Why the combined treatment was more effective than the individual treatments: The constant better performance of AOS + chitosan–alginate hydrogel over alginate hydrogel or AOS alone indicated that the effects were not attributable to a single mechanism. Alginate hydrogel mainly offers a physical advantage of water retention (Idrissi *et al.,* 2024). AOS delivers a more potent biological stimulation and may trigger stress-responsive pathways. The chitosan may provide both structural qualities to the hydrogel and extra elicitor activity (El-Araby *et al.,* 2024). The combined treatment therefore brings together several complementary functions:

Water retention + Root-zone modification + Stress signaling + Osmotic adjustment + Antioxidant defense

This multifunctional effect probably explains why the combined treatment was particularly effective under the most severe water deficit. The superior performance of the combined AOS + chitosan–alginate hydrogel treatment may reflect the complementary effects of water retention, root-zone modification, osmotic adjustment, and enhanced antioxidant defense. AOS may contribute to stress-responsive physiological signaling, whereas chitosan may provide additional elicitor activity. However, these mechanisms were not directly examined in the present study and therefore require further experimental verification. The present design does not include a chitosan-only treatment or a hydrogel + AOS treatment without chitosan. Therefore, the specific contribution of chitosan versus AOS versus the hydrogel matrix cannot be fully separated. The term 'complementary' is used here to describe the observed combined effect, not to imply a proven combined mechanism. Future studies with factorial separation of components are needed.

Relevance to Bougainvillea production under arid conditions: The findings are of particular significance for the use of Bougainvillea in dry and semi-arid environments. Bougainvillea is fairly tolerant to water restriction while it is difficult to keep excellent quality blooming plants alive under severe irrigation limits. Recently, it has been demonstrated that melatonin could enhance *B. glabra* drought tolerance by influencing physiological and biochemical responses and interacting with the microbial community in the rhizosphere (Wang *et al.,* 2026). This method directly addresses the root environment via the combination of water conservation and biological stimulation, unlike the other ways. In practice, this difference may be substantial. If stress intensifies, foliar feeding may be required multiple times. In addition, a root-zone hydrogel may be active in the substrate for a longer time and provide protection in close proximity to the roots (Idrissi *et al.,* 2024). This may be especially efficient for nursery production, Bougainvillea grown in containers and mature landscaping plants that are watered intermittently.

Conclusion

The data obtained imply that extreme water stress significantly affects the growth, flowering, photosynthetic performance and water status and increases oxidative stress and membrane damage in *Bougainvillea spectabilis*. Inclusion of AOS + chitosan–alginate hydrogel in a root-zone hydrogel substantially decreased these effects. The AOS + chitosan–alginate hydrogel was the most effective of all the treatments examined. It increased relative water content, photosynthetic activity and improved root development and osmotic adjustment. It enhanced antioxidant defense and lowered oxidative membrane damage. The treatment significantly boosted water usage efficiency and sustained flowering during severe water deficit. An interesting finding was the improved recovery following rewatering. Plants exposed to AOS + chitosan–alginate hydrogel recovered their water status and photosynthesis activity more rapidly and had less residual membrane damage than non-treated drought stressed plants. So the treatment might have a two-fold purpose, not only reducing drought damage, but also enhancing the plant's capacity to bounce back from dehydration. To conclude, the study proposes a new Bougainvillea drought management approach based on root-zone water conservation and physiological protection induced by biostimulants. This approach may be useful for ornamental production and landscaping in dry conditions, though field validation is needed. Future studies should investigate root hydraulic conductivity, aquaporin activity, ABA signaling, rhizosphere microorganisms, and several drought rewatering cycles to see whether the enhanced recovery is a real form of drought priming or physiological stress memory.

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