Vegetos
https://doi.org/10.1007/s42535-026-01708-5

RESEARCH ARTICLES

Effects of drought stress on morpho-physiological traits and mucilage yield in nine genotypes of *Alcea rosea* L

Arezoo Sadeghi[1] · Hassan Karimmojeni[1] · Timothy C. Baldwin[2] · Jamshid Razmjoo[1] · Abbas Ghafori[1] · Andrea Mastinu[3]

Received: 20 September 2025 / Revised: 19 April 2026 / Accepted: 23 April 2026
© The Author(s) 2026

Abstract
Hollyhock (*Alcea rosea* L.) is an important medicinal and ornamental species characterized by high genetic diversity and mucilage content; however, its response to drought stress remains insufficiently understood. In this study, nine genotypes ('Isfahan 1', 'Isfahan 2', 'Tabriz', 'Khomeini Shahr 1', 'Khomeini Shahr 2', 'Shahin Shahr', 'Shiraz 1', 'Shiraz 2', and 'Mahallat') were evaluated under three irrigation regimes using a randomized complete block design. Drought stress (I3 vs. I1) significantly reduced vegetative growth, 1000-seed weight, and seed yield, while enhancing mucilage accumulation by 26% in leaves, 93% in petals, and 291% in seeds. In contrast to typical stress responses, chlorophyll content increased under severe drought, suggesting a compensatory acclimation mechanism. Clear genotypic differences were observed. 'Khomeini Shahr 1', 'Tabriz', and 'Isfahan 2' exhibited greater drought tolerance, maintaining higher growth, physiological performance, and yield stability under water-limited conditions. In contrast, 'Shiraz 1' and 'Shiraz 2' were identified as the most sensitive genotypes. Importantly, increased mucilage production under drought stress was not consistently associated with reduced yield, indicating a genotype-dependent relationship rather than a strict trade-off. These findings highlight the potential for selecting drought-tolerant genotypes that combine yield stability with enhanced production of bioactive compounds, offering valuable implications for hollyhock cultivation in arid and semi-arid regions.

Keywords *Alcea rosea* · Drought stress · Genotypic variability · Mucilage · Gas exchange · Yield stability · Secondary metabolites

Introduction

Hollyhock (*Alcea rosea* L.), a member of the Malvaceae family, is a dicotyledonous ornamental and medicinal plant widely valued for its bioactive compounds, particularly mucilage, which is traditionally used for its anti-inflammatory and soothing properties (Nouraldini et al. 2010; Kumar et al. 2023). In addition to its ornamental importance, *A. rosea* represents a promising species for cultivation in arid and semi-arid regions due to its adaptability and economic value. However, climate change is expected to increase the frequency and intensity of extreme weather events, particularly prolonged drought, posing a serious threat to plant growth, productivity, and quality. In this context,

✉ Hassan Karimmojeni
kmojeni@iut.ac.ir

✉ Andrea Mastinu
andrea.mastinu@unibs.it

Arezoo Sadeghi
arezoo.sadeghi@ag.iut.ac.ir

Timothy C. Baldwin
t.baldwin@wlv.ac.uk

Jamshid Razmjoo
krazmjoo@cc.iut.ac.ir

Abbas Ghafori
a.ghafori@ag.iut.ac.ir

[1] Department of Agronomy and Plant Breeding, College of Agriculture, Isfahan University of Technology, Isfahan 84156-83111, Iran

[2] Faculty of Science and Engineering, University of Wolverhampton, Wulfruna St, Wolverhampton WV1 1LY, UK

[3] Department of Molecular and Translational Medicine, Division of Pharmacology, University of Brescia, Brescia, Italy

Published online: 11 May 2026

understanding how economically and medicinally important species such as hollyhock respond to water deficit conditions is becoming increasingly important (Sadeghi et al. 2024). Drought stress is one of the most critical abiotic factors limiting plant performance, as it reduces canopy development, restricts photosynthetically active radiation interception, and ultimately decreases biomass accumulation and yield (Cruz de Carvalho 2008; Cai et al. 2023). Morphological traits such as plant height, leaf number, and leaf area are particularly sensitive to water limitation due to the inhibition of cell expansion and division (Zhang et al. 2022; Hussain et al. 2019). In addition, plants regulate stomatal conductance and leaf development as adaptive strategies to minimize water loss under drought conditions. These morphological adjustments are closely linked to physiological processes. Drought stress often leads to reduced photosynthetic activity due to both stomatal and non-stomatal limitations, including restricted CO_2 diffusion, impaired enzymatic activity, and reduced electron transport efficiency (Flexas and Medrano 2022; Li et al. 2023). Consequently, dry matter accumulation declines, leading to significant reductions in yield and harvest index, particularly when stress occurs during reproductive stages (Zhao et al. 2023; Khan et al. 2021). Beyond its effects on growth and productivity, drought stress can also influence the synthesis of secondary metabolites. In *A. rosea*, mucilage represents a key bioactive compound with important medicinal applications. Previous studies have reported that mucilage accumulation tends to increase under water deficit conditions, suggesting a potential protective role in plant adaptation (Koocheki et al. 2007; Shekofteh et al. 2015). However, the relationship between drought stress, yield components, and mucilage production remains poorly understood, particularly in relation to genotypic variability. Despite the agronomic and medicinal importance of *A. rosea*, limited information is available on the integrated response of different genotypes to varying levels of water availability, especially under field conditions. In particular, it remains unclear how morpho-physiological traits, yield performance, and mucilage accumulation are coordinated under drought stress. Therefore, we hypothesized that drought stress would differentially affect morpho-physiological traits, yield components, and mucilage production among *A. rosea* genotypes, with more tolerant genotypes maintaining higher physiological performance and yield stability while enhancing mucilage accumulation as an adaptive response. Accordingly, the present study aimed to evaluate the response of nine *A. rosea* genotypes to different irrigation regimes, focusing on morpho-physiological traits, yield components, and mucilage production.

Materials and methods

Plant materials and growth conditions

Seeds of *Alcea rosea* L. were collected from naturalized populations in central Iran (Isfahan province), southern regions (Fars province, around Shiraz), and northeastern areas (Khorasan Razavi), as well as from montane habitats in East Azerbaijan (Tabriz region) and the Alborz mountain range (northern Iran). The nine genotypes (Shiraz 1, Shahin Shahr, Isfahan 2, Khomeini Shahr 2, Isfahan 1, Mahallat, Shiraz 2, Tabriz, Khomeini Shahr 1) evaluated in this study included both cultivated and wild types. Specifically, high-petal genotypes were obtained from the Agricultural and Natural Resources Research Center of Isfahan, while low-petal genotypes were collected from natural habitats across different regions, reflecting a broad range of genetic variability and potential adaptation to diverse environmental conditions. The experiment was conducted from February to September 2017 at the Lavark Research Farm, Isfahan University of Technology, Iran (32°43′ N, 51°31′ E; 1659 m a.s.l.). The site is characterized by a semi-arid climate, with a mean annual temperature of 14.5 °C and an average annual rainfall of approximately 90 mm (Fig. 1)

. The main physicochemical properties of the experimental soil are reported in Table 1. A randomized complete block design (RCBD) with three replications was employed. The experimental treatments consisted of a factorial combination of three irrigation regimes and nine *A. rosea* genotypes, resulting in a total of 81 experimental units ($3 \times 9 \times 3$). Each plot covered an area of 2 m^2 and consisted of five rows spaced 50 cm apart, with 20 cm between plants within each row, corresponding to a planting density of 25 plants m^{-2}. Prior to sowing, seeds were surface-sterilized in a sodium hypochlorite solution (5 g L^{-1}) for 5 min and thoroughly rinsed with distilled water. Sowing was performed directly in the field. Irrigation was applied using a drip irrigation system to ensure uniform water distribution.

Drought treatments

To ensure uniform crop establishment, all plots were irrigated equally during the early growth stage. Five irrigation events were applied until plants reached approximately 30 cm in height (about 40 days after sowing). Thereafter, differential irrigation treatments were imposed and maintained until the end of the growing season (early October).

Three irrigation regimes were defined based on permissible soil moisture depletion (P):

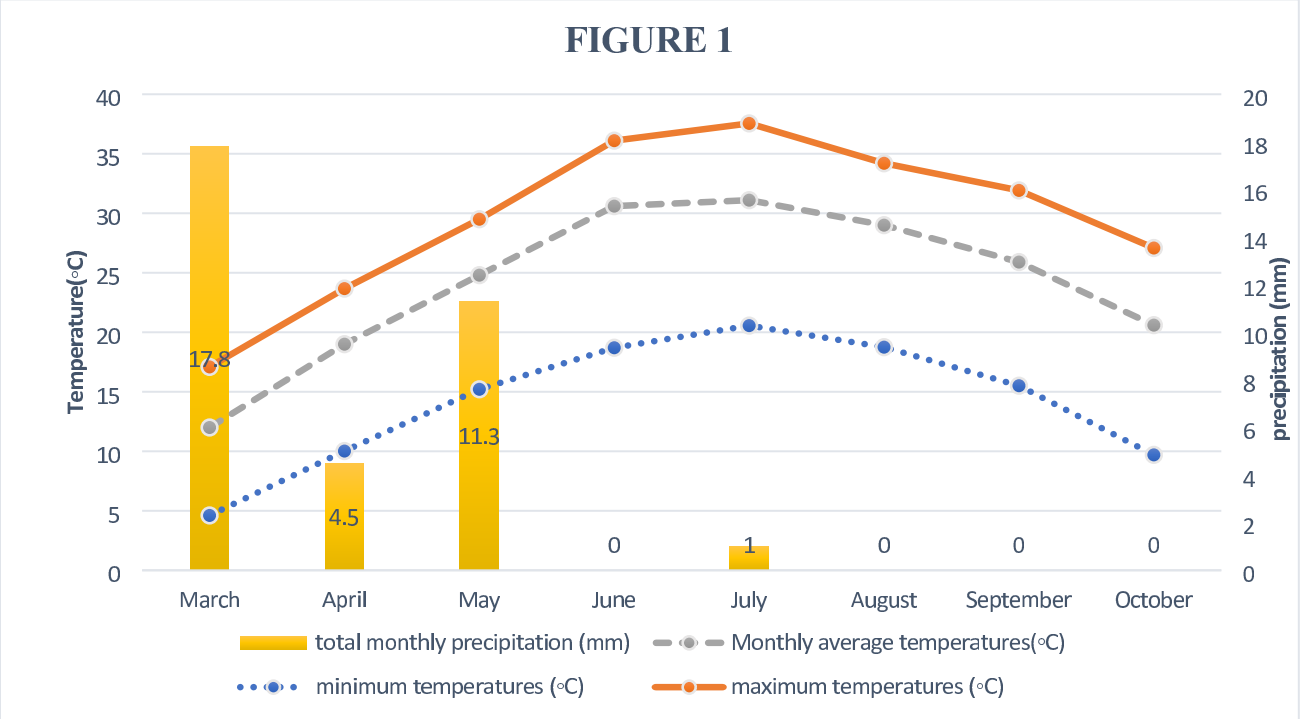


Fig. 1 Monthly average maximum and minimum temperature (°C) and total monthly precipitation (mm) in growing season in 2017

Table 1 Soil physico-chemical characteristics of studied soils

Depth (cm)	30 cm
Electrical conductivity (dS m^{-1})	11.18 ds m^{-1}
pH	7.5
Organic carbon percentage (OC %)	0.6%
Phosphorus (PPM)	7.14 mg kg^{-1}
Potassium (PPM)	44.2 mg kg^{-1}
Clay (%)	38.5
Silt (%)	36
Sand (%)	25.5

- I1: 30% depletion (well-watered control).
- I2: 60% depletion (moderate drought stress).
- I3: 80% depletion (severe drought stress).

Soil moisture content was monitored throughout the experiment using the gravimetric method. Soil samples were collected at a depth of 0–10 cm from two randomly selected points within each plot, approximately 72 h after irrigation. Samples were weighed, oven-dried at 105 °C until constant weight, and reweighed to determine soil water content. Irrigation scheduling was based on the measured soil moisture content and the predefined depletion thresholds for each treatment. When soil water content reached the assigned depletion level, irrigation was applied to restore soil moisture close to field capacity. The volume of irrigation water applied to each plot (Vw) was calculated based on soil available water (SAW), soil bulk density (Pb), root zone depth (D), and irrigation efficiency (Ea), according to standard soil water balance equations. This approach ensured that each treatment received a controlled and consistent level of water deficit corresponding to the intended stress intensity.

$$SAW = (\theta_{FC} - \theta_{PWP}) \times P_b \times V$$

Where:

- SAW (Soil Available Water): the amount of water that plants can use, calculated from soil moisture at field capacity and permanent wilting point.
- θFC: soil moisture content at field capacity (weight%).
- θPWP: soil moisture content at permanent wilting point (weight%).
- Pb: soil bulk density at the root development depth (g cm^{-3}).
- V: experimental plot volume used in soil moisture calculations.

$$V = (A \times D)$$

- A: surface area of the plot (m^2).
- D: depth of root development (cm or m, depending on calculation).

$$V_w = \frac{SAW \times P}{E_a}$$

- Vw: volume of water applied per irrigation (m³).
- Ea: irrigation water application efficiency (%).
- P: permissible soil moisture depletion for each treatment (30% for control, 60% for moderate stress, and 80% for severe stress).

$$\theta_{irr} = \theta_{FC} - (\theta_{FC} - \theta_{PWP}) \times P$$

θirr: volumetric soil moisture at the time of irrigation.

Measurements

At the 50% flowering stage, physiological and morphological traits were evaluated. Chlorophyll content, carotenoids, and gas-exchange parameters were measured on three representative plants per experimental plot. Growth attributes, including plant height, number of leaves, number of flowers, and leaf area, were also recorded at this stage. At full maturity, yield-related traits were determined, including aerial dry weight, number of seeds per capsule, 1000-seed weight, and total seed yield. In addition, mucilage yield was quantified separately for leaves, petals, and seeds. Leaf area was measured using a leaf area meter (Green Leaf Area Meter, model GA-5, Iran). Plant height and flower number were recorded on five randomly selected plants per plot and expressed as mean values. For pigment analysis, young fully expanded leaves from the upper canopy were collected from three plants per plot. Chlorophyll and carotenoid contents were determined spectrophotometrically according to the method of Lichtenthaler (1987). Gas-exchange parameters, including net photosynthetic rate (PN), transpiration rate (E), and stomatal conductance (Gs), were measured on the youngest fully expanded leaves using a portable photosynthesis system (LCA-4, ADC Bioscientific Ltd., UK). Measurements were conducted under stable environmental conditions at midday (11:00–13:00), with leaf temperature maintained at 28±2 °C, ambient CO_2 concentration at 370–390 µmol mol^{-1}, and photosynthetic photon flux density (PPFD) ranging from 350 to 500 µmol m^{-2} s^{-1}.

Mucilage measurement

Mucilage extraction was performed after two months of exposure to drought treatments. One representative plant per experimental unit was sampled, and leaves, petals, and seeds were collected separately. For each organ, 1 g of fresh tissue was homogenized with 10 mL of 0.1 N HCl using a mortar and pestle, followed by the addition of 10 mL of distilled water. The homogenate was filtered through a metal sieve and transferred into an Erlenmeyer flask connected to a vacuum filtration system. To precipitate mucilage, 60 mL of ethanol was added to the filtrate. The resulting mixture was filtered through pre-weighed filter paper, and the retained mucilage was dried to constant weight. Mucilage yield was calculated as the difference between the initial and final weight of the filter paper and expressed on a fresh weight basis.

Statistical analysis

All data were subjected to analysis of variance (ANOVA) using SAS software (version 9.2). The effects of irrigation regime, genotype, and their interaction were evaluated. Mean comparisons were performed using the least significant difference (LSD) test at the 5% probability level. Prior to analysis, data were checked for normality and homogeneity of variance to ensure the validity of ANOVA assumptions.

Results and discussion

Vegetative growth traits

The effects of irrigation regime, genotype, and their interaction were highly significant ($P \leq 0.01$) for all vegetative traits (Table 2),

indicating strong genotype × environment interactions under water deficit. Leaf number decreased by 20% and 28% under I2 and I3, respectively, compared with the control (I1) (Table 3),

Table 2 Values of mean squares in the analysis of variance of leaf number, leaf area (cm^{-2}), plant height (cm), flower number and plant dry weight (g) in 9 hollyhock (*Alcea rosea* L.) genotypes under reduced irrigation

S.O.V	df	Leaf number	Leaf area	Plant height	Flower number	Plant dry weight
Irrigation regimes (I)	2	105**	52,041**	11,524**	490**	0.25**
Genotype (G)	8	13.4**	7172**	118**	1637**	0.074 **
I × A	16	11**	3324**	3789**	1468**	0.072**
Error	48	3.70	349	42.5	103	0.00004

* and **: Significant at the 5% and 1% probability levels, respectively., ns: Non- Significant

Table 3 Comparison of the effects of different irrigation regimes and genotypes on leaf number, leaf area (cm^2), plant height (cm), flower number and plant dry weight (g) in 9 hollyhock (*Alcea rosea* L.) genotypes under reduced irrigation

	Leaf number	Leaf area (Cm^2)	Plant height (Cm)	Flower number	Plant dry weight (g)
Irrigation regimes					
I_1	13.8 [a]	45.5 [a]	120.9 [a]	52.3 [a]	0.202 [a]
I_2	11 [b]	27.6 [b]	86.1 [b]	30.7 [b]	0.043 [b]
I_3	10 [c]	18.1 [c]	69.6 [c]	22.5 [c]	0.029 [c]
LSD	1.86	11.6	7.25	1.89	0.004
Genotypes					
Shiraz 1	13.5 [ab]	22.4 [e]	79.1 [de]	34.8 [cd]	0.034 [df]
Shahin Shahr	12.6 [bc]	35.7 [b]	111 [b]	38.1 [c]	0.052 [d]
Isfahan 2	8.3 [e]	30.8 [d]	118 [a]	66.7 [a]	0.103 [c]
Khomeini Shahr 2	13.6 [ab]	30 [d]	78.2 [de]	22.8 [g]	0.056 [d]
Isfahan 1	11.4 [ed]	33.9 [d]	91.9 [c]	28.2 [f]	0.037 [e]
Mahallat	8.1 [e]	21.8 [d]	74.2 [e]	29.8 [ef]	0.054 [d]
Shiraz 2	10.4 [d]	33.4 [e]	74 [e]	41.5 [b]	0.028 [f]
Tabriz	11.3 [cd]	47.5 [a]	119 [e]	32.6 [de]	0.146 [b]
Khomeini Shahr 1	14.8 [a]	17.9 [f]	84 [a]	21.1 [g]	0.312 [a]
LSD	1.82	17.7	6.18	3.42	0.006

Means followed by the same letter are not significantly different by the LSD test at $P \leq 0.05$

confirming the high sensitivity of leaf initiation and expansion to soil moisture availability. This reduction reflects adaptive responses aimed at limiting transpirational surface area through reduced meristematic activity and accelerated senescence. Genotypic differences were evident: 'Khomeini Shahr 1' maintained the highest leaf number across irrigation regimes, suggesting superior vegetative resilience, whereas 'Mahallat' and 'Isfahan 2' showed greater sensitivity. Similar drought-induced reductions in leaf development have been reported in several crops, supporting the role of leaf area modulation as a conserved water-saving strategy (Zhang et al. 2023; Yemata and Bekele 2024). Leaf area declined by 39% (I2) and 60% (I3), with substantial variation among genotypes. The smaller reduction observed in 'Khomeini Shahr 1' (34%) compared with 'Isfahan 2' (87%) suggests more effective maintenance of cell expansion and turgor under stress. This may be associated with improved osmotic adjustment and water retention capacity, which are key determinants of drought tolerance (Hussain et al. 2019; Zhang et al. 2023). Plant height was significantly reduced under increasing drought intensity, with reductions ranging from 23% to 53% (Table 4).

This decrease is primarily attributed to limitations in cell elongation and reduced assimilate availability under stress conditions. Similarly, flower number declined across most

Table 4 Interactive effects of irrigation regimes and genotypes on leaf number, leaf area (cm^{-2}), plant height (cm), flower number and plant dry weight (g) in hollyhock (*Alcea rosea* L.)

Genotype	Irrigation regimes	Leaf number	Leaf area (Cm^2)	Plant height (Cm)	Flower number	Plant dry weight (g)
Shiraz 1	I_1	13	31.73	103	45	0.045
Shahin Shahr	I_1	18	48.26	142	47	0.095
Isfahan 2	I_1	15	69.9	164	147	0.174
Khomeini Shahr 2	I_1	12	36.7	102	41	0.097
Isfahan 1	I_1	13	45.7	128	34	0.054
Mahallat	I_1	12	27.5	86	30	0.089
Shiraz 2	I_1	9	54	91	66	0.032
Tabriz	I_1	15	73.8	168	35	0.36
Khomeini Shahr 1	I_1	18	21.6	100	27	0.87
Shiraz 1	I_2	11	18.9	73.2	38	0.023
Shahin Shahr	I_2	15	35.9	105	45	0.035
Isfahan 2	I_2	6	13.3	112	30	0.076
Khomeini Shahr 2	I_2	11	32	70.4	16	0.043
Isfahan 1	I_2	12	32.6	82	26	0.032
Mahallat	I_2	6	21.9	70.3	27	0.041
Shiraz 2	I_2	10	34.4	74.8	40	0.032
Tabriz	I_2	13	41.3	104	38	0.048
Khomeini Shahr 1	I_2	15	18.1	80.6	17	0.044
Shiraz 1	I_3	16	16.6	60.6	21	0.025
Shahin Shahr	I_3	5	23	84.46	23	0.025
Isfahan 2	I_3	5	91.5	77.33	23	0.061
Khomeini Shahr 2	I_3	18	21.9	62.4	14	0.028
Isfahan 1	I_3	9	23.6	64.46	25	0.025
Mahallat	I_3	6	16.1	66.06	23	0.034
Shiraz 2	I_3	13	11.8	55.73	19	0.020
Tabriz	I_3	7	27.6	84.2	26	0.025
Khomeini Shahr 1	I_3	12	14.1	71.33	19	0.023

Table 5 Values of mean squares in the analysis of variance of chlorophyll content (Chl.) (mg g^{-1} FW), carotenoids (mg g^{-1} FW), net photosynthetic rate (PN, µmol CO_2 m^{-2} sec^{-1}, transpiration rate (E, µmol H_2O m^{-2} sec^{-1}) and stomatal conductance (Gs, mol H_2O m^{-2} s^{-1}) in 9 hollyhock (*Alcea rosea* L.) genotypes under reduced irrigation

S.O.V	df	Chl.	G	Gs	E
Irrigation regimes (I)	2	0.009**	40.00**	37,729**	12**
Genotype (G)	8	0.006**	10.00**	24,523**	0.392 ns
I × G	16	0.0004**	26.50**	16,140**	6.79**
Error	48	0.00003	0.241	2066	0.532

* and **: Significant at the 5% and 1% probability levels, respectively., ns: Non- Significant

Table 6 Comparison of the effects of different irrigation regimes and genotypes on chlorophyll content (Chl.) (mg g^{-1} FW), *m* ratio, carotenoids (mg g^{-1} FW), net photosynthetic rate (PN, µmol CO_2 m^{-2} sec^{-1}, transpiration rate (E, µmol H_2O m^{-2} sec^{-1}) and stomatal conductance (Gs, mol H_2O m^{-2} s^{-1}) in 9 hollyhock (*Alcea rosea* L.) genotypes under reduced irrigation

	Chl.	G	Gs	E
Irrigation regimes				
I_1	0.064 c	7.28 b	273 a	5.92 a
I_2	0.077 b	5.24 c	298 b	4.60 a
I_3	0.1 a	2.9 a	365 c	3.38 a
LSD	0.01	0.57	53.47	0.69
Genotypes				
Shiraz 1	0.064 d	4.3 d	315 b	3.83 cd
Shahin Shahr	0.065 a	2.47 f	257 d	2.98 e
Isfahan 2	0.054 b	3.32 e	231 d	5.27 b
Khomeini Shahr 2	0.045 f	7.97 a	327 b	4.41 c
Isfahan 1	0.04 b	3.12 c	359 b	5.49 b
Mahallat	0.03 f	6.75 c	419 a	4.28 c
Shiraz 2	0.026 d	0.588 d	261 cd	3.5 de
Tabriz	0.02 c	3.96 d	218 d	5.18 b
Khomeini Shahr 1	0.013 e	7.01 bc	311 bc	6.75 a
LSD	0.003	0.27	25.09	0.4

Means followed by the same letter are not significantly different by the LSD test at $P \leq 0.05$

genotypes, although the increase observed in 'Mahallat' suggests a possible stress-escape strategy through accelerated reproductive allocation, as reported in drought-adaptive crops (Zhang et al. 2022).

Physiological traits

Chlorophyll and carotenoid contents were significantly affected by irrigation treatments (Table 5).

Notably, chlorophyll content increased under severe drought (I3), which contrasts with the commonly reported decline under stress. This response may reflect a compensatory acclimation mechanism, whereby increased pigment concentration per unit leaf area enhances light harvesting under reduced leaf expansion. Alternatively, it may result from a concentration effect due to reduced leaf water content (Rustioni and Bianchi 2021; Yan et al. 2024). Carotenoid accumulation further supports this interpretation, as these pigments play a critical role in photoprotection by dissipating excess energy and preventing oxidative damage. Such adjustments are consistent with enhanced stability of photosystem II and improved energy dissipation under stress conditions (Zait et al. 2021; Yan et al. 2024). In contrast, gas-exchange parameters (PN, E, and gs) declined significantly under I2 and I3 (Table 6),

indicating strong limitations to photosynthesis. These reductions are primarily driven by stomatal closure, which restricts CO_2 diffusion, but also involve non-stomatal factors such as decreased Rubisco activity, impaired electron transport, and reduced mesophyll conductance (Flexas and Medrano 2022; Li et al. 2023). These responses are part of a broader physiological adjustment to drought stress, involving osmotic regulation, antioxidant defense activation, and hormonal signaling pathways such as abscisic acid (ABA), which collectively contribute to stress tolerance (Ma et al.

Table 7 Interactive effects of irrigation regimes and genotypes on chlorophyll content (Chl.) (mg g^{-1} FW), *m* ratio, carotenoids (mg g^{-1} FW), net photosynthetic rate (PN, µmol CO_2 m^{-2} sec^{-1}, transpiration rate (E, µmol H_2O m^{-2} sec^{-1}) and stomatal conductance (Gs,, mol H_2O m^{-2} s^{-1}) in hollyhock (*Alcea rosea* L.)

Genotype	Irrigation regimes	Chl.	A	Gs	E
Shiraz 1	I_1	0.051	6.13	258	4.05
Shahin Shahr	I_1	0.095	4.02	217	4.22
Isfahan 2	I_1	0.084	12.8	191	6.36
Khomeini Shahr 2	I_1	0.041	4.34	282	6.29
Isfahan 1	I_1	0.096	13.6	218	7.43
Mahallat	I_1	0.033	5.55	290	5.67
Shiraz 2	I_1	0.050	8.23	240	4.66
Tabriz	I_1	0.060	4.56	183	7.03
Khomeini Shahr 1	I_1	0.055	8.56	255	7.54
Shiraz 1	I_2	0.063	4.8	286	3.91
Shahin Shahr	I_2	0.118	2.63	273	2.97
Isfahan 2	I_2	0.102	5.24	214	5.76
Khomeini Shahr 2	I_2	0.047	3.4	327	3.55
Isfahan 1	I_2	0.100	7.73	364	4.86
Mahallat	I_2	0.051	3.15	463	3.62
Shiraz 2	I_2	0.073	6.54	245	3.57
Tabriz	I_2	0.076	3.74	195	5.58
Khomeini Shahr 1	I_2	0.060	7.56	290	7.52
Shiraz 1	I_3	0.100	1.93	400	3.53
Shahin Shahr	I_3	0.178	0.76	283	1.78
Isfahan 2	I_3	0.130	3.97	261	3.66
Khomeini Shahr 2	I_3	0.056	2.23	371	3.4
Isfahan 1	I_3	0.111	2.53	496	4.18
Mahallat	I_3	0.065	0.68	504	3.55
Shiraz 2	I_3	0.082	5.49	300	2.27
Tabriz	I_3	0.111	3.6	278	2.93
Khomeini Shahr 1	I_3	0.064	4.91	390	5.18

Means followed by the same letter are not significantly different by the LSD test at $P \leq 0.05$

2020; Li et al. 2023). Genotypic variability was pronounced. 'Khomeini Shahr 1' and 'Shiraz 2' maintained relatively higher PN and E under drought, suggesting more efficient stomatal regulation and better preservation of biochemical photosynthetic capacity. In contrast, 'Mahallat' and 'Isfahan 2' exhibited stronger declines, indicating higher sensitivity to water deficit. These differences highlight distinct physiological strategies, with tolerant genotypes maintaining carbon assimilation despite reduced water availability (Table 7).

Yield traits

Irrigation regime and genotype significantly affected yield components (Table 8).

Severe drought reduced 1000-seed weight by up to 53%, confirming the strong sensitivity of reproductive processes to water deficit. Yield reductions are primarily associated with impaired assimilate translocation and reduced photosynthetic capacity during critical reproductive stages (Sallam et al. 2019; Zhao et al. 2023). Among genotypes, 'Isfahan 2' showed the highest seed and flower yield under optimal conditions, whereas 'Shiraz 1' exhibited the lowest performance (Tables 9 and 10).

However, under drought stress, genotypes such as 'Khomeini Shahr 1' and 'Tabriz' demonstrated greater yield stability, indicating superior tolerance. Yield stability under stress is considered a key trait for selecting drought-resilient genotypes in semi-arid environments (Zhang et al. 2023).

Mucilage content

Mucilage yield increased consistently with drought intensity, particularly in seeds (+291% under I3), suggesting a strong stress-induced activation of polysaccharide synthesis. This response may be interpreted as an adaptive mechanism contributing to osmotic adjustment, water retention, and cellular protection under dehydration (Rahimi et al. 2013; Ma et al. 2020). Genotypic variation was evident. 'Isfahan 2' exhibited the highest seed mucilage content, whereas 'Shiraz 1' and 'Shiraz 2' showed the lowest values. Similarly, 'Khomeini Shahr 1' showed the highest petal mucilage accumulation. These differences suggest that mucilage production is under genetic control and may contribute to differential drought tolerance. Importantly, the

Table 8 Values of mean squares in the analysis of variance of 1000 seed weight (1000-SW, g), seed yield (g), flower yield (g), leaf mucilage (g), petal mucilage (g) and seed mucilage (g) in 9 hollyhock (*Alcea rosea* L.) Genotypes under reduced irrigation

S.O.V	df	1000-sw	Seed yield	Flower yield	Leaf mucilage	Petal mucilage	Seed mucilage
Irrigation regimes (I)	2	0.922**	1,202,581**	203,733**	0.004**	489**	0.005**
Genotype (G)	8	1.598**	102,561**	30,072**	0.00004**	0.015 **	0.011**
I × G	16	0.143**	14,175**	2451**	0.02578**	0.001**	0.002**
Error	48	0.014	944	103	0.00001	0.00001	0.00001

* and **: Significant at the 5% and 1% probability levels, respectively., ns: Non- Significant

Table 9 Comparison of the effects of different irrigation regimes and genotypes on 1000 seed weight (1000-SW, g), seed yield (g), flower yield (g), leaf mucilage (g), petal mucilage (g) and seed mucilage (g) in 9 hollyhock (*Alcea rosea* L.) genotypes under reduced irrigation

	1000-sw	Seed yield	Flower yield	Leaf mucilage	Petal mucilage	Seed mucilage
Irrigation regimes						
I_1	2.02 ^{a}	729 ^{a}	353 ^{a}	0.042 ^{c}	0.023 ^{c}	0.009 ^{c}
I_2	1.7 ^{a}	456 ^{b}	277 ^{b}	0.055 ^{b}	0.034 ^{b}	0.025 ^{b}
I_3	1.54 ^{b}	313 ^{c}	179 ^{c}	0.054 ^{a}	0.045 ^{a}	0.037 ^{a}
LSD	0.065	46.72	9.125	0.004	0.004	0.002
Genotypes						
Shiraz 1	1.22 ^{e}	376 ^{e}	202 ^{f}	0.022 ^{g}	0.029 ^{c}	0.003 ^{e}
Shahin Shahr	2.14 ^{b}	563 ^{b}	314 ^{b}	0.033 ^{d}	0.028 ^{c}	0.004 ^{e}
Isfahan 2	2.34 ^{a}	659 ^{a}	340 ^{a}	0.025 ^{fg}	0.013 ^{e}	0.116 ^{a}
Khomeini Shahr 2	2.12 ^{b}	577 ^{b}	313 ^{b}	0.049 ^{c}	0.022 ^{d}	0.011 ^{d}
Isfahan 1	1.43 ^{d}	460	252 ^{d}	0.029 ^{fg}	0.142 ^{a}	0.005 ^{e}
Mahallat	1.98 ^{c}	554 ^{b}	306 ^{b}	0.03 ^{de}	0.038 ^{b}	0.013 ^{cd}
Shiraz 2	1.27 ^{e}	334 ^{f}	183 ^{g}	0.03 ^{dc}	0.031 ^{c}	0.003 ^{e}
Tabriz	1.41 ^{d}	421 ^{d}	183 ^{e}	0.091 ^{b}	0.006 ^{f}	0.045 ^{b}
Khomeini Shahr 1	1.86 ^{c}	520 ^{b}	302 ^{c}	0.186 ^{a}	0.007 ^{f}	0.016 ^{c}
LSD	0.123	29.13	9.66	0.004	0.003	0.002

Means followed by the same letter are not significantly different by the LSD test at $P \leq 0.05$

Table 10 Interactive effects of irrigation regimes and genotypes on 1000 seed weight (1000-SW, g), seed yield (g), flower yield (g), leaf mucilage (g), petal mucilage (g) and seed mucilage (g) in hollyhock (*Alcea rosea* L.)

Genotype	Irrigation regimes	1000-sw	Seed yield	Flower yield	Leaf mucilage	Petal mucilage	Seed mucilage
Shiraz 1	I_1	1.39	521	242	0.018	0.013	0.001
Shahin Shahr	I_1	2.31	857	422	0.031	0.026	0.004
Isfahan 2	I_1	2.57	949	452	0.013	0.011	0.02
Khomeini Shahr 2	I_1	2.36	875	421	0.031	0.008	0.006
Isfahan 1	I_1	1.6	637	329	0.021	0.083	0.009
Mahallat	I_1	2.19	846	413	0.025	0.036	0.01
Shiraz 2	I_1	1.7	486	219	0.028	0.023	0.001
Tabriz	I_1	1.96	560	271	0.053	0.003	0.028
Khomeini Shahr 1	I_1	2.1	827	405	0.163	0.005	0.005
Shiraz 1	I_2	1.17	342	221	0.022	0.022	0.002
Shahin Shahr	I_2	2.14	497	315	0.033	0.028	0.001
Isfahan 2	I_2	2.37	635	328	0.036	0.013	0.14
Khomeini Shahr 2	I_2	2.8	511	317	0.046	0.018	0.011
Isfahan 1	I_2	1.45	432	263	0.029	0.162	0.005
Mahallat	I_2	1.79	495	310	0.028	0.038	0.012
Shiraz 2	I_2	1.14	309	201	0.03	0.012	0.001
Tabriz	I_2	1.24	394	226	0.093	0.007	0.04
Khomeini Shahr 1	I_2	1.8	493	309	0.186	0.006	0.011
Shiraz 1	I_3	1.1	264	144	0.027	0.054	0.005
Shahin Shahr	I_3	1.96	334	206	0.035	0.03	0.007
Isfahan 2	I_3	2.07	394	240	0.037	0.015	0.187
Khomeini Shahr 2	I_3	1.82	346	201	0.07	0.04	0.017
Isfahan 1	I_3	1.25	312	165	0.036	0.18	0.002
Mahallat	I_3	1.97	320	196	0.036	0.04	0.017
Shiraz 2	I_3	0.98	209	129	0.033	0.027	0.003
Tabriz	I_3	1.03	310	143	0.126	0.009	0.066
Khomeini Shahr 1	I_3	1.69	329	191	0.21	0.011	0.033

Means followed by the same letter are not significantly different by the LSD test at $P \leq 0.05$

increase in mucilage content was not consistently associated with yield reduction, indicating that this trait may function as a complementary adaptive mechanism rather than a strict trade-off. This finding suggests that some genotypes are capable of coordinating carbon allocation between growth and protective metabolite synthesis under stress conditions (Ma et al. 2020; Zait et al. 2021).

Overall drought response and agronomic implications

The results demonstrate clear genotype-dependent differences in drought adaptation strategies in *A. rosea*. Tolerant genotypes such as 'Khomeini Shahr 1', 'Tabriz', and 'Isfahan 2' maintained higher vegetative growth, better physiological performance, and more stable yield under water deficit, while also exhibiting increased mucilage production. These findings indicate that drought tolerance in *A. rosea* is associated with an integrated response involving morphological adjustment, maintenance of photosynthetic function, and activation of protective biochemical pathways, as widely reported in drought-adapted crops (Flexas and Medrano 2022; Li et al. 2023). From an agronomic perspective, genotypes such as 'Khomeini Shahr 1' and 'Tabriz' represent promising candidates for cultivation in water-limited environments due to their ability to sustain yield performance while enhancing the production of valuable bioactive compounds. This dual advantage is particularly relevant for medicinal plant production systems in arid and semi-arid regions.

Conclusion

Our findings demonstrate that *Alcea rosea* exhibits pronounced genotype-dependent variation in response to drought stress, reflecting the presence of distinct adaptive strategies within the species. Water deficit significantly reduced vegetative growth, photosynthetic performance, and yield components, while markedly enhancing mucilage accumulation in leaves, petals, and seeds. Among the evaluated genotypes, 'Khomeini Shahr 1', 'Tabriz', and 'Isfahan

2' showed superior drought tolerance, maintaining higher vegetative growth, greater pigment stability, and more efficient gas-exchange performance under water-limited conditions. In contrast, 'Shiraz 1' and 'Shiraz 2' were identified as highly sensitive, exhibiting substantial reductions across morphological, physiological, and yield-related traits. A key outcome of this study is that increased mucilage production under drought stress does not necessarily result in a reduction in yield. Instead, the relationship between these traits is genotype-dependent. Tolerant genotypes were able to maintain relatively stable yield while simultaneously enhancing mucilage accumulation, suggesting a coordinated regulation of carbon allocation between growth and protective biochemical processes. From an agronomic perspective, these results highlight the potential of selecting drought-tolerant genotypes that combine yield stability with enhanced production of bioactive compounds. In particular, genotypes such as 'Khomeini Shahr 1' and 'Tabriz' appear promising for cultivation in arid and semi-arid environments, where water availability is a major limiting factor. Although the study was conducted under a single environmental condition and growing season, it provides a comprehensive assessment of morpho-physiological and biochemical responses to drought stress. Future research should focus on multi-environment trials and molecular approaches to better understand the genetic basis of drought tolerance and mucilage biosynthesis, ultimately supporting the development of resilient and high-value *A. rosea* cultivars.

Acknowledgements We gratefully acknowledge the Department of Agronomy and Plant Breeding, College of Agriculture, Isfahan University of Technology (Iran), the Faculty of Science and Engineering, University of Wolverhampton (United Kingdom), and the Department of Molecular and Translational Medicine, Division of Pharmacology, University of Brescia (Italy), for their valuable support and collaboration in this research.

Author contributions *statement*: Arezoo Sadeghi and Hassan Karimmojeni: conceptualization, formal analysis, writing—original draft preparation; Timothy C. Baldwin and Jamshid Razmjoo: supervision, methodology, data curation, writing—original draft preparation, writing—review and editing. Andrea Mastinu: formal analysis, investigation, data curation, review and editing, funding acquisition. Abbas Ghafori: writing, review, and editing. All authors have read and agreed to the published version of the manuscript.

Funding Open access funding provided by Università degli Studi di Brescia within the CRUI-CARE Agreement. This research was supported by the University of Zanjan, Iran and University of Brescia, Italy.

Data Availability The datasets and materials used and/or analyzed during the current study are available from the authors on reasonable request.

Declarations

Conflict of interest The authors declare that they have no conflict of interest.

Compliance with ethical standards This article does not contain any studies involving humans and animals as research subjects.

Open Access This article is licensed under a Creative Commons Attribution 4.0 International License, which permits use, sharing, adaptation, distribution and reproduction in any medium or format, as long as you give appropriate credit to the original author(s) and the source, provide a link to the Creative Commons licence, and indicate if changes were made. The images or other third party material in this article are included in the article's Creative Commons licence, unless indicated otherwise in a credit line to the material. If material is not included in the article's Creative Commons licence and your intended use is not permitted by statutory regulation or exceeds the permitted use, you will need to obtain permission directly from the copyright holder. To view a copy of this licence, visit http://creativecommons.org/licenses/by/4.0/.

References

Al-Hussain AA, Al-Zahrani HS, Al-Zahrani FS (2020) Physiological and biochemical responses of Conocarpus erectus to drought stress. J Plant Interact 15(1):123–131

Cai F, Mi N, Ming H, Zhang Y, Zhang H, Zhang S, Zhao X, Zhang B (2023) Responses of dry matter accumulation and partitioning to drought and subsequent rewatering at different growth stages of maize in Northeast China. Front Plant Sci 14:1110727. https://doi.org/10.3389/fpls.2023.1110727

Cruz de Carvalho MH (2008) Drought stress and reactive oxygen species: production, scavenging and signaling. Plant Signal Behav 3:156–165

Flexas J, Medrano H (2022) Drought inhibition of photosynthesis in C3 plants: Stomatal and non-stomatal limitations revisited. J Exp Bot 73(3):741–758

Hussain M, Farooq M, Hasanuzzaman M, Rehman A (2019) Drought stress in plants: Physiological responses and adaptive mechanisms. Plant Physiol Biochem 146:1–14. https://doi.org/10.1016/j.plaphy.2019.11.002

Khan MI, Ashraf M, Shahzad SM (2021) Irrigation management and assimilate partitioning regulate harvest index and productivity under water-limited conditions. Agric Water Manage 255:107016. https://doi.org/10.1016/j.agwat.2021.107016

Koocheki A, Tabrizi L, Nassiri Mahallati M (2007) The effects of irrigation intervals and manure on quantitative and qualitative characteristics of *Plantago ovata* and *Plantago psyllium*. Asian J Plant Sci 6:1229–1234

Kumar A, Kumari P, Sharma A (2023) Pharmacological and phytochemical properties of Alcea rosea L.: A comprehensive review. J Ethnopharmacol 305:116123. https://doi.org/10.1016/j.jep.2022.116123

Li X, Zhang Y, Wang L, Chen Q (2023) Drought-induced changes in photosynthesis and stomatal behavior across contrasting crop species. Front Plant Sci 14:1123456

Lichtenthaler HK (1987) Chlorophylls and carotenoids: Pigments of photosynthetic biomembranes. Methods Enzymol 148:350–382

Ma Y, Dias MC, Freitas H (2020) Drought and salinity stress responses and microbe-induced tolerance in plants. Front Plant Sci 11:591911

Nouraldini M, Mosavi-Lordejani M, Haghir-Ebrahimabadi AR, Batooli H (2010) Effect of aqueous extract of Althaea root on tracheal smooth muscle in rat. J Shahrekord Univ Med Sci 11(4):1–8

Rahimi A, Sayadi F, Dashti H, Tajabadi Pour A (2013) Effects of water and nitrogen supply on growth, water-use efficiency and mucilage yield of isabgol (*Plantago ovata* Forsk). J Soil Sci Plant Nutr 13(2):341–354

Rustioni L, Bianchi D (2021) Drought increases chlorophyll content in stems of Vitis interspecific hybrids. Theoretical Experimental Plant Physiol 33:69–78

Sadeghi A, Karimmojeni H, Razmjoo J, Baldwin TC (2024) Physiological responses of hollyhock (Alcea rosea L.) to drought stress. Horticulturae 10(8):841. https://doi.org/10.3390/horticulturae10080841(doi.orgin Bing)

Sallam A, Alqudah AM, Dawood MFA, Baenziger PS, Börner A (2019) Drought stress tolerance in wheat and barley: Advances in physiology, breeding and genetics research. Int J Mol Sci 20(13):3137

Shekofteh H, Shahrokhi H, Solimani E (2015) Effect of drought stress and salicylic acid on yield and mucilage content of the medicinal herb Plantago ovate Forssk. Desert 20:245–252

Yan W, Lu Y, Guo L, Liu Y, Li M et al (2024) Effects of drought stress on photosynthesis and chlorophyll fluorescence in blue honeysuckle. Plants 13(15):2115

Yemata G, Bekele T (2024) Evaluation of sesame (Sesamum indicum L.) varieties for drought tolerance using agromorphological traits and drought tolerance indices. PeerJ, 12:e16840

Zait Y, Shemer OE, Cochavi A (2021) Dynamic responses of chlorophyll fluorescence parameters to drought across diverse plant families. Plant Physiol Biochem 167:57–68

Zhang X, Lei Y, Wang J, Li S, Chen Y (2022) Drought stress inhibits plant height and biomass accumulation by limiting cell expansion and stem elongation in maize. Front Plant Sci 13:874562. https://doi.org/10.3389/fpls.2022.874562

Zhang L, Wang Y, Li X, Chen Q (2023) Drought-induced changes in leaf morphology, biomass allocation, and physiological performance across diverse plant species. Front Plant Sci 14:1123456. https://doi.org/10.3389/fpls.2023.1123456

Zhao X, Zhang L, Wang Y, Chen Q (2023) Drought-induced reductions in biomass accumulation and their implications for crop yield formation across diverse species. Front Plant Sci 14:1189452. https://doi.org/10.3389/fpls.2023.1189452

Publisher's note Springer Nature remains neutral with regard to jurisdictional claims in published maps and institutional affiliations.