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Effect of genotypes and foliar spray of methyl jasmonate and salicylic acid on andrographolide yield in *Andrographis paniculata* (Burm. f.) Wall. ex Nees. under semi-arid climate

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Abstract

Background *Andrographis paniculata* (Burm. f.) Wall. ex Nees. is an important medicinal plant grown in tropical and sub-tropical regions where semi-arid condition restricts economically viable cultivation. Foliar exogenous application of methyl jasmonate (MeJ) and salicylic acid (SA) was tested for their effectiveness in preventing yield loss in five different morphotypes of *A. paniculata* subjected to deficit soil moisture stress imposed during 90–140 days after transplanting.

Results Soil moisture content below 6% reduced chlorophyll and carotenoid content and upregulated antioxidant enzymes activity. The dry herbage yield was adversely affected by deficit soil moisture stress causing a 14% decline. The andrographolide concentration ranged between 1.40% and 1.54%. Though andrographolide concentration increased by 6%, andrographolide yield declined by 8.21% due to soil moisture stress. Moderately high doses of MeJ and SA reduced chlorophyll and carotenoid content and upregulated antioxidant enzymes activity, however, failed to prevent the loss in dry herbage yield or total andrographolide yield in any morphotype.

Conclusions Foliar application of MeJ and SA do not warrant any protection against stress induced yield loss in field grown *A. paniculata*. However, morphotype AP 13 (round canopy, open-type branch, long narrow leaf) and AP 35 (columnar canopy, closed-type branch, long broad leaf) for having comparatively high herbage yield remained at a better position for total andrographolide yield under deficit soil moisture stress in the semi-arid climate.

Keywords Andrographolide concentration, Antioxidant enzymes, Deficit soil moisture, Dry herbage yield, Leaf water potential

Background

Andrographis paniculata (Burm. f.) Wall. ex Nees. is one such medicinal plant which has gained considerable and progressive interest for decades not only in India but also worldwide. Because of its intensely bitter taste, it is commonly addressed as the "King of the bitters". In India, it is known as "*Kalmegh*". The native of this plant is India and Sri Lanka. It's distribution is

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broadly reported in Bangladesh, China, Hong Kong, Indonesia, Malaysia, Myanmar, Philippines and Thailand covering the Southern and South Eastern Asian regions (Hossain et al. 2016). This erect branched and annual flowering plant belongs to the *Andrographis* genus under the *Acantharea* family (Hossain et al. 2016). Traditionally, it is used in the form of powder, infusion, or decoction either alone or in combination with other medicinal plants. It is used for the treatment of diseases and alignment which includes alopecia, boils, chronic and seasonal fevers, cough, diabetes, dysentery, dyspepsia, enteritis, general debility, gonorrhoea, griping, helminthiasis, hemopathy, herpes, irregular bowel habits, jaundice, leprosy, liver complaints, loss of appetite, malaria, oedema, peptic ulcer, respiratory tract infections, scabies, skin eruptions and as a topical use in skin infections and snake-bites (Anjum et al. 2011).

As per reports, there are 78 ent-labdane diterpenoids, 41 flavonoids, eight quinic acid derivatives, four xanthones, five rare noriridoids, three steroids and three other compounds identified as secondary metabolites from *Andrographis paniculata*. The andrographolide, neoandrographolide and dehydroandrographolide; the lead bioactive compounds belonging to diterpene lactone are present in the whole parts of the herb and the leaf has a greater content of the said lead bioactive compounds (Sharma et al. 2018). At 110–130 days of cultivation, the andrographolides content is reported to be at the maximum level. Its growth and developments prefer moist and shady places, forests and wastelands. It is sensitive to abiotic stress like water deficit stress (Kalariya et al 2021) and salt stress (Hossain 2020) that adversely affects plant growth and drastically reduces crop productivity. Therefore, the plant part used, the harvest date and season, as well as climatic conditions affect both the qualitative and quantitative production of this crop.

By triggering a set of key physiological and biochemical processes, drought stress, more specifically the deficit soil moisture availability adversely affects plant growth and development by changing plant morphology (Jaleel et al. 2008). The impact at the cellular level includes changes in proline and other osmolyte content, leaf chlorophyll content, relative water content, chloroplast integrity and cell membrane stability. The generation of reactive oxygen species (ROS) causes damage to biomolecules by enhancing lipid peroxidation (Gao et al. 2018) degradation of protein and damage to the nucleic acid and thereby imposes limit on plant growth under drought stress (Jain et al. 2019). The physiological impacts of deficit soil moisture stress include reduced gaseous exchange capacity along with a downfall in the efficiency of photosystem I by interrupting the electron transport chain ultimately affecting compromised biomass gain (Kalariya et al. 2019).

In a plant, the toxicity of ROS can be mitigated through non-enzymatic and enzymatic ways (Gill and Tuteja 2010). The major components of non-enzymatic antioxidants are ascorbate (AsA), glutathione (GSH), tocopherol, flavanones, carotenoids and anthocyanins (Hasanuzzaman et al. 2019). The enzymatic antioxidant defence mechanism depends on the expression of some specific enzymes. This includes superoxide dismutase (SOD), ascorbate peroxidase (APX), glutathione peroxidase (GPX), catalase (CAT), monodehydroascorbate reductase (MDHAR), dehydroascorbate reductase (DHAR), glutathione reductase (GR) and glutathione s-transferase (GST) (Rajput et al. 2021).

Several researchers have studied the exogenous application of cellular protectants as a signalling molecule to enhance abiotic stress tolerance (Mir et al. 2018). Methyl jasmonate (MeJ) and salicylic acid (SA) both are preferred regulatory phytohormones known for their crucial role in plant signalling responses to environmental conditions in initiating defence mechanisms (Mir et al. 2018). In several plants, these two hormones have been shown to protect plants against abiotic stresses (Mohi-Ud-Din et al. 2021) and have been suggested at very low doses (5–20 µM MeJA and/or 0.5–2 mM SA) to effectively enhances the defence signalling routes to mitigate damage due to the abiotic stresses (Siboza and Bertling 2013).

Currently, limited information on the effect of soil water deficit stress on *A. paniculata* herbage yield and andrographolide yield is available. For example, under poly house conditions, foliar application of SA (200 mg L^{-1}) to a prolonged water deficit stressed (during 20–80 DAT) plants had been sown to up-regulate antioxidant defence mechanism and helped to manage photosynthetic efficiency in *A. paniculata* (Kalariya et al. 2018). In another study in the field aimed at the screening of *A. paniculata* genotypes for rain-fed conditions, it was revealed that dry herbage yield was reduced by nearly 30% along with a reduction in andrographolide yield with variable response among different genotypes due to soil moisture deficit stress (Kalariya et al. 2021). Under fully controlled conditions, a low dose of (5 µM) MeJA highly influenced the coordination of ISPH, GGPS and HGMS gene expression and yielded 5.25 times more andrographolide content after 24 h of treatment in cell cultures of *A. paniculata*. This has shown the involvement of MeJA in andrographolide biosynthesis (Sharma et al. 2015). This study examined the effectiveness of foliar application of MeJA and SA at moderately high doses on five different morphotypes in reducing andrographolide yield losses due to deficit soil moisture stress.

Methods

The experimental site and soil properties

The experimental field (latitude 07° 15′ N, longitude 78° 14′ E.) is located at the farm of the ICAR- Directorate of Medicinal and Medicinal and Aromatic Plants Research (DMAPR), Anand in Gujarat, India. Field experiments were conducted during *Kharif* season in two consecutive crop years 2019 and 2020. Sandy loam soil (pH 6.5–7.5) of the experimental plots holds nearly 23% water at field capacity. The field experiment was laid out in a split-plot design with three replications keeping genotype as the main treatment, water deficit stress as sub-treatment and foliar application of hormones as sub-sub treatment.

Plant materials, nursery and transplanting

Five genotypes, i.e. AP 2, AP 13, AP 24, AP 18 and AP 35 selected based on distinct morphological traits as per DUS descriptors (https://plantauthority.gov.in/crop-dus-guidelines) were used for the present investigation. The canopy shape of AP 13 is round and globular with narrow, long light green leaf lamina and open-type branch arrangement. AP 18 has a columnar canopy with a long broad light green leaf lamina with a closed branch arrangement. AP 02 and AP 24 are with pyramidal canopies with open branching patterns; however, AP 2 has long, broad green leaf lamina and AP 24 has short broad dark green leaf lamina. AP 35 has a columnar canopy with a long broad green leaf lamina with a closed branching pattern. Seeds were sown in the nursery to prepare seedlings for transplanting in the field. Transplanting of seedlings of these five varied morphotypes at the age of 45–50 days was carried out at 45 cm × 10 cm distance in 4 × 5 m sized plots.

Allocation of treatments

After receiving 58 mm rains during the 43rd standard week in crop year I (2019), i.e. at the crop age of 78 DAT, the irrigation was stopped to create deficit soil moisture stress which was rehydrated after 140 DAT through applying irrigation. In crop year II (2020), after receiving 10.2 mm rains in the 42nd standard week, the crop was raised with supporting irrigation till 75 DAT and then the irrigation was stopped for the next 60 days period to create deficit soil moisture stress which was rehydrated at the age of 140 DAT through applying irrigation. In both years, after resuming the stress condition, the crop was given irrigation at 10–12 days intervals till maturity harvesting at 160 DAT. Deficit soil moisture-stressed plants as well as control plants were treated with foliar application of 200 µM MeJ and 2.0 mM SA on the 15th, 30th and 45th day of stress condition. Thus, all five morphotypes were tested under six sets of treatment combination: (a) control (b) control + MeJ (c) control + SA (d) stress (e) stress + MeJ, (f) stress + SA. Sigma makes MeJ 95% was used to prepare a 200 µM working solution of MeJ and Loba Chemie make SA was used to prepare a 2.0 mM working solution of SA foliar application. The treated control plants were mock-treated with an equal amount of water spray. Physiological as well as biochemical parameters were recorded at 135–140 DAT and growth parameters were recorded at 155–160 DAT for two consecutive years (2019 and 2020). All the plots were kept weed free by three hand weddings and were supplemented with standard N: P: K doses at the rate of 80:40:40 kg ha^{-1}.

Weather parameters

Daily weather parameters were collected from meteorological observatory of Department of Meteorology, Anand Agricultural University, Anand. The mean of the standard meteorological week was calculated. The mean weather data of standard week coinciding crop growth period was considered as weather parameters at Anand location during crop season in year 2019 and 2020 is presented in Table 1.

Soil moisture content (SMC), relative water content (RWC %), leaf water potential and physiological parameters

Soil moisture content (SMC %) at 5–10 cm depth was measured periodically based on the gravimetric method. The relative water content (RWC) was calculated from the ratio of the difference between the fresh weight and dry weight to the difference between dry weight and turgid weight and multiplied by 100. Leaf water potential was measured as per standard methods (Kalariya et al. 2021) using a psychrometer (Wescor Inc, USA).

Pigment estimation and assays of activities of antioxidant enzymes

Chlorophyll *a* and *b* content and total carotenoid content and antioxidant enzymes activity were measured at 140 DAT. Chlorophyll content and total carotenoid from leaves were measured in the 80% acetone method (Arnon 1949) at 125 DAT. Extraction of antioxidant enzymes namely the SOD, CAT, APX and GPX enzymes was done from fresh leaf tissues with the help of a pre-chilled mortar and pestle. To 500 mg of properly homogenized fresh leaf tissues, 3 ml extraction buffer containing 1 mM EDTA and 1% (W/V) polyvinylpyrrolidone (PVP) in 50 mM sodium phosphate (pH 7.4) was added. These homogenates were centrifuged at 10,000 rpm for 20 min, and the supernatant was used for the assay. Inhibition in the photochemical reduction in nitrobluetetrazoilum (NBT) at 560 nm of the SOD

Table 1 Weather parameters at Anand location during crop season in year 2019 and 2020

Standard Week	ET_0 (mm)		Sunshine (hrs.)		Rain (mm)		Wind speed (Km/hr)		Temperature (°C)									
	Crop year		Crop year		Crop year		Crop year		Crop year I			Crop year II			Crop year I		Crop year II	
	I	II	I	II	I	II	I	II	T_{min} (C)	T_{max} (C)	T_{avg} (C)	T_{min} (C)	T_{max} (C)	T_{avg} (C)	I	II	I	II
33	3.5	1.6	3.0	0.1	9.2	371.0	5.6	6.2	31.5	25.1	28.3	28.1	25.4	26.8	91	71	97	90
34	5.7	2.4	7.0	1.6	0.0	98.0	5.9	6.3	33.4	25.0	29.2	29.9	25.9	27.9	89	63	93	86
35	2.8	3.3	3.5	5.7	162.6	23.4	4.6	5.8	30.9	24.8	27.9	31.4	25.6	28.5	94	77	94	72
36	3.4	3.8	3.7	6.6	56.4	2.0	3.5	3.3	32.9	26.0	29.5	33.9	26.6	30.3	95	74	89	65
37	1.3	3.9	0.1	4.8	35.4	28.0	4.2	3.6	29.9	25.8	27.9	33.8	26.5	30.2	96	86	90	66
38	2.5	3.2	2.6	4.2	79.8	36.0	2.9	4.1	32.2	25.3	28.8	33.6	25.9	29.8	95	80	91	73
39	2.8	3.0	3.5	4.5	72.0	3.2	3.8	4.0	31.4	25.1	28.3	32.1	25.3	28.7	93	79	91	66
40	3.7	4.2	6.4	8.8	60.4	13.2	4.1	4.6	32.1	24.9	28.5	33.9	24.9	29.4	93	63	87	52
41	4.4	4.5	8.9	8.0	0.0	0.0	2.1	2.3	34.6	22.7	28.7	35.9	23	29.5	87	44	77	39
42	4.0	4.3	8.2	7.0	0.0	10.4	2.2	3.7	34.5	22.2	28.4	35.1	26.9	31.0	88	44	88	57
43	3.6	4.4	4.7	9.5	0.4	0.0	4.7	2.2	31.5	21.7	26.6	35.3	20.3	27.8	83	57	80	30
44	2.7	4.0	5.3	9.1	58.6	0.0	2.4	2.4	32.5	23.7	28.1	34.0	17.6	25.8	90	66	80	30
45	3.4	3.4	6.4	9.3	4.4	0.0	2.2	1.9	32.1	22.4	27.3	33.4	15.8	24.6	89	61	80	30
46	3.1	3.1	8.1	8.5	0.0	0.0	1.9	2.6	32.1	19.7	25.9	32.6	19.1	25.9	90	45	79	40
47	2.6	3.7	7.3	8.7	0.0	0.0	1.5	3.1	31.4	17.8	24.6	30.4	15.2	22.8	91	46	81	39
48	2.4	3.6	6.7	8.4	0.0	0.0	2.4	4.3	31.2	19.6	25.4	30.4	17.6	24.0	91	54	77	42
49	3.4	2.8	5.3	9.5	0.0	0.0	5.1	1.5	28.2	18.1	23.2	32.7	15.1	23.9	73	51	88	36
50	2.5	2.0	8.7	4.8	0.0	16.4	2.8	2.3	28.2	13.6	20.9	27.4	17.9	22.7	87	46	94	62
51	2.5	2.7	8.0	8.8	0.0	0.0	4.0	3.7	27.2	15.5	21.4	26.6	12.6	19.6	83	56	82	44
52	2.7	3.1	6.1	9.2	0.0	0.0	3.4	3.7	22.5	11.2	16.9	26.8	12.3	19.6	70	38	74	37
1	2.4	2.6	6.7	4.1	0.0	0.0	3.3	3.4	25.5	12.7	19.1	25.7	14.9	20.3	89	48	77	48
2	3.2	2.9	8.0	5.6	0.0	0.0	4.6	3.9	26.6	13.8	20.2	27.5	16.1	21.8	80	44	89	59
3	2.9	2.8	8.9	8.6	0.0	0.0	3.4	1.8	24.6	9.7	17.2	29.2	13.6	21.4	87	48	91	44
4	3.2	3.3	9.0	9.4	0.0	0.0	3.0	3.1	27.8	13.0	20.4	26.9	11.1	19.0	84	44	81	38

ET_0 (mm)-evapotranspiration, T_{min} (°C) Minimum temperature, T_{max} (°C) Maximum temperature, T_{avg} (°C) Average temperature, RH_{avg} (%)-average relative humidity

(EC 1.15.1.1) was measured as the activity of SOD spectrophotometrically. The reaction mixture contained 13 mM methionine, 75 µM NBT, 2 µM riboflavin, 0.1 mM EDTA in 50 mM sodium phosphate buffer (pH 7.8). To 2.9 ml of the reaction mixture, 0.1 ml of the enzyme was added to start the reaction n under photochemical light. The quantity of enzyme required to inhibit the reduction in NBT by 50% in a reaction mixture was considered as one unit of SOD and enzyme unit of SOD was calculated (Adnan Nurhan et al. 2021).

According to the method of Aebi (1984) by measuring the decrease in absorption at 240 nm as H_2O_2 ($e = 39.4\ mM^{-1}\ cm^{-1}$) consumed by the 0.1 ml extract in 3 ml reaction mixture, total catalase (EC 1.11.1.6) enzyme activity was expressed as µ mol H_2O_2 oxidized $min^{-1}\ g^{-1}$ protein. Immediately in fresh extract, the APX (EC 1.11.1.11) activity was measured as described by (Adnan Nurhan et al. 2021). The method given by (Lowry et al. 1951) using folin-phenol reagent the protein concentration of enzymes extract was determined.

Harvesting and estimation of andrographolide content

Growth parameters

Observations on growth parameters were recorded at harvest maturity (150–160 DAT). To avoid errors in estimating biomass parameters caused by potential fluctuations of the moisture content of the plant material, a parallel determination of the dry matter content through drying at 105 °C in a hot air oven was performed to get a constant weight. A representative sample of the same plant material was kept on air drying to enable us to relate the results to dry matter without the detrimental effect of the high drying temperature on the andrographolide compound. These air-dried herbage samples

were finely powdered and used for the estimation of andrographolide content.

Determination of andrographolide content

To determine the andrographolide content, experimental samples were shade-dried, powdered and extracted in methanol. A modular HPLC (Shimadzu Corporation, Kyoto, Japan), consisting of two LC-20AD pumps, SPD-20A UV–Vis Detector, DGU-20A3 degasser, SIL-20AC HT autosampler, a CTO-10ASvp column oven, CBM-20 communications bus module was used for chromatographic separation of analytes on a Merck Rp-18 (250×4.6 mm, 5 um). The mobile phase consisted of methanol and water (65:35, v/v), and was delivered at a flow rate of 1.0 mL min-1 and the absorbance was set at 229 nm in a UV–Vis detector (Rajani et al. 2000). The column temperature was maintained at 40 ^{0}C for better resolution and the sample injection volume was kept at 10 µL.

Statistical analysis of data

For statistical analysis, the Microsoft (MS) office excel based add-in tool DSAASTAT programme was used and the data of both the crop years were subjected to a pooled analysis of variance appropriate to the experimental design keeping fixed effects under multi-factor analysis. Duncan's Multiple Range Test (DMRT) comparisons among the treatment combinations were performed at the $p < 0.05$ level. The correlation between relative leaf water content, leaf water potential, superoxide dismutase activity, guaiacol peroxidase activity, catalase activity, ascorbate peroxidase activity, chlorophyll a content, chlorophyll b content, total chlorophyll content, carotenoids, plant height, dry herbage yield, andrographolide concentration, and andrographolide yield was assessed by computing their linear relationships through Pearson's correlation coefficient. An interactive pairwise heat map based on Pearson's correlation coefficient value was prepared through an online module online http://www.heatmapper.ca/.

Results

Weather conditions during crop seasons

Weather parameters during the crop period revealed that there was evaporative demand of 22.6 mm and 23.4 mm in the crop year 2019 and 2020, respectively, during the stress period. Bright sunshine hours were 56.6 and 59.1 during the stress period in the crop years 2019 and 2020, respectively. The maximum temperature remained at 29.1 °C and 28.4 °C during the stress period in the crop year 2019 and 2020, respectively. The periodic measurement of soil moisture revealed that by stopping the irrigation, the soil moisture was depleted continuously and reached less than 6.0% in 50 days stress period (Fig. 1).

Soil water plant relationship

The mean of the leaf RWC was 87%, ranging between 86.6% and 87.6 with no significant variation among different morphotypes of *A. paniculata*. The RWC was significantly reduced due to soil moisture stress conditions. The soil moisture-stressed plants had a mean RWC below 81.6% whereas the control plants raised

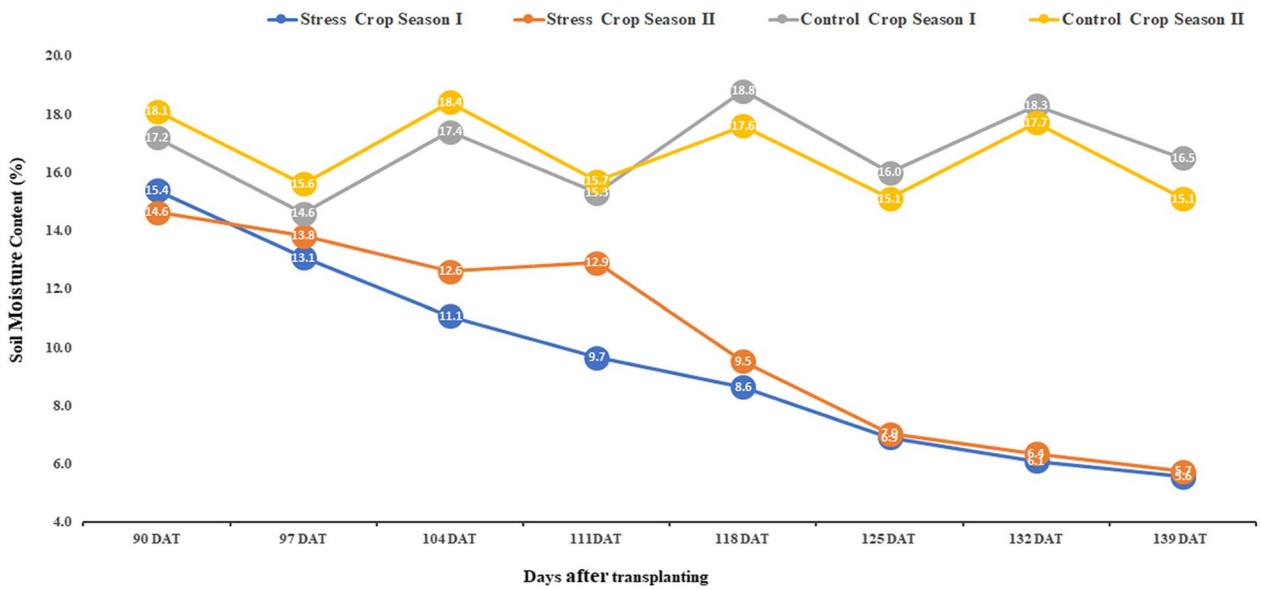

Fig. 1 Periodic soil moisture content (5–10 cm depth) of the experimental plots during 90–140 DAT of *Andrographis paniculata*

Table 2 Relative leaf water content, leaf water potential and activity of antioxidant enzymes in leaves of *Andrographis paniculata* at 140 DAT

Morphotype	Treatment	RWC (%)		LWP (MPa)		SOD U		GPOX uM/min/g		CAT µM/min/g		APX uM/min/g	
AP13	C	92.3	ab	−0.61	a	37.8	ijk	27.6	e	36.0	ghijk	132	lm
	C + MeJ	92.4	ab	−0.57	a	42.3	ghi	55.0	abcd	36.0	ghijk	149	ijkl
	C + SA	92.2	ab	−0.53	a	42.1	ghij	54.3	abcd	43.4	ghi	178	cdefg
	S	80.9	e	−1.36	ef	51.8	bcde	72.0	a	81.5	ab	191	abcde
	S + MeJ	81.1	e	−1.39	f	59.1	a	68.4	a	84.0	a	215	a
	S + SA	80.9	e	−1.27	def	59.8	a	59.3	abc	78.3	abcd	205	ab
AP18	C	93.0	a	−0.52	a	29.7	lm	21.2	e	34.8	hijk	108	mn
	C + MeJ	92.8	a	−0.52	a	35.3	jkl	39.8	cde	40.1	ghijk	147	jkl
	C + SA	92.8	a	−0.56	a	34.1	kl	37.5	de	41.3	ghijk	147	jkl
	S	81.9	de	−1.17	bcd	49.4	def	54.6	abcd	47.5	g	167	defghij
	S + MeJ	81.5	de	−1.23	cdef	49.5	def	60.7	abc	60.3	f	169	defghij
	S + SA	81.9	de	−1.07	b	53.7	abcd	58.0	abcd	67.8	def	159	ghijk
AP2	C	92.2	ab	−0.49	a	25.3	m	24.7	e	32.0	ijk	103	n
	C + MeJ	93.4	a	−0.59	a	38.8	hijk	60.1	abc	38.3	ghijk	160	fghij
	C + SA	93.1	a	−0.57	a	41.0	ghij	61.6	ab	42.2	ghij	114	mn
	S	82.4	d	−1.15	bcd	55.3	abcd	73.6	a	61.8	ef	177	cdefgh
	S + MeJ	80.7	e	−1.23	cdef	49.9	def	60.3	abc	80.3	abc	193	abcd
	S + SA	81.2	de	−1.20	bcde	56.0	abcd	28.8	e	59.8	f	207	ab
AP24	C	92.3	ab	−0.54	a	24.4	m	37.8	de	29.4	k	112	mn
	C + MeJ	91.4	b	−0.54	a	36.4	ijk	40.8	bcde	30.5	jk	134	klm
	C + SA	92.3	ab	−0.56	a	35.5	ijkl	57.7	abcd	41.2	ghijk	131	lmn
	S	81.9	de	−1.23	cdef	39.1	hijk	74.0	a	38.7	ghijk	176	defghi
	S + MeJ	81.4	de	−1.28	def	46.5	efg	71.7	a	45.7	gh	181	bcdefg
	S + SA	80.8	e	−1.24	cdef	51.5	cdef	66.9	a	71.6	bcde	203	abc
AP35	C	92.9	a	−0.53	a	41.6	ghij	40.2	cde	34.6	hijk	150	hijkl
	C + MeJ	92.9	a	−0.52	a	44.9	fgh	62.1	a	35.9	ghijk	165	efghij
	C + SA	92.3	ab	−0.51	a	41.2	ghij	60.1	abc	41.6	ghijk	168	defghij
	S	81.9	de	−1.28	def	58.3	ab	64.0	a	69.5	cdef	187	bcdef
	S + MeJ	81.8	de	−1.06	b	57.4	abc	68.5	a	82.5	ab	186	bcdefg
	S + SA	83.6	c	−1.08	bc	50.1	def	66.5	a	79.4	abc	159	ghijk

C Control, *S* Stressed, *MeJ* Methyl jasmonate, *SA* Salicylic acid, *RWC* Relative leaf water content, *LWP* Leaf water potential, *SOD* Superoxide dismutase, *GPOX*-guaiacol peroxidase, *CAT*-catalase and *APX*-ascorbate peroxiadase, means with the same letter in the columns do not showing significantly different ($P = 0.05$)—(Duncan Multiple Range Test)

at more than 15% SMC had a mean leaf RWC above 92.5% across the morphotypes (Table 2). The reduction in RWC content was nearly 12% under soil moisture stress conditions as compared to the control plants. The foliar application of MeJ and SA had no effect on leaf RWC content under control as well as soil moisture stress condition. The mean leaf water potential (LWP) was −0.88 Mpa. The LWP was minimum in AP 13 (−0.96 Mpa) and maximum in AP 35 (−0.83 Mpa). LWP was adversely affected by declined soil moisture content (Table 2). It was −0.54 MPa in control plants and decreased to −1.21 accounting a 124% decline due to lesser availability of soil moisture content. There was no effect of foliar application of MeJ and succinic acid on LWP either alone or in a combination of stress.

Antioxidant enzymes activities

The mean value for SOD activity was 47.0 U g protein^{-1} (Table 2). Among the genotypes, AP 35 and AP 13 had the maximum SOD activity. There was a significant variation in SOD activity concerning the stress condition. The SOD activity increased by 43% in soil moisture-stressed plants as compared to that of the control. The SOD activity in plants sprayed with SA and MeJ was upregulated by more than 10% of the control plants. The mean GPOX activity was 54.3 µM min^{-1} g^{-1}. GPOX activity ranged

between 45.3 μM min^{-1} $g^{-1}$to 60.2 μM min^{-1} $g^{-1}$with no significant variation among different morphotypes. There was a significant effect of treatment as the deficit soil moisture stress had resulted in 39% increase in GPOX activity as compared to the control pants. The GPOX activity was higher in MeJ, and SA-treated plants by 20% and 12%, respectively, as compared to the control plants.

The mean value for the CAT activity was 52.2 μM min^{-1} g^{-1} ranging between 42.9 μM min^{-1} g^{-1} and 59.7 μM min^{-1} g^{-1} among different morphotypes. The CAT activity was increased by 81% under deficit soil moisture stress conditions as compared to the control plants. The MeJ and SA had raised CAT activity by 15% and 22%, respectively, compared to that of the control plants. The APOX activity was maximum in AP 13 (178 μM min^{-1} g^{-1}) and minimum in AP 18 (149 μM min^{-1} g^{-1}). Under deficit soil moisture stress conditions, the APOX activity was upregulated by 32%. The MeJ and SA-treated plants had 10% higher activity of APOX.

Photosyntheic pigment content

Mean values of chlorophyll a and chlorophyll b, total chlorophyll content and carotenoids contents were 1.32 mg g^{-1} FW and 0.43 mg g^{-1} FW, 1.84 mg g^{-1} FW and 5.6 mg g^{-1} FW, respectively, (Table 3). There was a variation in pigment content under control as well as deficit soil moisture stress condition. There was a decline in chlorophyll *a* and total chlorophyll content by 4.5 and 4.6%, respectively, by stress condition. Among genotypes,

Table 3 Photosynthetic pigments content in leaves of *Andrographis paniculata* at 140 DAT

Morphotype	Treatment	Chl. *a* (mg g^{-1} FW)		Chl. *b* (mg g^{-1} FW)		Total Chl. (mg g^{-1} FW)		Carot. (mg g^{-1} FW)	
AP13	C	2.07	a	0.48	bcdefg	2.55	a	6.2	bcde
	C + MeJ	1.87	b	0.51	bcde	2.37	ab	4.7	ijk
	C + SA	1.64	c	0.65	a	2.29	b	4.7	ijk
	S	1.85	b	0.59	ab	2.44	ab	5.1	fghij
	S + MeJ	1.80	b	0.48	bcdefg	2.28	b	4.9	hijk
	S + SA	1.66	c	0.43	cdefgh	2.09	c	4.2	jk
AP18	C	1.34	e	0.37	defgh	1.71	defghi	5.0	ghijk
	C + MeJ	1.27	efg	0.38	defgh	1.65	efghijk	4.8	ijk
	C + SA	1.26	efgh	0.36	fgh	1.62	efghijk	4.1	k
	S	1.13	ghij	0.32	h	1.45	k	5.7	defgh
	S + MeJ	1.07	j	0.43	cdefgh	1.50	ijk	5.0	ghijk
	S + SA	1.06	j	0.45	bcdefgh	1.51	hijk	4.2	jk
AP2	C	1.28	ef	0.43	cdefgh	1.71	defghi	7.0	ab
	C + MeJ	1.06	j	0.52	bcd	1.58	fghijk	7.0	ab
	C + SA	1.07	j	0.42	cdefgh	1.49	jk	5.9	defg
	S	1.22	efghi	0.33	gh	1.55	ghijk	6.2	bcde
	S + MeJ	1.12	hij	0.37	defgh	1.49	jk	5.9	cdef
	S + SA	1.08	ij	0.36	efgh	1.44	k	6.3	abcde
AP24	C	1.50	d	0.37	defgh	1.87	d	6.2	bcde
	C + MeJ	1.24	efgh	0.50	bcdef	1.74	defg	4.2	jk
	C + SA	1.16	fghij	0.32	h	1.48	jk	4.6	jk
	S	1.28	ef	0.44	cdefgh	1.72	defgh	6.0	cde
	S + MeJ	1.16	fghij	0.36	fgh	1.52	hijk	4.8	hijk
	S + SA	1.14	ghij	0.44	cdefgh	1.58	fghijk	5.0	fghijk
AP35	C	1.30	ef	0.38	defgh	1.68	defghij	7.2	a
	C + MeJ	1.21	efghi	0.34	gh	1.55	ghijk	7.0	ab
	C + SA	1.25	efgh	0.54	abc	1.79	de	6.8	abc
	S	1.35	e	0.43	cdefgh	1.77	def	6.6	abcd
	S + MeJ	1.17	fghij	0.42	cdefgh	1.59	efghijk	6.4	abcde
	S + SA	1.09	ij	0.39	defgh	1.47	jk	5.5	efghi

C Control, *S* Stressed, *MeJ* Methyl jasmonate, *SA* Salicylic acid, *Chl. A* Chlorophyll a, *Chl. b* Chlorophyll b, *Total Chl*. Total Chlorophyll and *Carot*.-carotenoids, means with the same letter in the columns do not showing significantly different ($P = 0.05$)— (Duncan Multiple Range Test)

AP 13 had the highest chlorophyll content and AP 02 had the highest carotenoid contents. Foliar application of MeJ and SA had decreased chlorophyll *a* content by 11 and 12%, respectively, whereas the decline in total chlorophyll content by MeJ and SA was 7.7 and 8.0%, respectively. Foliar application of MeJ and SA reduced the carotenoid content by 12 and 14%, respectively.

Growth parameters

The mean plant height of all five morphotypes was 55.0 cm. AP 2 had the maximum plant height whereas AP 24 had the lowest plant height (Table 4). There was significant variation among the deficit soil moisture-stressed plants (52.5 cm) and the control plant (57.5 cm) for the plant height. Foliar application of MeJ, and SA imparted no distinguishable impact on plant height. The mean of the dry herbage yield of all five morphotypes across the years was 45.9 g plant^{-1}. Among the morphotypes, AP 35 had a significantly high herbage yield (56.3 g plant^{-1}) whereas AP 2 had the lowest (37.2 g plant^{-1}) dry herbage yield (Table 4). Results showed that the dry herbage yield was adversely affected by deficit soil moisture stress conditions accounting for a nearly 14% decline in herbage yield. Application of MeJ and SA did not show any effect on the dry herbage yield of any morphotypes.

Andrographolide content

The mean value for the andrographolide concentration in herbage was 1.50% on a dry weight basis. There was a significant variation among the different morphotypes

Table 4 Plant height, dry herbage yield, andrographolide concentration and andrographolide yield in leaves of *Andrographis paniculata* at 140 DAT

Morphotype	Treatment	Plant height (cm)		Dry herbage yield (g plant^{-1})		Andrographolide concentration (%)		Andrographolide yield (mg Plant^{-1})	
AP13	C	58.2	bcde	58.0	a	1.49	abcdefg	0.861	a
	C + MeJ	59.3	abcd	57.6	a	1.50	abcdefg	0.864	a
	C + SA	59.2	abcd	57.3	a	1.49	abcdefg	0.850	ab
	S	57.1	defg	49.5	bc	1.57	abcd	0.776	abc
	S + MeJ	52.8	hijk	48.9	cd	1.59	abc	0.778	abc
	S + SA	54.2	ghij	47.1	cde	1.59	abc	0.748	bc
AP18	C	60.5	abc	48.2	cd	1.32	g	0.633	de
	C + MeJ	61.0	ab	47.2	cde	1.33	g	0.627	de
	C + SA	58.9	abcde	44.3	e	1.37	efg	0.604	de
	S	59.5	abcd	40.2	fg	1.53	abcdef	0.613	de
	S + MeJ	53.8	ghij	39.6	gh	1.39	defg	0.548	e
	S + SA	55.7	efgh	37.9	ghij	1.47	abcdefg	0.556	e
AP2	C	62.0	a	38.4	ghij	1.41	cdefg	0.539	e
	C + MeJ	61.9	a	39.1	ghi	1.60	abc	0.623	de
	C + SA	61.4	ab	38.9	ghij	1.56	abcde	0.606	de
	S	54.9	fghi	36.4	ghij	1.47	abcdefg	0.535	e
	S + MeJ	54.5	fghij	34.6	j	1.62	ab	0.560	e
	S + SA	56.2	defg	35.7	hij	1.61	ab	0.575	e
AP24	C	48.0	mno	43.6	ef	1.61	ab	0.700	cd
	C + MeJ	49.8	klm	45.5	de	1.36	fg	0.618	de
	C + SA	49.4	lmn	43.5	ef	1.44	bcdefg	0.624	de
	S	46.5	no	38.1	ghij	1.48	abcdefg	0.564	e
	S + MeJ	45.5	o	35.0	ij	1.53	abcdef	0.536	e
	S + SA	41.6	p	36.3	ghij	1.66	a	0.603	de
AP35	C	58.8	abcde	59.7	a	1.48	abcdefg	0.880	a
	C + MeJ	57.5	cdef	59.9	a	1.43	bcdefg	0.853	ab
	C + SA	56.7	defg	59.9	a	1.41	cdefg	0.841	ab
	S	51.7	ijkl	52.7	b	1.54	abcdef	0.811	ab
	S + MeJ	51.3	jkl	52.9	b	1.53	abcdef	0.809	ab
	S + SA	52.8	hijk	53.0	b	1.55	abcdef	0.821	ab

C Control, *S* Stressed, *MeJ* Methyl jasmonate, *SA* salicylic acid, means with the same letter in the columns do not showing significantly different ($P = 0.05$)—(Duncan Multiple Range Test)

of *Andrographis paniculata* for the andrographolide concentration in herbage. The highest andrographolide concentration was in AP18 and AP 2 (1.54%) and the lowest in AP 13 (1.40%) (Table 4). The deficit soil moisture stress condition caused an increase in andrographolide concentration by 6% in comparison to the control plant. The effect of MeJ and SA as a foliar application on andrographolide concentration was non-significant with a negligible increase in the case of SA-treated plants. The mean value of andrographolide yield of all morphotypes was 0.68 mg plant^{-1}. AP 35 topped for having the highest andrographolide yield of 0.84 mg plant^{-1} whereas AP 2 had the lowest andrographolide yield of 0.57 mg plant^{-1}. Deficit soil moisture stress condition declined the andrographolide yield from 0.71 to 0.65 mg plant^{-1} accounting 8.21% reduction. Application of MeJ and SA did not show any significant effect on the dry herbage yield of any morphotypes.

Discussion

In irrigated cropping areas of the world, the water deficit stress is one of the most influential abiotic factors affecting crop yield. The difference between the water absorbed by the roots and lost through transpiration by the leaf is reflected by leaf RWC content. Reduced leaf RWC content showed the soil–plant water relationship was disturbed (Patanè et al. 2022). The reduced RWC is in accordance with the reduced soil moisture content of the soil. When the evaporative loss exceeds the uptake of water amid deficit soil moisture content, reduced RWC is reported (Tian et al. 2022). In response to decreased soil moisture availability, the plant closes its stomata to reduce transpiration. At the same time, the osmoprotectants produced in the cells together with the decreased RWC cause a decline in water potential (Ψ).

Under stress conditions, the overall growth is reduced and hence the leaf area is also affected negatively. To prevent transpiration losses, one of the mechanisms plants employ is to minimize the surface area exposed to the ambient air under stress condition. This can be achieved by reducing specific leaf areas as a kind of protective mechanism. This in turn helps the plants to withstand the adverse impacts of deficit soil moisture availability in plants (Kalariya et al. 2017).

The genotypic difference in chlorophyll and carotenoid content are based on genetic makeup. Under stress conditions, the hydrolysis of chloroplastic proteins causes the degradation of proteins. In our study, the declined chlorophyll content seems to be because of the chlorophyll destruction decreasing leaf pigments during the primary stress stage of degradation of proteins. A decrease in chlorophyll content under water deficit stress has been considered a typical symptom of pigment photo-oxidation and chlorophyll degradation (Sánchez-Reinoso et al. 2019).

Jasmonates in plants can modulate the growth and developmental processes and act as stress hormones that play an important role in plant tolerance to biotic and abiotic stresses. The growth and developmental processes can be modulated by Jasmonates in plants. Such modulation includes changes in the root, shoot and leaf growth, ripening of fruit, senescence of leaf, maturation of pollen along with biosynthesis of secondary metabolites (Jang et al. 2020). Under the stressed condition, the JA plays an important role in plant response to biotic and abiotic stresses (Wang et al. 2020).

The derivative of jasmonic acid, MeJ was first isolated from the flower of *J. grandiflorum*. Other derivatives along with jasmonyl isoleucine (JA-Ile) and oxylipins are collectively called jasmonates (Ali and Baek 2020). To perform the biological role, the MeJ need to be converted to JA by its conjugation to JA-Ile (Stitz et al. 2011).

Observing the trend of important genes of the photosynthesis process namely, ribulose bisphosphate carboxylase/oxygenase, chlorophyll a/b-binding protein and light-harvesting complex II in some crops revealed that JA application had downregulated these genes (Jung et al. 2007). In our case, the reduction in chlorophyll content would have the basis at the gene regulation level (Mohi-Ud-Din et al. 2021).

Salicylic acid (SA) plays important role in many physiological processes. It has a role to play in germination, absorption of ions, carbon fixation and general plant growth. This phytohormone; synthesized in root cells is translocated to other parts (El-Tayeb 2005). The antioxidant content is increased due to the eliciting effects of SA on physiological pathways producing secondary metabolites (Ghasemzadeh et al. 2012). Reduced chlorophyll content due to SA application helps better utilization of absorbed energy through reduced non-regulated energy dissipation. In this way, the amount of triplet excited state chlorophyll molecules available to generate singlet oxygen is reduced. Thus, it suppresses phototoxicity and reduces photoinhibition and photodamage by providing photoprotection to PSII in crop plants. It increases growth-regulating hormones, such as auxins and cytokinins and mitigates the impacts of environmental stresses (Sabagh 2022). It reduces the leakage of ions and accumulation of toxic ions in plants (Zhou et al. 2009). Plants intensify their defence mechanism through the overexpression of ROS-scavenging enzymes to detoxify ROS synthesized during stress. For being ubiquitous enzymes in aerobic organisms, the SOD has to play a key role. The other enzymes involved in this defence mechanism are CAT, POX, and APX (Foyer and Mullineaux 2019).

Increased SOD activity in *Andrographis paniculata* leaves indicated that SOD activity was allied to better protection against WD stress. CAT is the principal enzyme that scavenges harmful oxygen species in plants (Huang et al. 2022) and thus, increased CAT activities under WD stress suggested that it may be responsible for protection against oxidative damage. Many important reactions such as ascorbate oxidation, indoleacetic acid oxidation, lignification, phenol oxidation, pathogen defence and cell wall elongation are found to be associated with the activity of a large family of important plant enzymes; the peroxidases (Urs et al. 2006).

APX scavenges H_2O_2 and uses ascorbate as an electron donor in plants. The increase in APX activity in leaves is a response to the enhanced production of ROS and particularly H_2O_2 under water deficit stress. Local as well as systemic defence responses are conveyed by salicylic acid through a regulatory signal mediating plant response to water deficit stress (Munné and Peñuelas 2003) and osmotic stress (Liu et al. 2022). Salicylic acid improved the antioxidant system necessary to reduce oxidative damage and ion leakage from membranes and ameliorated the impact of heavy metal stress (Radwan et al. 2021). The action of salicylic acid is closely associated with the generation of various ROS. Treatment of wheat seedlings with salicylic acid caused a transitory enhancement of O_2^- and H_2O_2 production by plants and a simultaneous increase in the activity of SOD whereas the enhancement of the generation of H_2O_2 in salicylic acid-treated seedlings was accompanied by the activation of peroxidase (Alscher 2002).

Biomolecules are damaged by increased ROS under stress conditions which becomes a major factor limiting plant growth under drought stress (Jain et al. 2019). MeJ and SA regulatory phytohormones play a pivotal role in plant signalling responses. By regulating a range of physiological processes including photosynthesis and nitrogen and proline metabolism and by activating the antioxidant defence system, they protect plants against abiotic stresses (Mohi-Ud-Din et al. 2021). Though the foliar exogenous application of MeJ and SA showed their stress-mitigating effects through upregulating antioxidant enzymes activity, there were no significant effects on either dry herbage yield or the andrographolide yield in any morphotypes either alone or in a combination with soil moisture stress at maturity harvesting. This may be because MeJ and SA play a crucial role in the primary defence mechanism through cell signalling in plants under drought stress (Chhaya et al. 2021). Plants initiate various mechanisms to safeguard damage due to stress conditions. Such mechanisms include maintaining high tissue water potential, enhanced soil moisture mining capacity through a deep root system, preventing water loss through the closure of stomatal aperture, preventing transpiration losses, decreasing cell size, increasing cell elasticity and osmotic adjustment. Thus, the genetic control of tolerance is quite complex involving a change in the expression profiles of several genes. In such condition, though plants initiate various mechanisms to safeguard against damage ensuring survival, often fails to prevent loss of growth and yield. A previous study focused on the source-sink relationship in *A. paniculata* resulted in greater biomass partitioning towards the stem as compared to roots and leaves. Despite higher andrographolide content in leaves as compared to the stem, androgarpholide yield per plant basis was mainly decided based on dry weight accumulation pattern (Kalariya et al. 2021).

A heat map showing relationship among various parameters is presented in Fig. 2. A strong correlation between RWC and LWP was observed (Pearson's *r* value—0.98). It is also worth noting that the activity of antioxidant enzymes; SOD, GPX, CAT and APX were strongly and negatively correlated with RWC and LWP. The andrographolide concentration was negatively correlated with the RWC and LWP. Andrographolide concentration was negatively correlated with the andrographolide yield whereas a very strong and positive correlation between the dry herbage yield and the andrographolide yield (Pearson's *r* value—0.94) indicated that reduction in total andrographolide yield was majorly governed by the reduction in dry herbage yield under deficit soil moisture stress condition.

Conclusions

The present study revealed soil moisture stress in different morphotypes affected general plant growth resulting in herbage yield loss of up to 12%. Soil moisture stress during (75–140 DAT) reduced RWC and LWP, declined photosynthetic pigments and upregulated antioxidant enzymes activity. Dry herbage yield and andrographolide yield compromised due to soil moisture stress. It is also revealed that the impact of soil moisture stress was the same on all the morphotypes, and none of the morphotypes was superior for escaping yield loss. Though the andrographolide concentration in AP 35 was low as compared to that of AP 18, AP 24 and AP 2, AP 35, it was at a better position in gaining total andrographolide yield per plant because of its higher herbage yield. Foliar exogenous application of MeJ and SA upregulated antioxidant enzymes activity however, do not warrant any protection against stress induced yield loss in field grown *Andrographis paniculata.* Due to the relatively high herbage yield, the adverse effect of soil moisture stress on the total yield loss of andrographolide was less for morphotype AP 13 and AP 35 in semi-arid climate.

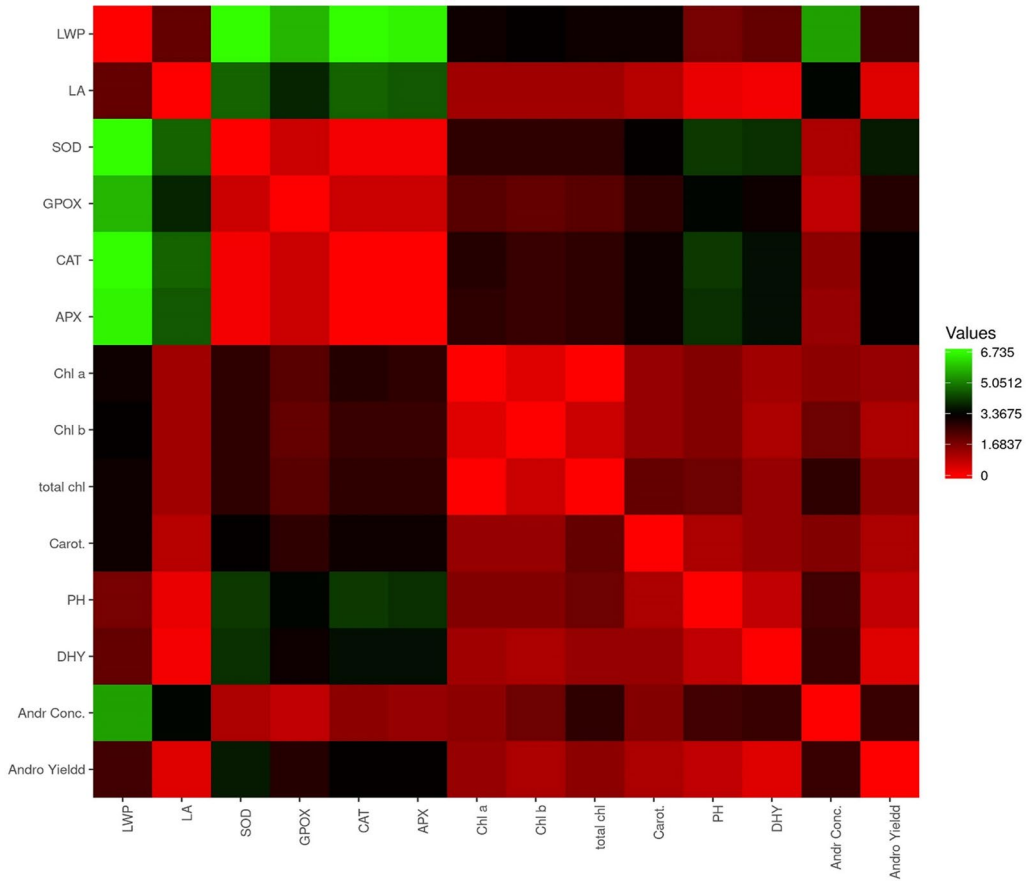

Fig. 2 The values of Pearson's *r* is indicated through a heat map. The Pearson's *r* (+ 1 to − 0.5) is indicated through colours. RWC % (Relative leaf water content), LWP MPa (Leaf water potential), SOD U (Superoxide dismutase), GPOX uM/min/g (Guaiacol peroxidase), CAT µM/min/g (Catalase), APX uM/min/g (Ascorbate peroxiadase), Chl a (Chlorophyll a mg^{-1} FW), Chl b (Chlorophyll b mg g^{-1} FW), total chl (Total Chlorophyll mg g^{-1} FW), Carot. (Carotenoids mg g^{-1}) FW, PH (Plant height, cm), DHY (Dry herbage yield, g plant $^{-1}$), Andr. Conc. (Andrographolide concentration %) and Andro Yield (Andrographolide Yield mg Plant.$^{-1}$)

Abbreviations

APX	Ascorbate peroxidase
AsA	Ascorbate
CAT	Catalase
DAT	Days after transplanting
DHAR	Dehydroascorbate reductase
DMRT	Dun-can's multiple range test
GGPS	Geranylgeranyl diphosphate
GPOX	Guaiacol peroxidase
GR	Glutathione reductase
GSH	Glutathione
GST	Glutathione s-transferase
HGMS	3-Hydroxy-3-methylglutaryl CoA synthase
HPLC	High performance liquid chromatography
H_2O_2	Hydrogen peroxide
ISPH	4-Hydroxy-3-methylbut-2-enyl diphosphate reductase,
LWP	Leaf water potential
MDHAR	Monodehydroascorbate reductase
MeJ	Methyl jasmonate
NBT	Nitrobluetetrazoilum
PVP	Polyvinylpyrolidone
ROS	Reactive oxygen species
RWC	Relative water content
SA	Salicylic acid
SMC	Soil moisture content
SOD	Superoxide dismutase

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Author contributions
KK involved in conceptualization and designing of the experiment set up, recording observations, acquisition of data, analysis and interpretation of data and writing original draft. DS involved in recording observations on antioxidant enzymes activity. PL made available weather data and involved in statistical analysis of the data and editing the MS. RP involved in statistical analysis of the data and editing the MS. NG and RS involved in extraction and analysis of bioactive compounds. GK involved in maintenance of plant genetic resources. All authors read and approved the final manuscript.

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Availability of data and materials
All the data generated or analysed during this study were included within the article.

Declarations

Ethics approval and consent to participate
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Consent for publication
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Competing interests
The authors report there are no competing interests to declare.

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References

Aebi H (1984) Catalase in vitro. Methods Enzymol. https://doi.org/10.1016/S0076-6879(84)05016-3
Ali MS, Baek KH (2020) Jasmonic acid signaling pathway in response to abiotic stresses in plants. Int J Mol Sci 21:621
Alscher RG (2002) Role of superoxide dismutases (SODs) in controlling oxidative stress in plants. J Exp Bot 53:1331–1341. https://doi.org/10.1093/jexbot/53.372.1331
Anjum SA, Xie X, Wang LC et al (2011) Morphological, physiological and biochemical responses of plants to drought stress. Afr J Agric Res 6:2026–2032
Arnon DI (1949) copper enzymes in isolated chloroplasts. Polyphenoloxidase in *Beta Vulgaris*. Plant Physiol 24:1–15. https://doi.org/10.1104/pp.24.1.1
El-Tayeb MA (2005) Response of barley grains to the interactive effect of salinity and salicylic acid. Plant Growth Regul. https://doi.org/10.1007/s10725-005-4928-1
Foyer CH, Mullineaux PM (2019) Causes of photooxidative stress and amelioration of defence systems in plants
Gao M, Zhou J, Liu H et al (2018) Foliar spraying with silicon and selenium reduces cadmium uptake and mitigates cadmium toxicity in rice. Sci Total Environ 631–632:1100–1108. https://doi.org/10.1016/j.scitotenv.2018.03.047
Ghasemzadeh A, Jaafar HZE, Karimi E (2012) Involvement of salicylic acid on antioxidant and anticancer properties, anthocyanin production and chalcone synthase activity in ginger (zingiber officinale roscoe) varieties. Int J Mol Sci. https://doi.org/10.3390/ijms131114828
Gill SS, Tuteja N (2010) Reactive oxygen species and antioxidant machinery in abiotic stress tolerance in crop plants. Plant Physiol Biochem 48:909–930. https://doi.org/10.1016/j.plaphy.2010.08.016
Hasanuzzaman M, Bhuyan MHMB, Anee TI, Parvin K, Nahar K, Mahmud JA, Fujita M (2019) Regulation of ascorbate-glutathione pathway in mitigating oxidative damage in plants under abiotic stress. Antioxidants (basel) 8(9):384. https://doi.org/10.3390/antiox8090384
Hossain MDS (2020) Proteomic studies: contribution to understanding plant salinity stress response. Glob J Botan Sci 8:1–10. https://doi.org/10.12974/2311-858X.2020.08.1
Hossain MS, Urbi Z, Evamoni FZ et al (2016) A secondary research on medicinal plants mentioned in the Holy Qur'an. J Med Plants 15:81–97
Huang L, Liu Y, Wang X, Jiang C, Zhao Y, Lu M, Zhang J (2022) Peroxisome-mediated reactive oxygen species signals modulate programmed cell death in plants. Int J Mol Sci 23:10087. https://doi.org/10.3390/ijms231710087
Jain M, Kataria S, Hirve M, Prajapati R (2019) Water deficit stress effects and responses in maize. In: Plant abiotic stress tolerance. Springer International Publishing, Cham, pp 129–151
Jaleel CA, Manivannan P, Lakshmanan GMA et al (2008) Alterations in morphological parameters and photosynthetic pigment responses of *Catharanthus roseus* under soil water deficits. Colloids Surf B Biointerfaces. https://doi.org/10.1016/j.colsurfb.2007.09.008
Jang G, Yoon Y, Choi YD (2020) Crosstalk with jasmonic acid integrates multiple responses in plant development. Int J Mol Sci 21:305
Jung C, Lyou SH, Yeu S et al (2007) Microarray-based screening of jasmonate-responsive genes in Arabidopsis thaliana. Plant Cell Rep. https://doi.org/10.1007/s00299-007-0311-1
Kalariya KA, Singh AL, Chakraborty K et al (2017) SCMR: a more pertinent trait than SLA in peanut genotypes under transient water deficit stress during summer. Proc Natl Acad Sci India Sect b: Biol Sci. https://doi.org/10.1007/s40011-015-0636-4
Kalariya KA, Gajbhiye N, Kumar J (2018) Evaluation of various genotypes of *Andrographis paniculata* for andrographolide yield under hormonal application. Acad J Biotechnol 6:029–036
Kalariya KA, Gajbhiye NA, Meena RP, Saran PL, Minipara D, Macwan S, Geet KA (2021) Assessing suitability of *Andrographis paniculata* genotypes for rainfed conditions in semi-arid climates. Inf Process Agric 8:359–368. https://doi.org/10.1016/j.inpa.2020.09.003
Kalariya KA, Gajbhiye NA, Deepa S, Meena RP, Saran PL (2019) Chlorophyll fluorescence: a physiological mechanism and a physical tool in plant eco-physiological studies In: Hemtaranja A (ed) Advanced Plant Physiology
Khalil R, Haroun S, Bassyoini F, Nagah A, Yusuf M (2021) Salicylic acid in combination with kinetin or calcium ameliorates heavy metal stress in *Phaseolus vulgaris* plant. J Agri Food Res. https://doi.org/10.1016/j.jafr.2021.100182
Liu J, Qiu G, Liu C, Li H, Chen X, Fu Q, Lin Y, Guo B (2022) Salicylic acid, a multifaceted hormone, combats abiotic stresses in plants. Life 12:886. https://doi.org/10.3390/life12060886
Lowry OH, Rosebrough NJ, Al F, Randall RJ (1951) Protein measurement with the Folin phenol reagent. J Biol Chem. https://doi.org/10.1016/s0021-9258(19)52451-6
Mir MA, John R, Alyemeni MN et al (2018) Jasmonic acid ameliorates alkaline stress by improving growth performance, ascorbate glutathione cycle and glyoxylase system in maize seedlings. Sci Rep 8:2831. https://doi.org/10.1038/s41598-018-21097-3
Mohi-Ud-Din M, Talukder D, Rohman M et al (2021) Exogenous application of methyl jasmonate and salicylic acid mitigates drought-induced oxidative damages in French bean (*Phaseolus vulgaris* L.). Plants 10:2066. https://doi.org/10.3390/plants10102066
Munné S, Peñuelas J (2003) Photo- and antioxidative protection, and a role for salicylic acid during drought and recovery in field-grown Phillyrea angustifolia plants. Planta 217:758–766. https://doi.org/10.1007/s00425-003-1037-0
Patanè C, Cosentino SL, Romano D, Toscano S (2022) Relative water content, proline, and antioxidant enzymes in leaves of long shelf-life tomatoes under drought stress and rewatering. Plants 11:3045. https://doi.org/10.3390/plants11223045
Rajani M, Shrivastava N, Ravishankara MN (2000) A rapid method for isolation of andrographolide from *Andrographis paniculata* Nees (Kalmegh). Pharm Biol. https://doi.org/10.1076/1388-0209(200007)3831-SFT204
Rajput VD, Harish SRK, Verma KK, Sharma L, Quiroz-Figueroa FR, Meena M, Gour VS, Minkina T, Sushkova S, Mandzhieva S (2021) Recent developments in enzymatic antioxidant defence mechanism in plants with special reference to abiotic stress. Biology (basel) 10(4):267. https://doi.org/10.3390/biology10040267
Sabagh ELA, Islam MS, Hossain A, Iqbal MA, Mubeen M, Waleed M, Reginato M, Battaglia M, Ahmed S, Rehman A, Arif M, Athar HUR, Ratnasekera D, Danish S, Raza MA, Rajendran K, Mushtaq M, Skalicky M, Brestic M, Soufan W, Fahad S, Pandey S, Kamran M, Datta R, Abdelhamid MT (2022) Phytohormones as growth regulators during abiotic stress tolerance in plants. Front Agron 4:765068. https://doi.org/10.3389/fagro.2022.765068
Sánchez-Reinoso AD, Ligarreto-Moreno GA, Restrepo-Díaz H (2019) Chlorophyll α fluorescence parameters as an indicator to identify drought susceptibility in common bush bean. Agronomy 9:526. https://doi.org/10.3390/agronomy9090526
Sharma SN, Jha Z, Sinha RK, Geda AK (2015) Jasmonate-induced biosynthesis of andrographolide in *Andrographis paniculata*. Physiol Plant. https://doi.org/10.1111/ppl.12252
Sharma AK, Basu I, Singh S (2018) Efficacy and safety of ashwagandha root extract in subclinical hypothyroid patients: a double-blind, randomized placebo-controlled trial. J Altern Complement Med 24:243–248. https://doi.org/10.1089/acm.2017.0183

Siboza XI, Bertling I (2013) The effects of methyl jasmonate and salicylic acid on suppressing the production of reactive oxygen species and increasing chilling tolerance in 'Eureka' lemon [*Citrus limon* (L.) Burm. F.]. J Hortic Sci Biotechnol 88:269–276. https://doi.org/10.1080/14620316.2013.11512965

Stitz M, Gase K, Baldwin IT, Gaquerel E (2011) Ectopic expression of *AtJMT* in *Nicotiana attenuata* : creating a metabolic sink has tissue-specific consequences for the jasmonate metabolic network and silences downstream gene expression. Plant Physiol 157:341–354. https://doi.org/10.1104/pp.111.178582

Tian H, Zhou Q, Liu W, Zhang J, Chen Y, Jia Z, Shao Y, Wang H (2022) Responses of photosynthetic characteristics of oat flag leaf and spike to drought stress. Front Plant Sci 13:917528. https://doi.org/10.3389/fpls.2022.917528

Urs RR, Roberts PD, Schultz DC (2006) Localisation of hydrogen peroxide and peroxidase in gametophytes of *Ceratopteris richardii* (C-fern) grown in the presence of pathogenic fungi in a gnotobiotic system. Ann Appl Biol 149:327–336. https://doi.org/10.1111/j.1744-7348.2006.00100.x

Wang J, Song L, Gong X et al (2020) Functions of jasmonic acid in plant regulation and response to abiotic stress. Int J Mol Sci 21:1446

Yadav B, Jogawat A et al (2021) An overview of recent advancement in phytohormones-mediated stress management and drought tolerance in crop plants. Plant Gene 25:100264. https://doi.org/10.1016/j.plgene.2020.100264

Yildirim AN, Şan B, Yildirim F, Celik C, Bayar B, Karakurt Y (2021) Physiological and biochemical responses of almond rootstocks to drought stress. Turk J Agric for 45(4):522–532. https://doi.org/10.3906/tar-2010-47

Zhang X, Zang R, Li C (2004) Population differences in physiological and morphological adaptations of Populus davidiana seedlings in response to progressive drought stress. Plant Sci 166:791–797. https://doi.org/10.1016/j.plantsci.2003.11.016

Zhou ZS, Guo K, Elbaz AA, Yang ZM (2009) Salicylic acid alleviates mercury toxicity by preventing oxidative stress in roots of Medicago sativa. Environ Exp Bot. https://doi.org/10.1016/j.envexpbot.2008.06.001

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