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Original Research Paper

Evaluation of antirrhinum genotypes for horticultural and phyto-chemical traits

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ABSTRACT

Snapdragon (*Antirrhinum majus* L.) is a commercially prized specialty cut flower in the international market. Among the inbred lines created, KTANT-2, KTANT-5, KTANT-6, KTANT-8, and KTANT-11 exhibited suitability for different horticultural traits. Total phenolic content in different genotypes ranged from 4.81 to 8.63 gallic acid equivalents (GAE)/100 g dry weight, while, total flavonoid content varied from 56.32 to 164.18 quercetin equivalents (mg/100 g QE). Antioxidant potential was determined through FRAP, DPPH, and MCA analysis. Additionally, FTIR analysis was conducted to identify the presence of various functional groups and compounds.

Keywords: Antioxidant, Antirrhinum, flavoniods, FTIR, phenolic

INTRODUCTION

Antirrhinum majus L. (snapdragon), family Plantaginaceae, and is native to northern Africa and southern Europe (Seo et al., 2020). It holds commercial significance as a specialty cut flower in the international market. It is commonly used as a pot plant, bedding, fillers and for herbaceous borders. Snapdragons bloom in spring, producing spikes with florets of varying colors and shades, including white, cream, yellow, rose, salmon, pink, mauve, red, magenta, and bicolored varieties (Lian et al., 2020). Beyond its ornamental uses, snapdragon has found applications in traditional medicine, with the whole plant decoction being employed for treating liver ailments, addressing watery eyes managing gum scurvy, various tumors, ulcers, and hemorrhoids (Lim, 2014). Jang et al. (2020) reported that extracts from Antirrhinum have high polyphenolic and flavonoid contents, as well as strong DPPH and ABTS radical-scavenging abilities. Fresh snapdragon flowers were found to possess a total antioxidant capacity (5.06 g ascorbic acid equivalents/kg), a total phenolic content (3.49 g gallic acid/kg), and a total flavonoid content (1.78 g rutin/kg) (Rop et al., 2012).

The consumption of flowers was popular during ancient times in Rome, Greece, and China, often used as alternatives to medicines or in combination with fruits, seeds, leaves, and root vegetables. Many of these flowers have a rich history of use in food preparation, adding aroma, flavors, and aesthetic value to dishes (Takahashia et al., 2020). These flowers can be enjoyed fresh or incorporated into savory dishes, including meat and fish, soups, wines, desserts, sweets, jellies, dyes, and spices. They are utilized in various forms, such as dried, powdered (Chen & Wei, 2017) and crystallized (Fernandes et al., 2019). While, historically edible flowers were primarily used for their fragrance and visual appeal, today's consumers are increasingly seeking new sources of natural bioactive compounds (Lysiak, 2022). Flowers are particularly rich in phenolic compounds, flavonoids, and anthocyanins (Mlcek & Rop, 2011) and play a crucial role in combating oxidative stress induced by various pathogens (Benvenuti & Mazzoncini, 2021).

In addition to culinary uses, edible flowers are known for being low in calories while rich in mineral compounds, vitamins, mucilage, amino acids, fibers, carbohydrates, essential oils, and proteins (Poonam et al., 2021). Numerous studies have highlighted the medicinal properties of edible flowers, including their potential as antidiabetic, anticancer, anti-anxiety, anti-inflammatory, antimicrobial, diuretic, and immunomodulatory agents (Jang et al., 2020). Consequently, the newly developed inbred lines were evaluated not only for their horticultural characteristics but also for their medicinal properties.

MATERIALS AND METHODS

The inbred lines of Antirrhinum were developed by selection from open pollinated seed at ICAR-Indian

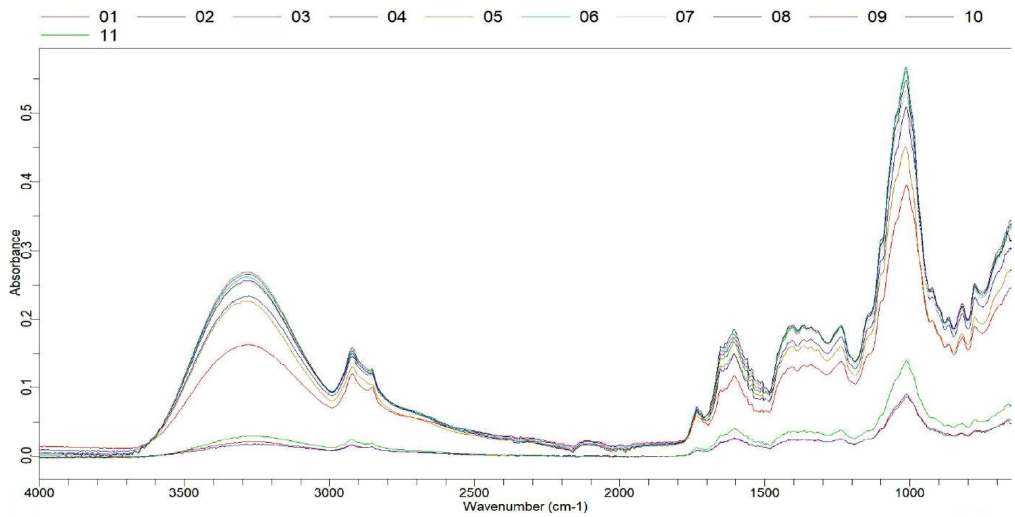

Fig. 1 : FTIR analysis of different Antirrhinum genotypes

Institute of Agricultural Research, Regional Station, Katrain, Himachal Pradesh and subjected to evaluation for various horticultural traits during 2020-2022 under mid-hill conditions (Fig. 1). Quantitative traits, including plant height, plant spread, days to flowering, duration of flowering, rachis length, number of flowers per stem, and stem thickness and qualitative traits such as growth habit, flower type, flowering intensity, and flower color were recorded under field conditions.

The freshly harvested flowers of 11 genotypes (Fig. 2) were directly transported to laboratory under controlled conditions to maintain their freshness and dried at 60±2°C in a hot air oven for 6 hours, finely ground, and then sealed in airtight pouches for refrigerated storage for further analysis. The total phenolic content (TPC) was calculated by following Singleton et al. (1999) and the results were expressed as mg/100 g GAE. Similarly total flavonoid content (TFC) was determined and expressed in mg/100g QE (Kiranmai et al., 2011).

The ferric reducing antioxidant power (FRAP) assay (Alvarez-Parrilla et al., 2019), metal chelating activity (MCA) determined (Chavan et al., 2013). Antioxidant activity (free radical scavenging activity), was assessed as per Brand-Williams et al. (1995) using DPPH as the source of free radicals. Antioxidant Activity (%) = (Ab (B) - Ab (S)) / Ab(B) × 100 (Here, Ab (B) represents the absorbance of the blank, and Ab(S) represents the absorbance of the sample). For qualitative analysis, optimized samples were examined for fourier transform infrared (FTIR) spectra using an Agilent Cary 630 FTIR spectrometer. The spectra were recorded within the wave number range of 4000-400 cm and interpreted by following the guidelines of Stuart & Thomas (1995). The data was subjected to a two-way analysis of variance (ANOVA) using a completely randomized design with SPSS 16.0 (IBM, SPSS Inc., USA) and the results are reported as means with standard deviations.

RESULTS AND DISCUSSION

Evaluation for horticultural traits

Among the different genotypes, plant height (mean±SE) ranged from 22.43±1.08 cm to 111.44±1.45 cm (F =12.19, $d.f.$ = 2, 10, $p \leq 0.01$), with the highest observed in the KTANT-5 genotype (111.44±1.45 cm). Maximum plant spread (F =15.67, $d.f.$ = 2, 10, $p \leq 0.01$) was observed in KTANT-2 (37.59 ± 1.67 cm), shortest time taken to flowering (F =21.07, $d.f.$ = 2, 10, $p \leq 0.01$) was noted in KTANT-6 (24.00 ± 1.16 days), and the longest duration of flowering (F =35.83, $d.f.$ = 2, 10, $p \leq 0.01$) was observed in KTANT-5 (80.00 ± 0.88 days). The rachis length (F =12.36, $d.f.$ = 2, 10, $p \leq 0.01$) was recorded longest in KTANT-8 (15.41±0.50 cm), while, KTANT-5 recorded highest number of flowers (F =8.01, $d.f.$ = 2, 10, $p \leq 0.01$)per stem (21.00±1.16), and KTANT-11 (F =15.87, $d.f.$ = 2, 10, $p \leq 0.01$) recorded thickest spike (7.34±0.08 mm) (Table 1). These observations are in line with findings of Lewis et al. (2021). For various qualitative traits, all the genotypes exhibited both upright and horizontal growth habits. Similar morphological studies have been conducted in various research projects, leading

Fig. 2 : Colour variations in different genotypes of Antirrhinum

to the classification of germplasm into different categories (Sekerci et al., 2017). Single, double, and unique trumpet-type flowers were observed, and a high flower density was observed among the genotypes. Color variations included red, red-purple, white, orange-red, yellow, and yellow-orange in the Antirrhinum genotypes (Table 1).

Evaluation for phytochemical traits

Phenols are the most abundant physiologically active compounds found in plants. They are classified into flavonoids and tannins, with flavonoids being the largest subgroup of polyphenols synthesized by plants (Jang et al., 2020). The total phenolic compounds (TPC) in different genotypes ranged from 4.50±0.29 to 8.35±0.24 gallic acid equivalents (GAE)/100 g dry weight (Table 2). The highest TPC (F =31.55, $d.f.$ = 2, 10, $p \leq 0.01$) was observed in KTANT-1 (8.35 ±0.24 mg/100 g GAE), which had red-purple-colored flowers. Flavonoids, a subclass of polyphenols, are characterized by a structure comprising two phenyl and one heterocyclic ring. They are predominantly found in flowers and offer various health benefits, including cancer inhibitory activities (Seo et al., 2020). Flavonoids are crucial constituents that determine the color of flowers. Total flavonoid content (TFC) ranged from 53.01±1.7 to 158.51±3.32 quercetin equivalents (mg/100 g QE), with the highest value (158.51± mg/100 g QE) was recorded in KTANT-7 (F =250.21, $d.f.$ = 2, 10, $p \leq 0.01$).

Table 1 : Evaluation of newly developed Antirrhinum genotypes for horticultural traits

Genotype	Plant height (cm)	Plant spread (cm)	Days taken to flowering (days)	Duration of flowering (days)	Rachis length (cm)	No. of flowers/ spike	Spike diameter (mm)	Growth habit	Flower type	Flower density	Flower colour (RHS colour)
KTANT-1	73.73±3.53	30.16±0.74	44 ±1.16	72±1.16	15.21±0.29	18.33±0.88	7.21±0.36	Upright	Single	High	Red Group 46 B
KTANT-2	108.47±2.25	37.59±1.67	48 ±1.16	61±1.76	14.07±0.43	16.00±1.16	5.08±0.27	Upright	Double	High	Red Purple 68 C
KTANT-3	72.88±3.18	25.69±1.87	46±1.20	65±1.76	13.42±0.62	16.67±1.33	6.37±0.27	Upright	Single	High	Red Purple 65 C
KTANT-4	65.91±1.90	26.16±1.47	50±1.16	62±1.16	12.56±0.48	14.00±1.16	6.09±0.27	Upright	Single	High	White Group 155 B
KTANT-5	111.44±1.45	26.26±1.49	46±1.16	80±0.88	14.62±0.63	21.00±1.16	5.36±0.27	Upright	Single	High	Red Purple Group 62 D
KTANT-6	22.43±1.08	28.52±1.03	24±1.16	30±1.16	5.00±0.38	7.33±0.67	0.00±0.00	Horizontal	Single	High	Orange Red Group 34 A
KTANT-7	110.81±0.77	27.13±0.89	45±1.76	61±1.76	13.20±0.38	12.33±1.20	5.16±0.33	Upright	Double	High	Red Group 53 A
KTANT-8	83.69±2.10	29.64±0.45	40±1.16	56±1.20	15.41±0.50	14.67±1.76	6.13±0.23	Upright	Single	Medium	Yellow Orange 23 A
KTANT-9	40.45±0.38	22.90±0.75	35±1.73	46±1.16	4.18±0.41	5.00±0.58	0.00±0.00	Horizontal	Trumpet	Medium	Purple Group 77 A
KTANT-10	75.78±1.81	23.72±1.73	40±1.16	50±0.88	13.39±0.64	16.33±1.20	6.43±0.15	Upright	Single	High	Red Purple Group 69 A
KTANT-11	90.48±1.00	20.41±0.39	44±2.31	49±1.53	15.07±0.64	20.33±1.20	7.34±0.08	Upright	Single	High	Yellow Group 4 A
C.D.	4.18	2.41	2.51	1.96	1.05	2.69	0.46	-	-	-	-
SE(m)	1.41	0.81	0.85	0.66	0.35	0.91	0.15	-	-	-	-
SE(d)	1.99	1.15	1.20	0.93	0.50	1.28	0.22	-	-	-	-
C.V.	3.13	5.19	3.48	1.99	4.95	10.65	5.30	-	-	-	-

Antioxidant activity

Antioxidant properties of antirrhinum were assessed through the FRAP, DPPH, and MCA tests. The FRAP test measures the reducing potential of an antioxidant by reacting with the ferric tripyridyltriazine (TPTZ) complex, producing a colored complex. FRAP activity ranged from 1.17±0.09 to 5.69±0.13 mg/100 mL, with KTANT-7 (F =64.20, $d.f.$ = 2, 10, $p \leq 0.01$) exhibiting the highest value for this activity at 5.69±0.13 mg/100 mL. MCA calculates the extract's capacity to chelate metal ions that may induce free radical-producing reactions. Metal chelating activity was highest (48.38±1.88%) in KTANT-2 (F =75.50, $d.f.$ = 2, 10, $p \leq 0.01$) and lowest in KTANT-11 (15.42±0.45%), having yellow-colored flowers. The DPPH test examined the ability of the antirrhinum extract to donate hydrogen and stabilize radicals (Ayob et al., 2021). The DPPH activity ranged from 35.10±1.39 to 92.90±1.35%. The highest DPPH activity (95.47%) was recorded in KTANT-10 (F =169.99, $d.f.$ = 2, 10, $p \leq 0.01$), characterized by red-purple-colored flowers, while, the lowest was found in KTANT-3 (35.10±1.39%).

FTIR analysis

Qualitative screening of various antirrhinum genotypes was conducted to confirm the presence of different functional groups and compounds, as depicted in Fig. 1 and Table 3. Among the samples, KTANT-3, KTANT-10, and KTANT-7 exhibited the highest peaks at all the wave numbers compared to the other samples. The peak within the wave-number range of 1047 to 11278 cm indicates C-O stretching, confirming the presence of phenols. Additionally, the peak at 3280 cm corresponds to O-H stretching, characteristic of alcohol and phenol. Furthermore, the presence of alkenes was suggested by the peak at 775 cm.

Table 2 : Phytochemical and antioxidant properties of antirrhinum genotypes

Genotype	Total phenolic content (mg/100 g GAE)	Total flavonoid content (mg/100 g QE)	FRAP (mg/100 mL)	Metal chelating activity (%)	DPPH (%)
KTANT-1	7.25±0.08	70.06±0.88	5.25±0.14	33.91±0.85	61.64±0.78
KTANT-2	6.62±0.19	59.37±1.31	4.81±0.08	48.38±1.88	74.63±1.16
KTANT-3	7.03±0.09	70.84±1.31	4.50±0.27	27.60±1.63	35.10±1.39
KTANT-4	6.93±0.05	61.58±1.49	4.90±0.06	41.11±0.80	85.80±2.10
KTANT-5	4.50±0.29	56.17±1.22	4.38±0.09	21.53±0.38	83.88±1.51
KTANT-6	6.26±0.21	62.91±1.17	5.06±0.07	30.16±0.68	51.41±1.36
KTANT-7	7.56±0.13	158.51±3.32	5.69±0.13	25.82±0.82	52.42±1.32
KTANT-8	6.22±0.12	62.46±2.07	1.17±0.09	21.39±0.90	88.81±1.18
KTANT-9	6.36±0.12	63.98±2.48	4.98±0.32	35.16±1.77	54.55±1.89
KTANT-10	8.35±0.24	53.01±1.75	5.05±0.04	21.88±0.93	92.90±1.35
KTANT-11	6.28±0.21	68.34±2.03	4.95±0.09	15.42±0.45	70.80±1.25
C.D.	0.51	5.48	0.44	3.30	4.23
SE(m)	0.17	1.86	0.15	1.12	1.43
SE(d)	0.25	2.67	0.21	1.58	2.03
C.V.	4.50	4.49	5.60	6.61	3.63

Table 3 : FTIR analysis of Antirrhinum genotypes

Wave No.	Functional compounds	Intensity										
		KTANT-1	KTANT-2	KTANT-3	KTANT-4	KTANT-5	KTANT-6	KTANT-7	KTANT-8	KTANT-9	KTANT-10	KTANT-11
775	Alkenes, =C–H out-of-plane bending	0.47	0.46	0.49	0.46	0.46	0.46	0.48	0.45	0.47	0.48	0.43
1237	Alcohol and phenols, C–O stretching	0.37	0.33	0.38	0.33	0.35	0.33	0.37	0.35	0.36	0.38	0.33
1341	Carbohydrates Coupled stretching and bending	0.32	0.31	0.34	0.31	0.31	0.31	0.33	0.30	0.31	0.33	0.29
1408	Aromatic phosphates (P-O-C) stretch, C-O epoxy and oxirane ring	0.31	0.30	0.34	0.30	0.29	0.30	0.33	0.29	0.30	0.34	0.28
1602	C=O stretch	0.28	0.27	0.30	0.27	0.27	0.27	0.29	0.27	0.28	0.29	0.26
2922	Carbohydrates, C-H stretching	0.28	0.28	0.31	0.28	0.27	0.28	0.29	0.26	0.27	0.30	0.25
3280	O-H stretching (alcohol))	0.40	0.39	0.43	0.39	0.38	0.39	0.41	0.38	0.39	0.42	0.36

CONCLUSION

This study found significant variations in plant height, spread, flowering duration, and other horticultural traits among the *Antirrhinum* genotypes. Phytochemical analysis identified essential compounds like phenols and flavonoids, with varying concentrations. Antioxidant assays (DPPH, FRAP, and MCA) showed diverse levels of antioxidant activity across the genotypes, with KTANT-10 exhibiting high DPPH activity, and KTANT-7 showing strong FRAP and metal chelating abilities. Qualitative screening confirmed the presence of functional groups linked to these phytochemicals. These results highlight the potential of *Antirrhinum* genotypes for valuable horticultural and phytochemical traits with possible health benefits.

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