Proc. Indian Acad. Sci. (Plant Sci.), Vol. 94, Nos 4-6, June 1985, pp. 683-694

Effect of different growth substances on the foliar stomata of *Tagetes erecta* L

VIDHU, R K SHARMA* and Y S MURTY

Botany Department, Institute of Advanced Studies, Meerut University, Meerut 250 005, India

*Present address: Botany Department, SSV (PG) College, Hapur, Ghaziabad, India

MS received 13 December 1983

Abstract. The response of abaxial and adaxial stomata to various growth substances were examined in leaves of *Tagetes erecta* L. From stomatal studies it is clear that growth substances produced several variations in the morphology of stomata. The anomalies recorded include persistent stomatal initial, loss of one or both guard cells in a significant percentage of stomata, contiguous stomata, cytoplasmic connections, one and a half contiguous stomata etc. Significant changes in frequency of epidermal cells and stomata, size of epidermal cells and stomata and stomatal index were noticed in various treatments.

Keywords. *Tagetes erecta*; stomata; growth substances.

1. Introduction

The influence of various growth regulators, environmental factors, mechanical injury etc. on the structure and development of stomata has been under study for the last three decades. Inamdar (1970) investigated the action of growth regulators on the development of stomata in the cotyledons of *Abelmoschus esculentus* Moench. Cooper *et al* (1972) studied the interaction of abscisic acid and kinetin on the stomata of barley. Effect of growth regulators—IAA and TIBA, on the structure and ontogeny of stomata is observed by Inamdar *et al* (1974). Kasat (1979) studied the effect of growth regulators on the stomatal development in *Vigna sinensis*. The growth regulators are also reported to affect the frequency of stomata, stomatal index and the size of epidermal and guard cells but not the ontogeny of stomata which seems to be a genetic character as also noticed by Sharma and Dunn (1968). For the present study *Tagetes erecta* L. of the family compositae was used. Observations are presented on the action of various growth substances—indole-3-acetic acid, gibberellic acid, 2,3,5-triiodobenzoic acid and kinetin.

2. Material and methods

The leaves that emerged after spray treatment with different growth substances were collected, washed thoroughly and fixed in FAA. The following growth substances were used: indole-3-acetic acid, gibberellic acid, 2,3,5-triiodobenzoic acid in 50, 100, 200 and 500 ppm concentrations and kinetin in 10, 25, 50 and 100 ppm concentrations.

Epidermal peels taken by the direct peel method from the middle region of the terminal leaf segments were stained in 1 % aqueous solution of safranin and mounted in glycerine. Camera lucida drawings were prepared showing the exact size, shape and number of stomata on the epidermal peels. The stomatal index, diversity of stomatal

types and stomatal frequency were calculated from ten different peels of each of the surfaces in each concentration at a uniform magnification. The stomatal index is calculated as defined by Salisbury (1927, 1932) *viz.* $100S/(E+S)$, where S is the number of stomata and E the number of epidermal cells over a given surface per unit area. The length and breadth are measured at random for 25 stomata in each concentration on both surfaces. Since the epidermal cells are variously oriented, their longest and broadest distances (here considered as length and breadth) are also measured in the same way as those of the stomata. Length/breadth ratio of stomata from both the surfaces is calculated (Solereder 1908).

3. Observations

3.1 *Mature epidermis*

The epidermal cells are polygonal isodiametric or elongated with mostly sinuous (figures $1A_1$, C_3, G_4; $2D_1$, D_2; $3C_1$), rarely straight (figures $1F_{10}$; $2C_7$, D_5; $3A_1$, A_2, B_1, E_1, F_1, F_4 and F_5) or arched anticlinal walls (figures $1B_4$; $2A_4$, D_1; $3D_1$). The stomata are distributed, all over the surfaces, except over the veins without any definite pattern of orientation.

3.2 *Mature stomata*

The great majority of stomata are anomocytic in the control and other treatments (figures $1A_1$, B_4, E_2, F_{10}; $2A_4$, C_7, D_1, D_2, D_5; $3C_1$, D_1, E_1). Diacytic stomata are also observed in GA_3 100 ppm on the abaxial side besides the normal type (figure $1G_4$). Sometimes anomocytic stomata surrounded by three epidermal cells simultating an anisocytic type, are observed on the abaxial surface of IAA 100 ppm, on both adaxial and abaxial surfaces of Kn 50 ppm and on adaxial surface of Kn 100 ppm treatments (figures $1C_3$; $3A_1$, A_2, B_1). The stomatal pore may be small lenticular (figures $1A_1$, A_2) or oval as observed in TIBA 50 ppm (figure $3C_2$) or abnormally big as observed in IAA 200 ppm, GA_3 50 ppm, Kn 50 ppm and TIBA 100 ppm (figures $1D_1$, F_2; $3A_1$, D_2). The anomocytic stomatal apparatus contains a pore enclosed by two reniform guard cells surrounded by 4–6 ordinary epidermal cells (figures $1A_1$, E_2, F_{10}; $2A_4$, D_1, D_3, D_5). The development of anomocytic stomata is as follows that the meristemoid directly functions as a guard mother cell without cutting off any subsidiary cell. It enlarges, becomes rounded and divides by a straight wall to form a pair of guard cells. A lenticular pore develops in between the two guard cells. In diacytic stomata the stomatal mother cell divides by a straight wall at right angles to the subsidiary cells to form a pair of guard cells.

3.3 *Abnormalities*

In addition to normal stomata various aberrant types are induced by growth substances. They are as follows:

3.3a *Persistent stomatal initial*: The persistent stomatal initials are observed in all concentrations of gibberellic acid used (figures $1F_5$, F_8, F_9, G_6; $2A_5$, B_2), in TIBA 200 ppm (figures $3E_2$, E_4) and 500 ppm (figures $3F_2$, F_4). These initials may occur

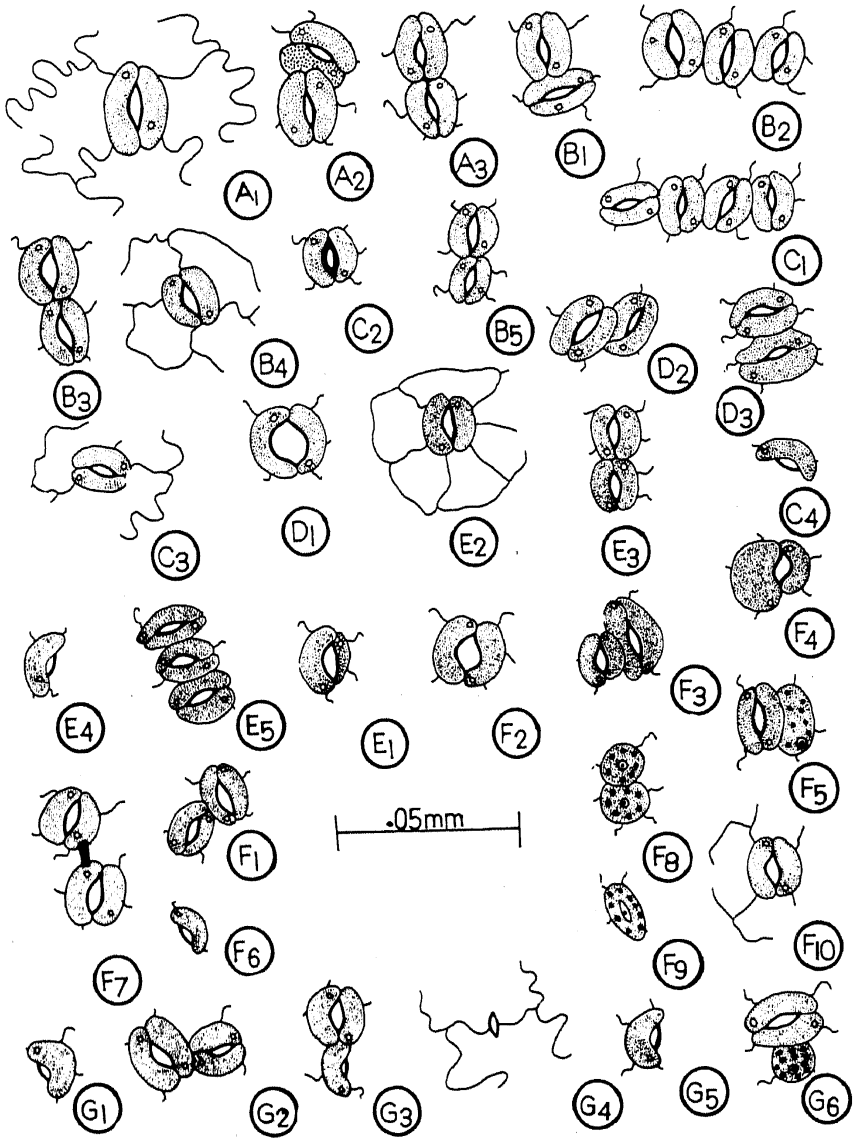

Figure 1(A–G). Effect of growth substances on stomata. A_1–A_3. Control. Portions of abaxial leaf epidermis. B_1–B_3. IAA 50ppm. Portions of adaxial leaf epidermis. B_4–B_5. IAA 50 ppm. Portions of abaxial leaf epidermis. C_1–C_2. IAA 100 ppm. Portions of adaxial leaf epidermis. C_3–C_4. IAA 100 ppm. Portions of abaxial leaf epidermis. D_1–D_2. IAA 200 ppm. Portions of adaxial leaf epidermis. D_3. IAA 200 ppm. Portions of abaxial leaf epidermis. E_1–E_3. IAA 500 ppm. Portions of adaxial leaf epidermis. E_4–E_5. IAA 500 ppm. Portions of abaxial leaf epidermis. F_7–F_{10}. GA_3 50 ppm. Portions of adaxial leaf epidermis. F_1–F_6. GA_3 50 ppm. Portions of abaxial leaf epidermis. G_1–G_3. GA_3 100 ppm. Portions of adaxial leaf epidermis. G_4–G_6. GA_3 100 ppm. Portions of abaxial leaf epidermis.

either solitary (figures $1F_9$; $2A_5$; $3E_4$, F_4) or contiguous with each other (figures $1F_8$; $2B_2$; $3E_2$) or with normal stomata (figures $1F_5$, G_6; $2C_8$; $3C_3$, E_1, F_2). The shape of the persistent stomatal initial may be spherical (figures $1F_8$, G_6; $2A_5$, B_2, C_8; $3C_3$) or ellipsoidal (figures $1F_5$, F_9; $2E_2$). Sometimes the persistent stomatal initial develops a much thickened wall (figure $3E_4$). Such initials may be caused because the meristemoid is inhibited in its further development. Such inhibited meristemoid cells fail to divide or give rise to a pair of guard cells with chloroplast and thick wall.

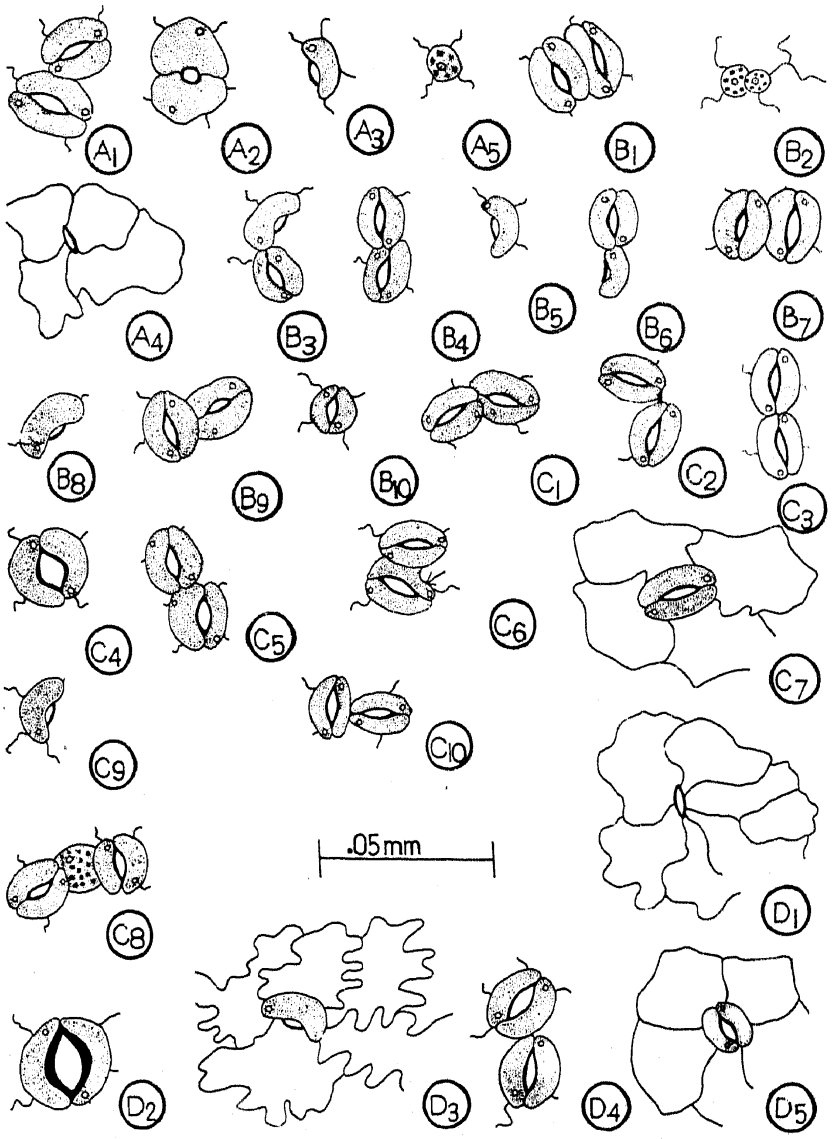

Figure 2(A–D). A_1–A_3. GA_3 200 ppm. Portions of adaxial leaf epidermis. A_4–A_5. GA_3 200 ppm. Portions of abaxial leaf epidermis. B_1–B_6. GA_3 500 ppm. Portions of adaxial leaf epidermis. B_7–B_{10}. GA_3 500 ppm. Portions of abaxial leaf epidermis. C_1–C_3. Kn 10 ppm. Portions of adaxial leaf epidermis. C_4–C_{10}. Kn 10 ppm. Portions of abaxial leaf epidermis. D_1–D_2. Kn 25 ppm. Portions of adaxial leaf epidermis. D_3–D_5. Kn 25 ppm. Portions of abaxial leaf epidermis.

3.3b *Contiguous stomata*: Variously oriented contiguous stomata were observed in control as well as in various treatments. Their frequency was more in IAA and TIBA treatments than in the control and other treatments. Two to four contiguous stomata (figures $1A_2$, B_2, C_1, E_5) were either juxtaposed (figures $1A_2$, B_1, B_2, C_1, F_1; $2A_1$, B_1, B_7; $3C_4$, F_6) superimposed (figures $1A_3$, B_3, B_5, E_3, E_5; $2B_4$, C_3, C_5, D_4; $3D_4$, E_3) or at right angles (figures $2B_9$, C_{10}) or obliquely oriented (figure $3B_4$) are also observed. Contiguous stomata may be equal (figure $1A_2$) or unequal in size (figure $1F_3$, G_2). Contiguous stomata develop after two or four stomata come in contact with each other and becomes contiguous as a result of readjustment of the epidermis during maturation.

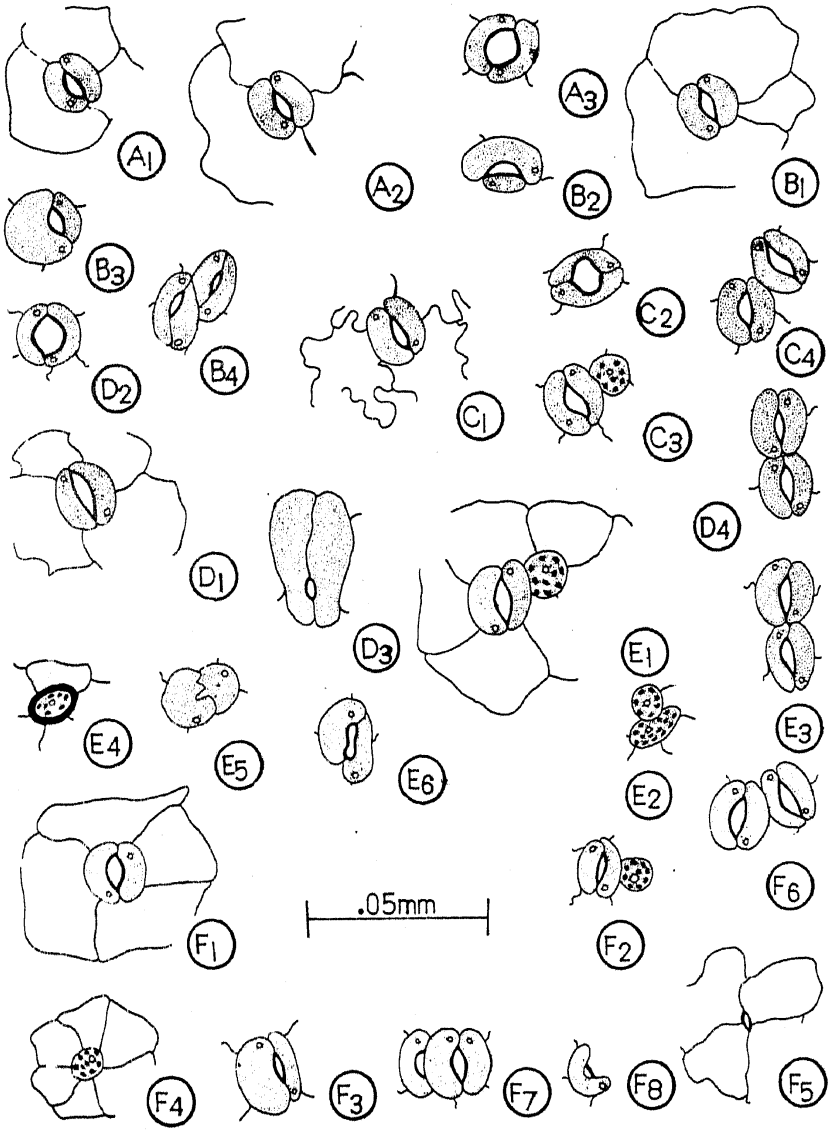

Figure 3 (A–F). A_1. Kn 50 ppm. Portions of adaxial leaf epidermis. A_2–A_3. Kn 50 ppm. Portions of abaxial leaf epidermis. B_1–B_2. Kn 100 ppm. Portions of adaxial leaf epidermis. B_3–B_4. Kn 100 ppm. Portions of abaxial leaf epidermis. C_1–C_3. TIBA 50 ppm. Portions of adaxial leaf epidermis. C_4. TIBA 50 ppm. Portions of abaxial leaf epidermis. D_1–D_2. TIBA 100 ppm. Portions of adaxial leaf epidermis. D_3–D_4. TIBA 100 ppm. Portions of abaxial leaf epidermis. E_1–E_3. TIBA 200 ppm. Portions of adaxial leaf epidermis. E_4–E_6. TIBA 200 ppm. Portions of abaxial leaf epidermis. F_1–F_3. TIBA 500 ppm. Portions of adaxial leaf epidermis. F_4–F_8. TIBA 500 ppm. Portions of abaxial leaf epidermis.

3.3c *Cytoplasmic connection*: Cytoplasmic connections connect the protoplasts of adjacent guard cells of nearby stomata. These were observed in GA_3 50 ppm (figure $1F_7$) and Kn 10 ppm (figure $2C_2$).

3.3d *One and a half contiguous stomata*: These were observed in various treatments in which a single guard cell is contiguous with a normal stomata (figures $1G_3$; $2B_3$, B_6; $3F_7$).

3.3e *Single guard cell*: Single guard cells were observed in various treatments (figures $1C_4$, E_4, F_6, G_1, G_5; $2A_3$, B_5, B_6, B_8, C_9, D_3; $3F_8$). In the development of single guard cell the meristemoid directly differentiates into a single guard cell instead of giving rise to a pair of guard cells. It enlarges, get notched on one side and becomes bean shaped with differential wall thickenings on the side of the notch.

3.3f *Degenerated guard cells*: Both the guard cells of stomata have degenerated and thus having only a pore were observed in GA_3 100 ppm (figure $1G_4$) GA_3 200 ppm (figure $2A_4$) TIBA 500 ppm (figure $3F_5$) and Kn 25 ppm (figure $2D_1$). During the process of degeneration the guard cell nucleus degenerates and disappears followed by cytoplasm and guard cell wall leaving only a pore with thickening.

3.3g *Other abnormalities*: Other abnormalities such as transversely divided guard mother cell with a small opening in the centre (figure $2A_2$), unequal guard cells (figures $1E_1$, F_4; $3B_3$, F_3), giant stomata (figure $2C_4$, D_2), stomata without pore (figure $3E_5$), obliquely oriented guard cells (figure $3E_6$), one guard cell encroached upon by an adjoining epidermal cell (figure $2C_6$), eccentrically situated pore on elongated guard cells (figure $3D_3$), one guard cell enlarged and reniform, the other small and straight (figure $3B_2$), pore side with thickened wall (figure $2D_2$) were also observed.

3.4 *Frequency of epidermal cells*

The number of epidermal cells on both adaxial and abaxial surfaces were counted in a unit area (table 1). A general pattern was observed on the abaxial surface where 50 and 100 ppm of IAA and GA_3 showed promotion in the frequency of epidermal cells while their 200 and 500 ppm showed an inhibition. However, TIBA and Kn had an inhibitory effect in 50 ppm and 10 ppm on frequency of epidermal cells while it progressively increased in 100, 200 and 500 ppm of TIBA and 25, 50 and 100 ppm of Kn. Maximum number of epidermal cells per unit area was found in Kn 100 ppm, *i.e.* 186·44 and minimum in IAA 500 ppm, *i.e.* 90·39. On the adaxial surface, frequency of epidermal cells showed a different pattern from that of abaxial surface. IAA decreased the frequency of epidermal cells as concentration increased. In an unit area 201·50 and 186·44 epidermal cells were found in GA_3 50 and 500 ppm while 109·22 and 101·69 cells were found in 100 and 200 ppm. In TIBA treated plant, the frequency of epidermal cells increased with increasing concentrations. In 10 ppm, only 94·16 epidermal cells were present in an unit area, while in 25, 50 and 100 ppm, frequency of epidermal cells increased with increasing concentration reaching 190·2 mm^{-2} in 100 ppm.

3.5 *Frequency of stomata*

It is defined as the number of stomata per unit area. On both the surfaces, it was found that all treatments increased the number of stomata per unit area except IAA 500 ppm where slight inhibition was found over control (table 1). IAA and kinetin increased the frequency of stomata but the frequency decreased with increasing concentrations on both the surfaces. GA_3 showed a different pattern with regard to the number of stomata on the abaxial and adaxial surfaces as far as concentration is concerned. On the abaxial surface it increased from 50 ppm to 500 ppm while on the adaxial surface it decreased from 50 ppm to 500 ppm. The frequency increased with increase in concentration of TIBA.

Table 1. Effect of different growth substances on epidermal characters in *T. erecta* L.

Treatment	Concentration (ppm)	Frequency of epidermal cells (mm^{-2})		Frequency of stomata (mm^{-2})		Stomatal index	
		Abaxial	Adaxial	Abaxial	Adaxial	Abaxial	Adaxial
Control	0	111·11 ±2·07	116·76 ±3·07	28·25 ±3·23	15·07 ±0·75	23·44	11·43
IAA	50	122·41 ±3·08	112·99 ±4·05	41·43 ±2·32	24·48 ±1·07	25·29	17·81
	100	112·99* ±1·09	111·11 ±5·05	35·78 ±1·04	22·60 ±0·99	24·05	16·90
	200	109·22* ±4·07	97·92 ±0·79	30·13 ±1·00	20·72 ±0·98	21·62	17·46
	500	90·36 ±2·69	90·39 ±1·89	26·37 ±0·97	13·18 ±0·34	22·58	12·73
GA_3	50	120·52 ±7·08	201·50 ±8·20	33·90 ±1·08	33·90 ±0·84	21·95	14·40
	100	111·11** ±2·08	109·22 ±0·99	38·25 ±1·07	30·13 ±0·73	25·31	21·62
	200	101·69 ±1·07	101·69 ±2·33	47·08 ±2·04	22·60 ±0·72	31·65	18·18
	500	97·92 ±6·04	186·44 ±3·86	54·61 ±1·06	22·60 ±0·81	35·80	10·81
TIBA	50	109·22 ±2·02	137·47 ±4·49	37·66 ±2·07	24·48 ±0·93	25·64	15·12
	100	111·11** ±3·04	150·65 ±12·23	48·96 ±1·09	20·72 ±0·46	30·59	12·09
	200	112·99* ±2·08	158·19 ±12·07	45·20 ±1·21	41·43 ±1·08	32·43	20·75
	500	141·24 ±3·07	167·60 ±15·05	60·26 ±3·27	43·32 ±1·07	29·91	20·54
Kn	10	109·22* ±4·89	94·16 ±1·08	52·73 ±2·09	54·61 ±2·03	32·56	35·90
	25	122·41 ±9·08	148·77 ±4·37	50·85 ±1·89	45·20 ±1·08	29·35	25·47
	50	139·35 ±2·34	167·60 ±10·85	43·31 ±1·25	37·66 ±0·94	23·71	20·54
	100	186·44 ±10·28	190·20 ±12·37	39·55 ±1·30	26·37 ±0·25	17·50	17·21

* Insignificant at 1%.
** Insignificant at 1 and 5%.

3.6 *Stomatal index*

It is more on the abaxial surface in all treatments than on the adaxial surface except in Kn 10 ppm where stomatal index was more on the adaxial surface. Maximum stomatal index was found in GA_3 500 ppm, *i.e.* 35·80 and minimum in Kn 100 ppm, *i.e.* 17·50 on the abaxial surface while on the adaxial surface maximum stomatal index was found in Kn 10 ppm *i.e.* 35·90 and minimum in GA_3 500 ppm, *i.e.* 10·81. (table 1).

3.7 *Epidermal cell size*

From the data it is clear that epidermal cell size was almost equal in control on both the adaxial and abaxial surfaces. On abaxial surfaces IAA 50 ppm decreased both length and breadth of epidermal cells, IAA 100 ppm decreased the length but breadth was not affected. In IAA 200 and 500 ppm epidermal cell size increased both length and breadth wise while on adaxial surfaces there was increase in size of epidermal cells as concentration increased. On abaxial and adaxial surfaces in GA_3 treated plants length of the epidermal cells gradually decreased with increasing concentration and ultimately became equal to that of control in 500 ppm. Breadth of epidermal cell was also affected by GA_3 treatment. In 50 ppm it was less while in 100 and 200 ppm it was more than that of control. In TIBA treatment, on both abaxial and adaxial surfaces both length and breadth decreased as concentration increased. In kinetin treatment all concentrations in the increasing order showed inhibitory effect on length and breadth on both the surfaces except Kn 10 ppm where a promotion in length and breadth of epidermal cells on both the surfaces was observed (table 2).

3.8 *Stomatal size*

Control plants had stomata which were 26·64 μ in length and 19·98 μ in breadth on the abaxial side while on the adaxial side stomata had an average length of 26·64 μ and breadth of 16·65 μ. On the abaxial surface, the length of stomata increased in 50 and 100 ppm IAA while it decreased in 200 and 500 ppm IAA. Breadth of the stomata remained same as in control except of 50 ppm. In general, both length and breadth of stomata on the adaxial surface showed reduction as concentration of IAA increased except in 50 ppm where there was no difference in comparison to control.

As far as stomatal size of GA_3 treated plants is concerned, on the abaxial side different concentrations except 500 ppm showed same length of stomata but width was greatly affected. On the adaxial surface length of stomata was reduced in all concentrations while breadth decreased in 50 and 100 ppm but was equal to control in 200 and 500 ppm. In TIBA, lower concentrations did not affect the length of stomata on the abaxial surface as it remained equal to that of control but the breadth showed reduction. On the adaxial surface as concentration increased, decrease in length was observed. In TIBA 50 ppm, the length and breadth were equal to that of control. In kinetin treatment, on both adaxial and abaxial surfaces length and breadth increased linearly as concentration increased except in Kn 10 ppm on the adaxial surface where breadth was reduced to nearly half of that of the control (table 2).

3.9 *Stomatal length/breadth ratio*

It was calculated for both the surfaces separately. In control plants, on the abaxial surface, the ratio was 1·33 and on the adaxial surface it was 1·6. The ratio was 1·63, 1·66, 1·16 and 1·33 on the abaxial surface and 1·6, 1·5, 1·5 and 1·75 on the adaxial surface in 50, 100, 200 and 500 ppm IAA respectively. In GA_3 treated leaf the ratio was 1·4, 1·75, 1·16 and 1·1 on the abaxial surface and 1·25, 1·5, 1·4 and 1·33 respectively on the adaxial surface in 50, 100, 200 and 500 ppm. TIBA and Kn concentrations increased the ratio on both the surfaces (table 2).

Table 2. Effect of different growth substances on epidermal characters in *T. erecta* L.

Treatment	Concentration (ppm)	Epidermal cell size				Stomatal size				Stomatal length/breadth ratio	
		Abaxial		Adaxial		Abaxial		Adaxial			
		Length (μ)	Breadth (μ)	Length (μ)	Breadth (μ)	Length (μ)	Breadth (μ)	Length (μ)	Breadth (μ)	Abaxial	Adaxial
Control	0	56·61 ±2·39	26·64 ±2·04	56·61 ±3·02	24·98 ±1·04	26·64 ±1·07	19·98 ±0·23	26·64 ±1·01	16·65 ±1·04	1·33	1·60
IAA	50	48·29 ±3·05	23·31 ±1·03	56·61** ±2·07	26·64 ±1·06	29·97 ±2·08	18·32 ±0·46	26·64** ±1·01	16·65** ±1·04	1·63	1·60
	100	53·28 ±2·07	26·64** ±1·04	59·95 ±2·09	24·98** ±1·04	33·30 ±2·30	19·98** ±0·54	26·31** ±1·02	16·65** ±1·04	1·66	1·50
	200	59·94 ±4·09	28·31 ±1·21	61·61 ±3·06	26·64 ±1·06	23·31 ±0·99	19·98** ±0·96	23·31 ±0·99	14·99 ±0·88	1·16	1·50
	500	63·27 ±5·95	28·31 ±1·22	63·27 ±4·04	28·31 ±0·99	26·64** ±1·07	19·96** ±0·96	23·31 ±2·08	13·32 ±0·44	1·33	1·75
GA_3	50	66·60 ±5·23	29·97 ±1·32	46·62 ±3·05	22·48 ±0·42	23·31 ±1·06	16·65 ±0·32	24·98 ±1·07	19·98 ±0·67	1·40	1·25
	100	64·94 ±4·98	28·31 ±0·97	56·61** ±3·07	26·64 ±0·94	23·31 ±1·08	13·32 ±0·21	29·97 ±2·39	19·98 ±0·67	1·75	1·50
	200	59·94 ±3·29	23·31 ±0·45	56·61** ±3·08	26·64 ±0·93	23·31 ±0·75	19·98** ±1·08	23·31 ±1·04	16·65** ±1·04	1·16	1·40
	500	56·61** ±3·29	26·64** ±0·98	53·28 ±3·05	23·31 ±1·02	19·98 ±0·45	19·98** ±1·08	19·98 ±0·97	14·99 ±0·89	1·00	1·33
TIBA	50	59·94 ±2·08	24·98 ±0·32	53·28 ±2·62	24·98** ±1·03	26·64** ±0·72	16·65 ±0·73	26·64** ±1·23	16·65** ±1·08	1·60	1·60
	100	56·61** ±3·07	24·98 ±0·43	49·95 ±1·82	23·31 ±1·02	26·64** ±0·45	18·32 ±0·62	23·31 ±1·24	16·65** ±1·09	1·45	1·40
	200	51·62 ±2·34	22·48 ±0·75	43·29 ±1·39	26·64 ±0·98	27·64 ±0·83	18·32 ±0·63	23·31 ±1·24	14·99 ±0·65	1·50	1·50
	500	49·95 ±3·02	21·65 ±0·69	46·62 ±2·60	21·65 ±1·07	29·97 ±1·42	19·98** ±1·08	23·31 ±1·24	19·98 ±0·88	1·50	1·60

Table 2 (*Continued*)

Treatment	Concentration (ppm)	Epidermal cell size				Stomatal size				Stomatal length/breadth ratio	
		Abaxial		Adaxial		Abaxial		Adaxial			
		Length (μ)	Breadth (μ)	Length (μ)	Breadth (μ)	Length (μ)	Breadth (μ)	Length (μ)	Breadth (μ)	Abaxial	Adaxial
Kn	10	57·44** ±3·04	26·64** ±1·01	59·94 ±2·90	25·81** ±1·08	21·65 ±0·82	16·65 ±0·94	19·98 ±0·97	13·32 ±1·02	1·30	1·50
	25	49·95 ±2·92	24·98 ±1·05	55·78** ±3·08	24.14* ±1·09	23·31 ±1·02	16·65 ±0·94	23·31 ±0·96	13·32 ±0·43	1·40	1·75
	50	49·12 ±3·03	23·31 ±0·97	53·28 ±3·02	23·31 ±0·98	26·64** ±1·21	18·33 ±0·78	23·31 ±1·24	16·65** ±1·04	1·45	1·40
	100	43·29 ±1·07	20·32 ±0·74	51·62 ±2·62	21·65 ±0·82	27·64* ±1·09	19·98** ±0·43	26·64** ±1·42	9·99 ±0·23	1·38	2·60

* Insignificant at 1%;
** Insignificant at 1 and 5%.

4. Discussion

The formation of persistent stomatal initials, single guard cells and other abnormalities observed here agree with those reported by Gangadhara *et al* (1977) who believe that the differentiation of a meristemoid into a stomatal complex may be controlled by the specific intrinsic factors at the region of the meristemoid and these factors are disturbed by the exogenous application of growth substances which ultimately lead to such aberrant developments. The present study shows that stomatal abnormalities are induced by the exogenous application of growth substances.

From this study it is concluded that in IAA treatment stomatal frequency increased up to 200 ppm while in GA_3, TIBA and Kn it increased in all concentrations on both the leaf surfaces. The present observations regarding stomatal frequency are in agreement with those of Gangadhara and Inamdar (1975) who also reported increase in stomatal frequency by GA_3 and IAA.

From the present findings it is clear that the number of epidermal cells are more on the adaxial surface than on the abaxial surface in control. All concentrations of IAA exhibited more epidermal cells on the abaxial side than on the adaxial side. In GA_3 treatment, the number of epidermal cells present on the adaxial surface is almost double that present on the abaxial side in 50 and 500 ppm while in 100 and 200 ppm it is the same on both the surfaces. It was observed that GA_3 treatment showed a gradual decrease in the number of epidermal cells as concentration increased. Such findings have also been recorded by Murty *et al* (1976). TIBA and Kn increased the frequency of epidermal cells on both the surfaces with increase in concentration.

Solereder (1908) suggested that length to breadth ratios may give the actual shape of the stomata. According to him the range of possible variation is limited, since the shape can only be (i) broader than long, (ii) roundish, (iii) broadly elliptical, (iv) narrowly elliptical and (v) angular. In this particular plant the third and fourth categories are common for all the treatments. The second is of more restricted occurrence, while the first and fifth are not observed in any case.

Control plants have stomata of different shapes on the two surfaces, which is clear from length/breadth ratio. The shape is broadly elliptical on the abaxial side and narrowly elliptical on the adaxial side. Among all the treatments used, roundish shape of stomata are observed in GA_3 500 ppm treatment on the abaxial surface. On the abaxial surface narrowly elliptical stomata are observed in 50 and 100 ppm of IAA, 100 ppm of GA_3, 50 ppm of TIBA, while in the remaining concentrations broadly elliptical stomata are observed. On the adaxial surface narrowly elliptical stomata are found in 50 and 500 ppm of IAA, 50 ppm of TIBA and 25 ppm of kinetin, remaining concentrations showed broadly elliptical stomata. In Kn 100 ppm a special feature observed on the adaxial surface is that stomata are very narrowly elliptical, *i.e.* the length is nearly three times the breadth.

Acknowledgements

One of the authors (Vidhu) acknowledges the UGC for a fellowship.

References

Cooper M J, Digby J and Cooper P J 1972 Effects of plant hormones on the stomata of barley: a study of interaction between abscisic acid and kinetin; *Planta* **105** 43–49

Gangadhara M and Inamdar J A 1975 Action of growth regulators on the cotyledonary stomata of *Cucumis sativus* L. structure and ontogeny; *Biol. Plant.* **17** 292–303

Gangadhara M, Rao J B, Inamdar J A and Patel R M 1977 Effect of growth regulators on the structure and development of cotyledonary and hypocotyledonary stomata of *Gossypium hirsutum* L var. 'digvijay'; *Phyton* **18** 9–28

Inamdar J A 1970 Action of growth regulators on the development of stomata of *Abelmoschus esculentus* Moench; *Flora* **159** 497–502

Inamdar J A, Gangadhara M and Sanjeeva Rao M 1974 Effect of growth regulators on stomata structure and ontogeny in the cotyledons of *Cucurbita maxima* Duch; *Geobios* **1** 113–117

Kasat M L 1979 Morphogenetic effect of various growth regulators on the foliar stomata of *Vigna sinensis; Geophytology* **9** 102–107

Murty Y S, Prakash G and Poonia O P 1976 Effect of gibberellic acid on cotyledonary epidermis of *Crotalaria juncea* L; *Sci. Cult.* **42** 438–440

*Salisbury E J 1927 On the cause and ecological significance of stomatal frequency with special reference to woodland flora; *Philos. Trans. R. Soc. Lond.* **B216** 1–65

*Salisbury E J 1932 The interrelations of soil, climate and organisms and the use of stomatal frequency as an integrating index of the water relation of the plant; *Beih. Bot. Zool.* **99** 402–420

Sharma G K and Dunn D B 1968 Effect of environment on the cellular features in *Kalanchoe fedchenkoi; Bull. Torrey. Bot. Club* **95** 464–473

Solereder H 1908 *Systematic anatomy of the dicotyledons* (Oxford: Clarendon Press)

---

* Original not consulted.