

CHARACTERIZATION OF SESAME (*Sesamum indicum* L.) GENOTYPES THROUGH CHEMICAL TESTS

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ABSTRACT

An experiment was carried out in the Seed Testing Laboratory of the Department of Seed Science and Technology, College of Agriculture, Junagadh Agricultural University, Junagadh, to characterize 25 sesame genotypes based on the chemical tests. The seeds were subjected to NaOH test, KOH test, coleoptile growth response to GA_3 and coleoptile growth response to 2, 4-D for differentiating the genotypes. On the basis of colour reaction with sodium hydroxide solution, the genotypes were grouped into four categories as no change (15 genotypes), light brown (5 genotypes), brown (4 genotypes) and dark brown (1 genotype) colour response. On the basis of colour reaction with KOH solution, the genotypes were categorized into four groups as light yellow (8 genotypes), light brown (10 genotypes), brown (5 genotypes) and dark brown (2 genotypes) colour response. Based on the differential growth response of coleoptile length to GA_3 , genotypes were grouped as low response (10-30 %) with four genotypes and moderate response (< 30 %) with twenty one genotypes. The per cent increase in coleoptile length over control ranged from 17.46 per cent (AT 213) to 49.49 per cent (AT 219). The genotypes showed varied response to 2, 4-D application (2 ppm). The per cent decrease in coleoptile length over control ranged from 76.90 per cent (RT 54) to 89.19 per cent (AT 285). Based on differential growth response of coleoptile length to 2, 4-D, the genotypes were grouped as susceptible (<85 %) with ten genotypes and highly susceptible (> 85 %) with fifteen genotypes.

KEY WORDS: SESAME, *Sesamum indicum* L., Genotypes, Sodium hydroxide

INTRODUCTION

Sesame (*Sesamum indicum* L., 2n = 26) is a very ancient oilseed crop grown next to groundnut and rapeseed and mustard in India. It belongs to the order *Tubiflorae*, family *Pedaliaceae*. It is basically considered a crop of tropical and sub-tropical regions, but it has also spread to the temperate parts of the world. Africa has been considered to be the primary centre of origin of sesame and it spread early through

West Asia to India, China and Japan, which themselves became secondary distribution centers (Weiss, 1983). The flower structure of sesame facilitates cross pollination, although it is considered as self pollinated crop. The extent of natural crossing ranges from 1 to 68 per cent (Abdel Aal *et al.*, 1976; Yermanos, 1980; Free, 1993; Sarker, 2004). *Apis florea*, *Apis dorsata*, *Megachile umbripennis*, *Andrena ilerreda*, *Ceratina sexmaculata* and *Trichometalle pollinosa*

were reported to be common pollinators on sesame (Rashad *et al.*, 1980). The amount of natural out-crossing depending upon the pressure of pollinating agents. In sesame, wind plays no part in natural cross pollination.

According to Kobayashi *et al.* (1990), 36 species have been identified in sesame, of which 22 species have been found in Africa, five in Asia, seven in both Africa and Asia and one species each in Crete and Brazil. Analysis of the chromosome number of 12 sesame species showed that there are three groups: $2n = 26$ (*S. indicum* and the wild *S. alatum*, *S. capense*, *S. schenckii* and *S. malabaricum*); $2n = 32$ (*S. prostratum*, *S. laciniatum*, *S. angolense* and *S. angustifolium*); $2n = 64$ (*S. radiatum*, *S. occidentale* and *S. schinzianum*). There is limited cross compatibility among the species mainly due to the differences in chromosomal numbers across the three cytotaxonomic groups. Therefore, it has been difficult to transfer desirable characteristics such as drought tolerance and resistance to diseases and pest, from wild relatives into cultivated sesame (Carlsson *et al.*, 2008).

The chemical tests are spot tests and useful in identification by change in seed colour as well as solution due to added chemicals. Simple biochemical tests *viz.*, phenol colour reaction, NaOH test, KOH test, seedling response to various chemicals *eg.* growth regulators, herbicides etc., have also been proved quite useful in detecting varietal mixtures as well as grouping large number of genotypes into distinct classes (Chakrabarthy and Agrawal, 1990). These tests do not much require virtually no technical expertise or training and can be completed in a relatively short time. The results of these tests are usually distinct, easily interpreted and help in grouping of the genotypes. In the light of the above facts,

the present study was planned to study the suitability of chemical tests for the identification of sesame genotypes.

MATERIALS AND METHODS

The experiment was conducted in the Seed Testing Laboratory of the Department of Seed Science and Technology, College of Agriculture, Junagadh Agricultural University, Junagadh, during *Kharif* 2016 to study the varietal characterization in 25 sesame genotypes *viz.*, GT 1, GT 2, GT 3, GT 4, GJT 5, GT 10, TKG 22, RT 54, PRAGATI, AT 164, AT 178, AT 213, AT 219, AT 222, AT 229, AT 235, AT 238, AT 242, AT 243, AT 252, AT 255, AT 264, AT 285, AT 287 and AT 290 based on chemical tests *viz.*, NaOH test, KOH test, coleoptile growth response to GA_3 , and coleoptile growth response to 2, 4-D.

Sodium hydroxide (NaOH) test

The seeds (one gram) of sesame genotypes were washed in distilled water and then soaked in 10 ml of five per cent NaOH solution in test tube for one hour at an ambient temperature. The solution was decanted and used for observation. Based on the change in colour of the solution, the genotypes were grouped as light brown, brown and dark brown.

Potassium hydroxide (KOH) test

The seeds (one gram) of sesame genotypes were washed in distilled water and then soaked in 10 ml of six per cent KOH solution for one hour in test tube at an ambient temperature. The solution was decanted and used for visual observation. Based on the change in colour of the solution, the genotypes were grouped as light yellow, yellow, light brown, brown and dark brown.

Seedling growth response to GA_3

The seeds of sesame genotypes were surface sterilized by washing in distilled water. Fifty seeds each in three repetitions were placed on moistened filter paper in petridishes with 25 ppm GA_3 solution and

incubated at $25 \pm 1^{\circ}\text{C}$ as per ISTA procedure (Anon., 1985). The water soaked blotter papers was used as the control. On seventh day, the coleoptile length of twenty five randomly selected seedlings was measured

and the growth response was recorded as per cent increase in coleoptile length over control. The per cent increase in coleoptile length over control was calculated using the following formula:

$$\text{Per cent increase in coleoptile length over control} = \frac{\text{Coleoptile length in GA}_3 - \text{Coleoptile length in control}}{\text{Coleoptile length in control}} \times 100$$

The genotypes were grouped based on per cent increase of coleoptile length over control as follows:

Category	Percentage increase in coleoptiles length over control
Very low response	: < 10 per cent increase
Low response	: 10-30 per cent increase
Moderate response	: > 30 per cent increase

Seedling growth response to 2, 4-D

The seeds of sesame genotypes were surface sterilized by washing in distilled water. Fifty seeds each in three replications were placed on moistened filter paper in petridishes with 2 ppm of 2, 4-D solution and then incubated at $25 \pm 1^{\circ}\text{C}$ as per ISTA procedure (Anon., 1985). The water soaked

blotter papers were used as control. On seventh day, coleoptile length of twenty five randomly selected seedlings was measured and the sensitivity response of genotypes was recorded as per cent decrease in coleoptile length over control. The decrease in coleoptiles length over control was calculated using the formula such as,

$$\text{Per cent decrease in coleoptile length over control} = \frac{\text{Coleoptile length in 2,4-D} - \text{Coleoptile length in control}}{\text{Coleoptile length in control}} \times 100$$

The genotypes were grouped based on per cent decrease of coleoptile length over control as follows:

Category	Percentage decrease in coleoptiles length over control
Susceptible	: < 85 per cent
Highly susceptible	: > 85 per cent

RESULTS AND DISCUSSION

Varietal identification by morphological characters is laborious, time consuming, tedious, cumbersome and costly affair. A number of chemical tests have been

developed for varietal identification such as sodium hydroxide test, potassium hydroxide test, gibberllic acid response test and 2, 4-D soak test. These chemical tests are very quick, easy and reproducible (Agrawal and

Sharma, 1989; Kumar *et al.*, 1995). Very often these tests provide supportive evidence for the morphological evaluation of the seeds (Vanderburg and Vanzood, 1991).

The seeds were subjected to NaOH, KOH, gibberllic acid response and 2, 4-D soak test for differentiating the genotypes (Table 1, 2 and 3; Plate 1 and 2). On the basis of colour reaction with sodium hydroxide solution, the genotypes were grouped into four categories as no change (15 genotypes), light brown (5 genotypes), brown (4 genotypes) and dark brown (1 genotype) colour response. On the basis of colour reaction with KOH solution, the genotypes were categorized into four groups as light yellow (8 genotypes), light brown (10 genotypes), brown (5 genotypes) and dark brown (2 genotypes) colour response. The varied coleoptile growth response of sesame genotypes to gibberllic acid (25 ppm) has been observed. Based on the differential growth response of coleoptile length to GA₃, the genotypes were grouped into two categories as low response (10-30 %) with four genotypes and moderate response (< 30 %) with twenty one genotypes. The per cent increase in coleoptile length over control ranged from 17.46 per cent (AT 213) to 49.49 per cent (AT 219). The genotypes showed varied response to 2, 4-D application (2 ppm). The per cent decrease in coleoptile length over control ranged from 76.90 per cent (RT 54) to 89.19 per cent (AT 285). Based on the differential growth response of coleoptile length to 2,4-D, the genotypes were grouped into two groups as susceptible (< 85 %) with ten genotypes and highly susceptible (> 85 %) with fifteen genotypes.

On the basis of seed and seedling response to chemical tests, genotype identification keys were prepared (Figure 1). The genotypes viz., GT 2, AT 222, AT 243, AT 285 and AT 290 were having similar response to chemical tests viz., no change in

colour reaction in NaOH test, light yellow colour reaction in KOH test, moderate differential growth response of coleoptile length to GA₃ and highly susceptible to 2, 4-D. The genotypes GJT 5 and AT 287 were differed from the above genotypes with respect to low differential growth response of coleoptile length to GA₃ and AT 242 with susceptible to 2, 4 - D.

The genotypes viz., GT 3, AT 255, PRAGATI, AT 229 and AT 238 were having similar response to chemical tests viz., no change in colour reaction in NaOH test, light brown colour reaction in KOH test, moderate differential growth response of coleoptile length to GA₃ and highly susceptible to 2, 4- D. Genotype AT 219 was differing from the above genotypes with susceptible to 2, 4-D, AT 213 with respect to low differential growth response of coleoptile length to GA₃ and susceptible to 2, 4-D, and genotype GT 10 with respect to dark brown colour reaction in NaOH and KOH test.

The genotypes viz., TKG 22, AT 264 and AT 178 were having similar response to chemical tests viz., light brown colour reaction in NaOH test, light brown colour reaction in KOH test, moderate differential growth response of coleoptile length to GA₃ and susceptible to 2, 4 - D.

The genotypes viz., RT 54 and GT 4 were having light brown colour reaction in NaOH test, brown colour reaction in KOH test, moderate differential growth response of coleoptile length to GA₃ and susceptible to 2, 4-D. Genotype AT 235 was differing from the above genotypes with respect to brown colour reaction in NaOH test and AT 252 with respect to brown colour reaction in NaOH test and dark brown colour reaction in KOH test.

The genotypes viz., GT 1 and AT 164 were having brown colour reaction in NaOH test, brown colour reaction in KOH test, low differential growth response of

coleoptile length to GA₃ and highly susceptible to 2, 4 - D. Genotype AT 164 was differing from the above genotypes with respect to moderate differential growth response of coleoptile length to GA₃.

The findings of the present investigation (NaOH and KOH tests) which are simple, quick, and cheap for determining the varietal differences in sesame genotypes could be used as routine genetic purity test. Similar observations and grouping was earlier reported by Suhasini (2006) and Mesfin *et al.* (2013) in sesame; Rao *et al.* (2002), Keshavulu *et al.* (2003) and Rao *et al.* (2013) in groundnut; Jawaharlal (1994), Ponnuswamy *et al.* (2003) and Reddy *et al.* (2008) in cotton; Biradarpatil *et al.* (2008) in safflower; Chavan (2010) in soybean; and Sathisha *et al.* (2012) and Kallihal *et al.* (2013) in sunflower.

Based on the seedling response to GA₃, observations and grouping was made by Suhasini (2006) and Mesfin *et al.* (2013) in sesame; Prakash and Lal (1968), Kurdikeri and Kurdikeri (1988), Singh and Afria (1990), Jawaharlal (1994), Ponnuswamy *et al.* (2003) and Reddy (2004) in cotton; Agrawal and Pawar (1990) in soybean; Rao *et al.* (2002) and Rao *et al.* (2013) in groundnut; Biradarpatil *et al.* (2008) in safflower; and Sathisha *et al.* (2012) and Kallihal *et al.* (2013) in sunflower.

Based on the seedling response to 2, 4 - D, observations and grouping was made by Suhasini (2006) and Mesfin *et al.* (2013) in sesame; Wax *et al.* (1974) in soybean; Shivakumar (2000) in rapeseed and mustard; Rao *et al.* (2002) and Rao *et al.* (2013) in groundnut; Ponnuswamy *et al.* (2003) and Reddy (2004) in cotton; Biradarpatil *et al.* (2008) in safflower; and Sathisha *et al.* (2012) and Kallihal *et al.* (2013) in sunflower.

CONCLUSION

Based on the results and discussion, it can be concluded that cultivar reaction to different chemicals like, NaOH and KOH solutions, differential growth response of coleoptile length to GA₃ and seedling response to 2,4-D was also found useful in grouping of sesame genotypes.

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Table 1: Identification and grouping of sesame genotypes based on sodium hydroxide (NaOH) and potassium hydroxide (KOH) test

Genotypes	Sodium hydroxide (NaOH) test	Potassium hydroxide (KOH) test
GT 1	Brown	Brown
GT 2	No change	Light yellow
GT 3	No change	Light brown
GT 4	Light brown	Brown
GJT 5	No change	Light yellow
GT 10	Dark brown	Dark brown
TKG 22	Light brown	Light brown
RT 54	Light brown	Brown
PRAGATI	No change	Light brown
AT 164	Brown	Brown
AT 178	Light brown	Light brown
AT 213	No change	Light brown
AT 219	No change	Light brown
AT 222	No change	Light yellow
AT 229	No change	Light brown
AT 235	Brown	Brown
AT 238	No change	Light brown
AT 242	No change	Light yellow
AT 243	No change	Light yellow
AT 252	Brown	Dark brown
AT 255	No change	Light brown
AT 264	Light brown	Light brown
AT 285	No change	Light yellow
AT 287	No change	Light yellow
AT 290	No change	Light yellow

Table 2: Identification and grouping of sesame genotypes based on coleoptile growth response to GA₃

Genotypes	Coleoptile growth (cm)		Per cent increase in coleoptile length over control	Groups
	Control	GA ₃		
GT 1	4.45	5.50	23.60	Low response
GT 2	4.22	6.18	46.54	Moderate response
GT 3	4.12	5.60	35.92	Moderate response
GT 4	4.54	6.32	39.30	Moderate response
GJT 5	4.50	5.70	26.58	Low response
GT 10	4.22	5.99	41.99	Moderate response
TKG 22	4.12	5.99	45.44	Moderate response
RT 54	3.55	4.80	35.21	Moderate response
PRAGATI	4.55	6.60	45.05	Moderate response
AT 164	3.95	5.60	41.77	Moderate response
AT 178	3.45	4.89	41.88	Moderate response
AT 213	4.25	4.99	17.46	Low response
AT 219	3.51	5.24	49.49	Moderate response
AT 222	4.25	5.97	40.52	Moderate response
AT 229	4.25	5.90	38.73	Moderate response
AT 235	4.22	5.95	41.04	Moderate response
AT 238	4.10	6.00	46.44	Moderate response
AT 242	3.88	5.44	40.21	Moderate response
AT 243	3.65	5.27	44.52	Moderate response
AT 252	3.89	5.67	45.71	Moderate response
AT 255	4.21	6.24	48.31	Moderate response
AT 264	3.95	5.20	31.65	Moderate response
AT 285	4.06	5.53	36.29	Moderate response
AT 287	4.87	5.87	20.49	Low response
AT 290	4.36	6.31	44.68	Moderate response
Mean	4.12	5.71	38.85	

Table 3: Identification and grouping of sesame genotypes based on coleoptile growth response to 2, 4-D

Genotypes	Coleoptile growth (cm)		Per cent decrease in coleoptile length over control	Groups
	Control	2, 4-D		
GT 1	4.45	0.42	86.08	Highly susceptible
GT 2	4.22	0.46	89.15	Highly susceptible
GT 3	4.12	0.45	85.55	Highly susceptible
GT 4	4.54	0.48	78.66	Susceptible
GJT 5	4.50	0.48	85.93	Highly susceptible
GT 10	4.22	0.36	89.04	Highly susceptible
TKG 22	4.12	0.47	81.06	Susceptible
RT 54	3.55	0.53	76.90	Susceptible
PRAGATI	4.55	0.44	86.17	Highly susceptible
AT 164	3.95	0.47	85.66	Highly susceptible
AT 178	3.45	0.52	84.93	Susceptible
AT 213	4.25	0.49	82.10	Susceptible
AT 219	3.51	0.54	84.68	Susceptible
AT 222	4.25	0.48	86.62	Highly susceptible
AT 229	4.25	0.47	85.80	Highly susceptible
AT 235	4.22	0.44	83.82	Susceptible
AT 238	4.10	0.45	85.73	Highly susceptible
AT 242	3.88	0.57	82.38	Susceptible
AT 243	3.65	0.49	86.44	Highly susceptible
AT 252	3.89	0.50	82.58	Susceptible
AT 255	4.21	0.50	85.21	Highly susceptible
AT 264	3.95	0.51	83.42	Susceptible
AT 285	4.06	0.44	89.19	Highly susceptible
AT 287	4.87	0.45	86.26	Highly susceptible
AT 290	4.36	0.51	88.23	Highly susceptible
Mean	3.24	0.48	84.86	

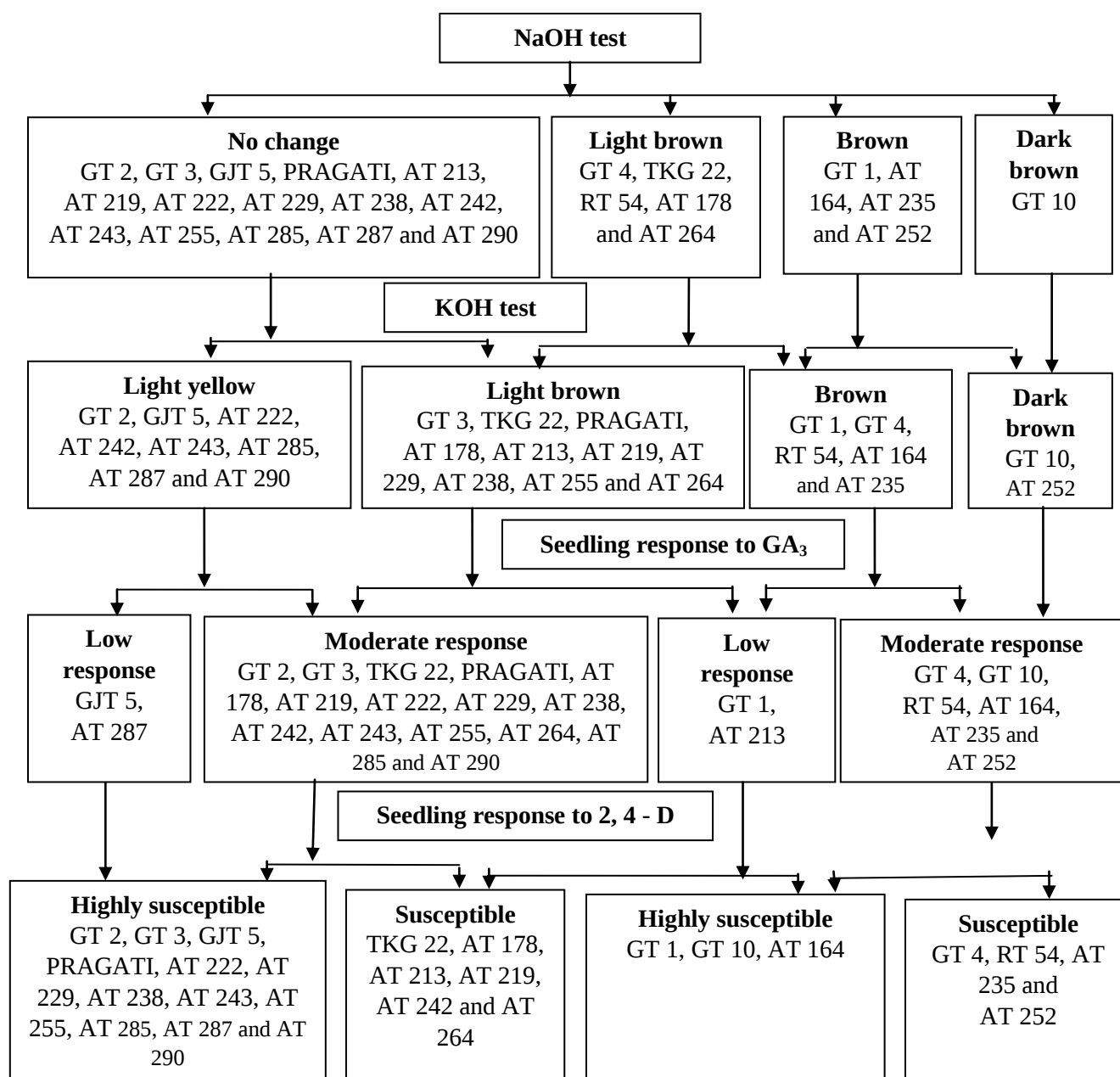


Figure 1: Sesame genotypes identification keys on the basis of seed and seedling response to chemical tests

SODIUM HYDROXIDE TEST



NO CHANGE



LIGHT BROWN



BROWN



DARK BROWN

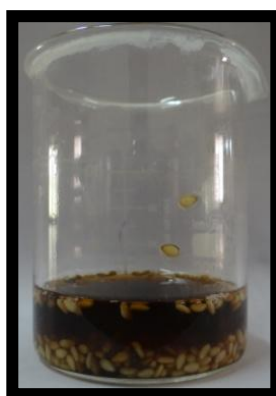
POTASSIUM HYDROXIDE TEST



LIGHT YELLOW



LIGHT BROWN



BROWN



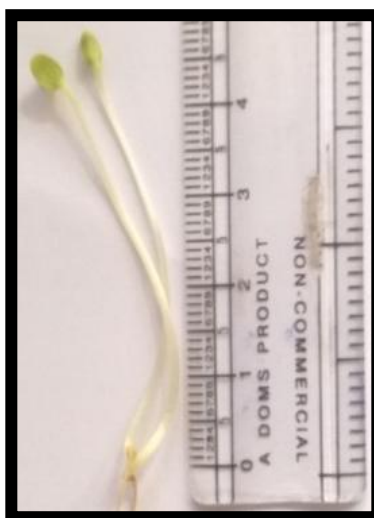
DARK BROWN

Plate 1: Sodium hydroxide (NaOH) and potassium hydroxide (KOH) test of sesame genotypes

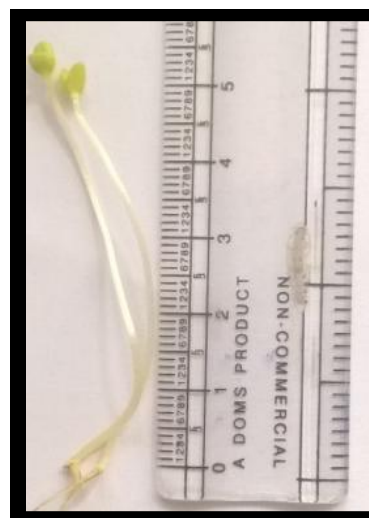
COLEOPTILE GROWTH RESPONSE TO GA_3



CONTROL



LOW RESPONSE



MODERATE RESPONSE

COLEOPTILE GROWTH RESPONSE TO 2, 4-D



CONTROL



SUSCEPTIBLE



HIGHLY SUSCEPTIBLE

Plate 2: Coleoptile growth response to GA_3 and 2, 4-D of sesame genotypes

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