

INDUCED VARIATION FOR YIELD AND QUALITY CHARACTERS OF GINGER (*Zingiber officinale* L.) USING ETHYL METHANE SULPHONATE

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ABSTRACT

Ginger (*Zingiber officinale* L.) is one of the important spice crops grown in different parts of the world. Unavailability of high yielding and good quality varieties is one of the major problems in ginger cultivation in Sri Lanka. Ginger is a vegetatively propagated crop and the conventional breeding procedure like hybridization cannot be adopted for its improvement. Moreover, the genetic variability available in the crop is very limited. The present study was conducted to create variability in ginger for yield and quality characters through induced mutation using Ethyl Methane Sulphonate (EMS) and to compare this variability with the variability available in the germplasm. Different concentrations of EMS with different soaking periods were used to induce mutation. High yielding mutants with better agronomical characters were identified in VM₁ generation. Selected mutants were grown as progeny lines in VM₂ generation with two replications. Out of 50 lines, 8 were selected for high yield with improved quality characters such as low crude fibre, high oleoresin and essential oil.

KEYWORDS: EMS, Ginger, Mutation.

INTRODUCTION

Ginger, *Zingiber officinale* Rosc. is an important spice crop grown in different parts of the world. It is a perennial, herbaceous, monocotyledonous plant belonging to the family Zingiberaceae, which is commonly cultivated as an annual (Purseglove *et al.*, 1981). The economic part of ginger is the underground rhizome, which is pungent and aromatic in nature. It is used mainly in medicine and for seasoning foods that gives an acceptable flavour and aroma. In Sri Lanka, ginger is grown in an area of 13,500 ha with an annual production of 6700t (FAOSTAT 2005). However, half the requirement is imported annually to meet the high demand in the country. Low yield and high disease incidence are major problems in Sri Lankan ginger cultivation.

Developing high yielding cultivar(s) having good quality attributes and resistance to rhizome rot disease is important to increase the production in the country. Ginger is a vegetatively propagated crop and the conventional breeding procedure like hybridization cannot be adopted for its improvement because of shy/no flowering behaviour of the plant and total absence of seed set due to stigmatic and stylar incompatibility. Consequently, the genetic variability available in the crop is very limited. Selection for yield, superior quality and resistance to diseases and pests has not shown fruitful

results, because of the low genetic variability. Therefore, the other alternative for improving the crop is to create variability through mutation. Induction of mutation has been considered as an established method for increasing genetic variability in many crops and according to FAO/IAEA database (2006) more than 2,540 varieties in different crops have been commercially released through mutation breeding. This also includes successful release of improved varieties in vegetatively propagated crops like garlic, cassava, turmeric and potato. Keeping this in view, the present studies on ginger were undertaken to study the effectivity of Ethyl Methane Sulphonate (EMS), a widely used chemical mutagen, to create variability in ginger and to study the variability of different horticultural characters, disease resistance and quality and identify the desirable mutants.

MATERIALS AND METHODS

The present investigations were conducted at the experimental farm of the Department of Vegetable Crops, Dr. Yaswant Singh Parmar University of Horticulture and Forestry, Nauni, Solan, Himachal Pradesh, India during 2004 and 2005.

The rhizome bits of Indian cultivar 'Himgiri' were used for the mutation studies. The seed rhizome bits weighing 5g each and with one/two buds were used for the studies. For inducing variability fifty rhizome bits in each treatment were soaked in ethyl methane sulphonate (EMS- $\text{CH}_3\text{SO}_2\text{OC}_2\text{H}_5$) solutions prepared in distilled water as per concentrations and soaking/dipping periods given below.

T1 - 0.2% EMS for 2 h	T2 - 0.2% EMS for 4 h
T3 - 0.2% EMS for 6 h	T4 - 0.2% EMS for 8 h
T5 - 0.4% EMS for 2 h	T6 - 0.4% EMS for 4 h
T7 - 0.4% EMS for 6 h	T8 - 0.4% EMS for 8 h
T9 - 0.6% EMS for 2 h	T10 - 0.6% EMS for 4 h
T11 - 0.6% EMS for 6 h	T12 - 0.6% EMS for 8 h

After treatment, the rhizome bits were thoroughly washed in running water and 2.0% Sodium hypochloride solution and dried in shade for 24 h. Fifty similar rhizome bits were used as control (without treatment).

The treated rhizome bits along with control were planted on raised beds of 3.0m x 1.0m x 0.15m to raise first vegetatively propagated mutagenic (VM1) generation. All the plants within treatments were observed for plant height, number of tillers and leaf size. Similarly, all the plants were harvested separately, cleaned and used for recording data on yield after maturity. These rhizomes were, then stored separately in pits with sand and

dried grass for raising second vegetatively propagated mutagenic (VM2) generation.

In 2005, stored rhizomes of selected progenies of VM1 were planted as progeny rows to raise VM2 generation. Sowing was done using similar plot size as in VM1. These were observed for growth, yield and rhizome quality characters. The standard cultural practices except the use of pesticides were followed during both seasons for raising the experimental materials.

RESULTS AND DISCUSSION

There was a significant influence of EMS on plant stand, morphological and yield characters of VM1 generation. EMS treatments drastically reduced percentage germination and values varied from 12 to 60% compared to 92% in control treatment (Table 1). Maximum germination of 60% was observed at the lowest concentration of EMS (0.2%) with the least time of soaking (2h). Increasing concentration and soaking period reduced germination accordingly. The reduction in germination by increasing concentration of EMS was also reported in various vegetative propagated crops like *Gladiolus* (Misra and Bajpai, 1983a), *Lycoris aurea* a tuberous flowering plant (Hsu *et al.*, 2001) and Chinese potato (Abraham and Radhakrishnan, 2004). Patil and Patil (2005) reported that mutagens induced chromosomal aberrations in treated buds of grape cuttings resulting in increased aberrant cells. Such aberrations are parallel with concentration and duration of the mutagenic treatments. Datta and Biswas (1985) also observed the frequency of aberrant cells increase with increasing concentration of mutagens in ginger. Higher chromosomal aberrations might be the reason for reduction of germination.

Changing plant height, number of tillers and leaf size, both in positive and negative direction was observed in different EMS treatments in VM1 generation (Table 2). Lower concentration of EMS with less soaking period showed increase in plant height, no of tillers and leaf size while higher concentration with maximum soaking period reduced these characters compared to the control. Abraham and Radhakrishnan (2004) had observed changes in plant height in both positive and negative direction by EMS treatments in Chinese potato. Misra and Bajai (1983c) recorded drastic reduction in plant height of various *Gladiolus* cultivars with different concentrations of EMS, DES (Diethyl sulfate) and NMU (Nitroso methyl urea). They proposed that it may be due to physiological disturbances and retarded cell division by arresting the mitotic division and adverse effect on auxin. Similarly, Gaul (1970) explained that the decrease in plant height by treating with higher doses of gamma rays and chemical mutagens may either be due to physiological damage or chromosomal aberrations. The enhanced

tillering in some plants is due to the creation of variability or rapid growth of unaffected tissues in treated plants replacing affected (mutated) ones during the course of growth and development. Similar observations were also made by Jayachandran and Mohankumaran (1992) in their study on ginger with γ irradiation.

Table 1. Effect of ethyl methane sulphonate on germination of rhizomes of ginger.

<i>Treatment</i>	<i>% Germination</i>
T ₁ - 0.2% EMS 2h	60.0
T ₂ - 0.2% EMS 4h	53.0
T ₃ - 0.2% EMS 6h	48.0
T ₄ - 0.2% EMS 8h	44.0
T ₅ - 0.4% EMS 2h	48.0
T ₆ - 0.4% EMS 4h	46.0
T ₇ - 0.4% EMS 6h	38.0
T ₈ - 0.4% EMS 8h	30.0
T ₉ - 0.6% EMS 2h	40.0
T ₁₀ - 0.6% EMS 4h	28.0
T ₁₁ - 0.6% EMS 6h	26.0
T ₁₂ - 0.6% EMS 8h	12.0
T ₁₃ - Control	92.0

EMS created higher variability for rhizome yield per plant as the population exhibited wider range (5 to 400g) for rhizome yield. In general, lower doses and shorter soaking periods gave high rhizome yield while higher dose and longer soaking period gave less yield compared to control. Abraham and Radhakrishnan (2004) had also observed positive and negative changes in yield of Chinese potato with different treatments of EMS. Stimulatory effect of EMS at lower concentration for corm weight and size of *Gladiolus* had been reported by Misra and Bajpai (1983b). The reduction of yield may be attributed to the fact that due to treatment damage, physiology of the plant was disturbed, which affected photosynthesis and respiration resulting in improper growth of plant (Misra and Bajpai, 1983b).

Fifty high yielding plants of VM1 generation were selected and planted separately for raising VM2 generation. Mean performances of these lines compared to control are presented in Table 3. Performance of selected progenies were higher than the control in all characters except dry matter content. The quality of the ginger is mostly adjudged by its aroma which is due to volatile compounds present in rhizomes. Therefore, essential oil and oleoresin are the major factors contributing for quality. In the present investigation selected mutants from VM₂ generation showed an increase in oleoresin content, a maximum of 6.62% being recorded which was 50% higher than the control.

Table 2. Effect of ethyl methane sulphonate on morphological characters and yield of VM₁ generation.

Treatment	Plant height (cm)		Number of tillers		Size of the leaves (cm ²)		Rhizome yield per plant (g)	
	Mean	Range	Mean	Range	Mean	Range	Mean	Range
T ₁ - 0.2% EMS 2h	52.85±3.12	20.10-70.10	8.12±0.66	3.00-14.00	49.32±3.34	18.80-58.60	150.95±20.74	5.00-400.00
T ₂ - 0.2% EMS 4h	50.86±2.37	22.10-62.30	6.50±0.52	2.00-14.00	44.57±2.56	16.00-52.10	135.88±15.31	20.00-230.00
T ₃ - 0.2% EMS 6h	41.54±2.86	11.00-61.00	6.42±0.64	1.00-13.00	38.43±2.47	15.30-49.10	94.74±13.24	10.00-220.00
T ₄ - 0.2% EMS 8h	40.50±2.12	18.00-58.50	5.29±0.53	2.00-12.00	31.21±2.34	15.10-42.60	81.39±12.85	5.00-250.00
T ₅ - 0.4% EMS 2h	38.74±1.84	21.00-56.00	5.61±0.58	2.00-13.00	39.63±2.41	20.70-51.30	87.07±11.13	5.00-190.00
T ₆ - 0.4% EMS 4h	37.18±2.77	18.10-56.30	5.50±0.42	1.00- 8.00	34.38±1.47	13.40-39.60	85.29±5.93	10.00-120.00
T ₇ - 0.4% EMS 6h	37.00±1.97	20.00-50.00	5.39±0.47	2.00-10.00	30.21±1.43	16.60-38.30	84.44±12.51	20.00-200.00
T ₈ - 0.4% EMS 8h	29.82±2.46	7.30-46.10	4.30±0.47	1.00- 9.00	28.96±2.30	5.50-32.60	38.29±7.68	5.00-120.00
T ₉ - 0.6% EMS 2h	37.57±2.70	18.10-52.00	5.20±0.43	2.00- 8.00	36.60±2.64	13.00-46.50	64.83±7.15	20.00-120.00
T ₁₀ - 0.6% EMS 4h	32.25±3.46	6.00-50.00	5.44±0.66	2.00- 9.00	28.73±1.32	17.10-38.60	70.15±14.92	20.00-210.00
T ₁₁ - 0.6% EMS 6h	25.80±3.30	5.00-46.20	4.86±0.62	1.00- 9.00	26.54±1.10	20.30-32.60	33.67±6.84	5.00-80.00
T ₁₂ - 0.6% EMS 8h	28.50±2.22	20.10-32.00	4.00±0.77	3.00- 5.00	22.11±1.95	5.60-28.90	22.50±10.75	10.00-70.00
T ₁₃ - Control	51.07±1.02	43.10-64.50	6.39±0.22	5.00-10.00	37.60±0.43	32.30-46.50	130.00±5.17	70.00-200.00

Table 3. Effect of ethyl methane sulphonate on mean characters of selected VM₂ progenies.

Character	Mean		Range		Coefficient of variation	
	VM ₂	Control	VM ₂	Control	VM ₂	Control
Plant height (cm)	63.30	63.20	54.30-70.10	43.1-64.50	13.35	7.12
Number of tillers	10.60	6.40	6.0-14.0	5.0-10.0	23.90	12.05
Size of the leaves (cm ²)	46.01	38.90	35.4-58.60	36.1-43.20	17.73	6.51
Yield in VM ₂ (g/plant)	145.47	136.00	80.00-290.00	60.0-180.0	33.38	28.52
Dry matter content (%)	19.15	21.34	16.03-26.43	20.12-21.58	10.58	2.34
Oleoresin (%)	4.99	4.40	3.75-6.62	4.10-4.61	12.72	4.92
Essential oil (ml/100g)	1.32	1.00	0.50-2.00	1.00-1.00	29.11	0.0
Crude fibre (%)	6.17	5.17	4.55-8.40	4.8-5.7	12.63	7.46

Table 4. Morphological, yield and quality characters of selected 8 progenies and control.

Progeny line	Treatment	Plant height (cm)	No of tillers	Leaf area (cm ²)	Rhizome yield per plant (g)	Dry matter %	Oleoresin %	Essential oil (ml/100g)	Crude fibre %
T1-11	0.2% EMS 2h	60.5	09	49.6	150.0	26.43	5.43	1.5	4.55
T1-17	0.2% EMS 2h	62.8	12	53.3	200.0	17.90	6.37	2.0	6.65
T1-20	0.2% EMS 2h	69.3	14	58.6	290.0	20.13	4.55	2.0	5.50
T2-4	0.2% EMS 4h	61.7	10	38.9	182.5	18.31	6.62	2.0	5.90
T2-8	0.2% EMS 4h	63.5	12	39.6	227.5	18.08	5.32	2.0	5.50
T2-14	0.2% EMS 4h	64.3	14	52.7	240.0	18.33	4.80	1.5	4.90
T4-7	0.4% EMS 2h	54.3	08	41.2	180.0	20.12	5.63	2.0	5.70
T9-3	0.6% EMS 2h	65.6	11	45.3	198.0	20.15	5.61	2.0	6.80
Himgiri	Control	63.2	07	46.01	136.0	21.34	4.40	1.0	5.17

Essential oil content of most of the selected mutants from different EMS treatments was higher than the control. Rekha *et al.* (2000) showed that in Davana (*Artemisia pallens* Wall.), there was an appreciable increase in oil content in EMS treated plants compared to the control and this increase was not linear to concentrations of EMS applied. Low crude fibre content is considered as one of the important quality attribute in dried ginger. The mean crude fibre content of selected VM₂ progenies was higher than the control. However, three mutants gave lower crude fibre content compared to the control. Sarkar *et al.* (1985) observed that mutants derived from EMS and DES (Diethyl sulfate) treated cuttings in sweet potato gave lower crude fibre compared to the control. Dry matter content of mutants varied from 16.03 to 26.43% and four mutants obtained gave dry matter content higher than the control. Mutants obtained through chemical mutagens having high dry matter content were also observed in potato (Kolesnikova and Maksimova, 1977) and sweet potato (Sarkar *et al.*, 1985).

There was a higher variability between these 50 lines for certain characters as shown by higher coefficient of variability of those characters. Therefore, eight promising lines were selected and the performance of these lines are presented in Table 4. All these eight lines were high yielding compared to control variety Himgiri and also showed good quality attributes. In various vegetative propagated crops, varieties have been developed through mutation breeding and released for cultivation, for example, sweet potato (Vasudevan *et al.*, 1996), potato (Love *et al.*, 1996) and turmeric (Raju *et al.*, 1980). Therefore, these 8 lines can be further evaluated and released for cultivation.

CONCLUSIONS

The results of the present study indicated that EMS treatment created variability in ginger. Selected lines of VM₂ can be used for further evaluation and released as varieties for cultivation.

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REFERENCES

- Abraham, M. and V.V. Radhakrishnan. 2004. Assessment and induction of variability in Chinese potato (*Solenostemon rotundifolius* Poir.) Journal of Root Crops 3(2):143-150.
- Datta, K. and A.K. Biswas. 1985. EMS induced mitotic consequences in three rhizomatic spice yielding plants. Chromosome Information Service 38:25-26.
- FAO/IAEA database, 2006. <http://www.mrd.iaea.org/MVD/default.html>.
- FAOSTAT, 2005. <http://www.faostat.fao.org/ste/default.aspx>.
- Gaul, H. 1970. Plant injury and lethality. In. Manual on Mutation Breeding. FAO IAEA, Vienna. pp 87-91.
- Hsu, W.S., L.C. Huaney, C. Liou, Y.H. Cheng, Y.M. Chang and C.H. Hsiao. 2001. Study on the mutation in *Lycoris aurea* via chemical mutagen treatment. Journal of Agriculture Research of China 50(2): 67-77.
- Jayachandran, B. and N. Mohankumaran. 1992. Effects of gamma irradiation in ginger. South Indian Horticulture 40(5): 280-288.
- Kolesnikova, L.G. and A.D. Maksimova. 1977. Effect of chemical mutagens on content of dry matter and starch in potato tubers. Genetica USSR 13(10):1742-1753.
- Love, S.L., T. Baer, A. Thomson Johns and B.K. Werner. 1996. Mutation breeding for improved internal quality and appearance of Russet Burbank. American Potato Journal 73(4):155-165.
- Misra, R.L. and P.N. Bajpai. 1983a. Mutational studies in Gladiolus. I. Effect of physical and chemical mutagens in sprouting and survival of corms. Haryana Journal of Horticultural Sciences 12(1-2):1-6.
- Misra, R.L. and P.N. Bajpai. 1983b. Mutational studies in Gladiolus. II. Effect of mutagens on corm multiplication and storage. The Punjab Horticultural Journal 23(3-4):233-237.
- Misra, R.L. and N. Bajpai. 1983c. Effect of mutagens on shooting, leaf number, heading, plant height and spike length in gladioli. Indian Journal of Horticulture 40 (1-4):107-111.
- Patil, S.G. and V.P. Patil. 2005. Mutation studies in Anabe shahi grape (*Vitis vinifera* L.). Indian Journal of Horticulture 62(3):223-226.
- Purseglove, J.W., E.G. Brown, C.L. Green and S.R.J. Robbins. 1981. Spices. Vol.2. Longman, London. pp 447-531.
- Raju, E.C., J.D. Patel and J.J. Shah. 1980. Effect of gamma irradiation on morphology of leaf and shoot apex of ginger, turmeric and mango ginger. Proceedings of the Indian Academy of Science 9(3):173.

INDUCED VARIATION IN GINGER USING EMS 17

- Rekha, K., S.N. Kak and A. Langer. 2000. EMS induced variability in *Artemisia pallens* Wall. Indian Journal of Plant Genetic Resources 13(1):37-41.
- Sarkar, K.K., O. Islam and S. Sen. 1985. Upgrading the tuber characters of sweet potato with chemical mutagen induced bud mutation. Experimental Genetics 1(2):95-101.
- Vasudevan, K., G. Padmaja and S. Jayakumar. 1996. Induced tuber colour mutants and their biochemical characteristics in sweet potato. Journal of Root Crops 22(2):137-139.