International Journal for Innovative Engineering and Management Research
A Peer Revieved Open Access International Journal
www.ijiemr.org

COPY RIGHT

ELSEVIER
SSRN

IJIEMR Transactions, online available on 20th Jan 2021. Link
:http://www.ijiemr.org/downloads.php?vol=Volume-10&issue=ISSUE-01

DOI: 10.48047/IJIEMR/V10/I01/34

Title: **Effect of Ethyl Methanesulphonate on Rice Varieties (Oryza sativa l.) of Assam**
Volume 10, Issue 01, Pages: 168-173.

Paper Authors

Ratan Chowdhury

USE THIS BARCODE TO ACCESS YOUR ONLINE PAPER

To Secure Your Paper As Per UGC Guidelines We Are Providing A Electronic Bar Code

International Journal for Innovative Engineering and Management Research
A Peer Revieved Open Access International Journal
www.ijiemr.org

Effect of Ethyl Methanesulphonate on Rice Varieties (Oryza sativa l.) of Assam

Ratan Chowdhury[1]
[1] Department of Botany, Rangapara College, Amaribari, Rangapara-784505 (India)
e-mail-Chowdhuryratan600@gmail.com

Abstract— Rice can serve as potential genetic resources for improving yield, resistance to pests and pathogens, and agronomic performance, can be grown practically anywhere even on a steep hill or mountain area with high rainfall and low labour costs. The present paper aims at finding out the mutagenic effects of the mutagen, Ethyl methanesulphonate in different traits selected such as differentiation in germination, seedling height and number of rootlets, root length and number of tillers in the seeds of two indigenous rice varieties of Assam, Ahu and Baismuthi. The experiments were conducted for both laboratory as well as field conditions. In concept with the laboratory observations, our findings show that the highest % germination as observed on the control samples as expected when compared to that of treatment. With increasing EMS amounts there has been a decrease in seedling height. Under field conditions, the plant height (in Ahu) observed during the late stages of seedlings and early stages of adult plants shows that it increase steadily with increase in concentration of EMS in comparison to that of the control whereas the number of root length and roots (in Baismuthi) shows a decrease in increasing EMS concentration in comparison to the control.
Key words: Ethyl methanesulphonate, rice, germination, Baismuthi

INTRODUCTION

Rice (*Oryza sativa*) is the staple food of more than half of the world's population. The importance of this crop lies in the fact that it has shaped the cultures, diets and economies of millions of people living particularly in Asia. Considering the role rice plays in providing food and nutritional security, and eradicating poverty, the United Nations designated the year 2004 as the International Year of Rice [12]. The high- yielding varieties of rice introduced from the plain or hill regions of the northern and southern parts of the country do not perform well in north eastern hill region [3]. The importance of local land races of rice in breeding programme lies in the evolution of gene complexes in the local cultivars that have co adapted to specific local environment through long period of natural selection. These gene complexes may not be readily reconstituted even by the modern recombinant DNA technology, we have to utilize the naturally occurring gene complexes for breeding of rice for specific local conditions [11]. Concerted efforts are required to understand the function of individual genes, and their interactions among themselves as well as with environment, in relation to variation in traits for a directed genetic manipulation of this important crop for the benefit of the mankind [24]. Mutation produces raw material for genetic improvement of crops [2]. Ethyl methanesulfonate (EMS) is a mutagenic teratogenic organic compound. EMS is one of the most frequently used alkylating agent for chemical mutagenesis in plants due to its potency and ease with which it can be used. EMS is more effective than physical mutagens [5, 4]. It is mainly used to produce GC to AT transition [17, 8]. EMS results in stable point mutations and thus produces an allelic series of missense changes that can provide a range of phenotypes. The present investigation was done to determine the effects of different doses of ethyl methanesulfonate on seed germination in two varieties of rice viz. Ahu (C1) and Baismuthi (C2).

International Journal for Innovative Engineering and Management Research
A Peer Revieved Open Access International Journal
www.ijiemr.org

MATERIALS AND METHODS

The experimental material consists of two indigenous rice varieties of Assam - Ahu (C1) and Baismuthi (C2). Ethyl methanesulfonate is used as mutagenic agent. 50 ml of 0.5%, 1%, 1.5%, 2% concentrations of Ethyl methanesulfonate (w/v) were prepared in water. The pH of the solutions was adjusted at 7.

Seeds of C1 (Ahu) and C2 (Baismuthi) were each placed in 16 petri dishes (8 petri dishes each of C-1and C-2) and double distilled water was added to a level above the seeds. Seeds were soaked overnight at room temperature for 20 hours. Subsequently, the water was decanted and 50 ml of 0.5%, 1%, 1.5%, 2% and a control dose of 0% concentrations of EMS (v/v) in water was added. Seeds were kept for 5 hours at room temperature followed by decantation of the EMS and rinsing with 100 ml of double distilled water (5 times, 4 minutes each). Seeds were then rinsed under running tap water for 4 hours before planting in Petri dishes. Following the next day of the treatments, the seeds were continuously assessed for the germination and developmental stages daily. The seedlings were then transferred in plastic pots and grown in the botanical garden under natural conditions.

The following parameters were studied for seedling growth under laboratory conditions

Germination percentage:

The seeds sown in the petridishes were grown under room temperature and observed daily until it reaches maximum generation. After seven days, the number of seeds germinated under this condition was recorded and was followed on 14th day from the day of sowing in laboratory.

The germination percentage was calculated by using the following equation:

$$\text{Germination(in percent)} = \frac{\text{No. of seedling emerged}}{\text{Total no. of seed sown}} \times 100$$

Shoot and root length (in cm) :

The growth of seedlings in terms of its height was also measured after 16 days from the day of sowing under laboratory conditions.

Root length (in cm):

The root length was measured on the 16th day from the date of sowing.

Number of roots:

The number of roots was counted on the 16^{th} day from the date of sowing.

The following parameters were studied for M1 seedling under field conditions.

The seedlings grown in the petridishes under room temperature were transferred to plastic pots on 17th day from the day to sowing. Seedlings from each EMS concentration applied and those of the control were transferred and planted in the rice field soil prepared in plastic pots. Also the plants were watered regularly with normal tap water. The seedling heights, number of tillers, number of root and the root length of the plants were measured from the day of transplantation.

Seedling height (in cm)

The effects of ethyl methanesulfonate on seedlings height of each individual from every treatment of both the varieties was measured on 12th day from the day to transplantation under field conditions.

International Journal for Innovative Engineering and Management Research
A Peer Revieved Open Access International Journal
www.ijiemr.org

Number of tillers

The alkylating agent EMS effects on number of tillers were observed under field conditions. The observation was done on the 12th day from the day of transplantation. Mean values of each treatment and control were calculated from the recorded data from each individual under the respective treatments. The mean values of different treatment were compared with that of the control to see the effects of mutagen.

Number of roots and root length

For observing root growth in terms of its number and length, the plants were uprooted slowly. It was done by loosening the soil under water with hand and pulling the plant up slowly one at a time. Care should be taken to avoid breakage of the roots observation were carried out once as in the case of all parameters.

Statistical analysis:

Statistical analysis was performed using one-way ANOVA (for $P<0.05$).

RESULTS AND DISCUSSION

Germination percentage:

Seed germination in both the varieties decrease due to the effects of EMS with considerably increase in the higher levels of doses. This reduction was found to be dose dependent in both the varieties. Similar observations were observed by many workers in *Vigna radiate* [18, 7]. Inhibition in seed germination after mutagenic treatment has been attributed to changes in biochemical and physiological system [23] and inhibitory effect of mutagen [19]. The reduction percentage in seedling germination ranges from 12.5% -67.5% in Ahu and 16.25%-71.25% in Baismuthi Thus, the stimulating effect of physical mutation on germination may be credited to the activation of RNA or protein synthesis. It may occur during the early stage of germination after the seeds are treated as also observed by Abdel *et al.,* 2008 [1].

Table 1: Effect of different treatment of EMS on seed germination of both the varieties

	Observation on 7th Day		Observation on 14th Day	
Mutagen conc(%)	Ahu	Baismuthi	Ahu	Baismuthi
Control	87.5	85	100	100
0.5	62.5	63.75	87.5	83.75
1	37.5	40	55	52.5
1.5	25	26.25	42.5	35
2	17.5	18.75	32.5	28.75

Seedling height

Table 2: Effect of different treatments of EMS on seedling height of both the varieties under laboratory and field conditions.

	Under laboratory conditions		Under field conditions	
Mutagen conc(%)	Ahu	Baismuthi	Ahu	Baismuthi
Control	4.18	5.26	42.58	38.98
0.5% EMS	5.29	4.9	46.54	42.96
1% EMS	5.02	4.61	38.02	31.56
1.5% EMS	3.76	4.48	49.32	27.88
2% EMS	3.66	5.82	49	41.4

The table shows that there is a decrease in seedling height with the increase in EMS concentration. The decrease is more significant in Baismuthi variety under field condition. But at 2% EMS the seedling height in both the varieties increased significantly. Under laboratory conditions there is a significant decrease in seedling height of Baismuthi at 0.5%, 1% and 1.5%. But there is an increase in Ahu variety at 0.5% and 1%. These findings are in close agreement with that of the earlier reports on chemical mutagen, effects of sodium azide [23, 21 and 10]. The enhancing effect may be due to sudden increase in the metabolic status of seedlings and increase in the activity of growth promoters. Reduction in biological criteria plant height may be attributed to a drop in auxin level, inhibition of auxin synthesis [20].

International Journal for Innovative Engineering and Management Research
A Peer Revieved Open Access International Journal
www.ijiemr.org

Root length:

The evaluation of root length under laboratory conditions was done on 16th day from the day of showing and under field conditions was done on 12th day from the day to transplantation under field conditions. A gradual reduction in root length was observed in both the varieties in all the treatments except at 2.0% EMS in the variety Baismuthi. Under field conditions the root length in both the varieties in all the treatments, except at 1.5% EMS in Baismuthi, is reduced significantly. A significant effect of the concentration of applied EMS on the root length in variety Ahu was observed under laboratory conditions. Reduction in root length occurred with each corresponding increase in the concentration of EMS. The effects observed under the low-or high-dosage treated plants are enhancement or inhibition of seedling/plant growth, root length and other biological responses. This is in contrast with the study by Nusrat et al., (2002)[12] that, an increase in chemical concentration, there was an increase in the rate of mutation leading to variations in rooting whereas under field conditions, the two varieties showed a more or less same response to root length reduction. Similar results have also been reported for *Capsicum annuum* and *Avena sativa* [14,9], when treated with EMS and dimethyl sulfate; EMS and azide, respectively.

Table 3: Effect of different treatments of EMS on root length of both the varieties under laboratory and field conditions.

	Under laboratory conditions		Under field conditions	
Mutagen conc(%)	Ahu	Baismuthi	Ahu	Baismuthi
Control	4.04	5.72	24.48	23.38
0.5% EMS	3.74	4.95	17.14	23
1% EMS	3.30	4.12	16.36	20.24
1.5% EMS	2.37	4.86	22.16	24.2
2% EMS	1.60	6.02	20.46	19.7

Number of roots:

Table 4: Effect of different treatments of EMS on number of rootlets under laboratory and field conditions

	Under laboratory conditions		Under field condition	
Mutagen conc(%)	Ahu	Baismuthi	Ahu	Baismuthi
0.5% EMS	3.6	4.93	75.8	75
1% EMS	4.13	3.8	34.6	54.6
1.5% EMS	2.53	1.93	67.2	36.4
2% EMS	2.53	3.86	83.4	59

Number of rootlets and roots increases significantly under lower doses of EMS in both varieties under laboratory conditions and gradually reduces its number with increase in EMS doses (i.e. from 1.5%- 2%) except for Baismuthi which increases again at 2% EMS. Thus this effect of mutagen on rootlets can led to the indication of effective mutagenesis that can result in formation of desirable traits. Under field condition, the root length in both the varieties in all the treatments, except at 1.5% EMS in Baismuthi, is reduced significantly. The highest reduction recorded in Ahu is 29.98% at 0.5% EMS while it was 20.46% at 2.0% EMS in Baismuthi.

Number of tillers per plant

On the 29th day after transplantation, there is an overall reduction in comparison to controls in number of tillers in both the varieties, where in Baismuthi, a decrease in the number of tillers with an increase in the concentration of the mutagen was observed. Reduced growth due to higher doses was also explained differently by different workers. It may be attributed to one or more of the following reasons:(i) the increase in growth promoters, (ii) the sudden increase in metabolic status of seeds at certain levels of dose, (iii) the increase in destruction of growth inhibitors, (iv) drop in the auxin level or inhibition of auxin synthesis and (v) decline of assimilation mechanism. The

variation in biological parameter viz. no of tillers may be attributed to a drop in auxin level [6] or due to decline of assimilation mechanism [14].

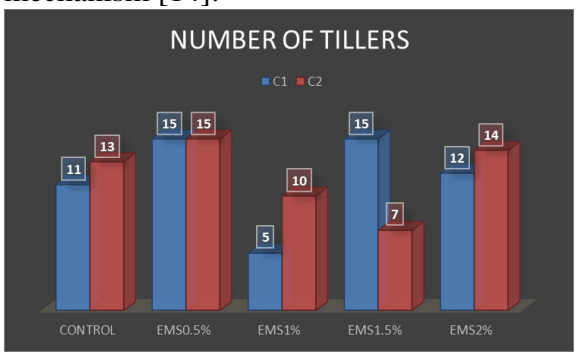

Figure 1: Maximum number of tillers observed in each concentration as on 29th day after plantation.

CONCLUSION

Morphological traits such as plant length and root numbers were decreased with concentration of EMS and treatment duration increase. Similar results were reported by Ramachandra *et al.*, (2008) who showed that there was a decrease in the growth parameters with increase in the concentration of EMS solution, which may be due to the fact that, the enzymatic activity associated with the biosynthesis of primary metabolic traits might have been reduced with increased concentration of EMS solution. Thus with the above observations and findings, we can compare the effects of mutagen EMS on the two rice varieties where variations were observed between them and under which concentration it acts more in enhancing various traits and this can also be interpreted with other different varieties of crops for generating new improved varieties.

ACKNOWLEDGEMENT

It is by God's grace and the will of the Almighty that I have this opportunity to express my deep sense of gratitude to all those who directly or indirectly have helped me throughout the project work.

I express my unbounded gratitude and indebtness to my supervisor, Dr. Md. Asad Ali, Assistant professor, Department of Botany, University of Science and Technology, Meghalaya, for giving me opportunity to do my project work under his supervision, and for his expedient advice, affection, critical evaluation, timely help, ever-encouraging suggestions and invaluable painstaking efforts taken towards my study, and above all for being an excellent human being during the most trying times in this tenure of project work.

I am sincerely thankful to Dr. Ranee Das, Head of the Department for providing me facilities to carry out my project work.

I also express my sincere thanks to the entire faculty members, Department of Botany, University of Science and Technology, Meghalaya, for their valuable guidance, inspiration and affection offered to me during my project works.

I take this precious moment to express my special heartfelt thanks to technical staff for their help, constant encouragement and motivation for stepping ahead, always.

Last but not the least; I am thankful to my parents, my friends and my brother for their constant support, blessings and encouragement.

Ratan Chowdhury
Place: Tezpur University.

REFERENCE

1. Abdel-Hady M.S, Okasha EM, Soliman SSA, Talaat M: Effect of Gamma Radiation and Gibberellic Acid on Germination and Alkaloid Production in *Atropa belladonna* L: Australian Journal of Basic and Applied Sciences. 2008; Vol. 2, No. 3, 2, pp. 401-405.
2. Adamu AK and Aliyu H: Morphological effects of sodium azide on tomato (*Lycopersicon esculentum* Mill): Science World Journal Vol. 2.2007; No.4, 9-12.
3. Asthana AV and Majumdar ND: Studies in rice germplasms of North Eastern Hill Region: 1981; Research Bulletin, no. 11.Http://dx.doi.org/10.5402/2013/651019
4. Bhat TA, Khan AH, Parveen S: Comparative analysis of meiotic abnormalities induced by gamma rays, EMS and MMS in *Vicia faba*

L: Journal of Indian Botanical Society.2005;84, 45-58.
5. Dhanayanth KPM, Reddy V: Cytogenetic effects of gamma rays and ethyl methanesulphonate in chilli piper (*Capsicum annum*): Cytology. 2000; 65, 129-133.
6. Gordon SA, Webber RP: Studies on the mechanism of phytohormone damage by Ionizing radiation: Plant Physiology. 1955; 30(3): 200-10.
7. Khan S, Wani MR, Parveen K: Induced genetic variability for quantitative traits in *Vigna radiata* (l) wilczek: Pakistan Journal of Botany. 2004; 36 (4): 845-850.
8. Koornneef M, Dellaert L, Veen J: EMS and radiation-induce mutation frequencies at individual loci in Arabidopsis thaliana: Mutation Research.1982; 93, 109–123.
9. Krishna-Murthy CS, Vasudevan K: Induced polygenic variation following single and combination treatment with azide, EMS and gamma rays in oat: Crop Improvement.1984; 11, 128–31.
10. Kumar, J.S., Selvaraj, R: Mutagenic effectiveness and efficiency of gamma rays and ethyl methanesulphonate in sunflower (*Helianthus annus* L.): Madras Agric. J.2003; 90(7-9): 574-576.
11. Medhabati K, Rajiv Das K, Rohinikumar M, Sunitibala H, Dikash Singh Th: Genetic Divergence in Indigenous Wild and Cultivated Rice Species of Manipur Valley. ISRN Genetics Volume. 2013; Article ID 651019.A.
12. Mohapatra T, Robin S, Sarla N, Sheshashayee M, Singh AK, Singh K, Singh NK, Amitha Mithra SV, Sharma RP: EMS Induced Mutants of Upland Rice Variety Nagina22: Generation and Characterization. Proceedings of the Indian National Science Academy. 2014 March; pp. 163-172.
13. Nusrat Saba and Bushra Mirza: EMS induced genetic variability in *Lycopersicon esculentum*. International Journal of Agricultural Biology. 2002; 4 (1): 208 – 213.
14. Patil AN, Meshram LD, Nandanwar RS: Induced quantitative variation in economic characters by chemical mutagens in chilli: Journal of Soils and Crops.1997; 7, 15–8.
15. Quastler H, Baer: Inhibition of plant growth by irradiation. V. Radiation effects on initiation and completion of growth: Cancer Research.1950; 10, 604-612.
16. Ramachandra KM, Srinivasappa KN, Farooqi AA: (2008). Mutagenic effect of ethyl methane sulphonate on growth, yield and composition of oil in patchouli (*Pogostemon patchouli* Pellet.): Biomed. 2008; 2 (4): 341-344.
17. Rao G: Efficiency and effectiveness of gamma rays and EMS in rice: Cytologia. 1977; 42, 443–450.
18. Rupinder S, Kole CR: Effect of mutagenic treatment with EMS on germination and some seedling parameters in mungbean: Crop Research.2005; 30 (2): 236-240.
19. Sahai: (1974). Cytogenetic, Mutational and Seed Protein Analysis of Some Pulses Materials. Ph.D. Thesis, IARI, New Delhi.
20. Skoog, F: The effects of X-irradiation on auxin and plant growth: Journal of Cellular Comparative and Physiology.1935; 7, 227-270.
21. Solanki IS, Sharma B: Induction and isolation of morphological mutations in different mutagenic damage groups in lentil (Lens culinaris Medik): Indian Journal of Genetics.1999; 59 (4): 479-48.5.
22. Sparrow AK, Woodwell GM: Prediction of sensitivity of plants to chronic gamma irradiation: Radiation botany.1962; 2, 9-26.
23. Wang PY, Yu BS: Preliminary study on gamma-rays chronic radiation for growing plants in soybean: Soybean Genetics Newsletter. 1988; 18, 82-85.
24. Yang, Lihui: Handbook of Chinese Mythology. New York: Oxford University Press.2005; p.198. ISBN 978-0-19-533263-6.