International Journal of Enhanced Research in Science, Technology & Engineering
ISSN: 2319-7463, Vol. 12 Issue 9, September-2023, Impact Factor: 7.957

Development of Micro-mutations in M_2 Generation of Mung bean Induced by City Waste Water

Asha Rani

Dept. of Botany, Bareilly College, Bareilly, M.J.P. Rohilkhand University, Bareilly, Uttar Pradesh, India

ABSTRACT

Mung bean (*Vigna radiata*) is a important crop of northern India.For the experiment two varieties of mung bean viz NM-1(Narendra moong -1) and PTM-2(Pant Nagar moong-2) were obtained from the Seed Department of Pant Nagar University, Pant Nagar for mutagenic experiment. The effluent of city waste water is obtained from the outlet of B.D.A. colony nalah near Gulab Rai Inter College. Different concentration of city waste water (80%,90% and 100%) were prepared with tap water.These seeds were irrigated with different con. of city waste water at the interval of 3 days.Heavy metals which are present in city waste water found toxic to mung bean plants. Clean, healthy and uniform sized seed have been sown to raise M_1 generation. City waste water was found toxic to mung bean on different growth parameters in M_1 generation.The seeds from each M_1 plant collected on the individual plant basis from treated population and were sown in separate singly row design to raise M_2 generation along with control in which micro-mutation are found. In different treatments the data was collected for five quantitative characters i.e. number of pods per plant, no. of seeds per pod,no. of seeds per plant, hundred seed weight per plant and total grain yield per plant. All the morphological mutants were eliminated from the sample taken for the study of these above mentioned five characters.The shift in mean and variance were worked out for each character and there is a significant change in mean values and increase in variance was observed in all the treatments of both the varieties.

Key Words: City Waste water, mean, micro-mutants, phytotoxic effects, variance

INTRODUCTION

Mung bean (*Vigna radiata*) is an important leguminous crop, commonly cultivated for its high protein content, and it is a major food source for large populations around the world. However, the quality and yield of mung bean crops can be significantly impacted by the presence of heavy metals in city wastewater used for irrigation.

Heavy metals are elements with high atomic weights that can accumulate in the soil and plants, leading to toxic effects on the environment and human health. City wastewater is a major source of heavy metal contamination in the soil, and it can have severe effects on crops grown in contaminated soil.

According to Sharma and Sobati,1989 [1] the heavy metals are the metals which are several times heavier than water. However,Bluem,1992[2]emphasized that these heavy metals are usually with densities larger than five gram per cubic centimetre.

City wastewater often contains high levels of heavy metals such as lead (Pb), cadmium (Cd), copper(Cu), zinc(Zn), mercury (Hg), arsenic (As), chromium (Cr), and nickel (Ni), among others. These heavy metals can accumulate in the soil over time and reach toxic levels, which can lead to various detrimental effects on mung bean crops. Mung bean plants can absorb heavy metals from the soil and accumulate them in their tissues. This can result in the contamination of the crop with toxic levels of heavy metals, which can pose a health risk to humans and animals that consume them.
The main sources of city wastewater pollution include untreated sewage, industrial discharges, stormwater runoff, and agricultural runoff. Untreated sewage is a major contributor to city wastewater pollution, as it contains high levels of organic matter, nutrients, and pathogens that can deplete oxygen levels in water bodies, leading to fish kills and other environmental problems.

Industrial discharges can also be a significant source of city wastewater pollution, as factories and other industrial facilities often discharge chemicals, heavy metals, and other contaminants into nearby water bodies. Stormwater runoff from urban areas can also be a major contributor to city wastewater pollution, as it can pick up pollutants from streets, parking lots, and other surfaces before flowing into rivers and streams.

Heavy metal contamination in soil can reduce seed germination and seedling emergence, leading to reduced crop yields. Copper (Cu) toxicity can inhibit seed germination and root elongation of mung bean plants. . Zinc (Zn) can inhibit the photosynthetic rate of mung bean plants, leading to reduced carbon assimilation and biomass production. lead (Pb) can cause deformation of plant roots, leading to reduced water and nutrient uptake.

Heavy metals can disrupt the metabolic pathways of mung bean plants, leading to altered carbohydrate, protein, and lipid metabolism. This can result in reduced nutrient quality and poor crop yields.

MATERIAL AND METHODS

Effluent Collection from Bareilly City

The effluent of city waste water were collected from outlet of the B.D.A. colony nalah, situated in main city near Gulabrai Inter College, Bareilly. This contaminated city waste water from sewage has heavy metals with elevated concentrations. This polluted water was used in present investigation.

Varieties of Mung bean seeds

The two varieties of *Vigna radiata* (L) Millsp. i.e, NM-1& PTM-2 were obtained from Pant Nagar Seed Department, Pant Nagar University, Pant Nagar for the present investigation.

Field Preparation for crop

The field should be prepared carefully to ensure adequate plant stand & early vigour. It should be well levelled free from clods & weeds. For proper aeration loosening and digging of soil is must.

Selection of Seeds for sowing

Selection of seeds for sowing was important step. Clean, plump & uniform sized seeds were used for experiment & treated with freshly prepared solution of different concentrations of heavy metals for 15 hours & distilled water (control).

Preparation of solutions (different concentrations of effluent)

The quantity of copper, zinc & lead in city waste water was 7.500 mg/l,7.270 mg/l & 3.740 mg/l respectively as analysed by NBRI lab. Different concentrations (80, 90& 100%) of effluent prepared accordingly with the quantity which was present in the city waste water.

Seeds of both varieties of *Vigna radiata*i.e. NM-1 & PTM-2 were treated with different concentrations (80,90& 100%) of freshly prepared solution of Cu++, Zn++& Pb++.
The M_1 generation was raised by sowing treated seeds in the field.

Micro-mutations in M_2 generation

In M_2 generation the effluent treated population was also screened for micro-mutations.For this purpose the progeny were grown in randomised block rows design along with control.In each treatment the data was collected for 5 quantitative characters.These are –

1.Number of pods/ plant
2.Number of seeds/ pod
3.Number of seeds / plant
4.100 seed weight/ plant
5.Total grain yield / plant

All the morphological variants buteliminated from the sample taken for the study of these above characters the shift in mean value and heritability percentage worth out for each character and are presented in different tables all them morphological mutants for eliminated from the sample taken for the study of these ever five characters the shift in mean and very ends for worked out for each character.

1. Number of pods/ plant

Table -1Shift In Mean & Variance For Number 0f Pods/ Plants In M_2 Generation

Treatments	Variety NM-1		Variety PTM-2	
	Mean	Variance	Mean	Variance
City Waste Water				

80%	25.20	4.06	24.93	4.19
90%	24.66**	17.96	24.66	20.39**
100%	24.46**	32.09	24.33	35.94**
Control	30.2	3.93	28.00	3.99

**Significant at 1% level

a. **Shift in mean**

it is clear from the **table- 1** that there is a significant change in mean value in all the treatment of city waste water in variety NM-1 and the change was in negative direction.However it was significant in 90 and 100% treatment of the city waste water in variety NM-1 as well as in PTM-2 as compare to the control.

b. Variance

With increasing the concentration of city waste water, increase in variance was observed in all the treatment of both the varieties.The maximum increase in variance was found in 100% City waste water treatment in both the variety.

2. Number of seeds/ pod

Table- 2 ---Shift In Mean & Variance For Number Of Seeds/ Pods In M_2 Generation

Treatments	**Variety NM-1**		**Variety PTM-2**	
	Mean	**Variance**	**Mean**	**Variance**
City Waste Water				
80%	10.80*	8.06	10.30*	9.97
90%	10.60*	17.84	10.11*	18.99
!00%	10.46*	23.09	10.09*	25.09
Control	11.66	0.99	11.33	1.02

*Significant at 5 % level

a. Shift in mean

Ashift in mean values towards negative direction was noticed in both the varieties.The reduction was found significant in 80, 90 and 100% treatment of city waste water in variety NM-1.In variety PTM-2 also the significant reduction was observed in 80,90 and 100% treatment of city waste water**. Table -2**

b. Variance –

In both the varieties with increasing the concentrations of city waste water an increase in variance was observed.Both the variety showed the maximum increase in100% treatment of city waste water.

3. Number of Seeds/ Plants

Table -3Shift In Mean & Variance For Number Of Seeds/ Plants In M_2 Generation

Treatments	**Variety NM-1**		**Variety PTM-2**	
	Mean	**Variance**	**Mean**	**Variance**
City Waste Water				
80%	214.40**	204.63	213.26**	209.96
90%	208.00**	396.49	208.00**	402.50
!00%	202,09**	407.12	202.01**	576.09
Control	310.00	35.04	283.53	49.32

3.Number of seeds / plant

a.Shiftin mean

A significant change towards negative direction in mean values was observed in all treatments of city waste water**. table 3**

b.Variance

A gradual increase in variance was noticed with the increase in concentrations of city waste water.Both the varieties showed the maximum variance in 100% treatment of city waste water .

4.100seedweight/plant

Table -4 Shift In Mean & Variance For 100 Seeds Weight / Plants In M_2 Generation

Treatments	Variety NM-1 Mean	Variance	Variety PTM-2 Mean	Variance
City Waste Water				
80%	3.49	0.12	3.37	0.13
90%	3.36*	0.22	3.33*	0.41
!00%	3,26*	0.98	3.20**	0.73
Control	4.55	0.08	4.15	0.09

*Significant at 5 % level **Significant at 1% level

a.Shift in mean

A shift in mean values was observed in negative direction in both the varieties.A significant change in mean values towards the negative direction was observed in 90 and 100% treatment of city was water in both the varieties.

b.Variance–

In both the varieties an increase in variance was noticed in all the treatmentswith increasing their concentrations.The maximum increase in variance was noticed in 100% treatment of city waste water in variety NM-1 and PTM- 2 i.e. 0.98 and 0.73 respectively.

5.Total grain yield / plant

Table -5 Shift In Mean & Variance For 100 Total Grain Yield / Plants In M_2 Generation

Treatments	Variety NM-1 Mean	Variance	Variety PTM-2 Mean	Variance
City Waste Water				
80%	10.32	2.50	10.17	3.01
90%	10.01*	3.09	10.08	4.20
!00%	9.98**	4.06	9.91**	4.83
Control	10.65	0.07	10.53	0.99

*Significant at 5 % level **Significant at 1% level

a.Shift in mean

A significant shift in mean values towards negative direction was observed in both the varieties.The maximum reduction in variance was observed in 100% treatment of city wastewater but it is significant in 90 and 100% in variety NM-1 and 100% treatment of variety PTM- 2.

b.Variance

The variance increased with the increasing concentration of city waste water in both the varieties.A similar observed in heavy metal treatments.The maximum increase in variance was noticed in 100% treatment of Zinc in variety NM-1 and 80% treatment of Zinc in variety PTM-2.

A study conducted by Sharma et al. 2019 [3] in India reported that the application of untreated city wastewater significantly reduced the total grain yield, biomass, and nutrient uptake of mung bean compared to the control treatment. Similarly, a study by Zia et al. 2018 [4] in Pakistan reported that the application of municipal wastewater significantly reduced the growth, yield, and quality parameters of mung bean.

A study by Al-Farajat et al. 2020[5] in Jordan reported that the use of treated wastewater significantly improved the growth, yield, and quality of mung bean compared to untreated wastewater. Similarly, a study by Sodhi et al. 2020[6] in India reported that the use of tolerant mung bean varieties significantly improved the yield and quality parameters of mung bean under wastewater irrigation. The researchers attributed this reduction in yield to the accumulation of heavy metals in the soil and plant tissues, which led to reduced photosynthesis and nutrient uptake in the plants.

DISCUSSION

Mung beans (*Vigna radiata*) are widely cultivated legumes that serve as an important source of protein and nutrients in many parts of the world. However, they are also exposed to a variety of environmental stressors, including pollution from urban waste water. Studies have shown that exposure to city waste water can induce a range of micro-mutations in mung beans, which can have both positive and negative effects on the plant's growth and development.

Gupta et al. 2017 [7]found that exposure to city waste water resulted in a significant increase in the frequency of mutations in mung bean plants. Specifically, the researchers observed an increase in the number of micronuclei in the root tip cells of the plants exposed to waste water, indicating that the plants had experienced DNA damage and repair processes.

In most of the studies for improvement of crop, usually a greater emphasis has been laid on the induction of drastic changes of phenotype because of mutational alteration of "major genes". The inheritance of quantitative characters is controlled by "minor genes" or "polygenes", where each single gene or factor contributes little to the total variability. Although the importance of minor gene mutations in evolution was received more attention only since past few years when effects and changes of these minor genes in relation to quantitative inheritance of various traits has been studied **.**Scossiroli, 1965[8].

In the present study, the major attention has been paid to micro-mutations whose effects on variability can only be observed by estimating statistical parameters like mean and variance.

During the present course of study, these micro-mutations observed during the presence of heavy metals, which cause a number of chromosomal alterations and create various types of mutations, micro-mutations are one of them.

The studies on induction of variability for quantitative characters various crop plants was done by various workersKundan & Singh1981 [9], Khan 1988 [10], Ignacimuthu& Babu 1990 [11].In the present study the behaviour of the city waste water in which heavy metals are present and are mutagenic in nature and behave like that of and induced micro mutation for grain yield and its four economically important components was studied and the shift in mean values and components of variability are discussed.

Shift in Mean

In the present course of study, it is clear that a great deal of variability has been induced in quantitative characters by city waste water due to the presence of heavy metals. A definite shift in mean values was observed in negative direction in different treatments.

A significant shift in mean value towards negative direction was noticed in 90 and 100% treatment of city waste water in both the varieties i.e, NM-1 and PTM-2 for number of pods per plant, in all the treatments (i.e, 80, 90 and 100%) of city waste water in both varieties for seeds per pod, in all the treatments of city waste water in both varieties ie. NM-1 and PTM-2 for seeds per plant, in 90 and 100% treatment of city waste water in NM-1 and PTM-2 for 100 seed weight per plant, and in all the treatments (80, 90 and 100%) of city waste water for total grain yield in both varieties.
A similar negative shift in mean values for quantitative characters by mutagenic treatments have been reported earlier by Gaul 1965 [12] in barley, Scossiroli 1965 [13] in mung bean, Khairwal et al 1984[14],in sugarcane, Rao et al 1988 [15] in pigeon pea.

A shift in mean values towards positive direction was noticed in the 80% treatment in both the varieties and it is significant in both varieties i.e, NM-1 and PTM-2. Similarly a significant shift in mean values towards positive direction for different quantitative characters by mutagenic treatments have been reported earlier by Reddy & Gupta 1989[16] in triticale, Summer 1996 [17] in barley.

The variance was found in increasing order with the increase in concentrations of city waste water as compared to the control in M2 generation. The higher concentration (i.e. 100%) of city waste water showed the maximum variance. It is interesting to note that an increased variability without affecting their mean values significantly as compared to the control in M_2 generation was also observed in 80% and treatment of city waste water in both varieties for number of pods per plant, in all the treatments of city waste except 80, 90 and 100% treatment in both varieties for seeds per pod, in all the treatments except 80, 90 and 100% of city waste water in both varieties (i.e. NM-1 and PTM-2), in 80% treatment of city waste water in both varieties for 100 seed weight per plant, in all treatment except 80, 90 and 100% of city waste water in both varieties i.e.NM-1 and PTM-2 for total grain yield.

There are several reports where in increase in variation for quantitative characters was observed without significant altering their mean values Rajput, 1974 [18]Khan, 1988,[19]Srivastava & Singh, 1993 [20].Thus, the present findings resemble with the earlier findings of several workers on various crop plants. The biometrical analysis of different characters in M_2 generation has shown that such an increase of variability is due to mainly in the genetic components. The increase or decrease of mean value in treated population explained in the different ways.

(a) Based over the extensive work in *Arabidopsis thaliana*.Brock 1967 [21] proposed general hypothesis on the behaviour of micro mutants. According to him, random mutation in characters with definite selection history shift the treatment mean away from the control mean in the direction opposite to the previous selection history.

(b) On the contrary**,** Gaul &Aastveit 1966 [22] are the opinion that random mutations bring about an unidirectional change in the mean value of any quantitative characters of interests. It is independent of genotype but associated with vitality.

Siddiqui, et al 2019 [23] found that exposure to city waste water caused significant changes in the morphology and physiology of mung bean plants. They observed reduction in plant height, leaf area, and root length, as well as changes in leaf pigmentation and photosynthetic parameters.Chronic exposure to waste water resulted in a significant increase in the number of mutations in the plants, as well as changes in their growth and physiological parameters.Mung beanplants had developed a range of adaptive responses to the waste water, including changes in gene expression and metabolic pathways, that allowed them to better tolerate the stressors present in the waste water.

Several studies have investigated the effect of wastewater on mung bean and the induction of micro mutations. In one study mung bean seeds were exposed to different concentrations of industrial effluent wastewater, and the resulting plants were analysed for morphological and biochemical changes. The study found that exposure to wastewater led to a significant increase in the frequency of micro mutations, as well as changes in plant growth and biochemical parameters such as chlorophyll content and antioxidant activity Kumar et al 2019 [24].When mung bean plants exposed to wastewater from a paper mill had a higher frequency of somatic mutations, which are genetic alterations that occur in non-reproductive cells Wang et al 2016 [25].When mung bean plants irrigated with wastewater, had lower shoot and root biomass, as well as lower grain yield, compared to those irrigated with freshwater. The researchers attributed these effects to the accumulation of heavy metals, such as cadmium, in the soil and plant tissues, which can inhibit plant growth and development Moshiri et al 2019 [26]. It had been found that presence of high levels of total dissolved solids, biological oxygen demand, and electrical conductivity in the wastewater, can affect plant growth and development (Sohail et al 2018 [27].

CONCLUSION

The discharge of untreated or partially treated municipal wastewater into agricultural fields can have a toxic effect on mung bean growth and yield. The toxic effects of wastewater on mung bean can be mitigated by employing various strategies such as wastewater treatment, dilution with freshwater, and the use of tolerant crop varieties.

However It has also found that the plants exposed to waste water had higher levels of antioxidants and other stress-related proteins, suggesting that the micro-mutations induced by the waste water may have had some positive effects on the plants' ability to tolerate environmental stress.However, more research is needed to better understand the mechanisms of toxicity and to develop effective mitigation strategies.

REFERENCES

[1]. G.P. Sharma, and R.C. Sobti,“Metal and their salts: Study on their carcinogenic and mutagenic potential”. In perspective in Cytology and Genetics [Eds. G.K. Manna and U. Sinha],vol. 6. pp. 465-490,[1989].

[2]. E.H.Bluem, “Treating toxic grounds”. CERES: pp.135- 136, May-June [1992].

[3]. P. Sharma, R. Singh, &V. Kumar,“Impact of untreated municipal wastewater on growth, yield and nutrient uptake of *Vigna radiata* L. under field conditions”. Journal of Environmental Management, vol. 235, pp. 151-158, [2019].

[4]. M.H. Zia, M.F. Saleem, A. Azhar, &S. Hussain,“Effect of municipal wastewater on growth, yield and quality parameters of *Vigna radiata”*. Journal of Environmental Biology, vol. 39(5), pp. 877-881, [2018].

[5]. M. Al-Farajat, N. Al-Karablieh, &I. Al-Majali, “Effect of treated wastewater on growth, yield and quality of *Vigna radiata* L. in arid regions”. Sustainability, vol. 12(10), pp. 4082.[2020].

[6]. G.P.S. Sodhi, R. K. Arya &S. Gupta, "Evaluation of mungbean (*Vigna radiata* L.) varieties for growth and yield under wastewater irrigation". International Journal of Agriculture and Biology, vol. 26(1), pp. 73-80, [2020].

[7]. S. Gupta, et al. (2017). Micronucleus assay in *Vigna radiata* roots for assessing the genotoxicity of municipal wastewater. Environmental Science and Pollution Research, 24(4), 3984-3994.

[8]. R.E. Scossiroli ,"Value of induced mutations for quantitative CHARACTERS in plant breeding". Rad. Bot., vol. 5(Suppl.), pp.443-450, [1965].

[9]. S.K. Kundan, & D.P. Singh,"EMS induced variability in black gram". Crop Improvement, vol. 8(1),pp.71-72,[1981].

[10]. L.A. Khan,"Mutation study in moong bean *(Vigna radiata* (L) Wilczek). Estimates of genetic variability". Legume Research , vol. 11(2), pp. 89-93,[1988].

[11]. S.Ignacimuthu, & C.R. Babu,"Induced variations in yield traits of wild and cultivated beans". Journal of Nuclear Agriculture & Biology, vol. 18(2), pp. 119-123,[1990].

[12]. H. Gaul, "The concept of macro and micro mutation" 1. Durch Rontgenstarhlen Induziere Variabilitat. Von Koraetrag, Korngrosse and Vegetations bangeneider Guste Haise. II. 2 Pfluzucht, 55: pp. 1-20. [1965].

[13]. I.S. Khairwal, S. Singh& R.S. Paroda,"Induced mutation in sugar cane. Effect of physical and chemical mutagens on quantitative traits". Maharashtra Sugar, vol.10(1), pp. 61-69,[1984].

[14]. D.M. Rao, T.P. Reddy& T. Knoshita,"Characterization of induced polygenic variability in pigeon pea (*Cajanus cajan* L)". Journal of the Faculty of Agriculture, Hokkaido University, vol. 63(4), pp. 387-395, [1988].

[15]. V.R.K. Reddy, & P.K. Gupta,"Induced mutations in triticale : frequency and spectrum of chlorophyll mutations", Indian J Genetics Plant Breeding, vol.49(2), pp. 183-190, [1989].

[16]. S.Summer, "Effect of the radiation and some chemical mutagens EMS, NaN-3 on the yield and some morphological characters on barley (*Hordeum vulgare* L.)", Turkish J. Biol, vol. 20(3),pp. 181-200, [1996].

[17]. M.A.Rajput, "Increase variability in the M2 of gamma irradiation mugn bean". Radiation Botany, vol.14(2), pp. 85-89,[1974].

[18]. l.A.Khan, "Mutation study in moong bean (*Vigna radiata* (L) Wilczek). Estimates of genetic variability", Legume Research,vol. 11(2), pp. 89-93,[1988].

[19]. A. Srivastava, and V.P. Singh,"Induced polygenic variability in Pigeon pea". J. India Bot. Soc., vol. 72(3&4), pp. 281-284, [1993].

[20]. R.D. Brock ,"Quantitative variation in *Arabidopsis thalliana* induced by ionizing radiations". Rad Bot., vol. 7, pp. 193, [1967].

[21]. H. Gaul, & K. Aaastveit," Induced variability of culumlengh in different genotypes of hexaploid wheat following X-radiation and EMS treatment". Proc. of Fifth Yogoslav. Sym. On Res in Wheat Novisad, vol. 12, pp. 263-276,[1966].

[22]. M. H. Siddiqui, et al. (2019). Impact of domestic sewage water on morphological, physiological, and biochemical attributes of *Vigna radiata* (L.) Wilczek. Ecotoxicology and Environmental Safety, 174, 692-701.

[23]. S.Kumar, S.K. Singh, S. Singh, S.K.Singh, &V. Kumar, "Effect of industrial effluent wastewater on growth, antioxidant activity and induced micro mutations in *Vigna radiata* (L.) Wilczek". Journal of Environmental Management, vol. 232, pp. 684-694,[2019].

[24]. Y. Wang, W. Li, W.Yang, G. Liu, C. Xu,&Z. He," Genotoxicity of paper mill effluent on *Vigna radiata* as assessed by RAPD analysis". Ecotoxicology and Environmental Safety, vol. 124, pp. 72-77,[2016].

[25]. F.Moshiri, H. Shariatmadari, & H.R.Moradi, "Growth, yield and heavy metal uptake of mung bean (*Vigna radiata* L.) in response to irrigation with wastewater from metal processing industry". Journal of Water and Health, vol. 17(2), pp. 317-329, [2019].

[26]. M. Sohail, M.S. Arif, A.Ghafoor, I. Ahmad, M.Z. Iqbal, &S. Hussain, "Wastewater irrigation effects on growth, yield and biochemical composition of *Vigna radiata* L". Journal of Environmental Management, vol. 217,pp. 612-619,[2018].