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Studies on yield, quality and physiological parameters in induced mutant population of M_2 generation in field bean [*Lablab Purpurea var. lignosus* (L.) Prain]

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Abstract
An experiment was carried out at Vegetable block, College of Horticulture, Anantharajupeta, Andhra Pradesh during 2019 with the elite mutant lines of the gamma irradiated population of the field bean variety TFB-2 to study the yield, quality and physiological parameters. The results revealed that $M_{2.10}$ observed a higher rate of photosynthesis. The mutant line $M_{2.21}$ recorded highest SCMR values. The better performance for 100 seed weight was recorded by the mutant line $M_{2.20}$. The mutant line $M_{2.24}$ is better performer for seed yield per plant. The intercellular CO_2 was noted to be highest in $M_{2.18}$ whereas the rate of transpiration was highest in $M_{2.32}$. Mutant line $M_{2.28}$ recorded the highest leaf temperature and stomatal conductance with the highest protein content.

Keywords: *Lablab Purpurea,* physiological parameters, stomatal conductance, Dolichos bean

1. Introduction
Field Bean [*Lablab Purpurea* var. *lignosus* (L.) Prain], also known as Dolichos bean, Hyacinth bean, Indian bean, Lablab bean, Bonavist bean, Egyptian kidney bean, *etc*., belongs to the family Leguminosae. Its basic chromosome number is x=11 and somatic chromosome number 2n=2x=22. Field bean is extensively cultivated as a high valued grain legume crop in India, Africa, and other tropical and sub-tropical countries. It is an important crop grown throughout the country and is also known as the poor man's bean.

Field bean is a self-pollinated crop with limited variability, which is a deterrent for any improvement programmer because genetic variability is imperative for the selection of better ide types. Mutation breeding is a powerful tool to enrich variation, particularly for attributes of economic importance in the crops like field beans where hybridization is difficult. Owing to the small flower size and low seed setting in emasculated buds, not much desirable genetic variability could be generated; therefore, efforts were directed to generate more purposeful variability through induced mutations in field beans.

Induced mutagenesis is one of the unconventional breeding methods, which can be applied to augment the variability and correct one or more defects in the cultivar or variety. The selection of effective and efficient mutagen (s) is very crucial for any mutation breeding programmer to recover a high frequency of desirable mutations (Solanki and Sharma, 1999) [25]. In the current study, an attempt has been made to know the extent of genetic variability induced for yield and quality characters in M_2 generations to select efficient and elite mutants of the field bean var. TFB-2 to be progressed to next generation.

2. Methodology
In a previous experiment taken up at the Department of Vegetable Science, College of Horticulture, Anantharajupeta, dry and healthy seeds of locally grown field bean variety TFB-2 (Tirupati Field Bean-2) were subjected to gamma irradiation treatments with Co_{60} (Cobalt-60) as a source at Bhabha Atomic Research Station, Trombay, Mumbai, Maharashtra and raised as M_1 generation along with untreated seeds of TFB-2 as control. Around 32 morphological variants, showing superiority in the number of primary branches per plant, pod yield, and seed yield per plant were isolated in the M_1 generation and used as experiment material for the present study.

For the present study, the seeds of the 32 mutants of M_1 generation along with TFB-2 as control were used to raise the M_2 generation (plant to progeny rows) during *Rabi*, 2019 in a Randomized Block Design.

Dry seed yield per plant was recorded by collecting and weighing total dry seeds from all the pods from the individual plant and weight was expressed in grams. From each plant, one hundred seeds were collected at random and weighed and the weight was measured in grams to be expressed as 100 seed weight. Physiological parameters *viz.*, photosynthetic rate, stomatal conductance, transpiration rate, leaf temperature and intercellular CO_2 were recorded using an LCi-SD portable photosynthetic system and observation was recorded from 9.00 am to 11.00 am. The data for physiological parameters were recorded for each mutant and control plant at the time of harvest. Protein content in both irradiated and untreated control field bean seeds was estimated as per the method developed by Lowry *et al.* (1951) [27] and expressed in mg / 100g. The chlorophyll content of 10 randomly selected leaves in each plant was measured using chlorophyll meter SPAD-502 plus, the mean was calculated and expressed as SCMR.
Observations were recorded and the entire recorded data were subjected to statistical analysis to get information on mean performance. The results are presented here under the following subheads with appropriate discussion.

3. Results and Discussion

In M_2 generation, 32 elite mutants of field bean var. TFB-2 were selected of which T_{14} mutant line showed no germination. This led to a total of 31 elite mutant lines of M_2 generation to be considered under present investigation and the results are depicted in Table 1, Graph 1 and Graph 2.

3.1 Dry seed yield per plant (g)

In M_2 generation, dry seed yield per plant ranged from 64.16 g to 178.67 g with a grand mean of 108.94 g. The maximum seed yield (178.67 g) was recorded in the mutant line M2.24 on par with M2.8 (174.89 g) followed by $M_{2.26}$ (167.81 g) while the minimum seed yield was recorded in $M_{2.15}$ (64.16 g). Seed yield per plant of untreated control was observed as 109.55 g. eleven mutant lines recorded significantly higher seed yield per plant when compared to the control.
The shift of yield and its components depends on the favourable association between the components in response to mutagenic treatments (Khan and Qureshi, 2006) [9]. Similar observations were reported by Harish Kumar *et al.* (2018) [6] in dolichos bean, Mahla *et al.*, (2010) [15] in cluster bean, Horn (2016) [7] in cowpea, Xu *et al.* (2020) [26] in pea and Elangovan and Pavadai (2015) [3] in okra.

3.2 100 seed weight (g)

The character 100 seed weight in M_2 generation ranged from 19.51 to 26.80 g with a total mean of 24.05 g. The mutant line $M_{2.20}$ recorded the maximum 100 seed weight (26.80 g) followed by $M_{2.28}$ (25.71 g) and the mutant line $M_{2.13}$ recorded the minimum 100 seed weight (19.51 g). The mean of 100 seed weight in control was recorded as 24.46 g. sixteen mutant lines produced a greater 100 seed weight when compared to control.
The decrease in the 100 seed weight may be due to induced mutation in meiotic cycle which affected the frequency of normal microspores up to greater extent and the megaspore to a lesser extent and hence the fruit set was directly affected (Amin *et al.*, 2015) [1]. Ogidi *et al.*, (2010) [19], Reena *et al.*, (2014) [20], Mohamed *et al.* (2020) [17] in cowpea, Elangovan and Pavadai (2015) [3] in okra and Kavera and Nadaf (2017) [8] in groundnut observed a similar trend in the variation induced across the mutant lines for the character of 100 seed weight.

3.3 Physiological parameters

3.3.1 Rate of photosynthesis (µmol m^{-2} s^{-1})

The rate of photosynthesis of the 31 mutant lines of M2 generation ranged from 6.17 µmol m^{-2} s^{-1} to 13.76 µmol m^{-2} s^{-1} with a mean value of 9.37 µmol m^{-2} s^{-1}. The highest (13.76 µmol m^{-2} s^{-1}) rate of photosynthesis was recorded in $M_{2.10}$ on par with M2.11 (12.87 µmol m^{-2} s^{-1}) followed by $M_{2.12}$ (11.46 µmol m^{-2} s^{-1}) and the lowest (6.17 µmol m^{-2} s^{-1}) rate of photosynthesis was shown in $M_{2.4}$. The control mean was registered as 10.47 µmol m^{-2} s^{-1}. Out of 31 mutant lines evaluated, five mutants *viz.*, M2.8 (10.67 µmol m^{-2} s^{-1}), $M_{2.10}$ (13.76 µmol m^{-2} s^{-1}), M2.11 (12.87 µmol m^{-2} s^{-1}), $M_{2.12}$ (11.46 µmol m^{-2} s^{-1}), and M2.32 (10.57 µmol m^{-2} s^{-1}) recorded a higher rate of photosynthesis than control mean. Singh and Singh (2007) [24] reported increase in photosynthesis in guava with increase in radiance.

3.3.2 Stomatal conductance (mmol m^{-2} s^{-1})

Significant variation was noticed among the mutant lines of M2 generation with respect to stomatal conductance. Stomatal conductance of the 31 mutant lines ranged from 0.51 mmol m^{-2} s^{-1} to 2.46 mmol m^{-2} s^{-1} with a mean value of 1.11 mmol m^{-2} s^{-1}. Stomatal conductance was maximum in $M_{2.28}$ (2.46 mmol m^{-2} s^{-1}) on par with $M_{2.3}$ (2.16 mmol m^{-2} s^{-1}), $M_{2.4}$ (2.10 mmol m^{-2} s^{-1}), $M_{2.17}$ (1.87 mmol m^{-2} s^{-1}) followed by $M_{2.12}$ (1.72 mmol m^{-2} s^{-1}) and minimum in $M_{2.32}$ (0.51 mmol m^{-2} s^{-1}). Stomatal conductance in untreated control recorded a mean of 2.22 mmol m^{-2} s^{-1}. One mutant line *viz.*, $M_{2.28}$ recorded higher stomatal conductance (2.46 mmol m^{-2} s^{-1}) when compared to control.
Stomatal conductance is an important biological determinate of carbon accumulation and transpiration by plants, because the flow of CO_2 into the leaf is controlled by stomatal regulatory processes. Stomatal conductance of guava cultivars significantly varied within the cultivars (Shiva *et al.*, 2017) [23].

3.3.3 Rate of transpiration (mmol m^{-2} s^{-1})

The rate of transpiration of the 31 mutant lines of M2 generation ranged from 2.45mmol m^{-2} s^{-1} to 6.93 mmol m^{-2} s^{-1} with a mean value of 4.124 mmol m^{-2} s^{-1}. Rate of transpiration was maximum in $M_{2.32}$ (6.93 mmol m^{-2} s^{-1}) on par with $M_{2.3}$ (5.76 mmol m^{-2} s^{-1}), $M_{2.6}$ (4.96 mmol m^{-2} s^{-1}), $M_{2.7}$ (5.51 mmol m^{-2} s^{-1}), $M_{2.9}$ (5.65 mmol m^{-2} s^{-1}), $M_{2.10}$ (4.94 mmol m^{-2} s^{-1}), $M_{2.11}$ (5.45 mmol m^{-2} s^{-1}), $M_{2.16}$ (5.09 mmol m^{-2} s^{-1}), $M_{2.21}$ (5.09 mmol m^{-2} s^{-1}) followed by $M_{2.4}$ (4.88 mmol m^{-2} s^{-1}) and minimum in $M_{2.15}$ (2.45 mmol m^{-2} s^{-1}). The mean value of the rate of transpiration in untreated control was recorded as 5.339 mmol m^{-2} s^{-1}. Five mutant lines *viz.*, $M_{2.3}$ (5.76 mmol m^{-2} s^{-1}), $M_{2.7}$ (5.51 mmol m^{-2} s^{-1}), $M_{2.9}$ (5.65 mmol m^{-2} s^{-1}), M2.11 (5.45 mmol m^{-2} s^{-1}), and $M_{2.32}$ (6.93 mmol m^{-2} s^{-1}) recorded a higher rate of transpiration when compared to control.
The stomatal control of transpiration rate is an important component of the leaf energy balance and can be of great importance for maintaining an optimal or appropriate leaf temperature for photosynthesis particularly under conditions of increasing or highlight intensity that are observed over a typical diurnal period. Transpiration often is seen as a cost for carbon fixation at the leaf level, but it is important to take into consideration its roles in the transport of solutes in the

different parts of the plant or for leaf cooling (Caird *et al.*, 2007) [2].

3.3.4 Leaf temperature (ºC)

Significant variation was noticed among the mutant lines of M_2 generation with respect to leaf temperature. Leaf temperature of the 31 mutant lines ranged from 29.02 ºC to 35.04 ºC with a mean value of 33.18 ºC. Maximum leaf temperature was recorded in $M_{2.28}$ (35.04 ºC) which was on par with all mutant lines except $M_{2.4}$, $M_{2.8}$, $M_{2.22}$, $M_{2.24}$, $M_{2.29}$, $M_{2.30}$, followed by $M_{2.3}$ (32.07 ºC) and minimum in $M_{2.10}$ (29.02 ºC). Leaf temperature in untreated control recorded a mean of 33.02 ºC. Nineteen mutant lines recorded higher leaf temperature when compared to control.

3.3.5 Internal cellular CO2 (µmol CO2 m^{-2} s^{-1})

In M2 generation, internal cellular CO2 ranged from 256.10 µmol CO2 m^{-2} s^{-1} to 338.85 µmol CO2 m^{-2} s^{-1} with a grand mean of 294.32 µmol CO2 m^{-2} s^{-1}. The maximum internal cellular CO2 (338.85 µmol CO2 m^{-2} s^{-1}) was recorded in the mutant line $M_{2.18}$ which was on par with $M_{2.11}$ (330.84 µmol CO2 m^{-2} s^{-1}) and $M_{2.29}$ (334.65 µmol CO2 m^{-2} s^{-1}) followed by $M_{2.28}$ (319.17 µmol CO2 m^{-2} s^{-1}), while the minimum internal cellular CO_2 was recorded in $M_{2.26}$ (256.10 µmol CO2 m^{-2} s^{-1}). Internal cellular CO_2 of untreated control was observed as 306.68 µmol CO2 m^{-2} s^{-1}. Eight mutant lines *viz.*, $M_{2.2}$ (310.95 µmol CO2 m^{-2} s^{-1}), $M_{2.7}$ (307.22 µmol CO2 m^{-2} s^{-1}), $M_{2.11}$ (330.84 µmol CO2 m^{-2} s^{-1}), $M_{2.18}$ (338.85 µmol CO2 m^{-2} s^{-1}), M2.19 (316.00 µmol CO2 m^{-2} s^{-1}), M2.20 (312.10 µmol CO2 m^{-2} s^{-1}), $M_{2.28}$ (319.17 µmol CO2 m^{-2} s^{-1}), $M_{2.29}$ (334.65 µmol CO2 m^{-2} s^{-1}) recorded significantly higher internal cellular CO2 when compared to the control.

3.4 Protein content (mg/ 100 g)

Protein content in field bean mutant lines of M2 generation ranged from 6.46 mg/ 100 g to 8.06 mg/ 100 g with a grand mean of 7.24 mg/ 100 g. The highest protein content (8.06 mg/ 100 g) was recorded in M2.28 on par with M2.11 (8.05 mg/ 100 g) and M2.17 (8.05 mg/ 100 g) followed by M2.26 (8.00 mg/ 100 g), while the lowest (6.46 mg/ 100 g) was recorded in $M_{2.2}$. Out of 31 mutant lines, nine mutant lines recorded the highest protein content over the grand mean. Seven mutant lines *viz.*, $M_{2.11}$ (8.05 mg/ 100 g), $M_{2.17}$ (8.05 mg/ 100 g), $M_{2.24}$ (8.04 mg/ 100 g), $M_{2.25}$ (7.92 mg/ 100 g), $M_{2.26}$ (8.00 mg/ 100 g), $M_{2.28}$ (8.06 mg/ 100 g) and $M_{2.31}$ (7.93 mg/ 100 g) had highest protein content when compared to control (7.88 mg/ 100 g).

Gamma rays were found to cause alteration in protein constituents by deamination of the proteins (Taha and Mohamed, 2004) [4] or by inducing appearance and/or disappearance of some protein bands (Rashed *et al.*, 1994) [14]. In addition, gamma rays caused aggregation of the low molecular weight proteins to a high molecular weight due to the formation of disulfide bridge between polypeptide chains resulting in decrease of protein solubility (Singh and Singh, 2015) [21].

The results are in agreement with that of More and Borkar (2016) [18] in French bean, Khan *et al.* (2018) [10] and Masry, *et al.* (2019) [16] in pea and Harish Kumar *et al.* (2018) [6] in dolichos bean.

3.5 SCMR

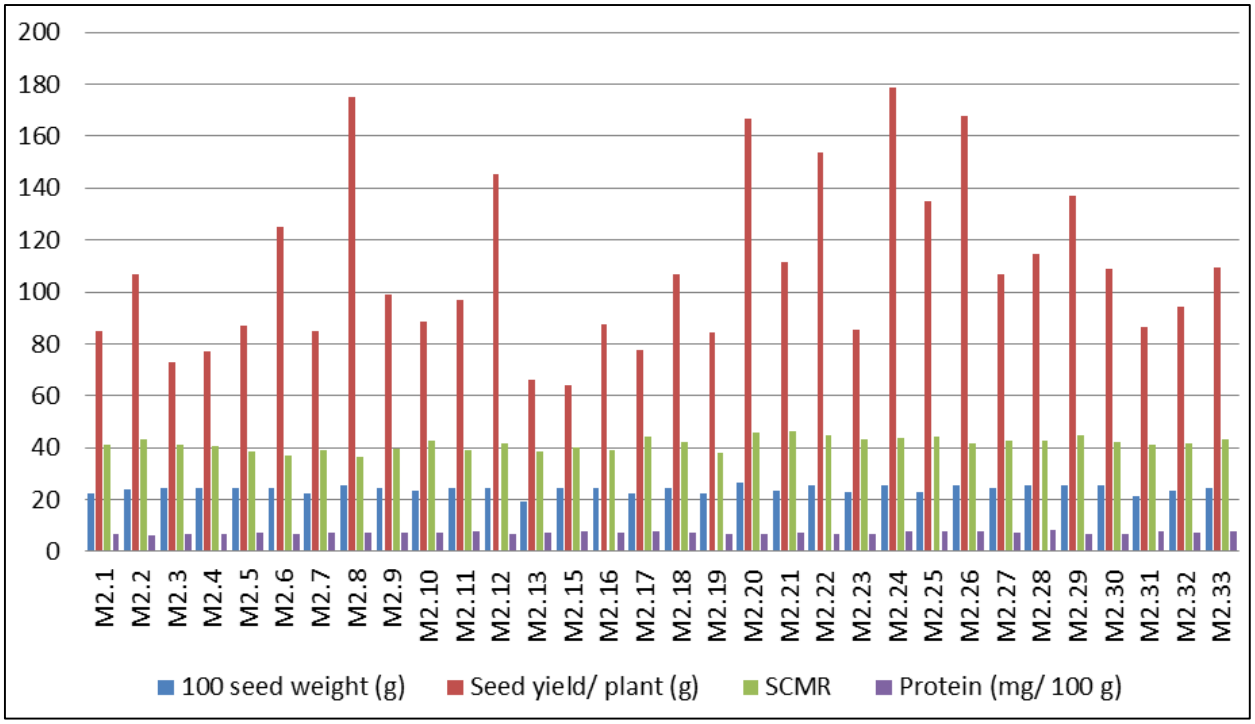
From the data recorded, the range for SCMR values in the mutant population of M2 generation ranged from 36.38 to 46.34 with a mean of 41.51. The maximum SCMR values were recorded in $M_{2.21}$ (46.34) on par with all other mutants except $M_{2.1}$, $M_{2.3}$, $M_{2.4}$, $M_{2.5}$, $M_{2.6}$, $M_{2.7}$, $M_{2.9}$, $M_{2.11}$, $M_{2.13}$, $M_{2.15}$, $M_{2.16}$ and $M_{2.19}$, followed by $M_{2.31}$ (41.13) while minimum SCMR values were recorded in $M_{2.8}$ (36.38) while the untreated control recorded 43.23. Seven mutant lines *viz.*, $M_{2.17}$ (44.42), $M_{2.20}$ (45.79), $M_{2.21}$ (46.34), $M_{2.22}$ (44.96), $M_{2.24}$ (43.88), $M_{2.25}$ (44.11), and $M_{2.29}$ (45.01) were observed to have higher SCMR than control.

Kiong *et al.* (2008) [12] reported reduction in chlorophyll which might be due to a more selective destruction of chlorophyll biosynthesis or degradation of chlorophyll precursors whereas Kim *et al.* (2000) [11] stated that the increased chlorophyll content can be correlated with stimulated growth at low doses of irradiation. The findings were in parallel with the recordings of Kozgar (2014) [13] in chick pea, Sharma *et al.* (2010) [22] and Xu *et al.* (2020) [26] in pea, Harish Kumar *et al.* (2018) [6] in dolichos bean, More and Borkar (2016) [18] in French bean and Girija and Dhanavel (2013) [5] in cowpea.

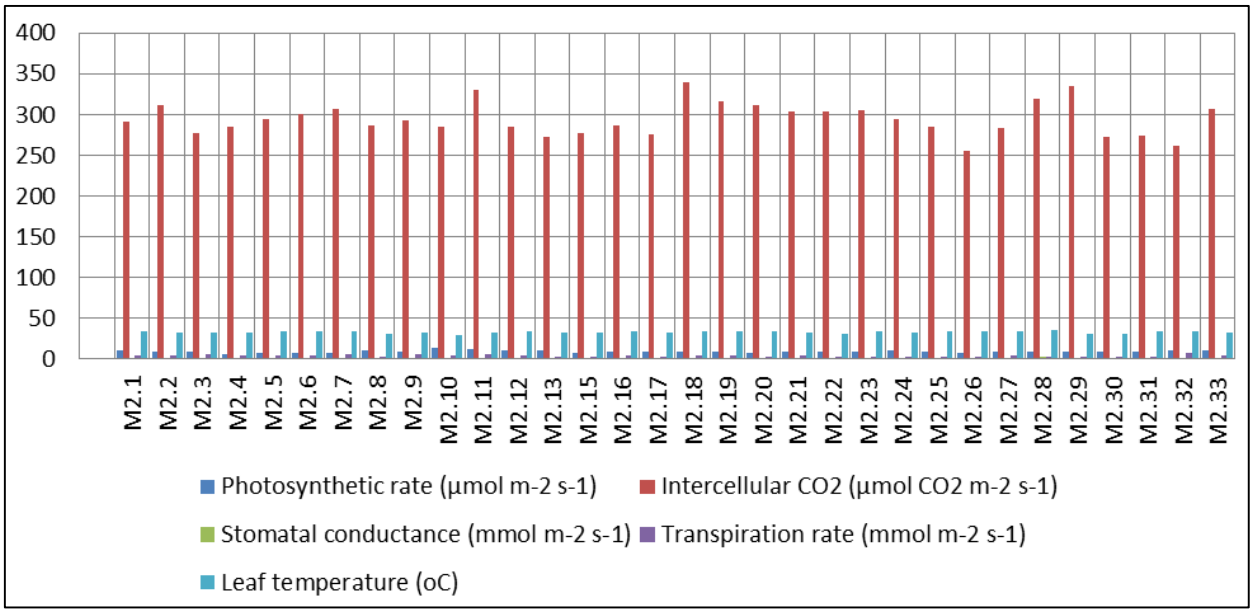
Table 1: Mean performance of the mutant lines of gamma irradiated field bean var. TFB-2 for yield, quality and physiological attributes in M_2 generation

Mutant Line	100 Seed Weight (g)	Seed Yield/ Plant (g)	SCMR	Protein (mg/100 g)	Photosynthetic Rate (µmol m^{-2} s^{-1})	Intercellular CO_2 (µmol CO_2 m^{-2} s^{-1})	Stomata Conductance (mmol m^{-2} s^{-1})	Transpiration rate (mmol m^{-2} s^{-1})	Leaf Temperature (ºC)
$M_{2.1}$	22.52	85.02	41.04	6.75	10.41	291.75	0.97	4.21	34.68
$M_{2.2}$	24.18	106.96	43.09	6.46	9.05	310.95	0.77	4.37	32.59
$M_{2.3}$	24.51	73.11	41.13	6.77	10.01	277.27	2.16	5.76	32.07
$M_{2.4}$	24.61	77.27	40.43	6.78	6.17	284.23	2.10	4.88	31.95
$M_{2.5}$	24.23	86.80	38.57	7.16	7.16	294.99	1.50	4.40	34.07
$M_{2.6}$	24.49	124.95	36.75	6.97	7.20	301.27	0.77	4.96	34.54
$M_{2.7}$	22.59	84.77	39.15	7.12	7.34	307.22	0.85	5.51	33.82
$M_{2.8}$	25.60	174.89	36.38	7.04	10.67	285.79	0.87	2.61	30.33
$M_{2.9}$	24.62	98.98	39.56	7.10	9.56	292.79	0.79	5.65	33.36
$M_{2.10}$	23.57	88.38	42.49	7.23	13.76	285.00	1.11	4.94	29.02
$M_{2.11}$	24.43	96.92	38.89	8.05	12.87	330.84	1.03	5.45	33.28
$M_{2.12}$	24.47	145.36	41.91	6.86	11.46	285.47	1.72	4.83	34.50
$M_{2.13}$	19.51	66.25	38.65	7.41	10.39	272.29	0.69	3.53	32.75
$M_{2.15}$	24.65	64.16	40.19	7.87	8.27	277.75	0.80	2.45	33.01
$M_{2.16}$	24.35	87.63	38.84	7.13	9.97	286.82	0.67	5.09	34.60
$M_{2.17}$	22.51	77.57	44.42	8.05	9.19	276.37	1.87	3.62	33.35
$M_{2.18}$	24.53	106.97	41.96	7.05	9.45	338.85	0.65	4.15	33.67

$M_{2.19}$	22.42	84.29	38.23	6.83	9.22	316.30	0.76	4.18	33.90
$M_{2.20}$	26.80	166.83	45.79	6.84	7.84	312.10	0.77	3.31	34.01
$M_{2.21}$	23.39	111.54	46.34	7.16	9.48	303.97	0.96	5.09	33.02
$M_{2.22}$	25.37	153.75	44.96	6.84	8.93	304.15	0.92	3.15	31.12
$M_{2.23}$	22.75	85.45	43.04	6.92	8.77	305.79	0.99	3.30	34.47
$M_{2.24}$	25.68	178.67	43.88	8.04	10.22	294.34	1.43	3.01	31.98
$M_{2.25}$	22.88	134.81	44.11	7.92	8.73	285.65	0.96	3.50	34.00
$M_{2.26}$	25.26	167.81	41.48	8.00	8.29	256.10	1.70	2.95	33.57
$M_{2.27}$	24.72	106.56	42.58	7.07	9.10	283.59	0.98	4.05	33.94
$M_{2.28}$	25.71	114.51	42.76	8.06	8.98	319.17	2.46	2.59	35.04
$M_{2.29}$	25.33	137.22	45.01	6.89	9.10	334.65	0.95	3.61	31.61
$M_{2.30}$	25.25	108.89	42.14	6.91	9.52	272.72	0.91	2.71	30.94
$M_{2.31}$	21.36	86.31	41.13	7.93	8.76	273.50	0.87	3.05	34.89
$M_{2.32}$	23.36	94.50	41.81	7.10	10.57	262.32	0.51	6.93	34.48
$M_{2.33}$	24.46	109.55	43.23	7.88	10.47	306.68	2.22	5.34	33.02
SE(m)±	0.21	2.04	1.743	0.016	0.639	3.23	0.225	0.701	0.860
CD 5%	0.59	5.90	5.050	0.046	1.851	9.37	0.652	2.032	2.493



Graph 1: Yield and Quality parameters of the elite mutants of gamma irradiated field bean var. TFB-2 in M_2 generation



Graph 2: Physiological parameters of the elite mutants of gamma irradiated field bean var. TFB-2 in M_2 generation

4. Conclusion

The results on the mean performance of mutant lines in the M2 generation revealed significant variability for all the observations.

Among all the mutant lines of M_2 generation, Mutant line $M_{2.10}$ observed a higher rate of photosynthesis. The mutant line $M_{2.21}$ recorded highest SCMR values. The better performance for 100 seed weight was recorded by the mutant line $M_{2.20}$. The mutant line $M_{2.24}$ was documented to be a better performer for seed yield per plant. The intercellular CO_2 was noted to be highest in $M_{2.18}$ whereas the rate of transpiration was highest in $M_{2.32}$. Mutant line $M_{2.28}$ recorded the highest leaf temperature and stomatal conductance with the highest protein content.

The mutant lines showing better performance for all the aforementioned traits can be selected as desirable mutants and progressed to M_3 generation for further improvement.

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