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Studies on Internode Length and Culm Anatomy of Induced Mutants of Aromatic Rice

K. M. Hasib

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ABSTRACT

Dry unhusked seeds of Tulaipanja, a tall aromatic *indica* rice, were irradiated with 200 Gy and 300 Gy doses of gamma rays for inducing short height mutants. A number of non-lodging true-breeding height mutants were studied in advanced generation. The number and length of the internodes of various mutants along with their parent Tulaipanja were studied for alteration in these characters for height reduction and culm anatomy for stiff-strawed non-lodging habit. There was no alteration in the number of identifiable internodes in the short culm mutants. All the mutants and the control Tulaipanja showed progressive decrease in individual internode length from the top (numbered in descending order) to downwards, with the panicle bearing internode (number one) being the longest. However, the mutants showed variation in the pattern and degree of reduction of individual internodes. All the mutant lines and control Tulaipanja except one mutant line (33-9-15) had almost similar number of vascular bundles. The thick band of sclerenchymatous tissue in the hypodermal region in the stiff-strawed mutant 88-8-3 may contribute to the rigidity of culm for non-lodging habit.

Keywords Internode, Culm anatomy, Induced mutants, Aromatic rice.

INTRODUCTION

The development of short statured non-lodging semi-dwarf plant is the important breeding objective in crop improvement program. A significant part of the success of the 'Green Revolution' in the 1960s resulted from the breeding of grain crops that had more efficient plant architecture (Khush 2001). The short statured non-lodging habit of the plant is associated with some morphological or anatomical features of stem which have a close relationship with yield. Lodging is the most important factor associated with reduction of grain yield and may reduce the grain yield significantly. The major constraints in increasing grain yield of traditional aromatic rice varieties are the lack of response to higher doses of fertilizers and susceptibility to lodging. Tulaipanja is such type of local aromatic tall cultivar which is susceptible to lodging. Mutants were isolated from such aromatic cultivar through induced mutation by gamma ray irradiation. These mutants are semi-dwarf in nature having superior plant type and are resistant

K. M. Hasib
Assistant Professor, Department of Botany, Sarat Centenary College, Dhaniakhali 712302, Hooghly, West Bengal, India

Email: kmhasib@yahoo.co.in

to lodging. The mutants responded to higher doses of fertilizers resulting higher yield than their parent. The number and length of internodes are directly associated with the culm length of rice plant and hence important for understanding height reduction in the semi-dwarf mutants. Internodes and the cell wall play important roles to attain suitable height and also to provide mechanical support to hold high yielding panicle (Sato Izawa *et al*. 2020). Some anatomical features like the size and arrangement of vascular bundles and the types of tissues which are directly associated with the culm strength are also important. In this context, an investigation was made on the morphology of internodes and some anatomical features of culm in true-breeding induced mutants including its parent Tulaipanja.

MATERIALS AND METHODS

The dry unhusked seeds of local aromatic rice landrace Tulaipanja were irradiated with 200 Gy and 300 Gy doses of gamma rays to induce mutations. Single plant selection following pedigree method was practised in segregating generations of mutated population, till apparent homozygosity was achieved. Finally some true-breeding short height promising mutants were isolated and maintained. Seeds of 11 selected mutants with variable plant height and the control Tulaipanja were sown in nursery bed. Twenty-five days old seedlings were transplanted in separate plot under puddled field condition with spacing of 20cm×15cm. Uniform management practices were followed. Ten plants after seven days of flowering were randomly sampled from the middle of the plot to exclude border effects. After removing the leaf sheath, numbers of developed internodes were counted from the primary tiller of ten randomly selected plants from each mutant families and their parent Tulaipanja. The length of internodes was measured in centimeter (cm). Internode length below 5 mm towards the base of the plant was not considered for measuring. Internodes were numbered from top to downwards in descending order i.e., internode bearing the panicle was numbered as the first one and so on.

Anatomical features of some non-lodging and semi-lodging aromatic mutants and their parent Tulaipanja were studied from transverse sections of the upper internode of stem. The sections were stained by double staining method. After passing through 30% and 50% alcohol grades for one minute, the sections were stained with safranine for 15 minutes. The sections were then washed with alcohol grades of 50%, 70% and 90% and were stained with light green which was dissolved with 90% alcohol. Sections were subsequently washed in alcoholic grades of 90% and 100% for one minute each and finally mounted in euparol. The sample sections were then observed under microscope. The arrangement of vascular bundles was also studied. Data on number of small and large vascular bundles were recorded.

RESULTS AND DISCUSSION

The number and length of internodes of various mutants and their mother Tulaipanja were studied to analyze the effect of these traits in plant height of the mutants (Table 1). There is a tendency of scented rice to lodge due to wind and rain because of its tall height (Mukherjee 2020). Because of the close relationship of number and length of internodes with lodging resistance, variation in these characters in induced short culm mutants have been extensively studied by Ehrenberg *et al*. (1956), Kawai *et al*. (1961) and Reddy *et al*. (1975). Reddy *et al*. (1975) viewed that the reduction in height may result either from reduction in internode number and/or internode length.

The number and length of internodes are associated with lodging resistance of short culm mutants. Zhao *et al*. (2021) opined that the internode length and stem diameter are the important characters affecting the lodging resistance of rice. Reddy *et al*. (1975) also reported induced variation in the number and length of internodes in rice. In the present investigation, there was a little difference in the number of internodes in the short culm mutants of different height groups of plants. The average number of identifiable internodes in the short culm mutants was 5.00 as compared to 6.80 in control. So, there is role of internode number in shortening the culm length in the short culm mutants. The results are in agreement with the observations of Ganashan and Whittington (1975) and Reddy *et al*. (1975) and Narahari (1988).

Table 1. Average number and length (in cm) of internodes of mother Tulaipanja and its mutants.

Parent/ Mutants	Average No. of Inter-nodes	Internode length (in cm) and per cent (%) contribution to total culm length							
		Internode-1		Internode-2		Internode-3		Internode-4	
		cm	%	cm	%	cm	%	cm	%
Tulaipanja	6.80	40.65	34.36	24.90	21.05	20.60	17.41	14.45	12.22
300Gy									
2-16-7	7.00	40.60	35.11	24.40	21.10	16.50	14.27	14.30	11.66
17-18-16	5.80	34.65	39.28	23.25	26.36	14.00	15.87	9.05	10.26
32-9-4	6.80	45.15	40.29	26.90	24.00	19.00	16.95	12.25	10.93
35-13-13	6.10	35.35	41.01	20.85	24.19	13.30	15.43	8.65	10.03
65-12-11	6.80	41.55	37.30	25.50	22.89	16.65	14.95	12.20	10.95
77-19-16	7.00	40.45	36.52	22.65	20.45	15.35	13.86	13.35	12.05
200Gy									
32-19-17	7.00	35.20	31.39	24.20	21.58	19.45	17.35	14.60	13.02
33-9-15	5.90	30.90	39.17	19.40	24.59	13.50	17.11	8.00	10.14
33-19-17	6.00	32.90	42.56	16.40	21.22	10.30	13,32	8.45	10.93
88-8-3	5.00	27.45	41.01	17.65	26.37	12.95	19.35	6.80	10.16
88-8-18	5.00	25.00	38.26	17.00	26.02	13.60	20.81	7.75	11.86

Table 1. Continued.

Parent/ Mutants	Average No. of Inter-nodes	Internode length (in cm) and per cent (%) contribution to total culm length						Culm length excluding panicle (cm)
		Internode-5		Internode-6		Internode-7		
		cm	%	cm	%	cm	%	
Tulaipanja	6.80	11.00	9.30	4.90	4.14	1.79	1.51	118.29
300Gy								
2-16-7	7.00	10.75	8.76	6.75	5.50	2.35	1.92	115.65
17-18-16	5.80	5.10	5.78	2.16	2.45	-	-	88.21
32-9-4	6.80	6.70	5.98	2.07	1.85	1.65	1.45	113.72
35-13-13	6.10	6.00	6.96	2.05	2.39	-	-	86.20
65-12-11	6.80	8.35	7.50	5.20	4.67	1.93	1.73	111.38
77-19-16	7.00	10.05	9.07	6.50	5.87	2.40	2.17	110.75
200Gy								
32-19-17	7.00	11.00	9.81	6.00	5.35	1.68	1.50	112.13
33-9-15	5.90	4.85	6.15	2.24	2.84	-	-	78.89
33-19-17	6.00	6.25	8.09	3.00	3.88	-	-	77.30
88-8-3	5.00	2.08	3.11	-	-	-	-	66.93
88-8-18	5.00	1.99	3.05	-	-	-	-	65.34

It was observed that all the mutant lines including control showed, in general, progressive decrease in individual internode length from the top to downwards, where the panicle bearing internode was the longest. Similar observations were also noticed by Reddy *et al*. (1975). The relative contribution of top two internodes to culm length was more than 50% in all the mutants as well as in control. Narahari (1988) also observed more than 50% contribution of top two internodes in the mutants.

In general, the contribution of 1st internode and 2^{nd} internode (from top) to the total culm length in the mutant with shorter culm was higher as compared

Table 2. Percentage reduction in the length of corresponding internode in the mutants of Tulaipanja. (-) indicates no reduction.

Mutant lines	Internode-1	Internode-2	Internode-3	Internode-4	Internode-5	Internode-6	Internode-7	Remarks
300Gy								
2-16-7	0.12	2.01	21.84	3.11	2.26	(-)	(-)	3>4>5>2>1
17-18-16	14.76	6.63	32.04	37.37	53.64	55.92	(-)	6>5>4>3>1>2
32-9-4	(-)	(-)	7.77	15.22	39.09	57.16	7.82	6>5>4>7>3
35-13-13	13.04	16.27	35.44	40.14	45.45	58.16	(-)	6>5>4>3>2>1
65-12-11	(-)	(-)	19.17	15.57	24.09	(-)	(-)	5>3>4
77-19-16	0.49	9.04	25.49	7.61	8.64	(-)	(-)	3>2>5>4>1
200Gy								
32-19-17	13.41	2.81	5.58	(-)	(-)	(-)	6.15	1>7>3>2
33-9-15	23.98	22.09	34.47	44.64	55.91	54.29	(-)	5>6>4>3>1>2
33-19-17	19.07	34.14	50.00	41.52	43.18	38.78	(-)	3>5>4>6>2>1
88-8-3	32.47	29.12	37.14	52.94	81.09	(-)	(-)	5>4>3>1>2
88-8-18	38.50	31.73	33.98	46.37	81.91	(-)	(-)	5>4>1>3>2

to the mutants with higher culm length and control. There was not much difference in the contribution of 3rd internode towards culm length among the mutants and control. The contribution of 4th internode to the total culm length was more or less similar in all the mutants and control. The contribution of 5th internode to the total culm length in the mutants with shorter culm length was lower as compared to the mutants with higher culm length and control. All the mutants with taller culm length had greater contribution in 6th internode to the total culm length which was more or less similar in control. There was not much difference in the contribution of seventh internode to the total culm length in the mutants and control.

In general, the pattern of reduction in internode length observed in the present study (Table 2) follows the pattern of reduction in having highly reduced basal internodes and a relatively little reduction of top internodes. Similar results were also reported by Reddy *et al*. (1975).

While, considering the degree of reduction of individual internode with respect to control, it was observed that maximum reduction took place in the two basal internodes in the semi-dwarf and semi-tall mutant lines except the semid-warf mutant 33-19-17. Zhong *et al*. (2020) reported that the short basal internodes are important for lodging resistance of rice. Rao *et al*. (2017) showed that lodging tolerant varieties in general have lower plant height and stem lengths along with lower length of basal internode compared to lodging susceptible varieties. In general, the contribution towards reduction in height was highest in 2nd internode followed by 1st internode. The reduction in 1st internode was slightly lower than the 2nd internode. Narahari (1988) observed that the internode below 3rd have major contribution in reducing the height of short culm mutants. Reddy *et al*. (1975) also reported greater per cent contribution of the lower internodes and lower percentage contribution of the upper internode in reducing the height of short culm mutants.

In other semi-dwarf mutant 33-19-17 and semi-tall-II mutant 2-16-7 and 77-19-16, the highest percentage contribution towards reduction was observed in the 3rd internode. Such variations in the pattern of reduction in individual internode have been observed by Ganashan and Whittington (1975).

Anatomical study of stem in the non-lodging and semi-lodging mutants 88-8-3, 33-9-15, 17-18-16, 35-13-14, 77-19-16 along with the parent Tulaipanja (Table 3) revealed that the mutant 88-8-3 had thicker band of sclerenchymatous tissue in the hypodermis than the control parent which might have contributed to the rigidity of this mutant as well as the non-lodging habit than the others including the control. The stem with a thick sclerenchymatous tissue and more vascular bundles were rigid. High numbers of sclerenchymatous cells are involved to impart lodging resistance.

The number of vascular bundles in the mutant

Table 3. Anatomical features of stem in selected short culm mutants and control Tulaipanja.

Mutants/ Control	Salient features	No. of vascular bundle Large	Small	Extra sclerenchymatous band
Control				
Tulaipanja	Tall indica traditional aromatic rice cultivar	9.77	13.33	Absent
Mutants				
17-18-16	Short statured culm, erect plant with higher number of tillers, semi-lodging plants	8.00	11.00	Absent
33-9-15	Short statured thin culm, erect, narrow green leaves with higher number of tillers, non-lodging plants	7.25	8.25	Absent
35-13-13	Erect plant, dark green leaves, semi-lodging plant	11.33	11.33	Absent
77-19-16	Tall, erect culm, semi-lodging plant	9.00	11.00	Absent
88-8-3	Short statured culm, erect, broad dark green leaves, higher no. of spikelets, awn less grain, non-lodging plants	12.50	10.67	Present

lines and control were more or less same except 88-8-3 and 33-9-15. The number of large vascular bundle was higher in the mutant 88-8-3 only, which was lower in the other mutant lines and control. It may contribute the stout nature of culm. The number of vascular bundle in the mutant 33-9-15 was lower than other mutants which were probably due to the thin diameter of the culm. Xu *et al*. (2000) opined that the number of vascular bundles had strong positive association with resistance to lodging. The high number of vascular bundles is important character to enhance the yield. The high yielding varieties had thicker internodes and the ground tissue had a large number of vascular bundles, consequently helping to create resistance to lodging. In this investigation, the non-lodging character of the mutant 88-8-3 might be due to the larger size of vascular bundles as well as thicker sclerenchymatous zone in the hypodermal region of stem.

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