

ORIGINAL ARTICLE



Colchicine induced alterations in plant architecture of common buckwheat (*Fagopyrum esculentum* Moench)

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Abstract

Colchicine treatment was used to create and characterize phenotypic variation in common buckwheat (*Fagopyrum esculentum* Moench), with emphasis on plant architecture and cytological traits. Seeds of cultivar VL-7 were treated with 0.05% colchicine for 8 and 12h, and the resulting C_1 population was screened for conspicuous morphological variation. Fifteen distinct plants were selected and categorized into five phenotypic groups: dwarf, highly branched, tall, thick-stemmed, and broad-leaved variants. Among the selected variants, plant height ranged from 17 to 153 cm, compared with 107cm in the untreated control. Stem diameter of the thick-stemmed variants ranged from 3.70 to 4.10 cm, compared with 1.70 cm in the control, while leaf area of the broad-leaved variants ranged from 23.93 to 25.17 cm², compared with 19.80cm² in the control. DW1 represented a distinct dwarf phenotype, attaining a height of 17cm and reaching maturity in 46 days compared with 52 days in the control, whereas TL1 was the tallest accession (153 cm). BR1, BR2, and BR3 exhibited increased primary branching, whereas TS1, TS2, and TS3 showed pronounced stem thickening. BL1, BL2, BL3, and BL4 exhibited broader leaves than the control. No evident differences in pollen morphology were observed between the selected accessions and the control. Cytological studies showed that all selected plants were diploid ($2n=16$), and stable chromosome doubling was not observed. These results demonstrate that colchicine treatment was associated with substantial morphological diversity in the C_1 generation despite the absence of confirmed chromosome doubling. The selected plants therefore represent colchicine-associated phenotypic variants rather than confirmed polyploids.

KEYWORDS

Phenotypic variation; Morphological variants; Plant architecture; Dwarf variants; Colchicine; *Fagopyrum esculentum*

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Introduction

Buckwheat (*Fagopyrum spp.*) is an important pseudocereal belonging to the family Polygonaceae and includes two major cultivated species, common buckwheat (*Fagopyrum esculentum* Moench) and Tartary buckwheat (*Fagopyrum tataricum* (L.) Gaertn.) [1,2]. It is valued for its nutritional and functionality properties, including high quality protein, dietary fibre, minerals, essential amino acids, and bioactive compounds, particularly flavonoids such as rutin and quercetin [3,4]. Its gluten free nature, short growth duration, and adaptability to diverse environments have further increased its importance as a food and crop species [5,6]. However, the production of buckwheat is limited by several agronomic limitations such as low and unstable seed yield, poor seed set, flower and seed abortion, susceptibility to lodging and seed shattering [7,8].

Improvement of plant architecture and yield related traits is therefore an important aim in buckwheat breeding. Induced phenotypic variation offers a way to expand the range of selectable traits for crop improvement. Colchicine is an anti-mitotic agent that is frequently used in plant breeding to induce chromosome doubling by disturbing spindle formation during cell division [9]. Chromosome doubling can result in morphological changes, including higher stem thickness, larger leaves and reproductive structures, altered plant stature, increased branching and other gigas like features [10]. In common buckwheat polyploid colchicine-induced changes in leaf characteristics, stomatal size, pollen size, floral characteristics and seed weight have been shown [11]. In spite of this treatment with colchicine does not always lead to stable chromosome doubling and the phenotypic responses may occur without any observable change in the chromosome number. Thus, the assessment of morphological variation after colchicine treatment must be made in combination with reproductive and cytological observations. In the present study, colchicine treated common buckwheat populations were screened in the C_1 generation for conspicuous phenotypic variants such as dwarf,

tall, highly branched, thick stemmed and broad leaved plants. Pollen morphology and chromosome number were studied to see if the phenotypic variation observed was correlated with any changes in the pollen characteristics or chromosome complement. The investigation therefore sought to characterize the colchicine-associated phenotypic variation in the C_1 generation and to determine if the selected variants showed detectable changes in pollen morphology or chromosome number following colchicine treatment.

Materials and Methods

Plant material and colchicine treatment

Seeds of common buckwheat (*Fagopyrum esculentum* Moench) variety VL-7 were obtained from National Bureau of Plant Genetic Resources (NBPGR), New Delhi, India. Seeds were soaked in distilled water for 12h and treated with 0.05% (w/v) colchicine for 8 or 12h. For each treatment period 300 seeds were treated and the same number of seeds soaked in distilled water for the same period served as untreated control. Following treatment, the seeds were thoroughly washed under running tap water and sown during the first week of May at the Kashmir University Botanical Garden (KUBG), Department of Botany, University of Kashmir, under natural agro-climatic conditions to raise the C_1 generation. The experiment was conducted in loamy soil without the application of chemical fertilizers or organic manure. Irrigation was provided at approximately 2–3-day intervals depending on prevailing weather conditions. The average temperature during the growing period (May–August) ranged from approximately 20–32 °C, and plants were exposed to natural day-length conditions. No fungicides were applied during the experiment, and plants were maintained under natural pest and disease conditions. The crop was harvested at the end of August. The experiment was laid out in a completely randomized design with three replications.

Screening and characterization of phenotypic variants

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The C_1 populations were screened during the growing season for obvious variation in plant architecture and growth habit. Fifteen plants with different phenotypic characteristics were selected and tagged individually and given accession numbers from the treated populations. The plants selected were grouped according to their most obvious morphological characteristics into dwarf, tall, highly branched, thick stemmed and broad leaved types. Plant height (cm), stem diameter (cm), number of primary branches per plant and leaf area (cm^2) were recorded for the selected plants and untreated control. Leaf area was measured using a leaf area meter, and the mean leaf area was calculated from ten randomly selected, fully expanded leaves from each selected plant. Representative photographs were taken to document the distinct phenotypes.

Pollen morphology

Fresh flowers from the selected C_1 accessions and untreated control plants were collected for mature anthers. Pollen grains were extracted by crushing the anthers in 1% acetocarmine on clean glass slides. Under a compound light microscope, pollen grains were studied for apparent differences in size and shape and representative microphotographs were taken.

Cytological analysis

Flower buds of the selected C_1 lines were separately collected and fixed in Carnoy's fixative (ethanol: acetic acid: chloroform, 6:3:1, v/v/v) for 24h and preserved in 70% ethanol. The anthers were crushed in 2% acetocarmine and observed under light microscope for chromosome analysis. The chromosome number was determined to check whether any of the selected phenotypic variants had undergone chromosome doubling after colchicine treatment.

Results



Figure 1. Colchicine induced selected plants variants in C_1 population, a (Control); b-dwarf (DW); c Branched (BR); d-tall (TL); e-thick stem (TS); f- broad leaved (BL).

Screening of the colchicine treated C_1 population revealed considerable variation in plant architecture and growth habit. The selected plants were grouped into five major phenotypic categories based on conspicuous morphological characteristics: dwarf, highly branched, tall and robust plants with thick stems and broader leaves. The selected variants field data are presented in Table 1. The phenotypic variants screened in the

C_1 generation are presented in the Figure 1.

Dwarf variants

The selected C_1 plants exhibited a clear dwarf and compact growth phenotype. The most severe dwarf phenotype (17 cm) was observed in accession DW1, whereas the untreated control was 107 cm. The plant had a dense and bushy growth habit, smaller leaves and seeds and attained seed maturity earlier (46 days) than the control (52 days). So DW1 was a specific dwarf variant in the colchicine treated C_1 population.

Highly branched variants

Several C_1 plants exhibited increased branching, as well as a more compact and robust plant architecture as compared to the control. The primary branches ranged from 6 to 7, compared to 2.9 branches per plant in the untreated control. BR1 had the highest number of primary branches (7) while BR2 and BR3 had 6 branches per plant each. These plants showed vigorous vegetative growth and were maintained as selected highly branched phenotypic variants for further evaluation.

Tall variants

The selected C_1 plants showed a great variation in plant height; some accessions were more taller than the untreated control (107 cm). The tallest accession was TL1 (153 cm), followed by TL2 (130 cm), TL3 (126 cm) and TL4 (121 cm). These accessions were characterized as tall variants as they exhibited an increase in plant height as compared to control.

Thick stemmed variants

Distinct thick stemmed phenotypes were observed among the selected C_1 plants. Stem diameter ranged from 3.70 to 4.10 cm, compared with 1.7 cm in the untreated control. The greatest stem diameter was recorded in TS1 (4.10 cm), followed by TS2 (3.90 cm) and TS3 (3.70 cm). These accessions exhibited visibly robust stems and stronger plant architecture.

Broad leaved variants

Marked variation in leaf size was observed among the selected C_1 plants. Leaf area ranged from 23.93 to 25.17 cm^2 , compared with 19.80 cm^2 in the untreated control. The greatest stem diameter was recorded in TS1 (4.10 cm), followed by TS2 (3.90 cm) and TS3 (3.70 cm). These accessions exhibited visibly robust stems and stronger plant architecture.

Pollen morphology

Microscopic examination of pollen from the selected C_1 accessions showed no obvious difference in pollen size or morphology when compared to the untreated control. The pollen grains in all the accessions studied were normal and similar in size and shape. Thus no obvious changes in pollen morphology were observed in the phenotypic variants selected.

Cytological analysis

Cytological examination of the selected C_1 accessions revealed no alteration in chromosome number following colchicine treatment. All examined plants retained the diploid chromosome complement ($2n=16$), identical to that of the untreated control. No evidence of stable chromosome doubling was detected in the selected variants. Therefore, the pronounced morphological variation observed in the C_1 generation was not accompanied by detectable chromosome doubling under the treatment conditions used.

Table 1. Morphological characteristics of selected phenotypic variants in the C₁ generation of common buckwheat following colchicine treatment.

Phenotypic variant	Accession(s)	Trait/characteristic	Observed value	Control
Dwarf	DW1	Plant height (cm)	17	107
Highly branched	BR1,	Primary branches per plant	7	2.9
	BR2,	Primary branches per plant	6	
	BR3	Primary branches per plant	6	
Tall	TL1	Plant height (cm)	153	107
	TL2	Plant height (cm)	130	
	TL3	Plant height (cm)	126	
	TL4	Plant height (cm)	121	
Thick stemmed	TS1	Stem diameter (cm)	4.10	1.7
	TS2	Stem diameter (cm)	3.90	
	TS3	Stem diameter (cm)	3.70	
Broad leaved	BL1	Leaf area (cm ²)	24,35	19.80
	BL2	Leaf area (cm ²)	25.17	
	BL3	Leaf area (cm ²)	24.71	
	BL4	Leaf area (cm ²)	23.93	

Discussion

Screening of the colchicine-treated C₁ population revealed substantial variation in plant architecture, including dwarf, tall, highly branched, thick-stemmed and broad-leaved phenotypes. The occurrence of contrasting phenotypes within a single treated population indicates a heterogeneous response to colchicine rather than a single morphological response. Similar colchicine-induced variation has been reported in common buckwheat and other crop species, with responses influenced by genotype, treatment conditions, plant material and the extent of chromosome doubling [9,11-13]. Importantly, all analyzed plants in the present study retained the diploid chromosome number (2n=16), indicating that the observed morphological variation was not associated with detectable stable chromosome doubling. The dwarf phenotype DW1 was particularly distinct, with a plant height of 17 cm and maturity at 46 days, together with a compact growth habit, smaller leaves and smaller seeds. This response contrasts with the colchicine-induced autotetraploid *Fagopyrum esculentum* reported by Srivastava and Kumar, in which confirmed tetraploids (2n = 4x = 32) showed slower growth and delayed flowering, along with thicker leaves, larger stomata, pollen grains and flowers [12]. Thus, the short stature and early maturity of DW1 differ from the growth response reported for the confirmed autotetraploids and suggest that colchicine treatment may produce growth responses independent of stable chromosome doubling. The occurrence of both dwarf and tall phenotypes in the same population further supports this differential response, with TL1, TL2, TL3 and TL4 showing greater plant height than the control. Such contrasting growth responses may be associated with differences in the sensitivity and recovery of treated meristematic tissues. Colchicine acts primarily by binding to tubulin and disrupting microtubule assembly, thereby interfering with spindle formation and chromosome segregation during mitosis [9,13]. Even when stable chromosome doubling does not occur, transient disruption of microtubule-dependent processes may influence cell division, cytokinesis, cell expansion and meristematic activity. Differences in

effects may consequently alter internode elongation, branching and overall plant architecture. Therefore, colchicine-induced morphological variation should not necessarily be interpreted as evidence of polyploidization. Variation in branching and stem development was also evident. BR1, BR2 and BR3 showed enhanced primary branching, with BR1 producing the highest number of branches, whereas TS1, TS2 and TS3 exhibited comparatively thicker stems. Increased branching may provide more potential sites for reproductive development, while thicker stems may contribute to greater structural support of the plant canopy [14]. Similarly, BL1, BL2, BL3 and BL4 showed greater leaf area than the control, with BL2 exhibiting the largest leaf area. Differences in leaf size may influence canopy development and light interception and consequently affect biomass accumulation and reproductive performance [15]. However, because these variants retained the diploid chromosome number and showed normal pollen morphology, these traits should be regarded as phenotypic responses to colchicine treatment rather than as evidence of polyploidy. The present findings therefore distinguish colchicine-induced phenotypic variation from stable chromosome doubling. In common buckwheat, Srivastava and Kumar confirmed chromosome doubling and associated morphological changes in autotetraploid plants [12], while related studies have similarly demonstrated morphological and cytological changes following colchicine-induced polyploidization [13]. In contrast, all selected C₁ plants in the present study retained 2n = 16 chromosomes. The dwarf, tall, highly branched, thick-stemmed and broad-leaved forms therefore appear to represent chromosome-doubling-independent responses to colchicine exposure. This finding highlights the importance of cytological confirmation when selecting colchicine-derived plants for polyploid breeding. From a breeding perspective, the phenotypic variation observed here may nevertheless provide useful material for selection. In particular, the combination of short stature and early maturity in DW1 could be valuable for developing compact and early-maturing buckwheat types, while highly branched and thick-stemmed variants may offer alternative plant architectures for further evaluation. Broad-leaved variants may also be useful for investigating

canopy development and productivity. However, these phenotypes should be evaluated in subsequent generations for yield, fertility, seed quality and stability before their breeding value can be established. Thus, although stable polyploidization was not detected, colchicine treatment generated phenotypically distinct diploid variants that may provide useful germplasm for future buckwheat improvement.

Conclusion

Treatment with colchicine caused considerable phenotypic variation in the C₁ generation of common buckwheat. Distinct variants included dwarf, tall, highly branched, thick stemmed, and broad leaved forms. Selected plants showed pronounced differences in plant architecture especially in plant height, branching, stem thickness and leaf size. Pollen morphology, however, was similar to that of the control, and cytological analysis showed that the diploid chromosome number (2n = 16) was retained in all variants examined. The morphological variation observed, therefore, was not associated with detectable stable chromosome doubling. The plants selected should therefore be considered as colchicine associated phenotypic variants rather than true polyploids. The persistence of these phenotypes and their breeding value will have to be tested in subsequent generations.

Author Contributions

All authors read and approved the final manuscript. Javaid Ahmad conceived the study, conducted the experimental work, and drafted the manuscript. Mohd Asgar Khan participated in the experimental work and contributed to data analysis. Wasim Javid contributed to data analysis and manuscript revision. Javaid Ahmad and Wasim Javid edited and revised the manuscript.

Declarations

Ethics approval

Not applicable.

Consent to participate

Not applicable.

Consent for publication

Not applicable.

Data availability

All data generated or analyzed during this study are included in this published article. The plant material used in this study was cultivated under experimental field conditions, and all experimental procedures complied with relevant institutional and national guidelines.

Conflict of interest

The authors don't have any conflict of interest.

Permissions to collect plant material

The seeds of common buckwheat used in this study were procured from the National Bureau of Plant Genetic Resources, New Delhi, India. The plant material was cultivated under experimental field conditions, and all procedures involving plant material were conducted with appropriate permission and in compliance with relevant institutional and national guidelines.

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References

- Ohnishi O. Search for the wild ancestor of buckwheat III. The wild ancestor of cultivated common buckwheat, and of tatar buckwheat. *Econ Bot.* 1998;52(2):123-133. <https://doi.org/10.1007/BF02861199>
- Chauhan RS, Gupta N, Sharma SK, Rana JC, Sharma TR, Jana S. Genetic and genome resources in buckwheat-present status and future perspectives. *Buckwheat.* 2010;2:33-44.
- Bonafaccia G, Marocchini M, Kreft I. Composition and technological properties of the flour and bran from common and tartary buckwheat. *Food Chem.* 2003;80(1):9-15. [https://doi.org/10.1016/S0308-8146\(02\)00228-5](https://doi.org/10.1016/S0308-8146(02)00228-5)
- Gimenez-Bastida JA, Zielinski H. Buckwheat as a functional food and its effects on health. *J Agric Food Chem.* 2015;63(36):7896-7913. <https://doi.org/10.1021/acs.jafc.5b02498>
- Gimenez-Bastida JA, Zielinski H. Buckwheat as a functional food and its effects on health. *J Agric Food Chem.* 2015;63(36):7896-7913. <https://doi.org/10.1021/acs.jafc.5b02498>
- Babu S, Yadav GS, Singh R, Avasthe RK, Das A, Mohapatra KP, et al. Production technology and multifarious uses of buckwheat (*Fagopyrum* spp.): A review. *Indian J Agron.* 2024;63(4):415-427. <https://doi.org/10.59797/ija.v63i4.5672>
- Halbrecq B, Romedenne P, Ledent JF. Evolution of flowering, ripening and seed set in buckwheat (*Fagopyrum esculentum* Moench): quantitative analysis. *Eur J Agron.* 2005;23(3):209-224. <https://doi.org/10.1016/j.eja.2004.11.006>
- Honda Y, Mukasa Y, Suzuki T, Maruyama-Funatsuki W, Funatsuki H, Sekimura K, et al. The breeding and characteristics of a common buckwheat cultivar, "Kitanomashu".
- Dhooghe E, Van Laere K, Eeckhaut T, Leus L, Van Huylenbroeck J. Mitotic chromosome doubling of plant tissues in vitro. *Plant Cell Tissue Organ Cult.* 2011;104(3):359-373. <https://doi.org/10.1007/s11240-010-9786-5>
- Sattler MC, Carvalho CR, Clarindo WR. The polyploidy and its key role in plant breeding. *Planta.* 2016;243(2):281-296. <https://doi.org/10.1007/s00425-015-2450-x>
- Srivastava A, Kumar G. Induced polyploidization in buckwheat (*Fagopyrum esculentum* Moench). *Indian J Genet Plant Breed.* 2022;82(3):333-341. <http://dx.doi.org/10.31742/ISGPB.82.3.8>
- Manzoor A, Ahmad T, Bashir MA, Hafiz IA, Silvestri C. Studies on colchicine induced chromosome doubling for enhancement of quality traits in ornamental plants. *Plants.* 2019;8(7):194. <https://doi.org/10.3390/plants8070194>
- Eng WH, Ho WS. Polyploidization using colchicine in horticultural plants: A review. *Scientia horticultrae.* 2019;246:604-617. <https://doi.org/10.1016/j.scienta.2018.11.010>
- Acquaah G, Acquaah G. Principles of plant genetics and breeding. Oxford: Wiley-Blackwell; 2012.
- Evans J, Poorter HJ. Photosynthetic acclimation of plants to growth irradiance: the relative importance of specific leaf area and nitrogen partitioning in maximizing carbon gain. *Plant Cell Environ.* 2001;24(8):755-767. <https://doi.org/10.1046/j.1365-3040.2001.00724.x>