



Irradiation of hexaploid wheat (*Triticum aestivum* L.) seeds with varying doses of gamma radiation leads to novel mutants

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Abstract

The plateauing wheat yield and its narrow genetic diversity warrants modifying wheat to meet the global food demand from the growing population. Gamma radiation-based mutagenesis is a widely used technique to develop or improve crops like wheat. However, optimizing radiation dose level and the response of wheat genetic background to radiation dose are the two key challenges. This study was conducted to assess the effect of different gamma radiations doses (0 Gy, 100 Gy, 150 Gy, 200 Gy and 250 Gy) on early growth, development and yield related attributes of six wheat genotypes (AZRC Dera, RK2022, landrace-49, landrace-52, black wheat and Kashmiri landrace), and to screen out mutants with promising growth, developmental and yield related attributes. Eighteen different traits (early growth, phenology and yield of the wild type and M₁ mutants) of the six wheat genotypes revealed differences ($p < 0.01$) among genotypes (wheat) and treatment (gamma radiation doses), underscoring the genetic diversity and differential radio-sensitivity of the wheat genotypes. Additionally, all the eighteen traits among all genotypes were negatively ($p < 0.01$) affected at high dose of 250 Gy, while optimum dose of 150 Gy positively impacted early growth and yield related traits. Kashmiri landrace exhibited improved superior yield related attributes such as early maturity, longer spike, greater spikelets per spike, greater number of seeds per spike and superior seed weight per spike, making it an ideal genetic material for mutation breeding programs. Hierarchical clustering revealed five distinct clusters of genotypes, Kashmir landrace and black wheat being the most distinct genotypes. Gamma radiation mutagenesis produced a vast array of radio morphs such as double spikes per tiller, high tillering, dwarfing and altered spike architecture. These mutants provide novel and valuable genetic resources for wheat breeding.

Keywords Mutagenesis · Gamma radiation · M1 mutants · Wheat landrace · Food production

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Introduction

Wheat is the third largest cereal crop after maize and rice in terms of global production, hence playing an important role in global food security (Ullah et al. 2018; Yue et al. 2019). Post Green Revolution, crop productivity has increased steadily due to advances in the field of genetics, hence enhancing yield potential, disease resistance and tolerance to various environmental anomalies (Shearman et al. 2005; Reynolds et al. 2012) as well as various agronomic attributes (Evenson and Gollin 2003). Genetic improvement is an ongoing process owing to the challenges of continuous variation in many factors surrounding crop production (Reynolds et al. 2006; Mirzoeva et al. 2024). Therefore, increased wheat production is needed to meet the demand of the growing population (Khan et al. 2013; Fischer and Rebetzke 2018).

By 2050, the world population is expected to reach nine billion which will ultimately make it challenging for current production to meet demand (Arif et al. 2022) and hence it becomes imperative to increase yield by 60% while maintaining nutritional quality. This could only be achieved by the application of modern genetic techniques like breeding with molecular markers (Hassan et al. 2020), genetic engineering and genome editing as well as mutagenesis techniques to accelerate wheat varietal development processes (Langridge 2013; Rangan et al. 2016). In this regard, non-targeted mutagenesis and targeted mutagenesis such as genome editing have emerged as promising tools to bring revolutionary changes to increase crop yield potential. Techniques such as single base substitution, targeted mutagenesis to knock out genes and gene/allele replacement *in vivo* are beneficial and promising (Li and Xia 2020; Zhan et al. 2021; Zulfikar et al. 2021).

Mutagenesis has long been known to modify traits for basic and applied research in different organisms. Induced mutations, caused by chemical agents such as ethyl methane-sulfonate (EMS) or physical agents like X-rays and gamma rays, are classified as gene, chromosomal, or genomic, which are passed to the offspring and serve as a key tool for developing improved crop varieties (Van Harten 1998; Solanki et al. 2011). Since the 1940s, mutation breeding has been widely used to introduce genetic variation, accelerating both evolutionary processes and breeding programs by artificially inducing desirable traits in plants. Mutation is a fundamental source of genetic diversity, enabling evolution and crop improvement. While single mutations can have significant effects, ranging from harmful to beneficial or neutral, natural mutation rates in crops are often too low to meet breeding needs (Raina et al. 2016; Yali and Mitiku 2022; Kharkwal 2023). To overcome this, breeders use induced mutations to accelerate genetic variation,

facilitating the development of crops with improved yield, resilience, and nutritional value. Induced mutagenesis techniques have emerged as a highly effective, fast, rapid and widely adopted method for the improvement of crops globally. Wheat variety development is a labor intensive, technically demanding and lengthy process. On the contrary, studies have demonstrated that mutation breeding decreases the time interval for varietal development and facilitates rapid varietal development compared with hybridization (Pandit et al. 2021; Kharkwal 2023).

Climate change is disrupting global food systems, leading to biodiversity loss and declining agricultural productivity. Extreme weather, shifting growing seasons, and emerging pests and diseases further threaten food security. To combat these challenges, continuous innovation in crop breeding is essential. Mutation breeding, alongside other biotechnological approaches, plays a crucial role in creating climate-resilient, high-yielding crop varieties, ensuring sustainable food production for a growing population (Wilde 2015; Hayat et al. 2025). Mutation drives genetic diversity essential for evolution and breeding. Single nucleotide mutations can have significant effects—ranging from harmful to beneficial or neutral. Mutation breeding has greatly contributed to crop improvement by creating genetic diversity, especially crucial as climate change threatens global food security and biodiversity. With declining crop yields, developing resilient varieties is vital. Since natural mutations are rare, induced mutagenesis helps generate desired traits, producing mutants with enhanced adaptability to harsh conditions (Wilde 2015; Zhan et al. 2021; Abdel-Ghaniy et al. 2022).

Mutation breeding techniques have led to the release of over 3000 crop varieties. Radiation-induced mutagenesis is one of the foremost cutting-edge techniques which has been very effective in developing mutant varieties. Among these 3000 crop varieties developed by radiation and chemical induced mutagenesis, 65% have been developed by gamma radiation and 22% by X-ray radiation (Çelik and Atak 2017; Dhole et al. 2024). Gamma radiation induced mutagenesis has broadened genetic diversity, abiotic stress tolerance and enhancing yield related components in several crops including bread wheat (Slade et al. 2005; Sing and Datta 2010; Cheng et al. 2015; Al-Naggar et al. 2015; Ahmed et al. 2017; Askari Kelestani et al. 2021), rice (Suzuki et al. 2008) and durum wheat (Albokari 2014). Previous studies have shown that response to radiation-based induced mutagenesis is dependent on the crop genetic background. Moreover, targeted traits improvement by radiation mutagenesis in crops like wheat require certain levels of optimum radiation dose (El-Degwy and Hathout 2014; Han et al. 2016; Shabani et al. 2022). Thus, it is important to optimize radiation doses to achieve maximum improvement in crops. Hence, this research aimed to evaluate the effect of

different gamma radiation doses on growth, development and yield related attributes of different wheat germplasm, and to select mutants with promising growth, developmental and yield related attributes.

Materials and methods

Materials

Six different wheat germplasm were used in this study. Two wheat varieties viz. AZRC Dera and RK2022 were kindly provided by Arid Zone Agriculture Research Institute, Dera Ismail Khan, Pakistan and Agriculture Research Institute (ARI) Ratta Kulachi, Dera Ismail Khan Pakistan, respectively. Three wheat landraces namely Kashmir landrace, landrace 49 (acc.#11234) and landrace 52 (acc.#11239) were acquired from National Agriculture Research Council (NARC), Islamabad, Pakistan. Black wheat was procured from Shanxi Academy of Agricultural Sciences, China (Table 1). Sufficient amount of true to type, healthy and pure seed of all the six accessions were sent to Nuclear Institute for Agriculture and Biology (NIAB), Jhang Road, Faisalabad for the purpose of irradiation at four different radiation doses i.e. 100 Gy, 150 Gy, 200 Gy and 250 Gy. NIAB is equipped with a Gamma ray source Cesium-137 (Cs- 137) whose power was 660 Gy/h at the time of purchase (in 2008). The source is kept in a facility equipped with ‘Gammacell 3000’ which is a purpose-built structure for research applications (Hayat et al. 2025). Upon irradiation, seeds were immediately brought to the Department of Plant Breeding & Genetics, Faculty of Agriculture, Gomal University Dera Ismail Khan (31°49' N, 70°55' E) where all the experiments were performed.

Methods

We conducted two separate experiments, one for the evaluation of early growth parameters such as germination rate (GR%), root length (RL), root density (RD) and coleoptile

length (CL) and the other one for the evaluation of agronomic parameters. The former experiment was performed in Petri-plates whereas the latter was performed in the field. In Petri plate experiment, completely randomized design (CRD) was followed with three replicates where approximately 25 seeds of each of the non-treated (referred to as A1, B1, C1, D1 and E1) and treated (A2-A6, B2-B6, C2-C6, D2-D6, E2-E6 and F2-F6) genotypes were washed with 10% NaOCl for 5 min followed by three washings with distilled water. Post washing, 25 seeds were placed on double layer of Whatman no. 1 filter paper moistened with distilled water in Petri plates. The growing conditions were kept at 28 ± 2 °C and a 10-h photoperiod in a growing chamber (Sanyo Galenkamp, United Kingdom). Ger% was recorded at different time intervals of 3 days post germination (DPG), 7 DPG, 9 DPG, 12 DPG and 14 DPG. The other early growth-related parameters were scored by sampling ten plants from each plate on the 14th day. RL (root length?) was measured by using a ruler to measure the length of primary root from the base of the seed to the tip of root. RD (root distribution, the measure of distribution of roots within given volume) was measured by dividing total root length by area of the petri plate. CL (coleoptile length, pointed sheath covering the first true leave) was measured from its base (the point where it emerges from the seed) to the tip using a ruler.

For field performance evaluation, the checks (wild of all six types of wheat germplasm) along with their mutants irradiated at four different doses (100 Gy, 150 Gy, 200 Gy, 250 Gy) were sown in randomized complete block design (RCBD) in three replications. Row length was kept at 5 m, row to row distance at 30 cm and plant to plant distance at 30 cm. A total of 500 seeds in triplicate of each of control and irradiated seeds at all doses were sown as single seed per hole at a depth of 1.5 cm using dibbler. Prior to sowing, land was prepared deep ploughing two to three times followed by rotavator activity for smooth experimentation. Necessary agricultural practices like weeding, hoeing and irrigation were applied from time to time when needed as well as fertilizer application at standard rate were carried out to meet demand of nitrogen and phosphorus. The measured traits included days to heading (DTH) was counted as number of days taken from sowing to when 50% spikes have left the flag leaf. Days to pollination (DTP) was counted as the date when the plant starts anthesis. Plant height (PH) of each genotype was recorded in centimeters with the help of meter rod by measuring the average distance from soil surface to the top of ear exclusive of the awns of five plants per genotype per replicate just before harvest. Peduncle length (PL) (cm) was estimated by measuring the stem between the flag leaf and head of the randomly selected plants. Flag leaf length (FLGLL), flag leaf sheath length (FLGLS) and flag leaf width (FLGLW) were measured in centimeters with a

Table 1 Details of germplasm used in this study

S. No	Genotype	Code	Acquired from
1	AZRC Dera	A	Arid Zone Research Institute (AZRI)
2	RK2022	B	Agriculture Research Institute (ARI)
3	Landrace 49 (Acc.#11234)	C	National Agriculture Research Council (NARC)
4	Landrace 52 (Acc.#11239)	D	National Agriculture Research Council (NARC)
5	Black wheat	E	Shanxi Academy of Agricultural Sciences, China
6	Kashmiri landrace	F	National Agriculture Research Council (NARC)

ruler using randomly selected plants of each the wild type and mutant genotypes. Number of tillers per plant (TLPP) were documented from randomly selected five wheat plants from individual block and their average was documented. Spike length (SPKL) (cm) of the randomly chosen five plants was calculated after measuring the spike from base to the pinnacle of spike exclusive of awns and further analyzed on their average calculated. Awn length (AWL) (cm) was measured with ruler from the base to the tip of awn from randomly selected plants and then mean was calculated. Number of spikelets per spike (SPKLPS) was recorded from the randomly chosen five plants and an average value was taken. Number of grains per spike (SDPS) was estimated hand threshing each of the selected plants' spikes manually and their number of grains were counted whose average was considered. Finally, seed weight spike⁻¹ (SDWPS) was determined by recording the weight of above-mentioned threshed grains in grams. Wheat leaf rust disease severity was scored using embedded image processing system (Xu et al. 2017) and yellow rust by machine learning approach (Jiang et al. 2023).

Statistical analysis

All statistical analyses were performed in R environment. The complete data was subjected to ANOVA (Analysis of variance) in R-package. Functions “cor” and “lm” were used to calculate Pearson correlation coefficient and linear regression among the studied traits, respectively. For

graphical visualization using “ggplot2”, “GGally,” and car functions employed. Dendrogram was constructed to explore the hierarchical clustering among genotypes using the “dendextend” function. Heritability was calculated using a custom R script. ANOVA, mean square, regression coefficient, and heritability tables were generated using the “Kable Extra” package.

Results

Analysis of variance (ANOVA) for early growth, phenology, rusts and yield related traits of six different wheat genotypes, exposed to varying doses of gamma irradiation revealed substantial variability (Table 2), reflecting the interplay of genetics, treatment (radiation dose) and the environment for the expression of the observed traits. All the measured traits in both experiments (except RD) were significantly different among genotypes at various *p*-levels (Table 2), highlighting considerable genetic variation among the evaluated wheat germplasm, GR%, RL, DTH, FLGLL and PH were significantly affected by the radiation dose demonstrating that gamma radiation has the potential to alter physiological, developmental and yield related traits in wheat genotypes. GR% and RL depicted higher sensitivity to radiation doses, making them as early indicators of radiation stress. The genotype x radiation dose interaction was also significant for several traits (GR%, DTH, PH, FLGLW, TLPP, PDL, AWL, SPKL, SPKLPS, SDPS and SDWPS) implying

Table 2 Analysis of variance (ANOVA) for early growth and yield related traits of six wheat genotypes irradiated with varying doses of gamma radiation

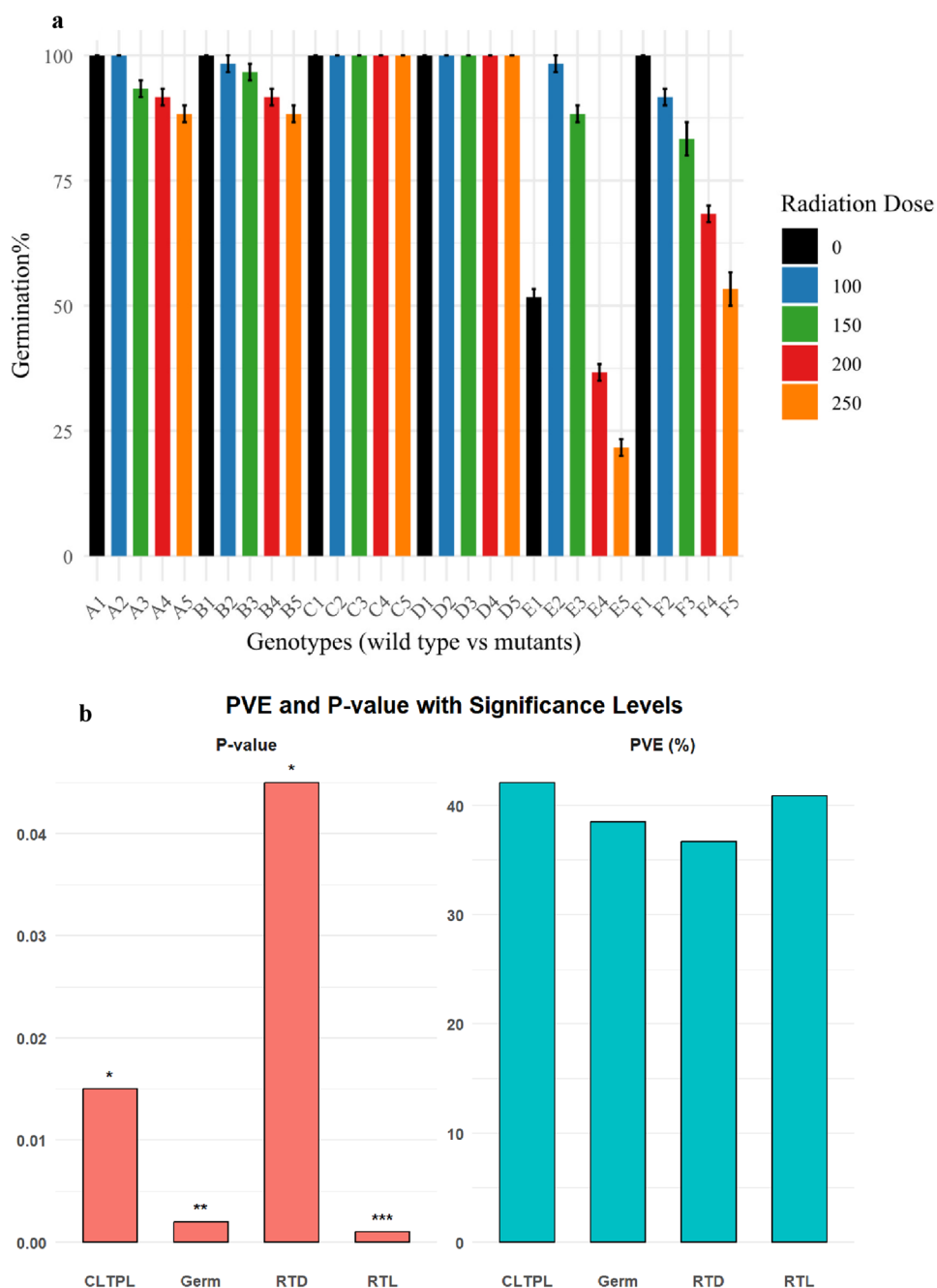
	Genotype	Replication	Dose	Genotype*Replication	Genotype*Dose	Replication*Dose	Genotype*Replication*Dose
Ger%	4793.68**	38.14*	2122.22*	12.84*	717.29***	8.05 ^{ns}	4.94 ^{ns}
CL	27.86*	0.043 ^{ns}	12.34 ^{ns}	0.02 ^{ns}	1.69 ^{ns}	0.09 ^{ns}	0.04 ^{ns}
RL	28.04***	0.43 ^{ns}	82.41***	0.06 ^{ns}	6.13 ^{ns}	0.09 ^{ns}	0.11 ^{ns}
RD	0.14 ^{ns}	0.09 ^{ns}	2.22*	0.15 ^{ns}	0.73 ^{ns}	1.94*	0.23 ^{ns}
DTH	505.34***	4.29*	6.60**	0.79 ^{ns}	28.39***	0.42 ^{ns}	1.62 ^{ns}
DTP	133.91*	4.01 ^{ns}	31.01 ^{ns}	6.93 ^{ns}	32.12 ^{ns}	3.70 ^{ns}	2.36 ^{ns}
FLGLL	133.91***	2.95 ^{ns}	95.02*	7.24 ^{ns}	37.82 ^{ns}	4.72 ^{ns}	6.37 ^{ns}
FLGLSL	133.91*	3.75 ^{ns}	17.10 ^{ns}	0.29 ^{ns}	4.97 ^{ns}	1.61 ^{ns}	1.56 ^{ns}
FLGLW	133.91***	0.007 ^{ns}	0.32 ^{ns}	0.02 ^{ns}	0.36*	0.01 ^{ns}	0.02 ^{ns}
TLPP	133.91*	1.73 ^{ns}	152.56 ^{ns}	0.33 ^{ns}	60.46***	3.07 ^{ns}	1.52 ^{ns}
PH	133.91***	21.77 ^{ns}	344.08**	47.11 ^{ns}	60.38*	26.07 ^{ns}	25.44 ^{ns}
PDL	133.91***	1.53 ^{ns}	54.45 ^{ns}	1.65 ^{ns}	30.01***	3.10 ^{ns}	5.35 ^{ns}
AWL	133.91***	0.41 ^{ns}	1.23 ^{ns}	1.11 ^{ns}	2.37**	1.36 ^{ns}	0.78 ^{ns}
SPKL	133.91***	0.84 ^{ns}	8.71 ^{ns}	0.69 ^{ns}	8.17***	1.46 ^{ns}	0.65 ^{ns}
SPKLPS	133.91***	1.89 ^{ns}	11.01 ^{ns}	1.34 ^{ns}	19.95***	0.65 ^{ns}	0.91 ^{ns}
SDPS	133.91***	5.49 ^{ns}	255.50 ^{ns}	3.41 ^{ns}	176.124***	3.10 ^{ns}	2.79 ^{ns}
SDWPS	133.91*	0.03 ^{ns}	1.74 ^{ns}	0.05*	1.10***	0.02 ^{ns}	0.01 ^{ns}
Yr-Lr	133.91***	0 ^{ns}	0.53 ^{ns}	0 ^{ns}	0.49 ^{ns}	0 ^{ns}	0 ^{ns}

Germ%; germination percentage, CLPTL; coleoptile length, RTL; root length, RD; root density, DTH; days to heading, DTP; days to pollination, FLGLL; flag leaf length, FLGLSL; flag leaf sheath length, FLGLW; flag leaf width, TLPP; tillers plant⁻¹, PH; plant height, PDL; peduncle length, AWL; awn length, SPKL; spike length, SPKLPS; spikelet's spike⁻¹, SDPS; seeds spike⁻¹, SDWPS; seed weight spike⁻¹, Yr-Lr; yellow rust-leaf rust. ns; non-significant, *, significant at *p*<0.01, **, highly significant at *p*<0.001, ***, highly significant at *p*<0.0001

that wheat genotypes responded differently to varying doses of gamma irradiation. On the contrary, replication showed non-significant effects for most traits. Likewise, genotype $\times$ replication $\times$ dose (higher level interactions) also remained non-significant, suggesting that the observed change in phenotype is due to main effect (gamma radiation treatment). Radiation, however, had minimal effects on several traits such as CL, FLGLS and disease resistance indicating genetic stability and potential value in breeding programs. The overall findings suggest that yield potential and wheat development were affected by both genetic makeup and gamma irradiation.

Gamma radiation had varying effects on the early growth parameters (GR%, RL, RD and CL). GR% of AZRC Dera, RK2022, black wheat and Kashmiri landrace was significantly reduced at higher doses of 200 Gy and 250 Gy. However, GR% of landrace-49 and landrace-52 remained unaffected (Figs. 1a and 2). The variance explained for GR% was 38%, indicating that one third of variation was due to the genetic background and the rest was caused by gamma radiation (Fig. 1b). A similar trend of gradual decrease in RL and CL at higher radiation doses (200 Gy and 250 Gy) was evident among all genotypes (Supplementary Figs. 1–3), while RD largely remained unaffected (Supplementary

Fig. 1 Effect of gamma radiation doses on days to germination%. **(a)** Response of germination% of six wheat genotypes to radiation doses of 0 Gy, 100 Gy, 150 Gy, 200 Gy and 250 Gy. A1, B1, C1, D1, E1 and F1 (shown by black bars) represent wild type and the rest are mutants of the respective genotypes. Numbers 2, 3, 4 and 5 at the Y-axis represent mutants developed at 100, 150, 200 and 250 Gy, respectively. **A:** AZRC Dera, **B:** RK2022, **C:** Landrace-49, **D:** Landrace-52, **E:** Black wheat, **F:** Kashmiri landrace. **(b)** PEV and p-values with significance for early growth traits. Germ: germination%, CLPTL; Coleoptile length, RTD; root density, RTL: root length



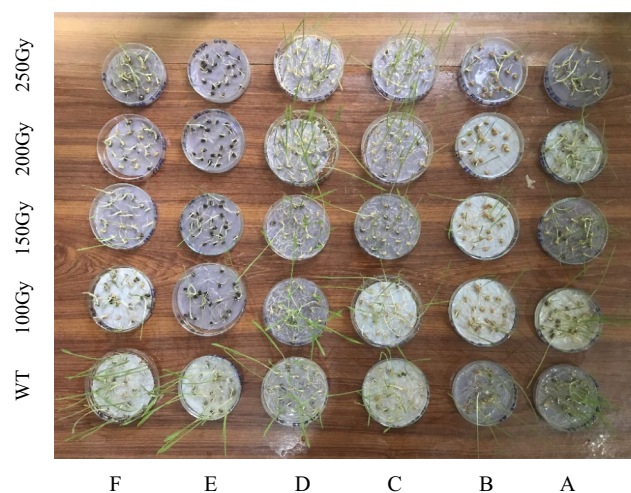


Fig. 2 Effect of varying doses of gamma radiation on germination% of different wheat genotypes. WT; wild type. A; AZRC Dera, B; RK2022, C; landrace-49, D; landrace-52, E; black wheat, F; Kashmiri landrace

Fig. 4). Agronomic and phenological traits are the adaptability traits that need to be considered initially in carving out a breeding program. Among the agronomic traits, our data trend emphasized significantly different DTH among various wheat genotypes exhibiting both stimulating and suppressing effects influenced by both radiation dose and genetic make up. A more pronounced induced early heading was exhibited by mutants of Kashmiri landrace (98 days as compared to 110 days in wild type) at higher doses of 200 Gy and 250 Gy (Fig. 3a) where the variance explained (PVE) was 45%, indicating a substantial genetic influence for this trait. Other phenological traits such as DTP, PH and PDL also showed a similar trend of decrease among all genotypes at higher radiation doses of 200 Gy and 250 G (Supplementary Figs. 5–7).

The current dataset revealed notable variations in SPKL across genotypes and radiation levels, reflecting genotype-dependent sensitivity and adaptive responses to gamma radiation exposure (Fig. 4a). Among six genotypes and their mutants, AZRC Dera showed moderate resilience to radiation, with maximum SPKL maintained at the intermediate dose of 200 Gy. RK2022 showed a stable and positive trend. This variety appears to possess enhanced tolerance and growth responsiveness, making it a promising candidate for mutation breeding aimed at improving yield traits. On the contrary, landrace-49 and landrace-52 exhibited irregular pattern of SPKL response to varying doses, suggesting radiation sensitivity and a narrow tolerance range. The Black wheat genotype displayed robust performance across all radiation doses indicating higher radiation tolerance and consistent spike development (Fig. 4a). Among all genotypes, the Kashmiri landrace exhibited the most dramatic response. With a control SPKL of 17.33 cm, it increased progressively at each radiation level- up to 18.33 cm

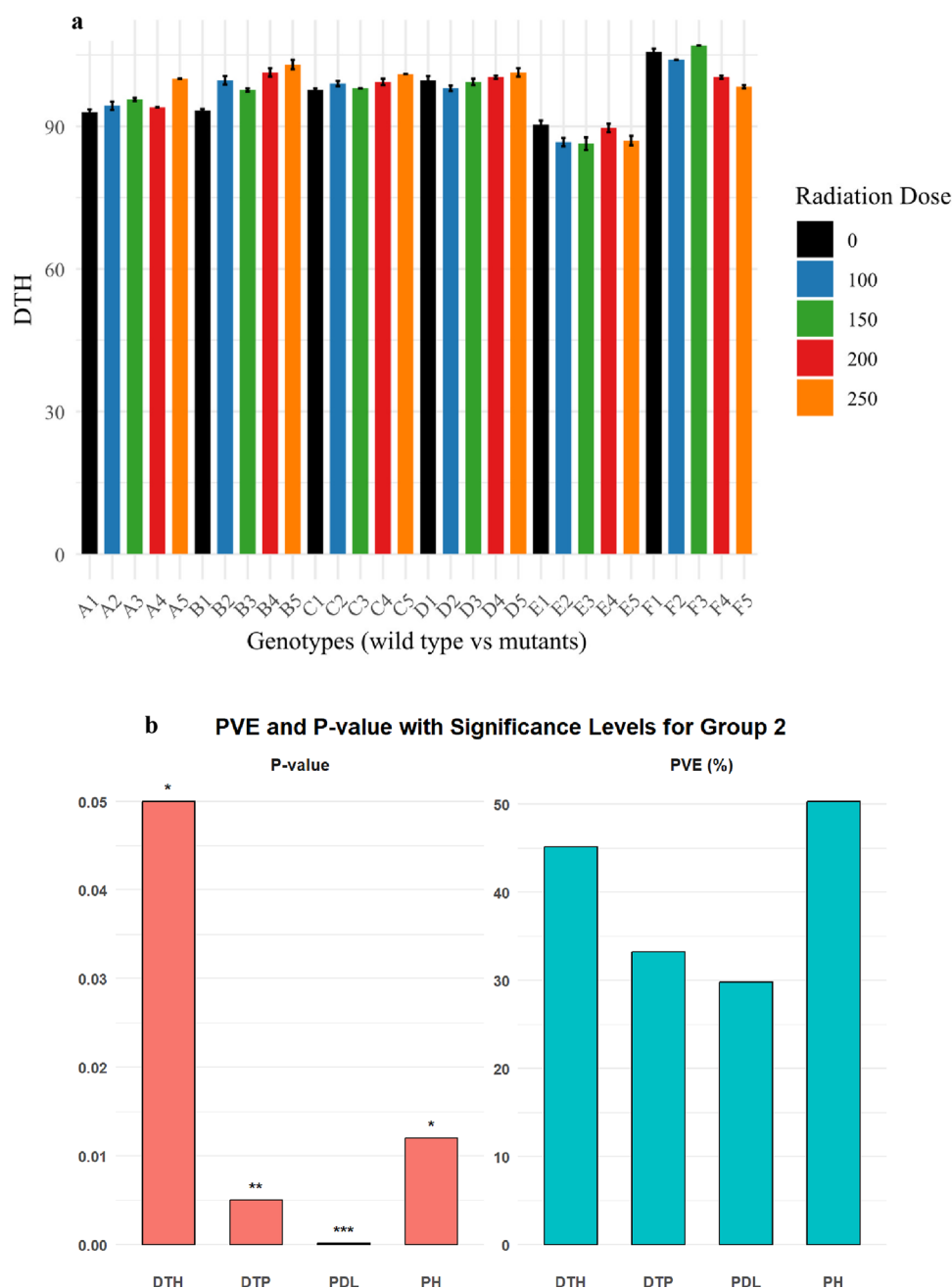
(100 Gy), 21 cm (150 Gy), 19.33 cm (200 Gy), and a remarkable 25.33 cm at 250 Gy. This substantial improvement, particularly at 150 Gy and 250 Gy, indicates a strong positive response to radiation, possibly due to enhanced cell division and elongation processes. This genotype also emerged as a top performer under mutagenic stress, making it highly valuable for future breeding strategies focused on increasing grain-bearing potential (Fig. 4a). The variance for SPKL was ~40%, explaining that gamma radiation is responsible for less than half of the observed variation in SPKL while remaining variation is due to the innate nature of the germplasm.

Correlation analyses showed strong negative correlation of GR%, RL, RD and CL with radiation dose, while all other traits showed positive correlation with radiation doses (Fig. 5). Based on p -values and R^2 regression values, the eighteen traits were grouped into three classes; those having statistically significant relationship with dose ($p < 0.05^*$), those having highly significant relationship ($p < 0.01^{**}$) and those with non-significant relationship ($p > 0.01^{ns}$ and 0.5^{ns}). Relationship between radiation dose and GR%, RL, CL and SWPS was highly significant ($p < 0.01^{**}$). While relationship between radiation dose and DTP, FLGLL, PL and PH was significant ($p < 0.05^*$). Half the traits including AWL, DTH, FLGLS, FLGLW, RD, SDPS, SPKL, SPKLPS, and TLPP showed non-significant relationship with radiation dose (Fig. 6).

The hierarchical cluster diagram (Fig. 7) illustrates the relationship among six different genotypes. The figure presents a dendrogram that grouped the genotype by similarity. The largest cluster in the dendrogram showed by the red color having genotypes AZRC Dera, RK2022 and landrace-49, indicating certain shared features among them. This clustering showed that, despite being non-identical, they are grouped together. These three genotypes also showed similar response to the gamma radiation doses (Fig. 7). High similarity noticed between the genotypes from same group (Black wheat and Kashmiri landrace) evidencing that they are genetically similar but still distinct from the other genotypes. Kashmiri landrace highlighted in blue, responded differently to the varying doses of gamma irradiation. Another compact cluster formed by the Black wheat, indicated a strong genotypic effect, largely independent of the radiation dose clearly different from the other genotypes. The gray cluster comprised of landrace-52 and RK2022, revealed some shared features in response to lower doses of gamma radiation, implying a relationship between resistant to sensitive genotypes.

Gamma radiation mutagenesis causes tissue specific mutations inherent to different types of developmental changes, especially in M_1 generation. In this study, the irradiation of six different wheat genotypes caused different

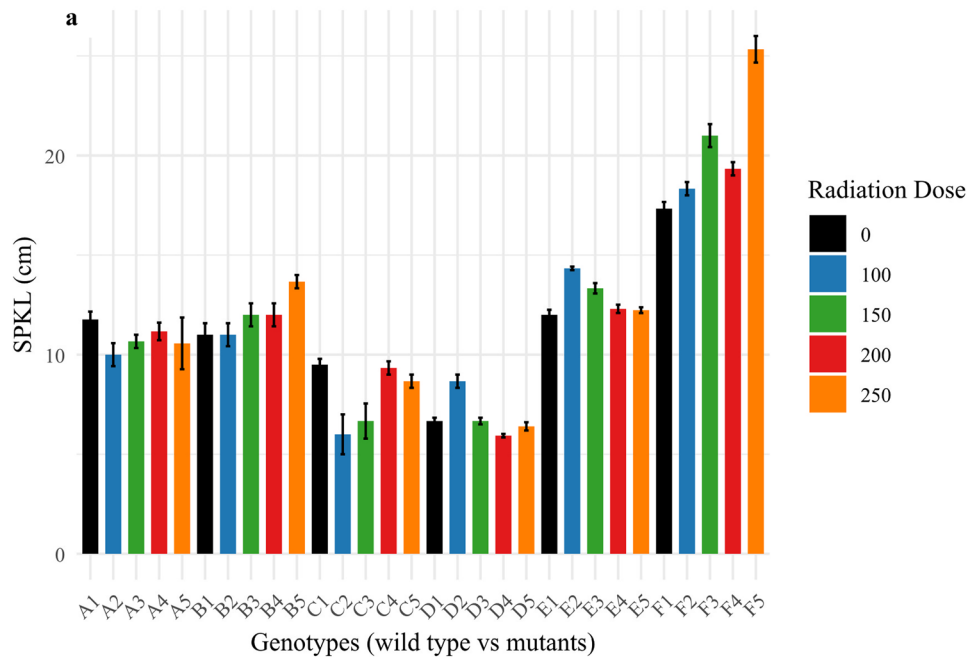
Fig. 3 Effect of gamma radiation doses on days to heading. (a) response of days to heading of six wheat genotypes to radiation doses of 0 Gy, 100 Gy, 150 Gy, 200 Gy and 250 Gy. A1, B1, C1, D1, E1 and F1 (shown by black bars) represent wild type and the rest are mutants of the respective genotypes. Numbers 2, 3, 4 and 5 at the Y-axis represent mutants developed at 100, 150, 200 and 250 Gy, respectively. **A:** AZRC Dera, **B:** RK2022, **C:** Landrace-49, **D:** Landrace-52, **E:** Black wheat, **F:** Kashmiri landrace. (b) PEV and p-values with significance for phenological traits. DTH: days to heading, DTP: days to pollination, PDL: peduncle length, PH: plant height



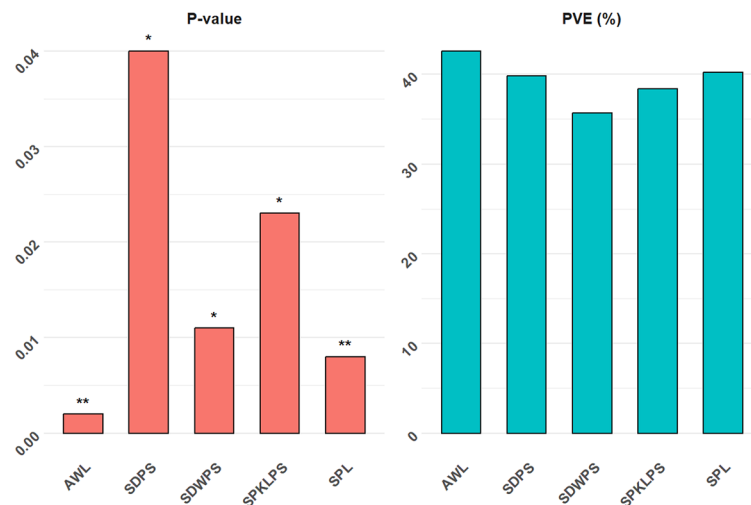
types of novel mutants in M_1 generation. It has long been known that gamma radiation can cause genetic variation which leads to the development of double spikes in wheat, this trait is defined by the development of two spikes per tiller instead of the normal single spike (Gill and Sethi 1969; Sethi and Gill 1969). The rare occurrence of double spike signifies the opportunity of increasing productivity through the use of mutagenesis. The outcome of this study resulted in eight double spike M_1 mutants in different wheat genotypes at the varying doses of gamma irradiation out of which five double spikes were found in AZRC Dera; one at 100 Gy, one at 150 Gy, two at 200 Gy, and one at 250 Gy. Another

double spike was observed in landrace-49 at 150 Gy. Landrace-52 also exhibited two double spikes, one at 150 Gy and other one at 250 Gy (Fig. 8). Besides, different radiation doses produced spikes with altered length, compactness and awns appearance or loss from wild type (Fig. 9). Besides, M_1 plants of AZRC Dera exhibited considerable variations in spike characteristics pertaining to spike length (long and short spike), supernumerary spikelets, awn variations (short awns) and loss of glume waxiness (Supplementary Fig. 11). Similarly, a higher dose (250 Gy) of gamma radiation displayed severe growth reduction and this effect was particularly evident from stunted height and fewer TLPP of the

Fig. 4 Effect of gamma radiation doses on spike length. **(a)** Response of spike length of six wheat genotypes to radiation doses of 0 Gy, 100 Gy, 150 Gy, 200 Gy and 250 Gy. A1, B1, C1, D1, E1 and F1 (shown by black bars) represent wild type and the rest are mutants of the respective genotypes. Numbers 2, 3, 4 and 5 at the Y-axis represent mutants developed at 100, 150, 200 and 250 Gy, respectively. **A:** AZRC Dera, **B:** RK2022, **C:** Landrace-49, **D:** Landrace-52, **E:** Black wheat, **F:** Kashmiri landrace. **(b)** PEV and p-values with significance for spike related traits. SPL: spike length, AWL: awn length, SPKLPS: spike-lets spike⁻¹, SDPS: seeds spike⁻¹, SDWPS: seed weigh spike⁻¹



b PVE and P-value with Significance Levels for Group 4 (Additional Traits)



mutant (Mu1C4-1) of landrace-49 exposed to 250 Gy (Supplementary Fig. 12a). On the contrary, an optimal dose of 150 Gy tremendously enhanced tiller plant⁻¹ as evident by Mu1D2-1 of landrace-52, showing enhanced tillering capacity relative to their wild types (Supplementary Fig. 12b). This result highlights the adaptive capacity of landrace-52 to increase tiller plant⁻¹ at optimum radiation dose.

Discussion

Early growth-related traits such as GR%, RL, RD and CL play a critical role in rapid emergence and establishment, rapid early growth, root development, nutrient uptake

forming the basis of overall plant yield (Ludwig and Asseng 2010). Radiation-based mutagenesis is a potential tool to modulate root related attributes and other early growth-related traits. Significant differences among all wheat genotypes and gamma radiation doses for the above-mentioned traits underscore the genetic diversity and differential radio-sensitivity of the wheat genotypes for germination and root length. The high sensitivity of GR% and RL to radiation doses implied that these two parameters can be used as early indicators of radiation stress. Previous studies found mean germination time, root and shoot length decreased with increasing dose of radiation (Iqbal et al. 2007; Borzouei et al. 2010).

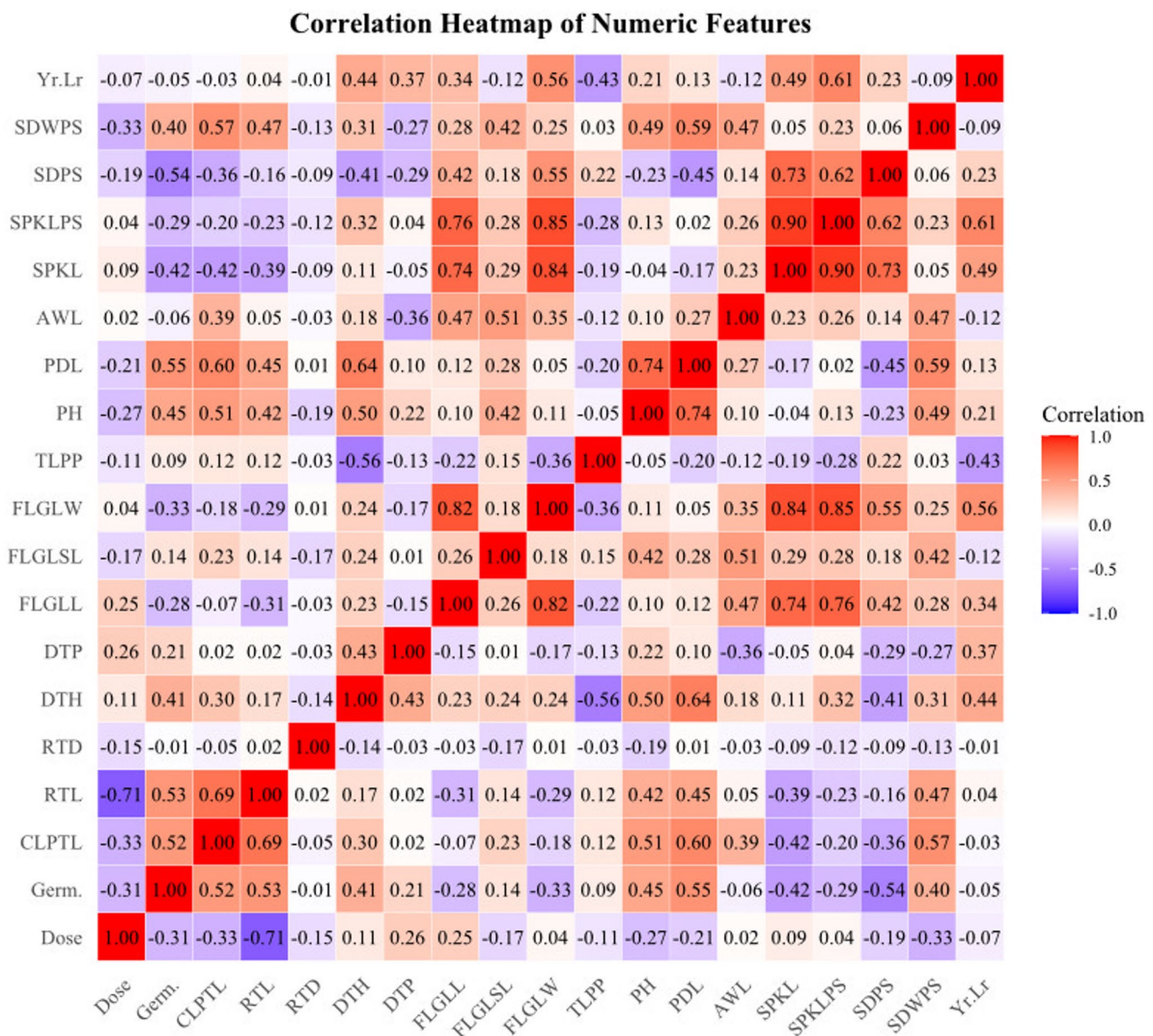


Fig. 5 Correlation between gamma radiation dose and different traits related to early growth, phenology and yield

Among agronomic traits, DTH is a critical developmental stage when the spike or ear emerges from its enclosing sheath (Akram et al. 2020). DTH (heading) and DTP (flowering) are determined by genes affecting the vernalization response, the photoperiod response and earliness per se (Eps) (Neumann et al. 2011). DTH and DTP are critical for plant early maturity and high yield under changing climate. Wheat genotypes with inherent capability of early maturity (less days to heading, pollination and maturity) tend to withstand heat stress during pollination, thereby producing more viable grains spike⁻¹ and attain high yield (Iqbal et al. 2007; Reynolds and Slafer 2012). In our study, DTH showed highly significant genotype and genotype x radiation effects, while DTP (flowering time) remained

non-significant at dose and genotype × dose levels. Previous reports suggested that gamma radiation may delay or advance flowering depending on genetic background of wheat cultivar and radiation dose (Khan et al. 2010; Shabani et al. 2022). This variation in DTH could also be due to the genetic changes imparted by gamma radiation in the Eps genes. To add to it, physical mutagenic agents (gamma rays) may bring out genetic variation in crops with novel but important agronomic properties that were otherwise absent in nature (Perez-Jimenez et al. 2020). One of the six genotypes used in our study, Kashmiri landrace possessed superior traits like long spikes, more SPKLPS and SDPS and SDWPS. However, these traits are compromised in hot climatic conditions of Dera Ismail Khan owing to

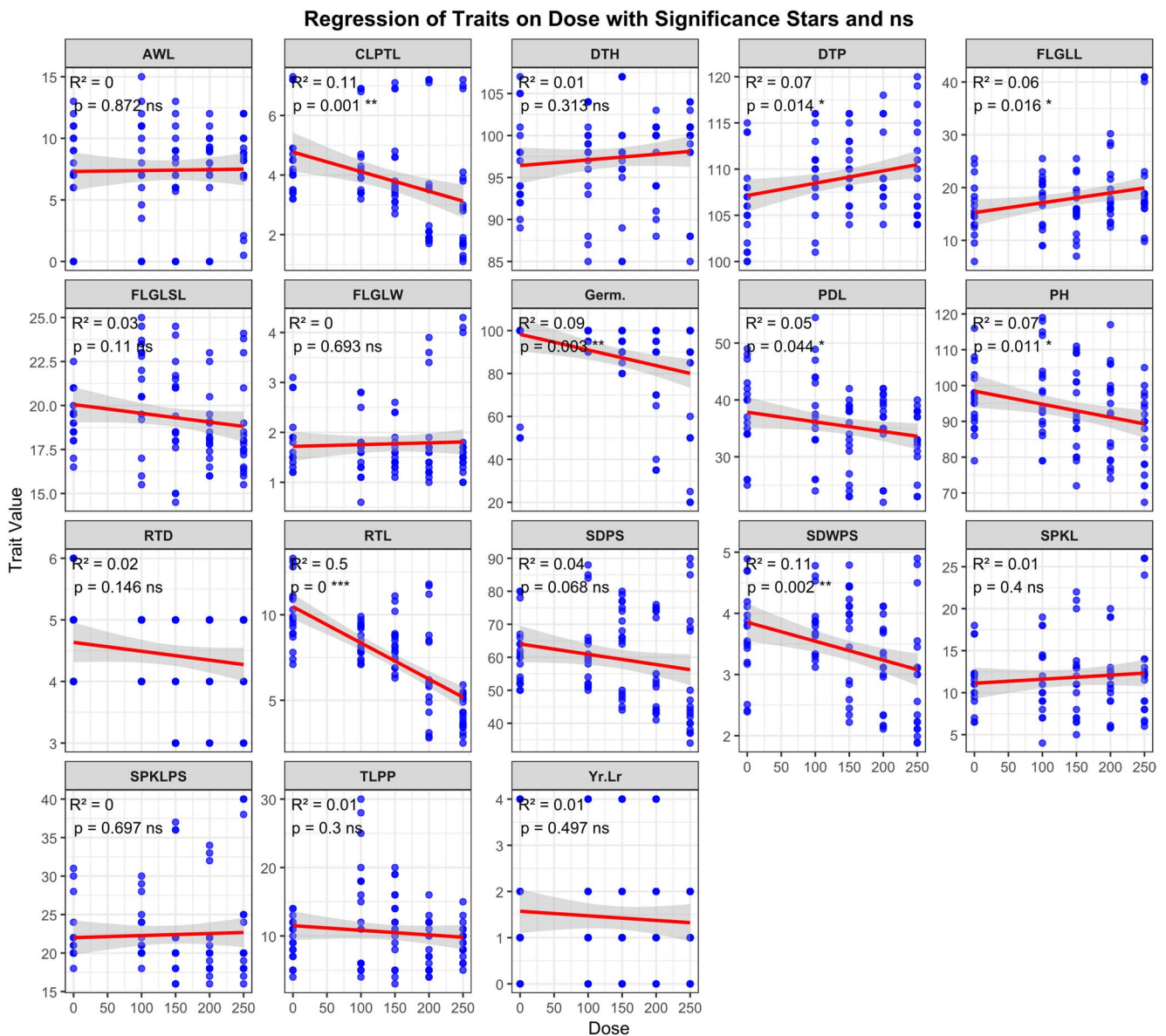


Fig. 6 Regression analysis of different traits of six wheat genotypes irradiated with varying doses of gamma radiation

delayed heading, pollination and maturity of this landrace. In this investigation, Kashmiri landrace displayed a marked decline in days to heading from 116 to 98 with the elevated gamma rays doses. Thus, mutation breeding improved and established Kashmiri landrace as an exemplary genotype by inducing early maturation suitable to survive under terminal heat stress-prone condition of Dera Ismail Khan.

Yield is a complex quantitative trait in wheat (Arif et al. 2021). Yield and its contributing traits are controlled by multiple genes interacting with environmental factors (Arif et al. 2020). Among yield contributing traits, SPKL is the most crucial as it directly influences the number of spikelets and potential grain yield. Additionally, various yield related components such as PH, tillering, stem and spike related

attributes interact with each other, and with environment to determine over all wheat yield and productivity in diverse agro-climatic conditions (Fischer and Rebetzke 2018; Ray et al. 2019). Gamma radiation is an effective mutagenic agent, which when applied at optimal dose, has been effective to improve various yield related attributes and overall biological and grain yield of wheat, rice, barley and legumes. Previous studies have utilized gamma radiation and improved various yield related components, for instance dwarf height and lodging resistance (Manjaya 2023), TLPP (Dhole et al. 2024), SPKL and SPKLPS (Cassata 2024), SDPS (Cheng et al. 2015) and thousand grain weight (Cheng et al. 2015; Rana et al. 2025). M_1 mutants of our genotypes significantly varied for PH, PL, TLPP, SPKL, AWL, SPKLPS,

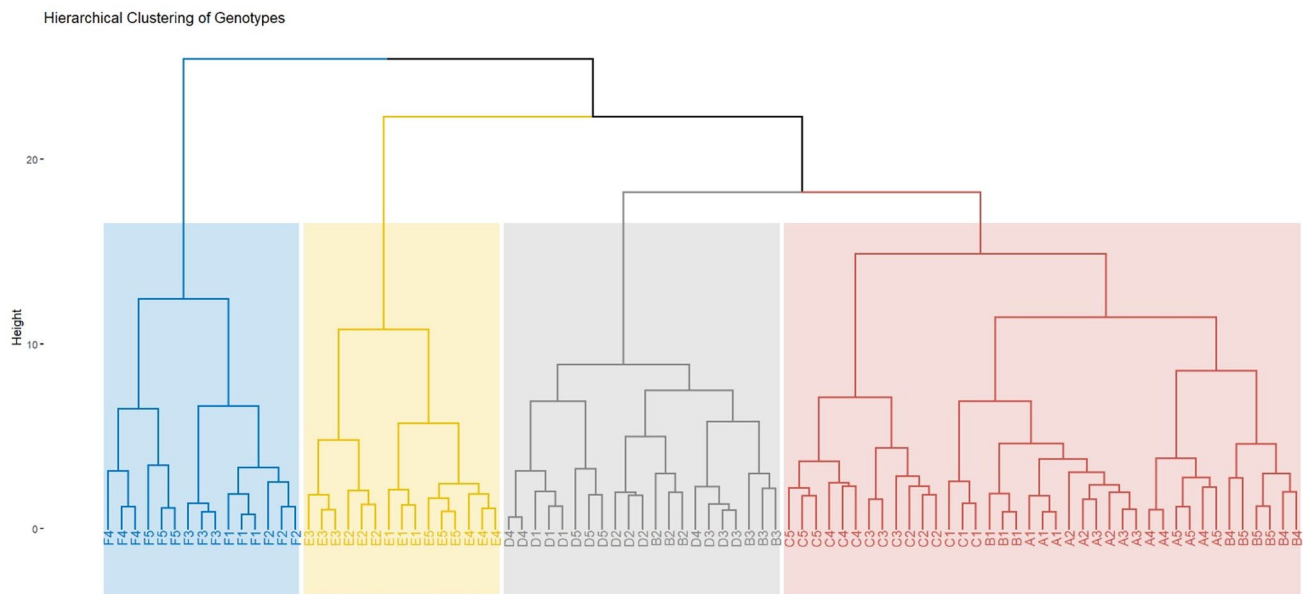


Fig. 7 Hierarchical Cluster analysis of six wheat genotypes irradiated with varying doses of gamma radiation. **A:** AZRC Dera, **B:** RK2022, **C:** Landrace-49, **D:** Landrace-52, **E:** Black wheat, **F:** Kashmiri landrace

Fig. 8 Novel double/branched spike mutants obtained at different gamma dose. WT; wild type, BSM: branched spike mutant, **A:** AZRC Dera, **B:** landrace 49, **C:** landrace 52



SDPS and SDWPS. Gamma rays are known to ionize water molecules in cells and other alterations that interfere with the genetic code resulting in the expression of a wide range of phenotypic changes, some of which could be utilized in plant breeding (Riviello- Flores et al. 2022) for specific purposes. Notable variations in SPKL across genotypes and radiation levels reflect genotype-dependent sensitivity and adaptive responses to gamma radiation exposure. All genotypes, except RK2022 and Kashmiri landrace, showed either stability or reduction in SPKL with increasing radiation doses. Previously, radiation-based improvement in SPKL and SPKLPS was genotype dependent (Ahmed et al. 2017) which corroborates our findings as we also found

a clear positive trend for SPKLPS in Kashmiri landrace mutant (39.3 SPKLPS at 250 Gy) relative to wild type (29.6 SPKLPS at 0 Gy). This robust enhancement implies extensive stimulation of reproductive structures, likely due to beneficial mutagenic effects that enhanced cell division in developing spikes. Among all six genotypes, Kashmiri landrace exhibited most pronounced improvement in SDPS (87.6 in M_1 at 250 Gy relative to 65.6 in wild type), SDWPS (4.7 g in M_1 at 250 Gy relative to 3.5 g in wild type), and SPKL (25.33 cm at 250 Gy as compared to 17.33 cm in wild type). This genotype emerged as a top performer under mutagenic stress, making it highly valuable for future breeding strategies focused on increasing grain-bearing potential.

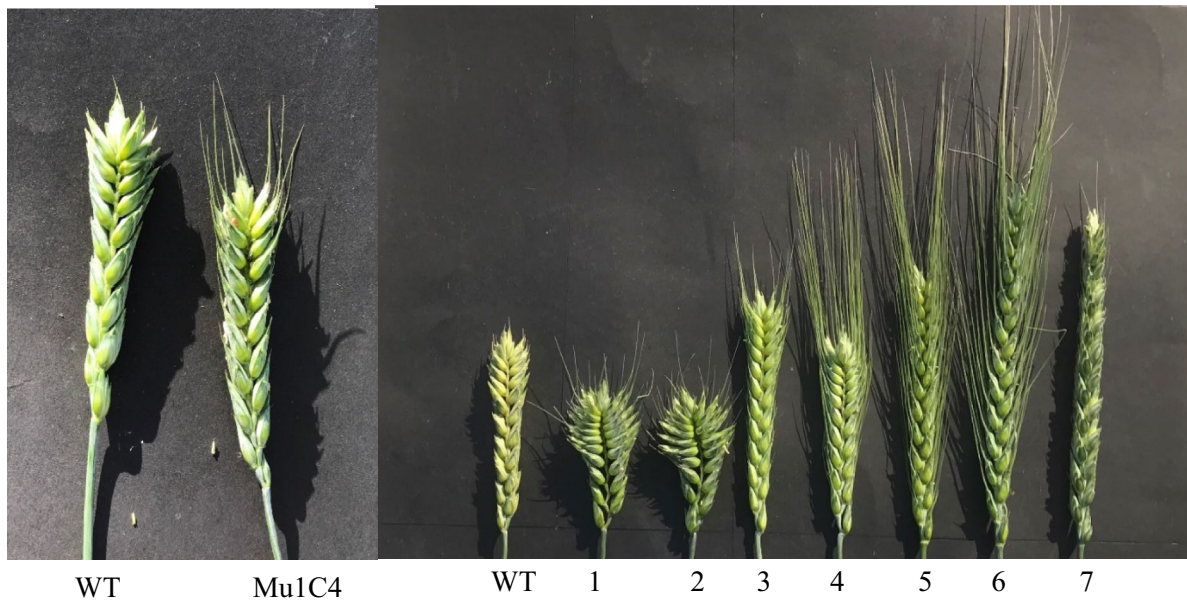


Fig. 9 Variation in spike architecture of M1 and M2 plants of the landrace 49. WT; Wild type, Mu1C4; Short hairy-spike M1 mutant of landrace 49 irradiated at 200 Gy, 1–7; spikes of M2 mutants plant originated from the seeds of Mu1C4 short hairy-spike mutant

The regression and correlation analysis demonstrated a general negative effect of gamma radiation on all yield-related traits in wheat, especially at higher doses. While traits like TLPP, SPKL and AWL show mild to moderate declines, attributes such as SDPS and SDWPS exhibited strong negative correlations, indicating their higher sensitivity to radiation. This pattern suggests that gamma radiation impaired both vegetative and reproductive development, but its most profound impact is on reproductive success and grain filling, which are crucial for final yield. Therefore, while low to moderate doses may be used for mutation breeding, high radiation doses are detrimental to yield-related attributes and should be carefully optimized. Additionally, some wheat genotypes showed improvement in yield related attributes at a lower doses (100–150 Gy), other showed improvement for certain traits at higher doses (200–250 Gy). These data suggest that radiation based yield related trait improvement is dependant on genetic background of wheat genotype (El-Degwy et al. 2014; Ahmed et al. 2017; Balkan et al. 2019; Shabani et al. 2022), and careful selection of optimal dose be opted for tartgetted traits improvement in different wheat genotypes. Similar inferences have been drawn for quinoa in a recent study (Hayat et al. 2025).

Radiomorphosis is a phenomenon in which mutagens such as gamma radiation induce different morphological and genetic changes as a consequence of mutation in DNA that ultimately lead to changes in plant growth, structure and function. Gamma radiation mutagenesis causes tissue specific mutations inherent to different types of developmental changes, especially in M₁ generation (Von Well et al. 2023). Gamma radiation can induce changes in gene that

are associated with the floral initiation, spike development or spikelet formation, thereby leading to the development of double spikes (Din et al. 2003; Su et al. 2011). In our study, different types of novel radiomorphs in M₁ generation were observed. Double spike is a characteristic that contributes to the higher grain yield. The rare occurrence of double spike signifies the opportunity of increasing productivity through the use of mutagenesis. The outcome of our research resulted in eight double spike M₁ mutants in different wheat genotypes at the varying doses of gamma irradiation. However, the advancement of double spike M₁ mutants into M₂ generation failed to inherit this trait which is supported by (Gill and Sethi 1969; Sethi and Gill 1969). Besides, M₁ plants of AZRC Dera exhibited considerable variations in spike characteristics pertaining to SPKL (long and short spike), supernumerary spikelets, awn variations (short awns) and loss of glume waxiness. This study demonstrated that hexaploid wheat produce supernumerary spikelet's that may contribute to grain yield. Screening of EMS mutant library of Chinse wheat genotype KN9204 produced 114 mutant lines with spike-related radiomorphs; paired spikelets were the most common alteration (81.57%, 93 of 114 lines), followed by altered auxiliary organs such as awns and glumes (Wang et al. 2023). These data suggest that spike related radiomorphs at certain concentration of EMS and specific lower doses of gamma radiation have the ability to produce a vast array of spike related radiomorphs which are important genetic materials both for basic and applied research.

Conclusion

The current study investigated the creation and characterization of novel mutants of hexaploid wheat (*Triticum aestivum* L.) induced by varying doses of gamma radiation. Ger% of AZRC Dera, landrace-49 and landrace-52 were not affected by the application of 100 Gy and 250 GY, making them radiation insensitive towards Ger%. Lower doses and higher radiation doses increased and decreased the RL. Additionally, the gamma rays induced early heading in Kashmiri landrace. Among all tested genotypes, Kashmiri landrace conferred improved superior yield related traits like (early maturity, longer spikes, spikelets per spike, seeds per spike and seed weight per spike) even at the highest tested dose (250 Gy) making it an ideal genetic material for mutation breeding programs. Finally, our efforts provided valuable insights into the gamma rays effects on various morphological and developmental traits in wheat. This study also addressed the widening of narrow genetic diversity in the available germplasm. Generation advancement of novel M₁ mutants developed here to M₅ generation, and their physio-morphological and molecular investigation are important to determine the stability and heritability of beneficial traits induced by radiation.

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Author contributions Conceptualization was done by NUK; methodology by NUK and IA; software work by BTN; validation was done by DL; formal analysis by MY and DKYT; investigation by MAR; data curation by WE; writing-original draft by NUK and BTN; writing-review and editing by MY and DKYT; visualization by MAR; supervision and funding by WE and MY; All authors have read and agreed to the published version of the manuscript.

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Data availability All data are available in the manuscript file and supplementary file.

Declarations

Competing interests The authors declare no competing interests.

Institutional Review Board Statement Not applicable.

Informed consent Not applicable.

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References

- Abdel-Ghaniy R, Hassan A, Ghonaim M (2022) Mean performance, genetic diversity and DNA analysis in M3 generation of some wheat mutants under water stress. *J Prod Dev* 27:417–442
- Ahmed S, Khan WM, Khan MS, Akhtar N, Umar N, Ali S, Hussain S, Shah SS (2017) Impact of gamma radiations on wheat (*Triticum aestivum* L.) varieties (Batoor and Janbaz). *Pure Appl Biol* 6:218–225
- Akram S, Arif MAR, Hameed A (2020) A GBS-based GWAS analysis of adaptability and yield traits in bread wheat (*Triticum aestivum* L.). *J Appl Genet* 62:27–41
- Al-Naggar AMM, Kh F, Al-Azab SES, Sobieh AM (2015) Use of gamma rays and hybridization to create new drought tolerant wheat genotypes. *Int J Plant Soil Sci* 5:282–299
- Albokari M (2014) Induction of mutants in durum wheat (*Triticum durum* Desf. cv. *samra*) using gamma irradiation. *Pak J Bot* 46:317–324
- Arif MAR, Attaria F, Shokat S, Akram S, Waheed MQ, Arif A, Börner A (2020) Mapping of QTLs associated with yield and yield related traits in durum wheat (*Triticum durum* Desf.) under irrigated and drought conditions. *Int J Mol Sci* 21:2372
- Arif MAR, Shokat S, Plieske J, Lohwasser U, Chesnokov YV, Kumar N, Kulwal P, McGuire P, Sorrells M, Qualset CO, Börner A (2021) A SNP-based genetic dissection of versatile traits in bread wheat (*Triticum aestivum* L.). *Plant J* 108:960–976
- Arif MAR, Waheed MQ, Lohwasser U, Shokat S, Alquddah AM, Volkmar C, Börner A (2022) Genetic insight into the insect resistance in bread wheat exploiting the untapped natural diversity. *Front Genet* 13:898905
- Balkan A, Bilgin O, Başer İ, Göçmen DB, Demirkan AK, Deviren B (2019) Improvement of grain yield and yield associated traits in bread wheat (*Triticum aestivum* L.) genotypes through mutation breeding using gamma irradiation. *Tekirdag Ziraat Fak Derg* 16:103–111
- Borzouei A, Kafi M, Khazaei H, Nasiryan B, Majdabadi A (2010) Effects of gamma radiation on germination and physiological aspects of wheat (*Triticum aestivum* L.) seedlings. *Pak J Bot* 42:2281–2290
- Cassata F (2024) Atomic Spaghetti Durum Wheat and Mutation Breeding in Italy (1950s–1970s). *Agric Hist* 98:572–606

- Çelik Ö, Atak Ç (2017) Applications of ionizing radiation in mutation breeding. *NIGR* 6:111–132
- Cheng X, Chai L, Chen Z, Xu L, Zhai H, Zhao A, Ni Z (2015) Identification and characterization of a high kernel weight mutant induced by gamma radiation in wheat (*Triticum aestivum* L.). *BMC Genet* 16:1–9
- Dhole, VJ, Jegadeesan S, Punniyamoorthy D (2024). Use of Gamma Rays in Crop Improvement. In: Kumar, N. (eds) Plant Mutagenesis. Sustainable Landscape Planning and Natural Resources ManagementDi (pp. 135-157). Cham. Springer Nature Switzerland. https://doi.org/10.1007/978-3-031-50729-8_11
- Din R, Khan MM, Qasim M, Jehan S, Khan MI (2003) Induced mutability studies in three wheat (*Triticum aestivum* L.) varieties for some morphological and agronomic characteristics. *Asian J Plant Sci* 2:1179–1182
- El-Degwy IS, Hathout MS (2014) Influence of gamma rays on the performance and genetic parameters for grain yield and yield attributes of bread wheat. *Egypt J Agron* 36:41–55
- Evenson RE, Gollin D (2003) Assessing the impact of the Green Revolution, 1960 to 2000. *Science* 300:758–762
- Fischer RA, Rebetzke GJ (2018) Indirect selection for potential yield in early-generation, spaced plantings of wheat and other small-grain cereals: a review. *Crop Pasture Sci* 69:439–459
- Gill KS, Sethi GS (1969) Double spike: a radiomorph in wheat and barley. Punjab Agricultural Univ., Ludhiana, India
- Han B, Gu J, Zhao L, Guo H, Xie Y, Zhao S, Liu L (2016) Factors affecting the radiosensitivity of hexaploid wheat to γ -irradiation: radiosensitivity of hexaploid wheat (*Triticum aestivum* L.). *PLoS ONE* 11:e0161700
- Hassan R, Waheed MQ, Shokat S, Arif MAR, Tariq R, Arif M, Arif A (2020) Estimation of genomic diversity using sequence related amplified polymorphism (SRAP) markers in a mini core collection of wheat germplasm from Pakistan. *Cereal Res Commun* 48:33–40
- Hayat U, Afzal I, Arif MAR, Shahbaz M, Rasul F (2025) Determination of Gamma Rays Lethal (LD50) and Growth Reduction (GR50) Dose in Chenopodium quinoa Varieties (UAF-Q7 and UAF-QS) for Mutation Breeding. *Plant Breed* 1:16. <https://doi.org/10.1111/pbr.70000>
- Iqbal M, Navabi A, Salmon DF, Yang RC, Spaner D (2007) Simultaneous selection for early maturity, increased grain yield and elevated grain protein content in spring wheat. *Plant Breed* 126:244–250
- Jiang Q, Wang H, Wang H (2023) Severity assessment of wheat stripe rust based on machine learning. *Front Plant Sci* 14:1150855
- Kelestani A, Ramezanpour S, Borzouei A, Soltanloo H, Navabpour S, Saadati S (2021) Application of gamma rays on salinity tolerance of wheat (*Triticum aestivum* L.) and expression of genes related to biosynthesis of proline, glycine betaine and antioxidant enzymes. *Physiol Mol Biol Plants* 27:2533–2547
- Khan SA, Marak HC, Gupta A, Sarmah K (2010) Effects of dates of sowing on phenology, thermal and radiation regimes, and yield of wheat. *J Agrometeorology* 12:53–57
- Khan N, Hussain J, Hassan A, Shah MM (2013) PCR-based identification of Rye translocations in Pakistani wheat germplasm. *Minerva Biotechnol* 25:87–94
- Kharkwal MC (2023). History of Plant Mutation Breeding and Global Impact of Mutant Varieties. In: Penna, S., Jain, S.M. (eds) Mutation Breeding for Sustainable Food Production and Climate Resilience (pp. 25-55). Springer, Singapore. https://doi.org/10.1007/978-981-16-9720-3_2
- Langridge P (2013) Wheat genomics and the ambitious targets for future wheat production. *Genome* 56:545–547
- Li S, Xia L (2020) Precise gene replacement in plants through CRISPR/Cas genome editing technology: current status and future perspectives. *Abiotech* 1:58–73
- Ludwig F, Asseng S (2010) Potential benefits of early vigor and changes in phenology in wheat to adapt to warmer and drier climates. *Agric Syst* 103:127–136
- Manjaya JG (2023) Contribution and impact of mutant varieties on food security. *advanced crop improvement. Theory Pract* 1:47–73
- Neumann K, Kobiljski B, Denčić S, Varshney R, Börner A (2011) Genome-wide association mapping: a case study in bread wheat (*Triticum aestivum* L.). *Mol Breed* 27:37–58
- Pandit R, Bhusal B, Regmi R, Neupane P, Bhattarai K, Maharjan B, Poudel MR (2021) Mutation breeding for crop improvement: a review. *RFNA* 2:31–35
- Perez- Jimenez M, Tallon CI, Perez- Tornero O (2020) Inducing Mutations in Citrus spp.: Sensitivity of Different Sources of Plant Material to Gamma Radiation. *Appl Radiat Isot* 157:109030
- Raina A, Laskar RA, Khursheed S, Amin R, Tantray YR, Parveen K, Khan S (2016) Role of mutation breeding in crop improvement-past, present and future. *Asian Res J Agric* 2:1–13
- Rana A, Rana V, Bakshi S, Kumar SV (2025) Isolation and characterization of gamma rays induced mutants for improved agromorphological performance and harder grain texture in wheat (*Triticum aestivum* L.). *Int J Radiat Biol* 101:85–100
- Rangan P, Furtado A, Henry RJ (2016) New evidence for grain specific C4 photosynthesis in wheat. *Sci Rep* 6:31721
- Ray DK, West PC, Clark M, Gerber JS, Prishchepov AV, Chatterjee S (2019) Climate change has likely already affected global food production. *PLoS ONE* 14:e0217148
- Reynolds MP, Borlaug NE (2006) Impacts of breeding on international collaborative wheat improvement. *J Agric Sci* 144:3–17
- Reynolds M, Foulkes J, Furbank R, Griffiths S, King J, Murchie E, Slafer G (2012) Achieving yield gains in wheat. *Plant Cell Environ* 35:1799–1823
- Riviello-Flores MD, Cadena-Iñiguez J, Ruiz-Posadas LD, Arévalo-Galarza MD, Castillo-Juárez I, Soto Hernández M, Castillo-Martínez CR (2022) Use of Gamma Radiation for the Genetic Improvement of Underutilized Plant Varieties. *Plants* 11:1161. <https://doi.org/10.3390/plants11091161>
- Sethi GS, Gill KS (1969) Doubling-tendency morphological changes induced by different mutagens in barley. *Rad Bot* 9:415–419
- Shabani M, Alemzadeh A, Nakhoda B, Razi H, Houshmandpanah Z, Hildebrand D (2022) Optimized gamma radiation produces physiological and morphological changes that improve seed yield in wheat. *Physiol Mol Biol Plants* 28:1571–1586
- Shearman VJ, Sylvester-Bradley R, Scott RK, Foulkes MJ (2005) Physiological processes associated with wheat yield progress in the UK. *Crop Sci* 45:175–185
- Singh B, Datta PS (2010) Gamma irradiation to improve plant vigour, grain development, and yield attributes of wheat. *Radiat Phys Chem* 79:139–143
- Slade AJ, Fuerstenberg SI, Loeffler D, Steine MN, Facciotti D (2005) A reverse genetic, nontransgenic approach to wheat crop improvement by TILLING. *Nat Biotechnol* 23:75–81
- Solanki RK, Gill RK, Verma P, Singh S (2011) Mutation breeding in pulses: an overview. *Breeding of pulse crops*. Kalyani, Ludhiana, pp 85–103
- Su YH, Liu YB, Zhang XS (2011) Auxin–cytokinin interaction regulates meristem development. *Mol Plant* 4:616–625
- Suzuki T, Eiguchi M, Kumamaru T, Satoh H, Matsusaka H, Moriguchi K, Nagato Y, Kurata N (2008) MNU-induced mutant pools and high performance TILLING enable finding of any gene mutation in rice. *Mol Genet Genomics* 279:213–223
- Ullah KN, Li N, Shen T, Wang P, Tang W, Ma S, Zhang Z, Jia H, Kong Z, Ma Z (2018) Fine mapping of powdery mildew resistance gene Pm4e in bread wheat (*Triticum aestivum* L.). *Planta* 248:1319–1328
- Van Harten AM (1998) Mutation breeding: theory and practical applications, Cambridge University Press, UK

- Von Well E, Booyse M, Fossey A (2023) Gamma irradiation as tool for mutation breeding in wheat. In *Wheat Research and Utilization*. <https://doi.org/10.5772/intechopen.111713>
- Wang D, Li Y, Wang H, Xu Y, Yang Y, Zhou Y, Chen Z, Zhou Y, Gui L, Guo Y, Zhou C (2023) Boosting wheat functional genomics via an indexed EMS mutant library of KN9204. *Plant Commun* 4:100593. <https://doi.org/10.1016/j.xplc.2023.100593>
- Wilde HD (2015) Induced mutations in plant breeding. *Advances in Plant Breeding Strategies: Breeding, Biotechnology and Molecular Tools*. Springer International Publishing, Cham, pp 329–344
- Xu P, Wu G, Guo Y, Yang H, Zhang R (2017) Automatic wheat leaf rust detection and grading diagnosis via embedded image processing system. *Procedia Comput Sci* 107:836–841
- Yali W, Mitiku T (2022) Mutation breeding and its importance in modern plant breeding. *J Plant Sci* 10:64–70
- Yue Y, Zhang P, Shang Y (2019) The potential global distribution and dynamics of wheat under multiple climate change scenarios. *STOTEN* 688:1308–1318
- Zhan X, Lu Y, Zhu JK, Botella JR (2021) Genome editing for plant research and crop improvement. *J Integr Plant Biol* 63:3–33
- Zulfiqar S, Ishfaq S, Ikram M, Nawaz MA, Rahman MU (2021) Characterization of gamma-rays-induced spring wheat mutants for morphological and quality traits through multivariate and GT Biplot analysis. *Agronomy* 11:2288. <https://doi.org/10.3390/agronomy11112288>
- Mirzoieva T, Stepasyuk L, Diachkov D, Nitsenko V, Velychko O, Lozynska T, Kapelista I (2024) Prospects for the Production of Niche Grain Crops in the Context of the Need to Ensure Food Security. *Rocznik Ochrona Środowiska* 26:568–586