

Selection Of Mutant Lines Developed With Two Mutagenic Agents In Triticum Polonicum L.In The Mantaro Valley – Peru

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Abstract

Wheat is one of the most important crops in the Andean zone of Peru. It is a food and economic source for the population, especially Triticum polonicum L in the Mantaro Valley, which develops at altitudes where other wheat crops do not thrive, tolerating phytosanitary diseases and resisting frost. One of the limitations of T polonicum L lies in the overturning of the main stem, and being a self-pollinating species. For this reason, it was proposed, as study objectives, to select mutant lines developed with two mutagenic agents gamma Co-60 radiation and colchicine with characters of agronomic yield components and desirable characteristics and then detect through cytogenetic analysis the variability in somatic cells for each treatment. The DBCR design was used to evaluate the treatments at field level in 3 doses for each mutagenic agent and a control, being a total of 7 treatments (T1:T6 and T0). At the phenotypic level, quantitative agronomic traits were identified in the mutant lines, such as: yield increase T3 (637500 ears ha⁻¹), thousand kernel weight T2 (53.4 g), height frequency 80-90 cm T4 (0.45), ear length (9.8) and qualitative traits such as presence of anthocyanins in the stem auricle T2, T4, T5 and T6 and earliness of 6 months (T2 and T4). Cytogenetic evaluations were performed to detect variability in the lines obtained. Cytogenetic variations were observed in all treatments: increased micronuclei (58.3), binucleated cells (0.98), increased metaphase 46.5, etc. These lines of T polonicum L, will be an interesting alternative for flour production in Peru, so this species could be of great value as a genetic resource and for the improvement of new wheat varieties.

Keywords: mutagenic agents, Triticum polonicum L mutant lines, genetic variability.

Introduction

The progressive decrease in diversity has led to genetic erosion and led plant breeders to look for new alternatives to obtain crops with better yields and resistance to phytosanitary diseases in less time and cost. This is reflected in durum wheat, which plays the most important role in agricultural production, such as Triticum turgidum ssp. and Triticum polonicum L. MacKey (2005), as opposed to flour wheat, such as Triticum aestivum L. Triticum durum L., Triticum durum L.; current production does not cover the total needs, increasing imports, which currently reach 96% with a national production of 223 thousand MT. Peru is the fourth country with the highest per capita consumption of wheat in Latin America, with an average annual consumption of 63 kg per person, surpassed by Chile, Argentina and Uruguay with consumption of over 100 kg per person (SIN, 2014).

Triticum polonicum L. is a species that has not yet been reported to have been introduced to Peru, but it is reported to have originated in Italy, western Ukraine, warm regions of Asia and in Algeria and Ethiopia (Belay and Bekele, 2006) and is a crop widely used by farmers in the Mantaro Valley for their own consumption. The Mantaro Valley is located in the Junin Region, at 3316 masl and its main production lies in small grade cereals roots and tuberous, farmers face inclement weather such as "frosts", which occur between the months June to August, recording temperatures below -1±2 °C with SENAMHI reports (2019), this wheat presents good resistance to low temperatures the agronomic characteristics of this wheat are: grains are very elongated, plump the weight of 1,000 grains can reach 80 g and are difficult to separate from the straw, it was also demonstrated that genotypes of T polonicum L (Polish wheat) so far, there is no information on the technological quality of the grain this species could be a valuable starting material for the improvement of new wheat cultivars according to Suchowilska et al. (2019). High yielding crop cultivars often have low nutritional quality due to low levels of protein, minerals in particular Fe and Zn and selected nutraceuticals Zhao et al. (2009)as its grain content has high zinc, iron, copper, manganese, and molybdenum contents than common wheat, which contributes to its nutritional value and health benefits such as considerably higher concentrations of fat and dietary fiber (Bieńkowska, Suchowilska, Kandler, Krska, and Wiwart, 2018; Wiwart et al. (2013).

It should be taken into consideration that tetraploid wheat (AABB, 2n= 4x =28) contains a diversity of storage proteins, which contribute to increase the number of grains per ear, with high gluten content in T. Polonicum L, according to Kang (2012). One of the problems with this wheat is its tall growth habit, which is disadvantageous because the plants can reach up to 2 m in height, causing lodging. T. polonicum L., Kan (2012), another characteristic that is noticed, is its behavior with late maturation of 7 months, compared to flour wheat of 6 months, but with the advantage that T. polonicum L., after finishing its phenological development of 7 months, it resprouts tillers, which are usually used as fodder and use of the new ears that are reborn from these tillers, which is a good characteristic of this wheat for fodder for grazing animals for the villagers, and being a self-pollinating species.

The objective of the study was to select mutant lines using two mutagenic agents and identify the lines with the best agronomic characteristics, corroborating the mutations with cytogenetic analysis, which is why an interesting proposal was made in the central region of Mexico with the improvement of flour wheat, focusing on precocity, resistance to lodging and quality of soft gluten grain. Huerta et al. (2011) have tested lines in up to 90 different environments and know characteristics such as response to the diversity of races of rusts (Puccinia spp.), to limited irrigation, their yield potential, stability, consistency and behavior in certain

producing regions, which has facilitated decision making (Villaseñor et al., 2015) T. polonicum L, presenting good tolerance to this fungal disease and aphid pests.

They also present favorable morphological characteristics, such as: (i) large grains, (ii) ears with large glumes and edges, (iii) good yield for production at altitudes above 3300 masl Mont (2008), A semi-dwarf accession GA3 of the cross between lines Triticum polonicum IC12196 of the F2 was found as a potential line for backcrossing and generating wheat varieties with better establishment in plant height and higher leaf biomass in unfavorable environments watanabe (2004) optimizing yields require breeding methods such as gamma and neutron, methodologies have been established to induce variation, which are used in the improvement of important seed-propagated crops such as rice, wheat, barley, and cotton. Mutation induction techniques, in combination with tissue culture and molecular methods, allow having a methodology to improve vegetatively propagated plants such as musaceae, potato, cassava and pineapple (Ahloowalia and Maluszynski, 2001).

According to IAEA (2015), ionizing radiation has been widely used to increase genetic variability. One method to induce and identify beneficial mutations involves repeated irradiation of the material. This study was based on studies based on radiosensitivity of plant tissue, using as the most appropriate dose to have viable seeds are under 100 to 60 Gray Co γ-ray equivalent to 1J Kg-1 or 100 rads (Zamorano and Javier, 2017), and as a result, mutations accumulate in the material and the selection of beneficial mutations is encouraged Alvarez (2014). Crossing between two flour wheat lines by cytogenetic analysis of T. spelta and T. aestivum revealed similarities in their chromosomal structure. The F1 cross has revealed heterozygosity effects on grain yield, number of grains per ear and grain weight Suchowilska et al. (2019).

In this study cytogenetic analysis helped to corroborate the presence of somatic abnormalities in the treatments this is corroborated with that reported with (Predieri et al., 2001; Sakin et al., 2005; Tomlekovaet al., 2010), in which they concluded that mutagenic agents are responsible for causing lesions or alterations, conformational modifications of chromosomes as a result of incorrect repairs of the hereditary material, or without repairing alterations called aberrations, according to Cardone etal. (2010), among other changes observed in plants they point out the presence of chromosomal mosaic, abnormal meiosis and albinism.

For Fuch (2002), colchicine can be applied to germinating seeds, seedlings or adult plants Lacadena (2001), inducing the generation of aberrations and polyploidy during metaphase, as a consequence of the lack of adhesion of chromosomes to the achromatic spindle. The effects of mutagens in plants can be used for qualitative assessment of desirable and viable morphogenetic individuals (Misik et al., 2006). As some mutations are lethal for phenological development such as the presence of micronuclei reported by Alves et al. (2014), T. polonicum L. may constitute a valuable genetic material for breeding new wheat varieties characterized by high nutritional value and satisfactory resistance to Fusarium and Puccina s. (Wiwart et al., 2013).

For this reason, there are still no lines for crossing lines compatible with T. polonicum, being the use of mutagenic agents, a useful and fast alternative widely used in genetic improvement processes, according to Poehlman (1965), and with the purpose of inducing genetic variability associated with tolerance characters to biotic and abiotic factors. In this way, a BCR experimental design was applied up to generation 1 to obtain desirable mutant lines. To obtain

and detect desirable improvements, it is necessary to cultivate until the second generation, very numerous populations. In addition, in mutation breeding, the probabilities of success are greater when applied to characteristics for which efficient selection techniques exist, according to Mainero (2015). The results obtained will allow from the new lines generated and the genetic diversity present in the germplasm of ancient wheat as an important reservoir of economically useful genes for Cuadrado et al. (2008) with characteristics of resistance, higher yield, precocity and flour quality and if achieved as a crop for the highlands of Peru will allow the establishment of production and exchange relationships between producers of the middle and high agroecological floors of the inter-Andean valleys.

Materials and Methods

In the present study, two mutagenic agents were used at three different doses: with Colchicine (N-(7S)-5, 6, 7, 7, 9-tetrahydro-1, 2, 3, 10-tetramethoxy-9 oxobenzo heptalen-7-yl) acetamide) three treatments: T1:0.5 %, T2:1.0 %, T3:1.5 %. With Co-60 gamma radiation three treatments: T4:0.05 kGy, T5:0.10 kGy, T6:0.15 kGy and T0: seeds without treatment, seeds were obtained from the Germplasm Bank PC-UNCP-SPCGP (cereal program, small grain cereals sub program) of the Agricultural Experimental Station El Mantaro, which were standardized in size 1 cm in length.

The three standardized samples were irradiated, according to the protocols established by the Institute of Nuclear Energy of Peru, each sample of 500 grams per dose, in double polyethylene bags sealed, packed and labeled hermetically, the samples were irradiated with a type I irradiator Brand: Gammacell 220, Excel model: C-198, series: GS-401 gamma kGy Co-60 at doses of 0, 05kGy/1min, 0, 10kGy/2min, 0, 15kGy/3min, by irradiation field method; ET01-APLI10-SERA, at ambient conditions of19, 6°C; 968 h Pa.

For the method with colchicine 0.5 mg SKU: 7750304000109, 300 seeds of T polonicum L were sterilized at 24 h inhibition, placed in Petri dishes, with sterile distilled water and filter paper inoculated with colchicine dose at germination stage II at 0.5 %, 1.0 %, 1,5 % in each treatment of 20 seeds as experimental unit in 5 replicates for each treatment, the seeds were soaked in the second and third germination phase with each of the three doses for 24 h respectively and rinsed three times after the end of the inoculation time with colchicine.

In the experimental field, the treatments were distributed in experimental units according to the completely randomized block design (DBCR) for the agronomic management of the crop began with sowing, comparing the treatments in different conditions, so for its spatial distribution, 20 seeds were sown with a distance between plants every 15 cm and the control were installed in the Mantaro Valley at a temperature of $13 \pm 1^{\circ}$ C, 74 ± 2 % RH, the agronomic tasks necessary for its normal development were maintained, emphasizing the necessary irrigations in the critical phenological stages of the crop, carrying out quantitative and qualitative morphological evaluations of the phenotypes and taking samples for cytogenetic analysis.

Statistical analysis

For the completely randomized block design DBCR analysis of variance (ANOVA) for the yield components and for the cytogenetic analysis (MULTIVARIATE ANOVA) of two factors with three

levels was used, evaluating the seven (7) treatments with five (5) replications per treatment. The data according to Chura (2014) were analyzed by tukey ($p \le 0.05$) using the statistical program SAS 9.2 Inc.

Cytogenetic analysis

Using seeds obtained from T polonicum L. lines, Generation 1, the germination of the treated seeds was carried out in the Cytogenetics Laboratory of the Faculty of Biological Sciences at the Universidad Nacional Mayor de San Marcos, using Petri dishes, in which 10 seeds of each treatment were placed at 90% RH, temperature 21°C, 10 hours of light for 24 hours. 1 ml of distilled water was added every 12 h to hydrate the seeds until they reached 1 cm root length. Germination rate was calculated as follows:

Germination rate% = Number of germinated seeds × 100 %.

Total number of seeds

For cytogenetic analysis, roots were fixed in acetic acid ethanol (3:1) for 15 min, immersed in 1N HCl solution 5 min to clean the cytoplasm, softened with Targa solution (acetic acid, lactic acid and distilled water in 9:5:6 ratios) for minutes15. The cells were stained with 2 % acetic orcein acetate. Then a slide was placed at 45° in relation to the slide holder and squash was performed. The cells were observed under the microscope trying to identify cells that were in metaphase and that presented anomalies as a result of the application of the treatments observed at 10x, 40x and 100x. Subsequently, with the help of a Zeiss optical microscope, with a 30.3 Mpx Canon EOS camera incorporated, the cells with somatic and metaphase abnormalities were photographed at a magnification of 1000x, considering 40 cells per plate López et al. (2014).

Results

On average, 77 spikes per square meter were established Table1. The results indicate that T2 and T4 with 111.21 grains per spike and 53.5g thousand grains weight, quantitative results that exceed the control (T0), the establishment and density of grains with coefficient of variability of 2.75%, 2.73 % and 3.32% for spikes per square meter, grains per spike and thousand grains weight respectively, for resistance to stem lodging average heights 0.88 in the treatments and spike length of 9.8, finding highly significant differences between treatments.

Treatments	Spikes (m²)	Grains per ear	Weight of thousand grains (g)	Plant height (m)	spike length (cm)
ТО	106.63	26.2	48.4	1.57	13.44
T1	89.75	21	47.2	0.74	8.63
Τ2	111.21	30.65	53.4	0.75	9.6
Т3	68.79	14.15	32.2	0.73	9.06

Table 1. Analysis of variance of yield components and desirable characteristics of selected lines.

T4		72.0	72.0 19.65 53		0.78	9.01		
Т5		60.5	12.6	51.7	0.85	9.00		
Т6		30.0	9.3	42,6	0.84	10.00		
Source of variation	G.L.	СМ						
Blocks	4	3.0715 ns	2.61428 ns	0.2429 ns	0.01186 ns	2,971 ns		
Treatmen t	6	3142.8905 **	156.7142**	* 271.066** 0.39114*		13.6667**		
Error	24	2.56309	1.54762	2.2929	0.01308	1.15476		
Media	Media		19.07	47	0.88	9.8		
C. V. (%)		2.75	2.73	3.32	13	10.97		
S		1.6009	1.244	1.5142	0.1144	1.07460		
CM: mean square								
Table 2. Sel	ection	of mutant line	S.					
Mutant li	ine		Phenotypic characteristics					
TPVEM-0	C1	Good spike albinism performance, superior resistance to stem flare smooth edges (ArLI)						
TPVEM-0	C2	Superior yield; average ear length with frizz, presence of anthocyanins in atrium and stem, Erecta vertical Intermedia (EI) ear, resistance to puscina striformis (stripe rust), increased resistance to stem blight, early wheat						
TPVEM-(C3	Good yield, spike length and width, increased stem flare resistance, erect spike (EE), short ridge-ridge (ArCc)						
TPVEM-I	R4	High yielding, small albino spikes, Intermediate Erect (EI) vertical spike, wide basal spikes, resistance to stem lodging, short semidwarf short-ridge early wheat (ArmCc)						
TPVEM-I	VEM-R5 Resistance to Puccinia striiformis (yellow rust), resistance to stem lodging, and smooth edges (ArLI).					em lodging, long		

TPVEM-R6 Resistance to stem blight, presence of anthocyanins in stem auricles, albinism of spikelets, resistance to Puccinia striiformis (yellow rust), short ridge crest (ArCc).

TPVEM-C1: TP: Triticum polonicum L, VEM: El Mantaro valley, C: colchicine treatment, R: treatment with Co-60 gamma radiation, 1: number of doses per treatment.

Changes in the coloration of the auricles were observed due to an increase in anthocyanins in the treatments compared to the control, which presented amber colored auricles, T1 and T2 presented purple coloration at the ends of the auricles and the stem, T3 auricles with wine coloration at the ends, T4 and T6 auricles with purplish blue coloration and T5 with purplish red coloration extending to the base of the leaf as shown in Figure 1.



Figure 1. Color changes in atria from treatments in T. polonicum.

In the treatments of T. polonicum in Figure 2, size variations in diameter and coloration were observed by treatment compared to the control (a). Color variations ranging from beige to light pink shades in spikes and edges compared to (a), which is spike and light brown edge color, which coincides with the descriptions of Mont (2008). Regarding ridge shape, modifications were found in a mutant line of T4 (e) presenting a short semi-smooth ridge (ArCc) compared to the control (a) which are long and smooth ridge (ArLl), T1, T2, T3, T5 and T6 (ArLl), (ArSmCc), (ArMCc) and (ArLc) respectively.



Figure 2. Morphology and color of spikelets of treatments by treatment compared to the control A: (T0), B-D: (T1, T2, T3), E-G: (T4, T5, T6).

Cytogenetic analysis with colchicine (Fig. 3) showed polyploidy (c, e, f), multipolarity (c, f, k, l) in treatments T1, T2 and T3, monoploidy (d, o) in T1, and the presence of buds. Micronuclei in T2 and T3, in T2 only binucleated cells (g, h) were observed. In T3, there was a significant increase of micronuclei, buds, anaphase bridges.



Figure 3. Cytogenetic analysis of the 0.5% colchicine treatments: (a-e), 1%: (f-k), 1.5%:(l-q).MN: micronuclei, Y: buds, M: multipolarity, P: bridges, CA: unattached chromosome, CB: binucleated cells, CR: broken chromosome, A: asynchronism, MP: monoploidy, MF: metaphases.

In the treatments with Co-60 gamma radiation (Fig. 4), individual micronuclei or a greater number of micronuclei per cell were observed, as shown in Fig. 2; also higher frequency of buds in (b, c, m, n, p and s), accompanied with MN and P in (b, c) in T4, (g, r) in T5 and T6 micronuclei, unattached chromosomes, buds in T5, high frequency in comparison to the control buds and micronuclei in T6. Arrows indicate the presence of abnormalities found in each treatment.



Figure 4. Cytogenetic analysis of Co-60 gamma radiation treatments, 0.05kGy: (A-E), 0.10kGy: (F-N), 0.15kGy: (Ñ-U), control:(V-W).MN: micronuclei, Y: buds, M: multipolarity, P: bridges, CA: unattached chromosome, CB: binucleated cells, CR: fragmented chromosome, A: asynchronism, MP: monoploidy, MF: metaphases.

The six treatments evaluated (Fig. 5) show that there is a highly significant difference ($p \le 0.001$) in the frequency of micronuclei (MN) in both mutagenic agents, compared to the control in which there was no somatic alteration; in addition, significant differences were found in Y, P, CB, CF, A, MP and MF. But the results were not significant in CA for both mutagenic agents. The cells evaluated in the control presented an average of 11.84 metaphases, which in comparison with the colchicine treatment increased significantly in frequency, while26.44, the opposite happened with Co 60 gamma radiation where the average was 7.43 metaphases.

The analysis of variance showed that there are significant differences (p<0.05) between treatments T6, T5 (kGy) and T1, T2 (%). Significant variations were observed in micronuclei frequency T6 (58.33%), T5 (48.67%), T4 (35.3%); an increase of metaphases in T1 (46.5%), T2 (22.3%) for colchicine treatment and T5 (9.8%) for gammaCo-60 radiation Tukey's multiple mean comparison frequency values.





Figure 5. Somatic abnormalities in root meristem cells of Triticum polonicum L., after 48 h exposure to (A): colchicine (%) and (B): gamma radiation (kGy). MN: micronuclei, Y: buds, M: multipolarity, P: bridges, CA: unattached chromosome, CB: binucleated cells, CF: fragmented chromosome, A: asynchronism, MP: monoploidy, MF: metaphases. T0: control, T1: 0.5%, T2: 1%, T3: 1.5%, T4: 0.05kGy, T5: 0.10kGy, T6: 0.15kGy.

In the Tukey multivariate analysis of means, T6 statistically outperformed the other treatments with a mean of 3.17 buds in somatic cells, as well as the presence of micronuclei 58.33, followed by T5 for multipolarity no significant differences were found, but a higher frequency in treatments T1, T2 and T5, similar presence in ploidy, CA, CB, CR, A and MP. Treatments T1 and T2 were statistically superior to the other treatments with means (46.5 and 22.83) respectively.

Table 3. Multivariate analysis of variance of the seven treatments in somatic cells of Triticum polonicum L. mutant lines.

Treatments (%)										
Somatic abnormalities	T1	Т2	Т3	Т4	Т5	Т6	то	CV ^a	SD	Pr>F
Y	0.83b	0b	1.17b	2.83a	3.84ª	3.17a	0b	40.02	0.68	**
MN	3.33d	0.17e	1.66of	35.3c	48.67b	58.33a	0e	5.90	1.23	***
М	1.17a	1.33a	0.67a	0a	0.82ª	0.17a	0a	124.48	0.74	*
Р	0b	0b	0b	0b	1.17ª	0b	0b	105.73	1.18	**
CA	0.17a	0.49a	0a	0a	0.33ª	0.33a	0a	208.76	0.39	NS
СВ	0.5a	0.82a	0.98a	0a	0a	0a	0a	149.03	0.49	**
CF	0b	0b	0b	0b	0.50ª	0b	0b	241.52	1.17	**
А	0b	0b	0.5a	0b	0b	0b	0b	241.52	1.17	**
MP	1.33a	0b	0b	0b	0b	0b	0b	222.78	0.42	**
MF	46.5a	22.83b	10cd	5d	9.8cd	7.5cd	11.84c	19.57	3.17	**

MN: micronuclei, Y: buds, M: multipolarity, P: chromosome bridges, CA: unattached chromosomes, CB: binucleated cells, CF: fragmented chromosome, A: asynchronism, MP: monoploidy, MF: metaphases. *** Significance at 0.001 ** significance at 0.01, * significance at 0.05 NS: not significant. Different letters within columns present significant differences (p<0.05) Tukey, a=0.05.

Discussion

The results in Table 1 agree with the reports of Pavón (2011), who mentions that the category of physical and chemical mutagens is capable of generating genetic variability, altering the hereditary material, increasing mutations above the natural level. The increase in the frequency of phenotypes with lower height compared to the control T0 could possibly be explained by the presence of the semi-dwarfism gene reported in a semi-dwarf accession GA3 of the cross between lines of Triticum polonicum IC12196 watanabe (2004), to which the increase in the frequency of phenotypes with lower height compared to the control T0 could be explained. The increase in yield in T2 and T4 compared to the control are clear evidence of what was reported by Huerta et al. (2011) that the improvement of flour wheat is focused on earliness, resistance to lodging and quality of soft gluten grain tested in different wheat lines and environments, which generates resistance and activation of ancestral genes of T. polonicum L.

Within the field observation, it was observed that the treatments of T. polonicum L present resistance to stripe rust, since this infection at severity levels affects the yield by reducing

the photosynthetic area up to 100% and at the same time synthesizes photosynthates important for grain filling and gleaning, important factors mentioned by CIMMYT (2006) and that were one of the new characteristics to identify the mutant lines. SnRK2 (plant-specific protein kinase) are involved in several biological processes, such as plant defense and environmental challenges (Want et al., 2019).

At four days of ear emergence which is an indicator of earliness it was observed that T2 was the earliest and the Co-60 gamma radiation treatment was T4, compared to the control of which T2 surpassed it in earliness at 6 months and which agrees with what was mentioned by Poehlman (1965) since the use of mutagenic agents is a useful and fast alternative widely used in genetic improvement processes and the crop adapts efficiently to biotic and abiotic factors. The treatments analyzed cytogenetically in meristematic roots of T. polonicum L., which showed somatic abnormalities as micronuclei compared to the control in both mutagenic agents, coincided with Cardone et al. (2010), a strong highly significant variation in the frequency of micronuclei T6 (58.33%), T5(48.67%), T4(35.3%) of Co 60 gamma radiation and metaphases in T1(46.5%), T2(22.3%) for colchicine treatment and T5(9.8%) for Co 60 gamma radiation.

The increase in the frequency of micronuclei, metaphases and other anomalies in somatic cells of T. polonicum L., after exposure to 48h (colchicine) and Co-60 gamma radiation (1 to 3 min) shows a significantly higher frequency, because they suffered a mutagenic effect, resulting from unrepaired or improperly repaired damage in the parental cells (Leme and Morales, 2009).The dose and response values indicate higher somatic abnormality in seeds treated at low doses for Y, MN, MF, in treatments T1 and T2 but the opposite is presented in binucleate cells exposed to colchicine which does not coincide with what was reported by (lavicoli et al. 2014) demonstrating that an increase of aberrations in somatic cells in T. polonicum L., which are generally observed ultrastructural changes and could be associated with apoptosis, necrosis, vacuolar deformation, morphological modifications that do not significantly affect their viability and phenological development Doorm et al. (2011).

Conclusion

Six mutant lines TPVEM-C1, TPVEM-C2, TPVEM-C3, TPVEM-R4, TPVEM-R5, TPVEM-R6 were selected, developed with the two mutagenic agents: gamma radiation Co-60 and the chemical agent colchicine with desirable agronomic characteristics such as high yields T3 (637500 ears. ha^{-1}), thousand kernel weight T2 (53.4 g), resistance to lodging frequency of height 80-90 cm in treatment T4 (0.45), ear length (9.8) and earliness of 6 months (T2 and T4).

In addition, tolerance to puccina striformis sp, and as new characteristics, the presence of anthocyanins in T2, T4, T5 and T6 and good performance against frost in the Mantaro Valley. Cytogenetic analysis also identified changes at the cellular level, mainly a high frequency of micronuclei (58.3%) evidenced in all treatments with mutagenic agents, treatments T2 and T4 being one of the best lines as a genetic resource for breeding and crossbreeding with the species Triticum polonicum L. The two mutagenic agents developed in T. polonicum L. proved to be

suitable for producing genetic variability for new mutant lines with agronomic characteristics for improvement in self-pollinated plants.

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