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PARAMETERS OF WINTER WHEAT GROWING AND DEVELOPMENT AFTER MUTAGEN ACTION

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Abstract: *Dry seeds of seven winter wheat varieties and one line were exposed by gamma-rays and chemical mutagens to determine their effect on plant grows and development. Next parameters have been investigated – plant survival, pollen fertility, components of yield structure. Informative value by mutation depression evaluation of all parameters has been developed. The most informative parameters for mutagenic depression in M_1 generation of winter wheat plants were survival of plants, pollen fertility, indicators such yield structure parameters as plant height, grain weight per spike, thousand grains weight. Insensitive and most sensitive for mutagen actions varieties have been developed.*

Key words: *mutagen depression, winter wheat, chemical mutagens, gamma-rays.*

1. Introduction

There are three reasons for the study of plant mutant generation M_1 after mutagenic effect. The first one is the determination of the mutagen-polluted area suitability for agriculture. More than 70% of soil in Ukraine used for agriculture is constantly exposed to chemical and physical mutagens. Ukrainian government planned to use some areas with high level of pollution for forestry and production of grain. However, data on the negative consequences in form of reduced grain productivity of plants cultivated on such soil (effects of mutagenic depression in the first generation) have been obtained [12 - 15].

The second reason is the fact that the amount of material, obtained from the first generation, limits the opportunities of mutation breeding programmes (especially using the lethal doses). Induce of mutations in crop plants contribute by increasing genetic variability and enrich plants germplasm for direct selection and cross-breeding [11, 19]. More than 3000 mutant varieties have been directly or indirectly derived through mutation induction, including 200 bread wheat varieties [2]. Induced mutations have been applied to produce mutant varieties by changing the plant characteristic for a significant increase in production and improve quality [1]. Wheat is the top food crop in Ukraine as well as in the whole world and the biggest part of grain is

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obtained primarily from winter wheat. Wheat is the stable food of millions of people globally. This crop is widely adapted to wide range of climatic conditions. A large variety of food that include bread, cakes, noodles, crackers, breakfast food, biscuits, cookies and many other confectionary items are prepared from wheat. The total area for winter wheat cultivation in Ukraine covers 6.8 mln ha with actual productivity of 24 mln tons and average capacity of 2.8 t/ha [12].

The third reason is the use of obtained M_1 population to extract valuable mutant strains in further generations. The study is devoted to the survival ability and productivity of plants exposed to mutagenic effect at different doses, concentrations and mutagen nature. Mutagenesis reduces plant growth and other crop yield structure components, increases the pollen sterility and cuts the germination and survival abilities of plants by means of chemical agents and gamma rays; sometimes the great part of population is killed with the critical doses [9, 10, 16, 20]. Depression increases with the increase of dose [6, 7, 9]. But sometimes we observe stimulating effect (in case of low doses) or absence of depression (at medium concentrations of some chemical mutagens) [14, 15, 18, 19].

Thus, there is a contradiction between the mutagen efficiency (for breeding purposes) in the induction of valuable mutations, which often occur at high doses and small population of M_1 plants obtained when applying such doses [17, 19].

High doses are more successful in obtaining large quantity and a wide range of mutations. Some genotypes have total or partial resistance to mutagenic effect. These genotypes are important both for obtaining new varieties of plants and cultivation in case of mutagenic pollution.

Gamma rays, nitrosomethiureas, methylsulphat and diazoatsetilbutan are the most important and frequently used mutagens well known for their effect on the plant growth and development and the appearance of morphological, cytological and physiological changes in cells and tissues; they are also traditional in breeding. Most commercial varieties are obtained by means of gamma irradiation [20].

The development of direct mutants into commercial varieties is still a common practice in seed propagated crops [8]. In first generations for wheat as cultivar composed from three genomes we observed only some dominant mutations in our investigations, but we continued our experiments on next generations (M_2 – M_6). Positive desirable mutant will be selected and be incorporated in future breeding programs.

2. Material and methods

Dried seeds (approx. 14% moisture content) of Favoritka, Lasunya, Hurtovina, Kolos Mironovschiny Sonechko, Kalinova, Voloshkova varieties and line 418 of winter wheat (Triticum aestivum L.) were subjected to 100,150, 200, 250 Gy gamma irradiation, nitrosomethilurea (NMU) 0,0125 and 0,025 %, nitrosoethilurea (NEU) – 0,01 та 0,025 %, 1,4-bisdiazoatsetilbutan (DAB) – 0,1 and 0,2 %, dimethylsulphat (DMS) – 0,0125, 0,025 and 0,05 % presoaked. Each treatment was comprised of 1000 wheat seeds. Exposition of chemicals mutagens was 18 hours. These concentrations and exposure are optimal for the breeding process that has been repeatedly established earlier [1, 5, 11, 15]. Non-treated varieties were used as a check.

Treated seeds were grown in rows with inter and intra-row spacing of 50 and 30 cm, respectively, to raise the M_1 population. The untreated seeds of mother varieties (parental line/variety) were also

planted after every ten rows as control for comparison with the M_1 population [17]. M_1 plant rows were grown in three replications with check-rows of untreated varieties in every ten-row interval. The planting was implemented in 2010 – 2014 (five years of estimation) growing season in DSAEU science-educational center, Aleksandrovka, Dnepripetrovsk region, Ukraine. Black soil of the center is characterized by clay loam containing high organic matters. The center are lying 245 above the sea level with 8 - 11 °C temperature during wheat growing season (September/July) and the average rainfall is about 400 - 550 mm in vegetation season respectively. Normal cultural practices including fertilization were done whenever it is necessary. Data on seed germination and surviving plants were recorded considering whole plots of M_1 population. Data on yield structure components (plant height, general number of culms, number of productive culms, spike length, spikelets per spike, number of grain per spike, grain weight per spike and plant, 1000 grains weight) were taken from 50 randomly selected plants of each treatment representing more or less all types of morphological plants [3, 4].

Mathematical processing of the results was performed by the method of analysis of variance; the variability of the mean difference was evaluated by Student's t-test. Factor analyses were conducted by module ANOVA. In all cases standard tools of the program Statistica 6.0 were used.

3. Results and Discussion

In M_1 population, observations were recorded seed germination and plant surviving, pollen fertility, plant height, spikes/plant, spike length, kernels/spike, 1000-grain weight, yield/plant (Table 1 and 2). Standard error (±SE) values of the treated populations are at tables too.

The results on germination of seeds, survival rate of plants derived from treated and untreated seeds are tabulated (Table 1).

Germination and survival abilities of seeds reduce compared to untreated seeds of the initial variety in all cases except one (Sonechko, NEU). Plant survival ability ranges from 55 (Voloshkova) to 5.3% (Sonechko) at 250 Gy, from 70 (Sonechko, Kalinova) to 63% (Lasunya) at 0.025% NMU, from 85 (Line 418) to 79% (Voloshkova) at 0.2% DAB, and from 45% (Hurtovina) to 38% (Line 418) at 0.05% DMS, while it ranged from 98 to 92% under untreated control. As for the impact of gamma rays on the germination and survival abilities, it is the usual effect in plants for most crops previously observed by many researchers in wheat as well. However, we can see that chemical mutagens are more specific at the comparable effect.

In general, the correlation between the dose value and survival abilities of plants is at the level of 0.9 for gamma rays and 0.7-0.8 for chemical mutagens. *Sonechko* was extremely sensitive to gamma rays, but its sensitivity to nitrosoalkilureas together with *Kalinova* was weak. Plants of all varieties showed higher level of depression being processed with DMS and gamma rays.

Correlation between the dose value and pollen sterile was -0.9 for any type of mutagens. As we can see, the highest level of this indicator was observed after the mutagenic effect on *Kolos Mironivschini* seeds. Frequency of pollen sterility was of the highest level after gamma irradiation, primarily, in the varieties obtained by processing with gamma rays. Only these varieties showed extremely low fertility at a dose of 250. For this parameter, sequence of mutagens by depressive consequences is

as follows: gamma-rays →DMS →NMU →NEU →DAB from the highest to the lowest value of depression. Pollen sterility is the more reliable parameter for monitoring depressive consequences compared to germination and survival rates. The Table 2 shows that resistance to mutagenic effect directly depends on the genotype of processed material.

All parameters of the crop yield structure have been studied. Components such as plant height, 1000 grain weight, grain weight per plant, number of grains per spike, grain weight per spike, general number of culms, number of productive culms, spike lengths have been developed. But only three (plant height, grain weight per spike and1000 grain weight) showed statistically difference level of mutagen depression under any dose action.

Main parameters of grown of winter wheat plant at M_1 generation Table 1

Trial	Germination, %	Survival g after winter, %	Germination, %	Survival after winter, %
	Variety: Kolos Mironivschini		Variety: Kalinova	
Check	98±0,57	91±0,93	94±0,94	88±0,98
gamma-rays, 100 Gy.	66±0,76*	76±1,01*	75±1,07*	70±1,11*
gamma-rays, 150 Gy	69±1,09*	66±1,13*	71±1,15*	66±1,18*
gamma-rays, 200 Gy	58±1,48*	54±1,71*	47±1,24*	44±1,43*
gamma-rays, 250 Gy	38±1,26*	36±1,34*	37±0,83*	35±1,10*
NMU, 0,0125%	80±1,05*	78±1,01*	74±0,61*	73±0,56*
NMU, 0,025%	66±1,01*	65±0,87*	69±0,49*	68±0,48*
NEU, 0,01 %	81±0,94*	81±0,94*	82±0,92*	82±0,92*
NEU, 0,025 %	74±0,70*	73±0,67*	76±0,80*	76±0,78*
DAB, 0,1%	85±1,03*	84±0,96*	84±0,51*	83±0,68*
DAB, 0,2%	82±0,63*	80±1,01*	81±0,79	81±0,78
DMS 0,0125%	76±0,88*	76±0,96*	77±0,99*	75±1,10*
DMS 0,025%	65±0,72*	65±0,64*	67±1,82*	64±1,23*
DMS 0,05 %	41±0,47*	40±0,43*	45±1,10*	42±0,98*
	Variety: Voloshkova		Variety: Sonechko	
Check	92±0,57	87±0,93	94±0,94	89±0,98
gamma-rays, 100 Gy.	73±0,76*	69±1,01*	65±0,57*	62±0,93*
gamma-rays, 150 Gy	64±1,09*	60±1,13*	43±0,57*	40±0,93*
gamma-rays, 200 Gy	55±1,26*	52±1,34*	31±1,14*	29±1,72*
gamma-rays, 250 Gy	55±1,48	51±1,71	5,6±1,07*	5,3±1,39*
NMU, 0,0125%	78±0,90*	78±0,81*	79±1,02*	78±0,82*
NMU, 0,025%	69±0,70*	67±0,69*	69±1,04*	67±0,94*
NEU, 0,01 %	90±0,81	88±0,74	87±1,10*	87±1,0
NEU, 0,025 %	82±0,78*	80±0,68*	79±1,40*	78±1,25*
DAB, 0,1%	84±0,96*	84±1,10*	84±1,02*	84±1,10*
DAB, 0,2%	80±0,74*	79±0,90*	84±1,06	83±1,10
DMS 0,0125%	77±1,03*	76±0,85*	75±1,11*	74±0,98*
DMS 0,025%	64±0,88*	63±1,20*	68±0,43*	66±0,63*
DMS 0,05 %	40±1,12*	39±1,17*	40±0,34*	40±0,82*
	Variety: Favoritka		Variety: Hurtovina	
Check	98±0,57	91±0,93	92±0,94	84±0,98
gamma-rays, 100 Gy.	82±0,76*	76±1,01*	73±1,07*	67±1,11*

Trial	Germination, %	Survival g after winter, %	Germination, %	Survival after winter, %
gamma-rays, 150 Gy	58±1,09*	54±1,13*	52±1,15*	48±1,18*
gamma-rays, 200 Gy	49±1,26*	45±1,34*	55±0,83*	50±1,10*
gamma-rays, 250 Gy	39±1,48*	36±1,71*	36±1,24*	33±1,43*
NMU, 0,0125%	79±0,93*	78±0,87*	78±1,01*	77±0,93*
NMU, 0,025%	70±1,30*	69±1,0*	68±0,76*	68±0,74*
NEU, 0,01 %	88±0,82*	87±0,74*	86±0,90*	85±0,36
NEU, 0,025 %	82±1,04*	80±0,98*	81±1,02*	79±0,99*
DAB, 0,1%	84±0,92*	84±0,98*	85±0,71*	84±1,10
DAB, 0,2%	86±0,98	85±1,11	81±0,86*	80±1,10*
DMS 0,0125%	73±0,84*	73±1,13*	74±0,93*	73±0,73*
DMS 0,025%	68±0,92*	67±1,26*	61±1,07*	60±0,97*
DMS 0,05 %	45±0,67*	44±2,13*	47±0,65*	45±1,08*
	Variety: Lasunya		Variety: Line 418	
Check	98±0,57	94±0,93	93±0,94	92±0,98
gamma-rays, 100 Gy.	54±0,76*	52±1,01*	74±1,07*	67±1,11*
gamma-rays, 150 Gy	48±1,09*	46±1,13*	70±1,15*	55±1,18*
gamma-rays, 200 Gy	42±1,26*	41±1,34*	48±1,24*	36±1,43*
gamma-rays, 250 Gy	37±1,48*	35±1,71*	39±0,83*	35±1,10
NMU, 0,0125%	76±1,20*	75±1,0*	82±1,40*	80±1,2*
NMU, 0,025%	64±1,50*	63±1,1*	73±0,81*	68±0,34*
NEU, 0,01 %	88±1,10*	85±0,88*	88±1,12*	85±1,02*
NEU, 0,025 %	81±1,30*	79±1,1*	78±1,50*	78±1,04*
DAB, 0,1%	86±1,11*	86±0,90*	89±1,14*	88±1,10*
DAB, 0,2%	84±1,70	83±1,16	85±0,61*	84±1,09*
DMS 0,0125%	75±0,80*	74±1,16*	76±1,02*	74±0,67*
DMS 0,025%	68±1,00*	67±0,95*	64±1,12*	62±0,33*
DMS 0,05 %	41±1,36*	39±0,34*	39±1,13*	38±0,92*

* - difference is statistically significance from check at $t_{0.05}$

Pollen fertility after mutagen action, % Table 2

Trial	Kolos Mironivschini	Kalinova	Voloshkova	Sonechko	Favoritka	Hurtovina	Lasunya	Line 418
Check	93,1	95,0	89,7	96,7	95,7	98,6	96,8	93,0
gamma-rays, 100 Gy	82,9*	91,2*	81,3*	84,5*	79,9*	82,3*	84,8*	89,1*
gamma-rays, 150 Gy	74,6*	82,7*	74,5*	70,9*	64,7*	67,8*	71,2*	81,6*
gamma-rays, 200 Gy	69,8*	71,2*	69,2*	64,5*	50,7*	59,9*	61,3*	73,4*
gamma-rays, 250 Gy	52,5*	64,6*	61,6*	42,3*	42,5*	47,9*	43,8*	66,1*
NMU, 0,0125%	89,1*	84,3*	87,6	79,8*	90,1*	88,5*	89,2*	89,6*
NMU, 0,025%	85,2*	72,3*	84,3*	64,2*	85,4*	84,4*	86,6*	85,4*
NEU, 0,01 %	90,2*	88,0*	88,9*	84,6*	93,0*	95,4*	93,2*	90,1*
NEU, 0,025 %	88,4	84,2*	87,0*	80,1*	90,6*	88,7*	90,8*	88,3*
DAB, 0,1%	95,4	90,0*	73,4*	90,5*	93,1	95,9	94,2	89,7*
DAB, 0,2%	88,3*	88,7*	70,0*	88,8*	89,4*	90,8*	91,5*	88,1*

Trial	Kolos Mironivschini	Kalinova	Voloshkova	Sonechko	Favoritka	Hurtovina	Lasunya	Line 418
DMS 0,0125%	93,8	89,8*	82,3*	83,0*	80,0*	85,3*	83,1*	83,6*
DMS 0,025%	92,0*	83,7*	77,3*	78,6*	75,7*	78,6*	70,9*	70,2*
DMS 0,05 %	88,7*	78,0*	69,1*	71,2*	65,2*	64,7*	62,6*	58,3*

* - difference is statistically significance from check at $t_{0.05}$

Regarding the plant height, correlation between the dose and the indicator constituted -0.89, (high invert correlation). This parameter decreases if the dose increases. However, the differences between versions can be statistically unreliable. Gradual decrease in height is a tendency. We have not observed any differences between the varieties except for the stimulating effect of NEU at a concentration of 0.01% for *Kalinova* and *Sonechko*. These varieties have been obtained by means of chemical mutagenesis.

The indicator of grain weight per spike was more informative, weight was falling statistically valid with every increase in dose. Here we have the same situation with the varietal specificity by depression as in the previous case. *Sonechko* responded to mutagenic effect in the worst manner. The same stimulatory effect as in the previous case with *Kalinova* was observed. The correlation coefficient was -0.92.

The housand grain weight is the most informative indicator. Depression value at each dose is clear and statistically valid. *Sonechko* was not the worst variety according to this indicator. The correlation coefficient was - 0.96.

The high variability by these indicators gives greater opportunities for the selection of desirable genotypes.

The most informative parameters to determine the degree of mutagenic depression in the first generation for plant growth and development were germination and survival rates, pollen fertility rate, indicators such yield structure parameters as plant height, grain weight per spike, thousand grains weight.

Factor analysis showed that the formation of all crop yield structure values primarily depends on the genotype factor and secondly on the mutagen dose and nature for chemical agents, and on the dose, genotype and mutagen nature for gamma rays respectively.

6 mutant forms have been developed in M_1 populations (21 sterility or semi-sterility, 11 by changes in plant height, 14 by early maturity, predominantly by 200-250 Gy). All these forms were sown in next generations.

4. Conclusions

The most informative parameters to determine the degree of mutagenic depression in the first generation for plant growth and development were germination and survival rates. Thus, the greatest depression among all varieties was observed in *Sonechko* under all parameters (except thousand grains weight). *Kolos Mironivschini* was the most resistant to mutagenic effect. We recommend it for the cultivation on soil polluted with mutagens.

The indicators of plant germination and survival rates are recommended to be used as model parameters of mutagenic depression. Varieties obtained by action of

chemical mutagenesis show specificity in demonstration of mutagenic depression based on the presence of stimulating effect or the absence of depression for some indicators of crop yield structure.

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