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Effect of gamma radiation of 60Co on sunflower plants (*Helianthus annuus* L.) (Asteraceae), from irradiated achenes

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Abstract

In order to know the effect of 60Co gamma irradiation, in the sunflower crop, were irradiated achenes in the Transelektro LGI-01 in the Instituto Nacional de Investigaciones Nucleares. The data was evaluated under a completely randomized design, where the treatments were 0, 100, 200, 300, 400, 500, 600, 700, 800 and 900 Gy and four repetitions (10x4) = 40 experimental units. The response variables were: plant height, root length and volume, dry biomass. The results indicated that germination and sprouting decreased as the radiation increased, adjusting these to a quadratic model. Plant height, length, root volume and dry biomass decreased at high doses. From this investigation it was concluded, that doses of 100 and 200 Gy, have a stimulating effect on plant height and root length, being an important agent, to induce genetic variability in sunflower.

Keywords: genetic variability; mutation; phenotype.

1. Introduction

Sunflower (*Helianthus annuus* L.), it is an oleaginous crop that for many years was considered to be of European origin, particularly from the old Russia, although its center of origin is northeast of Mexico and southeast of the United States of America (Torretta *et al.*, 2010; Bye *et al.*, 2009), where there are the ancestors of this, such as the genera *Helianthus* and *Tithonia*, with the latter has genetic compatibility and can be crossed due to their equal chromosome number 2n = 34 (Luévanos *et al.*, 2010). Among the uses of this crop has been: ornamental, fodder and bioremediator plant of soils affected by hard water, due to its great capacity to absorb cations, such as Ca^{++} (Díaz *et al.*, 2017). Regarding gamma irradiation, it is a type of ionizing radiation with great penetration power, due to its short wavelength. This is emitted by some radioactive elements such as ^{60}Co, which is the most used in breeding programs such as radioinduced mutagenesis, to induce genetic variability that almost does not exist in nature (Iglesias *et al.*, 2010). The prior art, has its foundation in the great penetration power of gamma radiation 7.5 MeV (1.2 x 10^{-12} J) (OIEA, 2017), which causes direct damage to the DNA molecule, since it breaks the hydrogen bonds that join the purine and pyrimidic bases, including causing changes in the mating of the same, causing in the soma mutations known as molecular mutations, which can be heritable (Lyaz and Naz, 2014). These changes, in most cases, can cause unwanted changes in the phenotype of the mutant, but in some cases, those mutations may be of interest to the breeder, thus, radio-induced changes can be: high yield, resistance to some adverse factor for the crop, new color shades, etc. (Gómez *et al.*, 2017). Regarding the application of ^{60}Co gamma radiation in crops, Díaz *et al.*, (2003), mention that ^{60}Co gamma rays, induce genetic variability in bulbs of

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Tigridia pavonia (L. f.) DC, in the M_1V_1 generation, when *Tigridia* is destined to gardening. On the other hand, Fe *et al.* (2000), mention that the application of 350 Gy of radiation, inhibit the production of pods in the cultivation of soybean (*Glicine max* L.). In horticultural crops such as potatoes (*Solanum tuberosum* L.), Salomon *et al.* (2017), they indicate that dose of 20 Gy of gamma irradiation of Co ^{60}Co, stimulate the germination of the botanical seed Atzimba x TPS-13 whose germination is low. As you can see, in the sunflower crop, Work on this tendency to induce genetic variability is limited.

Therefore, the general objective of the present study was: to evaluate the effect of ^{60}Co gamma radiation, from seedlings from irradiated achenes.

2. Materials and methods

Study area

The present investigation was carried out in the experimental field of the Universidad Tecnologica de Tehuacan, located at 18°24'51" north latitude, 97°20'00" west longitude and 1409 meters of altitude.

Germplasm and radiation dose

The genetic material consisted of achenes of sunflower cv. Victoria, which were donated by the germplasm bank, of “Ecofisiologia de Cultivos” Colegio de Postgraduados Campus Montecillos, México. The achenes were irradiated, in the Transelektro LGI-01, of Instituto Nacional de Investigaciones Nucleares (ININ), located in Marquesa Ocoyoacac, Mexico.

Experimental design and treatments

The experimental design was completely randomized (DCR) with four repetitions (10x4) = 40 experimental units, following the mathematical model $Y_{ij} = \mu + T_i + \varepsilon_{ij}$ where: Y_{ij}, is the response variable of the i-th radiation dose in the j-th repetition; μ, is the true general average; T_i, is the effect of the i-th radiation dose and ε_{ij}, is the experimental error of the i-th radiation dose in the j-th repetition (Cochran and Cox, 2010; Infante and Zarate, 2005; Steel y Torrie, 1985). The treatments consisted of radiation doses: 0, 100, 200, 300, 400, 500, 600, 700, 800 and 900 Gy, which were previously determined for sunflower, as established by Díaz *et al.* (2017).

Sowing and handling of the experiment

The irradiated achenes were planted in 200-well polystyrene trays, using Peat moss as substrate, depositing an achene for each cavity. In order to obtain a good germination, the substrate during sowing was maintained at field capacity. To avoid damage by solar radiation, the tray with the genetic material, was placed in the incubation area of a saw-type greenhouse. To avoid damage by pathogens, when the seedlings emerged, they were treated with streptomycin sulfate at a rate of 0.60 g L^{-1}, using running water as a vehicle.

Response variables

Germination percentage

It was determined for each dose of radiation, by means of the relationship GP = $\frac{GS}{SS} x\, 100$ where: PG, is the percentage of germination; GS, germinated seeds and SS, seeds sowing.

Sprouting percentage

It was determined for each dose of radiation, by means of the relationship SP = $\frac{SP}{SS} x\, 100$ where: SP, is the percentage of sprouting; SP, sprouted plants and SS, seeds sowing.

Plant height

This variable was measured, starting from the epicotyl of the plant, up to the apical bud, using a ruler three meters high and reporting the result in centimeters.

Length and root

A representative sample of five seedlings was taken, to later measure, from the cap of the root, to the beginning of the epicotyl. To achieve the above, a digital vernier was used and the results were reported in centimeters.

Root volume

The root volume was calculated using the Archimedes principle. Using a 100 mL test tube, a volume of water known in this case was added, 50 mL. The volume of water displaced, when introducing the sunflower root, will be the root volume in cm^3.

Dry biomass

Three seedlings were taken, which were introduced in paper bags, to be introduced in a foamed air oven, for 72 hours, until reaching the constant weight and measuring its weight, in a model analytical balance USS-DBS3-3 (Barrios *et al.*, 2014). The variables were subjected to an analysis of variance (ANOVA), in order to determine significant differences between them.

When the response variables were significant, they were applied Tukey's multiple comparison test, at a level of significance of 5% probability of error.

3. Results and discussion

Percentage of germination and sprouting

Germination as well as sprouting, were adjusted to a third-degree polynomial model, with a highly significant coefficient of determination. The highest percentages of these variables occurred with doses of 100 and 200 Gy, whose values were 95 and 93% respectively, to decrease progressively as the radiation dose was increased (Figure 1). These mathematical models, differ with those presented by Loeza *et al.* (2016), who mention that the germination in seeds of *Hibiscus sabdariffa* L., subjected to gamma irradiation of ^{60}Co, decreases as the radiation dose increases from 5 Gy to 50 Gy and whose model had a linear fit. Similarly, Antúnez *et al.* (2017), mentions that the application of 100, 200 and 300 Gy of gamma radiation, does not affect the germination percentage of irradiated seeds of *Physalis peruviana* L., which differs with the present study. This is mainly due to the different species used in both studies, as well as the irradiation ranges used.

Morphological variables

The highest plant height, as well as root length and dry biomass, were induced with 100 and 200 Gy, while in the range of 400 to 700 Gy, plant height and root length were statistically equal. The lowest cumulative biomass was found in the high doses of radiation, 800 and 900 Gy, with 3.6 and 3.3 g respectively (Table 1). This response was due to a lower height of the plant, which caused the plant to accumulate a lower dry biomass. This same trend was observed in Leucaena leucocephala plants from irradiated seeds, where doses of 20, 80 and 100 Gy of irradiation, inhibit root growth, by 5 and 10% in relation to the control individual (Cepero *et al.*, 2001).

Table 1
Analysis of variance and multiple comparison test, for four response variables in sunflower (*Helianthus annuus* L.), subjected to ten levels of gamma radiation of ^{60}Co

Treatment Gy	PH cm	RL	RV cm^3	DB g
0	5.5 b	3.0 ab	2.0 a	4.0 b¶
100	8.0 a	3.9 a	3.0 a	5.3 a
200	7.0 a	3.4 a	2.0 a	5.0 a
300	6.5 b	3.2 ab	2.0 a	4.6 b
400	6.0 b	3.0 b	2.0 a	4.5 ab
500	5.6 b	3.0 b	2.0 a	4.5 ab
600	4.8 b	2.9 b	2.0 a	4.2 b
700	4.3 b	2.6 b	2.0 a	4.0 b
800	3.6 c	2.0 c	1.0 ab	3.6 bc
900	2.5 c	1.3 d	1.0 ab	3.3 bc
HSD	1.20**	0.4**	1.50*	0.50**
CV%	8.50	5.10	7.90	10.70

Values within column with the same literal, statistic mind are equal to P≥0.05 probability of error, according to Tukey; PH, plant height; RL, root lenght; RV, root volume; DB, dry biomass; HSD, honest significant difference; CV, coefficient of variation; *;**; n.s, significant to 0.05; 0.01 y not significant.

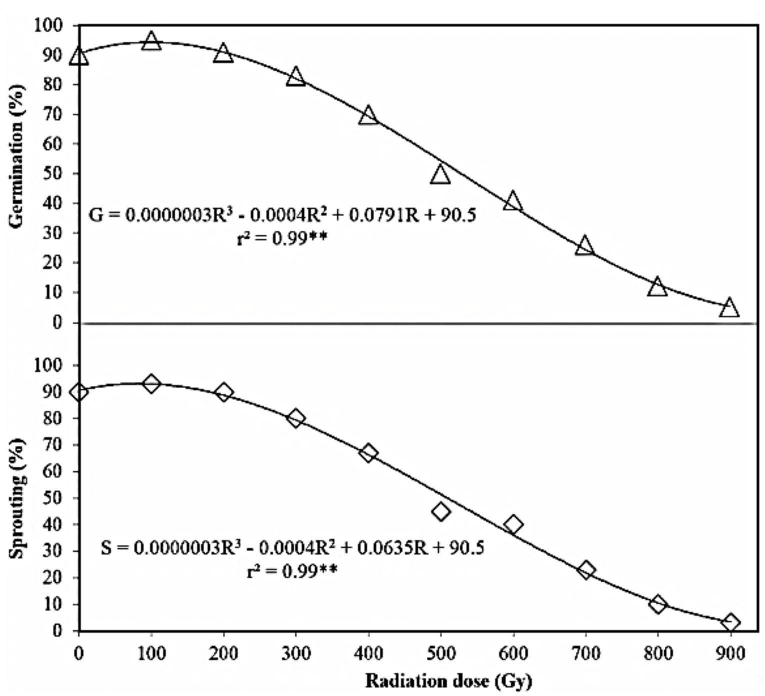

Figure 1. Percentage of germination and sprouting, of achenes of sunflower (*Helianthus annuus* L.), irradiated with gamma of ^{60}Co. Universidad Tecnologica de Tehuacan. 2017. G, germination; S, sprouting; *; **; n.s, significant to 0.05; 0.01 and not significant.

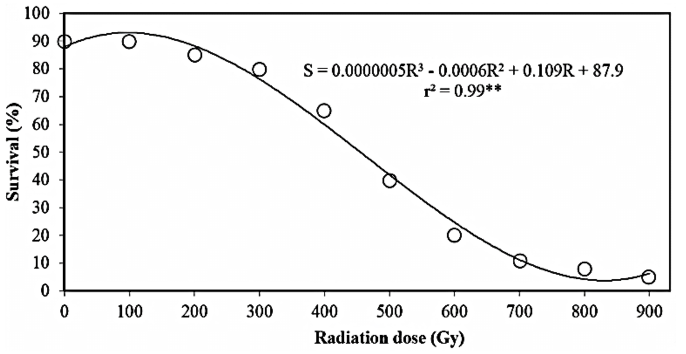

Figure 2. Survival percentage of sunflower seedlings (*Helianthus annuus* L.), subjected to ten levels of ^{60}Co gamma radiation. Universidad Tecnológica de Tehuacán. 2017. S, survival; R, radiation; **;*; n.s, significant al 0.01; 0.05 y not significant.

Percentage of survival

Survival percentage was a consequence of germination and sprouting together, having a third order polynomial behavior (Figure 2), and whose coefficient of determination was high 0.99 and highly significant. As the radiation dose increased from 200 to 900 Gy, the percentage of survival decreased, indicating that the sunflower is a sensitive species at high levels of radiation, thus in this way the application of 500 Gy decreases survival by 54.9% (Figure 3). This same response was found in Polianthes, where the percentage of survival, decreases, by increasing the radiation dose from 10 to 30 Gy, despite being a tuberous plant (Estrada *et al.*, 2011). In contrast Lemus *et al.* (2002), worked with two varieties of vigna bean (*Vigna unguiculata* L.), in similar ranges of irradiation, found that the application of 600 Gy, causes a drastic fall in the percentage of survival. This response to gamma irradiation was also studied by Ramírez *et al.* (2006), who when working with tomato seeds (*Lycopersicum esculentum* Mill.), mention that the first effects of gamma radiation, affect physiological processes such as germination and seedling growth, submitted to these agents and report, then in tomato, the germination begins to decrease in 500 Gy, checking what was found in this investigation.

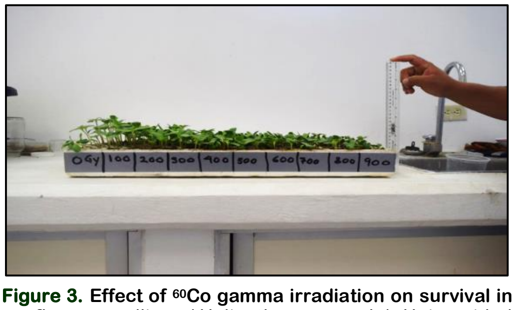

Figure 3. Effect of ^{60}Co gamma irradiation on survival in sunflower seedlings (*Helianthus annuus* L.). Universidad Tecnológica de Tehuacán. 2017.

4. Conclusions

The following conclusions were derived from the present investigation. Germination, sprouting as well as the percentage of survival in sunflower, are affected as the gamma radiation of ^{60}Co is increased from 300 to 900 Gy. Doses of 100 and 200 Gy have a stimulating effect on plant height and root length. The sunflower, is a species sensitive to gamma irradiation of ^{60}Co, for this reason this physical agent can be used to induce genetic variability in this crop and in future studies of genetic improvement. for future work, It is recommended not to use radiation greater than 500 Gy, because 50% of the population dies, therefore, superior radiations are not necessary for a work of genetic variability.

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References

Antúnez, O.O.M.; Cruz, I.S.; Sandoval, V.M.; Santacruz, V.A.; Mendoza, O.L.E.; de la Cruz, T.E.; Peña, L.A. 2017. Induced variability in physiological characters of *Physalis peruviana* L. by 60Co gamma rays applied to the seed. Revista Fitotecnia Mexicana 40(2): 211-218.

Barrios, B.M.; Buján, A.; Debelis, S.P.; Sokolowski, A.C.; Blasón, A.D.; Rodríguez, H.A.; López, S.C.; Grazia, J.; Mazo, C.R.; Gagey, M.C. 2014. Root Biomass/Total Ratio in Soybean (*Glycine max*) Under Two Tillage Systems. Terra Latinoamericana. 32: 221-230.

Cepero, L.; Mesa, R.A.; Lajonchere, G.; Prieto, M. 2001. Stimulation of the growth of *Leucaena leucocephala* cv. Cunningham with 60Co gamma rays. Pastos y Forrajes. 24: 235-240.

Cochran, G.W.; Cox, M.G. 2010. Experimental designs. Ed. Trillas. Mexico, D. F. 661 pp.

Bye, R.; Linares, E.; Lentz, D.L. 2009. Mexico: Center of origin of sunflower domestication. Revista Especializada en Ciencias Químico Biológicas 12(1): 5-12.

Díaz, L.E.; Morales, R.A.; Báez, R.I.; Vargas, R.E.J.; Hernández, H.P.; Olivar, H.A.; Rosales, M.B.E.; Loeza, C.J.M. 2017. Potential absorption of calcium in sunflower (*Helianthus annuus* L.) and its effect on pH and electrical conductivity. International Journal of Current Microbiology and Applied Sciences 6(6): 3299-3304.

Díaz, L.E.; Pichardo, R.J.C.; De la Cruz, T.E.; Norman, M.T.; Sandoval, R.F.; Vázquez, G.L.M. 2003. Induced variability in *Tigridia pavonia* (L. f.) D.C. var. Sandra by irradiation of bulbs with 60Co gamma rays. Revista Chapingo Serie Horticultura 9(2): 235-241.

Estrada, B.J.A.; Pedraza, S.M.E.; De la Cruz, T.E.; Martínez, P.A.; Sáenz, R.C.; Morales, G.J.L. 2011. Effect of gamma rays 60Co in tuberose (*Polianthes tuberosa* L.). Revista Mexicana de Ciencias Agrícolas 3: 445-458.

Fe, D.C.; Romero, M.; Ortíz, R.; Ponce, M. 2000. Radiosensitivity of soybeans to gamma rays 60Co. Cultivos Tropicales. 21(2): 43-47.

Gómez, L.; Aldaba, G.; Ibañez, M.; Aguilar, E. 2017. Development of advanced mutant lines of barley with higher mineral concentrations through radiation-induced mutagenesis in Peru. Peruvian Journal of Agronomy 1(1): 14-20.

Iglesias, A.L.G.; Sánchez, V.L.R.; Tivo, F.Y.; Luna, R.M.; Flores, E.N.; Noa, C.J.C.; Ruiz, B.C.; Moreno, M.J.L. 2010. Effect of gamma radiation on Abies religiosa (Kunth) Schltd. et Cham. Revista Chapingo Serie Ciencias Forestales y del Ambiente 16(1): 5-12.

Infante, G.S.; Zarate, D.L.G. 2005. Statistical methods: a multidisciplinary approach. Ed. Trillas. Mexico, D. F. 643 p.

Lemus, Y.; Mendez, N.J.R.; Cedeño, J.R.; Otahola, G. V. 2002. Radiosensibilidad de dos genotipos de frijol (*Vigna unguiculata* (L.) Walp.) a radiaciones gamma. Revista UDO Agricola 2(1): 22-28.

Loeza, C.J.M.; Díaz, O.M.T.; De la Cruz, T.E.; García, A.J.M.; Olivar, H.A., Vargas, R.E.J.; Reséndiz, M.R.C.; Díaz, L.E.; Morales, R.A. 2016. Genetic improvement of some attrinutes agronomic in roselle (MALVACEAE) with gamma radiation of 60Co. International journal of development research 6(7): 8295-8298.

Lyaz, S.; Naz, S. 2014. Effect of Gamma irradiation on morphological characteristics and isolation of curcuminoids and oleoresins of *Curcuma longa* L. Journal Animal Plant Science 24(5): 1396-1404.

Luévanos, E. M. P., Reyes, V. M. H., Villareal, Q. J. A. Y Rodríguez, H. R. 2010. Obtaining intergeneric hybrids *Helianthus annuus* L. x *Tithonia rotundifolia* and its morphological and molecular analysis. Acta Botánica Mexicana 90: 105-118.

Organización Internacional de Energía Atómica-OEIA. 2017. Manual of good practices for food irradiation. Colección de informes técnicos. No. 481. 91 p.

Ramírez, R.; Gonzalez, L.M.; Camejo, Y.; Zaldivar, N.; Fernandez, Y. 2006. Radiosensitivity study and selection of the range of X-ray stimulant doses in four tomato varieties (*Lycopersicon esculentum* Mill). Cultivos Tropicales 27(1): 63-67.

Salomón, D.J.L.; Gonzalez, C.M.C.; Castillo, Hernández, J.G.; Varela, N.M. 2017. Effect of gamma rays on the germination of botanical potato seed (*Solanum tuberosum* L.). Cultivos Tropicales 38(1): 89-91.

Steel, D.R.G.; Torrie, H.J. 1985. Bioestadística: principios y procedimientos. 2ª. Edición. Mc Graw-Hill. Bogotá, Colombia. 640 p.

Torretta, J.P.; Medan, D.; Roig Alsina, A.; Montaldo, N.H. 2010. Day visitors of the sunflower (*Heliantus annuus* L. Asterales: Asteraceae) in Argentina. Revista Sociedad Entomológica Argentina 69(2-1): 17-32.