THE CYTOGENETIC EFFECTS WITH LEAD NITROGENOUS TREATMENT AT *HELIANTHUS ANNUUS* L. FAVORIT AND PR64A83 CULTIVARS

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Abstract: The analysis of some cytogenetic parameters shows that the nitrogenous lead application on Favorit and PR64A83 cultivars of sunflower exercises a light mutagenic effect on the cells of the root apex. At same time an intensification of the frequency of the ana-telophases with aberration takes place, especially of those with bridges, with lagging chromosomes, but also of those with fragments, demonstrating the perturbatory action that this substance exercises on the division axle. The apparition of a great number of metaphases with displayed chromosomes proves the perturbation of well function to division axle, effect signalized especially at the Favorit cultivar.

INTRODUCTION

The lead nitrogenous is obtained through the action of an excess of diluted nitric acid on the metallic lead, oxyde or carbonated lead. The isomorphic nitrogenous lead crystallizes with that of calcium and strontium in the cubic system. The thermographic analysis showed that in the decomposition process resulted basics salts. The nitrogenous lead has a moleculary weight of 331.23 and a density of 4.53. It is found as incolore, cubic, monoclinic crystals. It is used as a caustic in the textile industry, as a source for other lead salts and explosive mixtures (Bălănescu G., 1964).

In the present paper we proposed to investigate the cytogenetic alterations which appeared after the nitrogenous lead application, in different dilutions, to two of the *Helianthus annuus* L cultivars: Favorit and PR64A83.

MATERIALS AND METHODS

The biological material is represented by sunflower seeds belonging to two hybrid early cultivars: Favorit and PR64A83 proceeded from the Botanical Garden in Iasi. The germination was ensured in Petri boxes with moistened filter paper at a temperature of 22 ± 2^0C. After germination when the little roots are between 10-15mm the treatment was made like this:

- the control variant –the seeds and the embryonic roots have been maintained in distilled water for 3 hours.
- the treatment variants were made by placing 25 germinated seeds in solutions of nitrogenous lead with the following concentrations: 0.10; 0.25 and 0.50. The dilutions were made in distilled water.

For removing the lead nitrogenous traces, the little roots were maintained for 2 hours in distilled water at the environment temperature. The settlement of the vegetal material was made in absolute ethylic alcohol: glacial acetic acid (3:1) for 20 hours. Until the moment of processing the little roots were kept in refrigerator, in an ethylic alcohol solution of 70%. The microscopic preparations were made using the Squash method (Cîmpeanu et al., 2002). For this the little roots were subdued to hydrolysis in 50% HCl (v:v) for 8 minutes and after this were coloured with 10% Carr solution for 24 hours.

For each variant five preparations were analysed and the photos were taken with the Nikon Eclipse 600 microscope, the 100x objective with immersion, with the aid of Nikon Cool Pix 950 digital camera.

RESULTS AND DISSCUSIONS

The mitotic index

The reactivity (as seen in figure 1) of the two varieties to the lead nitrogenous treatment is similar only at the minimal concentration (0.10%). At the PR64A83 cultivar the mitotic index increases proportionally to the increasing lead nitrogenous concentration, being two times greater at the 0.50% lead nitrogenous variant, in comparison to the control variant.

For the Favorit cultivar the mitotic index grows more significantly with small and medium concentrations, meanwhile at the maximal concentration this value is close to that of the control variant (figure 1).

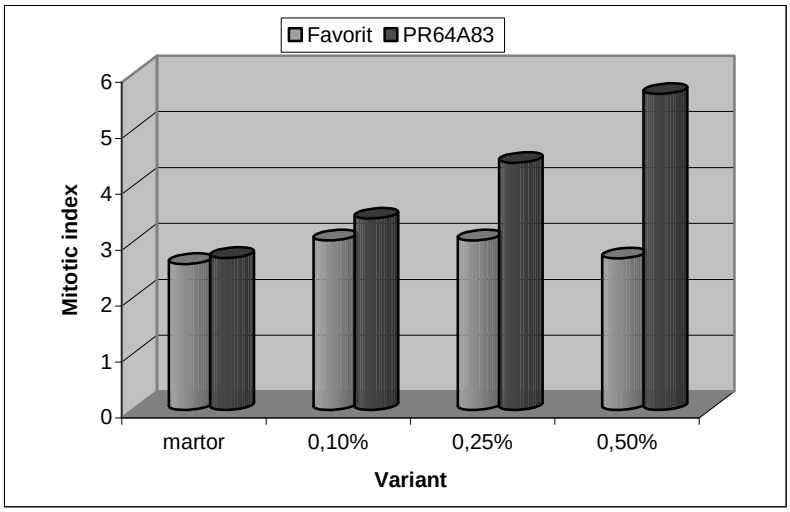

Figure 1. The mitotic index variation at the Favorit and PR64A83 cultivars after the lead nitrogenous treatment

The lead nitrogenous treatment application to the both cultivars determines the frequency of the cells that are in prophases to grow. At the PR64A83 cultivar prophases weight grows proportionally to the lead nitrogenous concentration, meanwhile at the Favorit cultivar , the treatment variants have resembling values, being between 2,06% (0,10% nitrogenous variant) and 2,19% (0,25% nitrogenous).

The percent of the cells that are in metaphases to both cultivars is similar to that of the control variant, but diametrally opposite at the treated variants. So, if at the Favorit cultivar the number of cells that are in metaphases decreases proportionally to the increasing lead nitrogenous concentration, at the PR64A84 cultivar, the metaphases frequency is superior to that of the control variant for all the treated variants (figure 2)

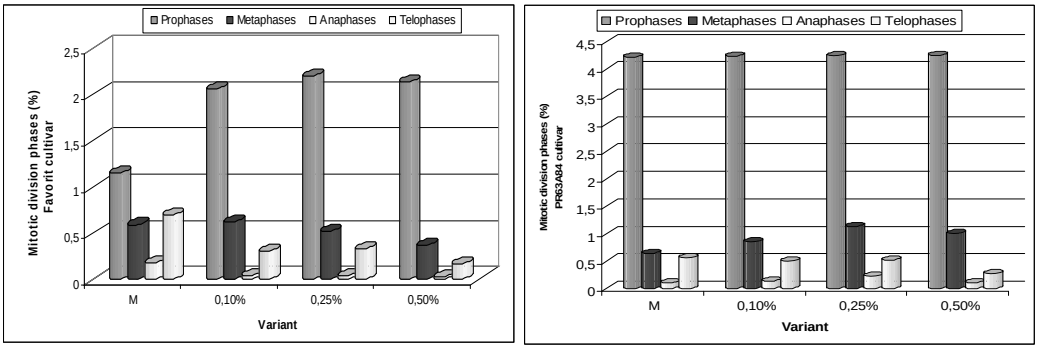

Figure 2.The frequency of cells that are in prophases at the Favorit and PR64A83 cultivars after lead nitrogenous treatment

A similar situation of that mentioned above is recorded for the cells that are in anaphases which, at the Favorit cultivar are in a much smaller weight at the treated variants, in comparison to the control variant. In the case of PR64A83 cultivar the frequency of cells that are in anaphases grows proportionally to the lead nitrogenous concentration, the only exception being recorded with the minimal nitrogenous dilution, where the value of this parameter is close to that of the control variant: 0.11% anaphases (0.50% nitrogenous lead), and 0.10% cells that are in anaphases (control variant).

The two studied cultivars have a similar behavior regarding the frequency of cells that are in telophases, meaning that with the increasing lead nitrogenous concentration a correspondent decreasing of the number of cells takes place, the lowest value being recorded at the maximal nitrogenous concentration (0.50%). At this variant the most important decrease of the number of telophases is recorded at the Favorit cultivar being four times smaller, in comparison to the control variant (from 0.70 to 0.17) which is in correlation with the decrease in the number of anaphases being presupposed that this substance has a direct action on the well function of the division axle and on the fragmoplast formation.

According to these results we can say that the frequency intensification of cells in divizion to treated variants is realised mostly through the accumulation of prophases and more less of cells that are in metaphases (PR64A83 cultivar).

The frequency of cells with aberrations

Another analysed cytogenetic parameter was represented by the establishment of frequency and of the main types of aberrations recorded in mitosis ana-telophases. Analysing figure 3, it results that the weight of these is variating from one dilution to another, but is almost identically at the both cultivars. Thus to the both cultivars are recorded two peaks, at minimal and maximal lead nitrogenous concentration .At the medium dilution (0.25%) the frequency of cells with aberrations is close to that recorded at control variant, this probably resulted from repairing processes which could have happened at macromolecule ADN level.

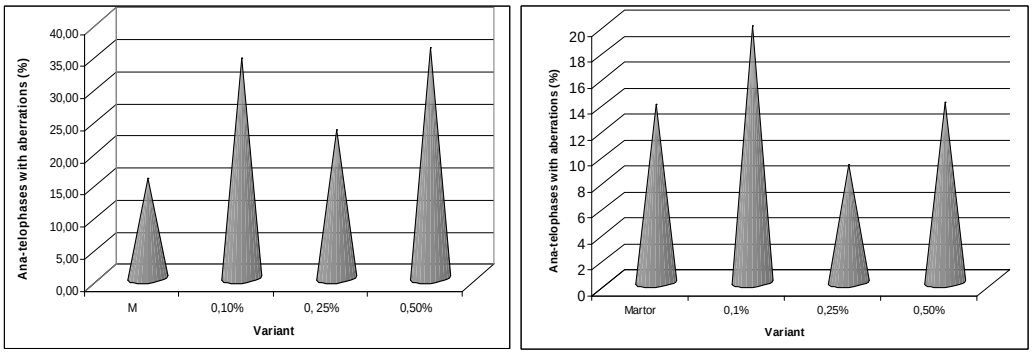

Figure 3. The frequency of ana-telophases with aberrations Favorit (left) and PR64A83 (right) cultivars

The signalised aberrations spectrum is large enough being represented by the ana-telophases with simple and multiple bridges, fragments, as well as a reduced number of complex aberrations(ana-telophases with bridges and fragments) At the two cultivars it was revealed one aberration category that is not found at the other cultivar. Thus the ana-telophases with lagging chromosomes are shown at Favorit cultivar and ana-telophases with bridges and fragments only at PR64A83 cultivar (figure 4).

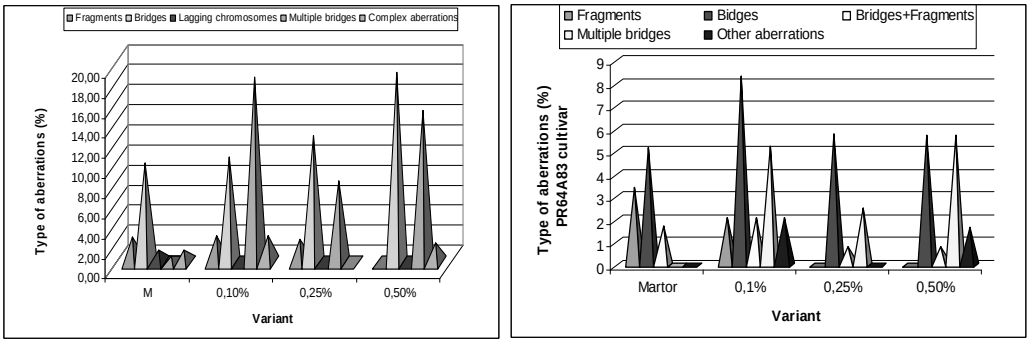

Figure 4.The main types of aberrations and the frequency of those at the Favorit (left) and PR64A83 (right) cultivars

At the both studied cultivars the biggest weight had the ana-telophases with bridges (simple and multiple), these being present in a pretty large number at the control variant as well (figure 5).

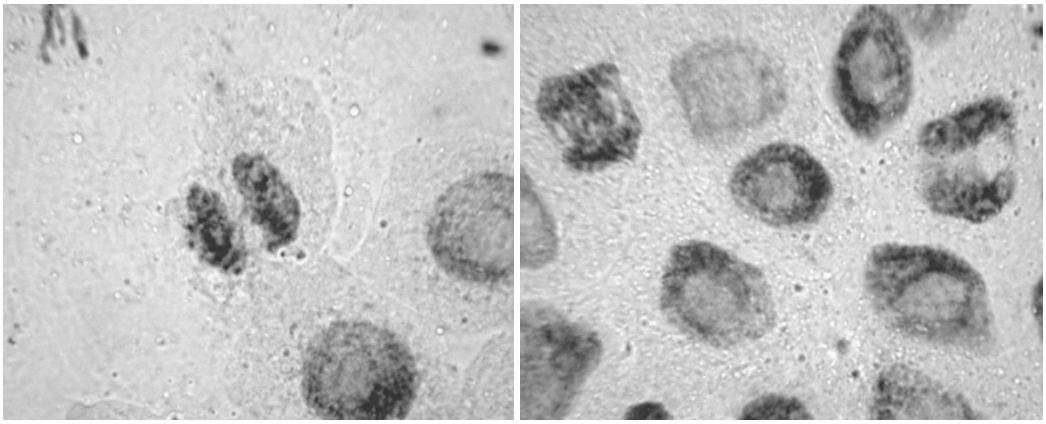

Figure 5. Simple bridge (left) and multiple bridge (right) at the Favorit cultivar, after lead nitrogenus treatment

In the case of Favorit cultivar there were shown metaphases with dispersed chromosomes in all cytoplasmatic mass(C-mitosis) similar to those obtained through colchicine treatments, from which results that lead nitrogenous exercises a strong effect on the well function of the division axle. The frequency of C-mitosis grows proportionally to the increasing lead nitrogenous concentration so that at the big concentrations (0.25% and 0.50%) this represents over 40% from the total analysed metaphases (figure 6 and 7).

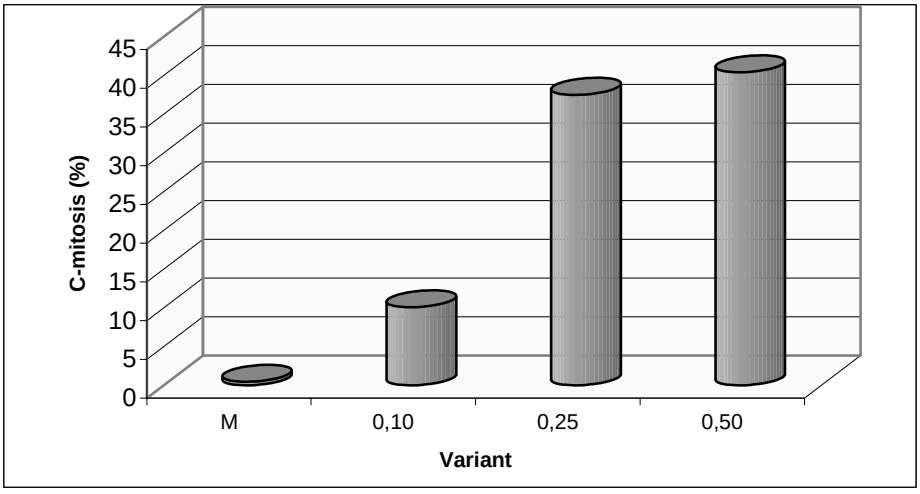

Figure 6. The C-mitosis frequency at the Favorit cultivar after lead nitrogenous treatment

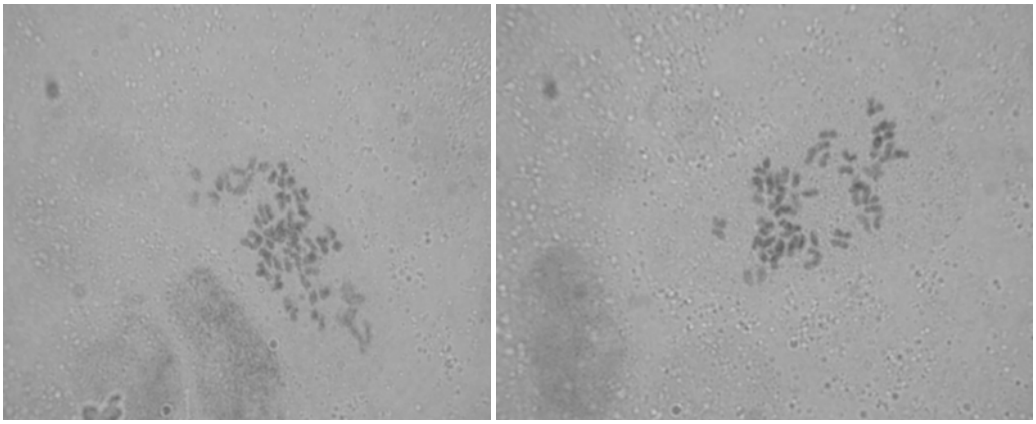

Figure 7. C-mitosis at the Favorit cultivar after lead nitrogenous treatment

CONCLUSIONS

The mitotic index, after applying lead nitrogenous treatment is superior at the treated variants in comparison to the control variant. The reactivity at this substance is varying from one cultivar to another, meaning that at the Favorit cultivar the big concentrations reduce the frequency of cells that are in division, while at the PR64A83 cultivar the value of this parameter grows propotionally to the lead nitrogenous concentration.

The biggest weight of cells with aberrations at both cultivars was recorded at the minimal lead nitrogenous concetration.

The aberrations spectrum in mitosis ana-telophases is large enough and is getting larger at the same time with the lead nitrogenous treatment application. The most important aberrations types were: bridges (simple and multiple), ana-telophases with lagging chromosomes, as well as a reduced number of complex aberrations. At the Favorit cultivar appear C-mitosis with a greater weight at greater lead nitrogenous concentrations, a fact that proves the colchicomimetic effect of this substance.

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