CYTOGENETICS EFFECTS INDUCED BY NITRATE OF LEAD ON MITOTIC DIVISION AT *ALLIUM CEPA* L.

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Key words: nitrate of lead, *Allium cepa* L., root meristem, cells in mitotic division, chromosomial aberrations.

Abstract: The paper presents the influence of nitrate of lead upon the mitotic division of *Allium cepa* L. The treatment with nitrate of lead has determined the lessening of the mitotic index and the chromosomial mutations. Also nitrate of lead determined in little proportion cells autopoliploid. The experiment prowed that nitrate of lead, known as a polluting agent has a mutagenic potential on the plants.

INTRODUCTION

It is known that the lead is polluting agent very toxic for plants and animals (Ciplea, Ciplea,1978; Heggestad,1968; Kihlman,1966)

At plants, action of the lead demonstrated on various chromosomial aberrations (Pădureanu, 2004; Pădureanu, 2004). Our investigations focused the determination of the mitotic index, the determination of the frequency of the types of chromosomial aberrations from metaphases and aberrant ana-telophases.

MATERIAL AND METHODS

The biological material used in the experiment, was represented by seeds of *Allium cepa* L., harvested from a local population cultivated at the Experimental Didactic Station “V. Adamachi” from the University of Agricultural Sciences and Veterinary Medicine, Iaşi.

The seeds were put to germination in lab conditions. When the roots reached 15 – 17 mm in length, they were treated with acetate of lead.

Nitrate of lead was used in the form of watery solutions in three concentrations: 5%, 1%, 0.1%.

The time of action of the respective solutions on the radicular meristems was differentiated as follows: 5% solutions acted for 48 hours, 24 hours, 4 hours, 2 hours; 1 % and 0.1% solutions acted for 4 hours and 2 hours.

Taking into account the concentration and the time of action of the solutions 8 variants have resulted.

Besides these eight experimental variants, there was also used a control plot and in this case no treatments were applied to the radicular meristems.

For further cytogenetic investigations, the treated and non/treated roots (control) were fixed in Carnoy fixing solution for 24 hours at 4°C then hydrolised with HCl and coloured with the basic colouring matter Carr.

The radicular meristem was displayed using squash technique.

15 preparations and 10 microscopical fields/preparation were examined for all the variants and control.

The microscopical examination was carried out using the optic microscope Nikon Eclipse 600.

The microphotographies were made with the camera from the endowment of the microscope.

RESULTS AND DISCUSSIONS

The analysis of the mitotic index

The first aspect investigated, correlated with the mutagenic capacity of the treatments by nitrate of lead on the onion it was represented by the effect of nitrate of lead on mitotic division’s stages.

The number of cells in division diminish correlated with the increase the concentrated and time of action of nitrate of lead (fig.1). The situation for each phase of mitotic division is represented in figures 2, 3, 4 and 5. The percentage of the cells in prophase is greater when the concentration is 5%, 2 hours, and other variants is small by comparison with control (fig.2).

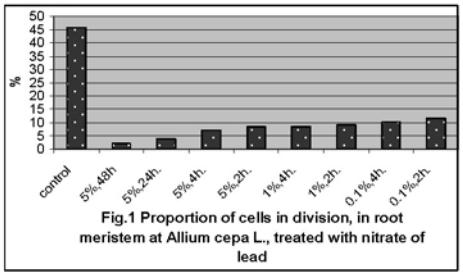

Fig.1 Proportion of cells in division, in root meristem at Allium cepa L., treated with nitrate of lead

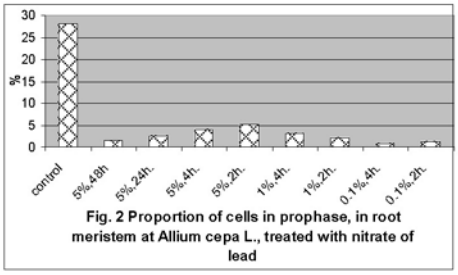

Fig. 2 Proportion of cells in prophase, in root meristem at Allium cepa L., treated with nitrate of lead

The high percentage of cells in metaphase at the variants with 1% concentration (2hours) and 0.1% (4 hours, 2 hours) is surprising (fig.3).

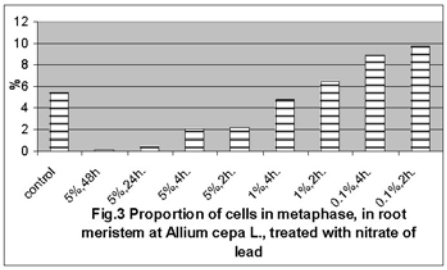

Fig.3 Proportion of cells in metaphase, in root meristem at Allium cepa L., treated with nitrate of lead

The proportion of cells in anaphase and in telophase remain much low by comparison with control in all cases (fig. 4 and 5).

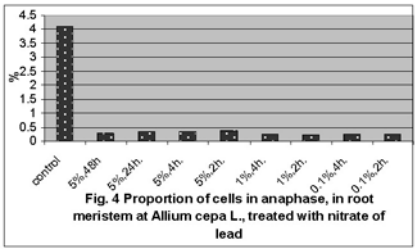

Fig. 4 Proportion of cells in anaphase, in root meristem at Allium cepa L., treated with nitrate of lead

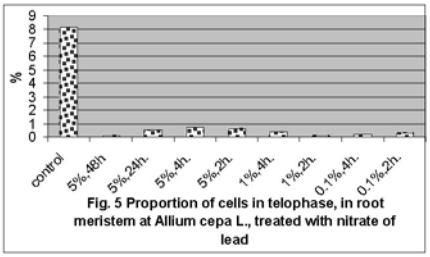

Fig. 5 Proportion of cells in telophase, in root meristem at Allium cepa L., treated with nitrate of lead

The analysis of the cells in aberrant metaphase and aberrant ana-telophase

In figure 6 is notice that the highest proportions of aberrant metaphases were induced by 1% and 5% concentrations. Aberrant ana-telophases were produced in high percentages(0.85%, 1.05%, 1.01%) at 5% concentration. On constate that at most cases is not never registered normal metaphases (5%: 48 hours, 24 hours; 1%: 4 hours, 2 hours; 0.1%: 4 hours) and not even normal ana-telophases. This aspect it might due on small number of mitogen cells of respectively variants.

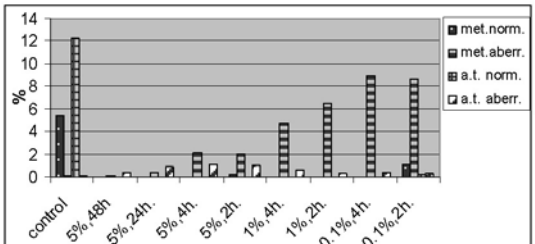

Fig. 6 Proportion of cells in normal and aberrant metaphases and ana-telophases in root meristem at Allium cepa L., treated with nitrate of lead

The analysis of the types of chromosomial aberrations

The proportion of the types of chromosomial aberrations induced by nitrate of lead on onion root meristem is graphically represented in figure 7.

This polluting agent induced the thick chromosomial bridges between nuclei by the variant with 5% concentration.

The chromosomial fragments were present at majority variants, excepting the variants with 5% concentration, 2 hours and 1% concentration, 2 hours.

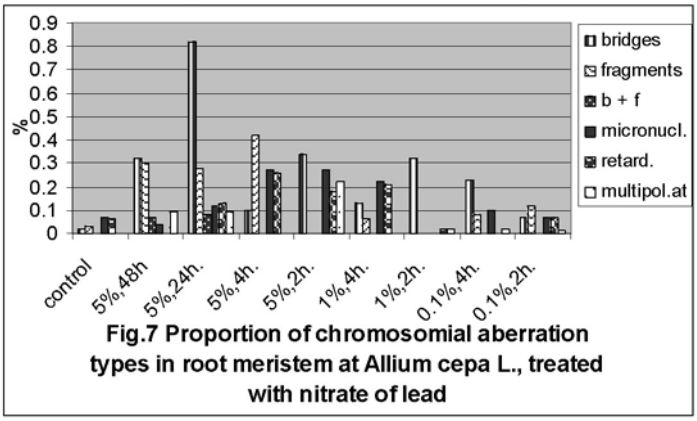

Fig.7 Proportion of chromosomial aberration types in root meristem at Allium cepa L., treated with nitrate of lead

The associations between bridges and fragments appeared only at variants with 5% concentration, 48 and 24 hours.

Micronuclei appeared in interphases, were registered in small percentage at majority variants excepting the variant with 1% concentration, 2 hours.

Retardatary chromosomes appeared at majority variants, excepting the variants with 5% concentration, 48 hours, and 0.1%, 4 hours.

Multipolar ana-telophases were registered at all three concentration of the polluting, but at certain times of action (5%: 4 hours; 1%: 4 hours) were not induced.

A special effect constated at the polluting consist in the presence of many metaphases with picnotic chromosomes sparse in all mixoplasma. Such aberrant metaphase appeared exclusiveness at majority variants, excepting the variants with 5% concentration, 2 hours and 0.1%, 2 hours, when was registered normal and aberrant metaphases.

As a result at this aberrant metaphases appeared c-anaphases characterized by monocromatidic chromosomes pairs wich not separated between that two cellular pols. The

denomination “c-anaphase” is explained by specific behaviour at the chromosome’s cells treated with colchicine (Natarajan, Ahnström, 1969).

Existence the metaphases with picnotic chromosomes and the c-anaphases might be explained owing to the whole inhibition the division shaft by influence the natrium of lead, feeld effect equally under treatments with colchicine.

As a results at this pseudo-anaphases (c-anaphases) appeared much pseudo-telophases characterized by mononucleate cells, wich big nucleus, whose size is double by comparision with diploide normal cells. Such the nuclei are autopoliploides. On surprised diverses stages by formation at this nuclei.

It is fitting much prudence when to affirm the presence of the autoploides nuclei, but existence of the nuclei with four nucleolus at onion is an strong argument in that sense.

By beside the types of aberrations presenced, on registered other atipic aspects concretized by very prolonged, bacilliform nuclei (21-62 μm), and picnotic, not-functionally nuclei.

It is known that cells at the caliptra has prolonged nuclei, but the nuclei named by us aberrant is not the same with those from caliptra, because they have a specific form, and the nucleolus inspire on aspect at bacillus with spores.

Different aspects of chromosomial aberrations induced by nitrate of lead are presented in figures 8-15.

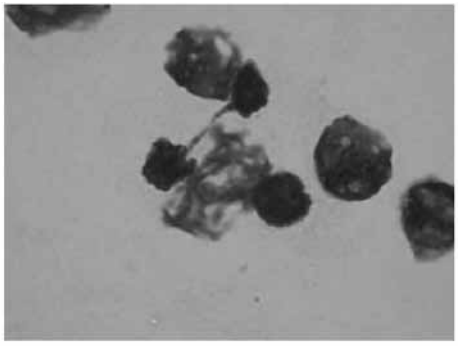

Fig. 8 Telophase with a bridge in root meristem at *Allium cepa* L. treated with nitrate of lead 5%, 24 hours (1000X)

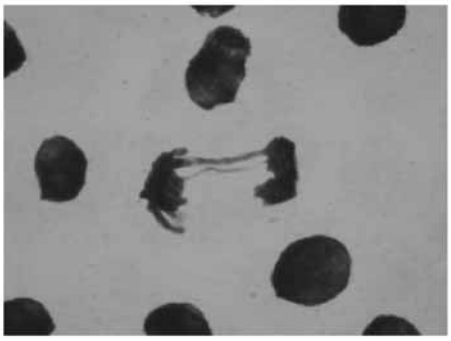

Fig. 9 Ana-telophase with two bridges in root meristem at *Allium cepa* L. treated with nitrate of lead 5%, 24

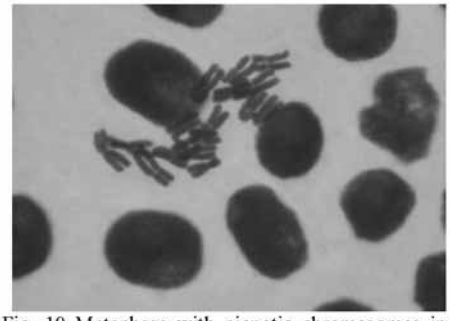

Fig. 10 Metaphase with picnotic chromosomes in root meristem at *Allium cepa* L., treated with nitrate of lead 5%, 4 hours (1000X)

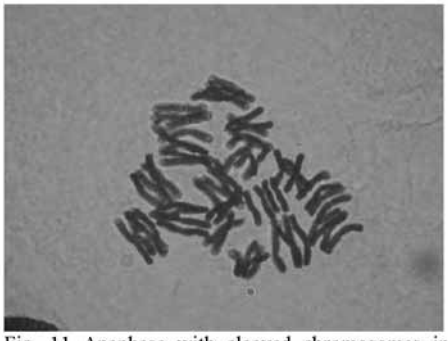

Fig. 11 Anaphase with cleaved chromosomes in root meristem at *Allium cepa* L., treated with nitrate of lead 5%, 2 hours (1000X)

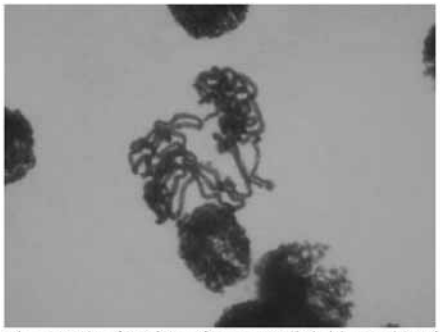

Fig. 12 The forming of a autopoliploide nucleus in root meristem at *Allium cepa* L., treated with nitrate of lead 5%, 4 hours (1000X)

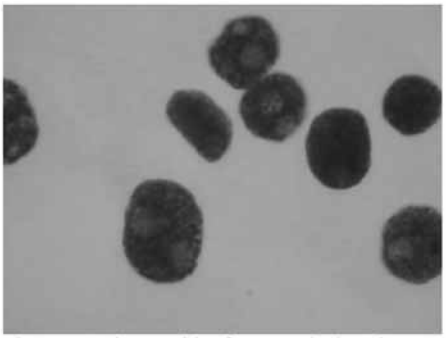

Fig.13 Nucleus with four nucleolus in root meristem at *Allium cepa* L., treated with nitrate of lead 0.1%, 4 hours (1000X)

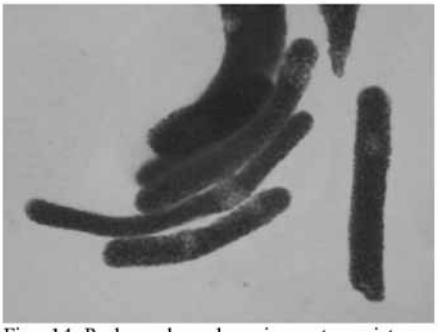

Fig. 14 Prolonged nucleus in root meristem at *Allium cepa* L., treated with nitrate of lead 5%, 4 hours (1000X)

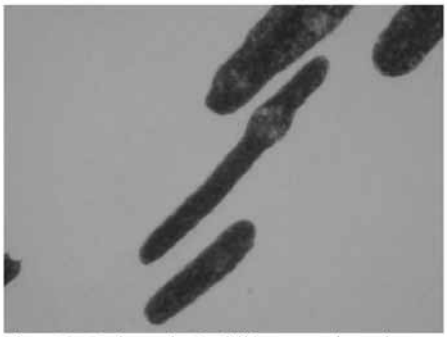

Fig. 15 Prolonged, bacilliform nucleus in root meristem at *Allium cepa* L., treated with nitrate of lead 0.1%, 4 hours (1000)

CONCLUSIONS

Nitrate of lead, known as a polluting agent has a strong inhibitory effect on mitotic division of *Allium cepa* L.

Nitrate of lead has a real mutagenic potential, proof is diverse chromosomial aberrations.

Nitrate of lead seems to be total inhibitory action at mitotic shaft, perturbeing the behaviour of the chromosomes in the same manner as the effect of colchicine.

Nuclei with four nucleolus are one argument that nitrate of lead has to be autopoliploid effect.

The side of habitual prolonged nuclei at the caliptra, appeared bacilliform nucleus and picnotic nuclei consequence of natrium of lead.

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