CYTOGENETIC EFFECTS INDUCED BY ACETATE OF LEAD ON MITOTIC DIVISION AT *ALLIUM CEPA* L.

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

Abstract: The paper presents the influence of acetate of lead on the mitotic division at *Allium cepa* L..The effect of acetate of lead on onion radicular meristems were expressed by chromosomial mutations, whose rate was differentiated depending on the concentration function and time of action of respective substance.The experiment pointed ont that the lead, wh is known as a polluting agent has a mutagenic potential on the plants.

INTRODUCTION

It is known that the lead and all its combination are polluting agents causing severe intoxications, especially when they are introduced into the digestive tract.

The dangerous dose for an aduet, ranges between 30 and 60 g acetate of lead (Nenițescu, 1985).

The slow intoxications causing severe disturbances with lead are called saturnism.

At plants, the lead proves its toxicity causing metabolic disturbances, even death (Rosen, 1964).

Some other polluting agents are also responsible for metabolic deficiency in plants, due to chromosomial reorganizations (Amoore, 1963; Fiskesjö, 1969; Heggestad, 1968; Ramel, 1969; Tamarin, 1990).

THE AIM OF INVESTIGATIONS

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.

Acetate 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 DISCUSIONS

Table1 presents the results obtained after treating onion radicular meristem with acetate of lead.

The analysis of the mitotic index

The inhibitory effect of this polluting agent is expressed by the reduced percentage of the cells in division in direct correlation with an increased concentration and time of action of the acetate of lead (fig. 1).

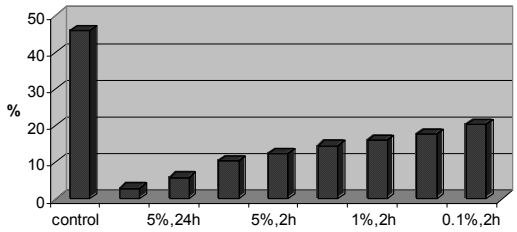

Fig. 1 Proportion of cells in division, in root meristem at Allium cepa L., treated with acetate of lead

The percentage of the cells in prophase is low v. s. control, ranging between 1.60-6.69%.

The number of the cells in metaphase is even smaller (0.17-4.40%), as well as the cells from anaphase (0.15-4.41%) and telophase (0.98-5.10%).

In all the phases of mitosis, the percentage of the cells in division was in inverse proportion to the increased concentration and time of action of the polluting agent.

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

Acetate of lead induced between 0.10 and 0.56% aberrant metaphases v. s. control, in which only 0.09% cells in aberrant metaphases were recorded. The variant with 5% concentration and time of action of 4 hours was the most efficient in inducing aberrant metaphases (fig. 2).

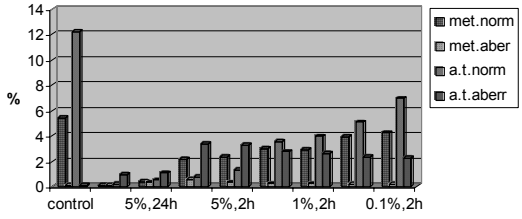

Fig. 2 Proportion of cells in normal and aberrant metaphases and ana-telophases in root meristem at Allium cepa L.treated with acetate of lead

The proportion of the cells in aberrant ana-telophase registered supraunitary percentages in almost all variants (0.94-3.36%), while control induced only 0.08% aberrant ana-telophases . The maximum percentage of the cells in aberrant ana-telophase was induced by the variants with 5% concentration and time of action of 4 hours and 2 hours (fig. 2).

The analysis of the types of chromosomial aberrations

The proportion of the types of chromosomial aberrations induced by acetate of lead on onion root meristem is graphically represented in figure 3.

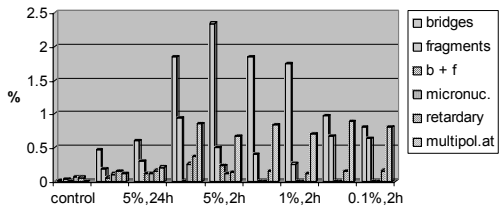

Fig. 3 Proportion of chromosomial aberration types in root meristem at Allium cepa L., treated with acetate of lead

The chromosomial bridges in aberrant ana-telophases were induced in maximum proportion (2.34%) by the variant with 5% concentration and time of action of 2 hours.

The chromosomial fragments met in metaphase and aberrant ana-telophase were present in subunitary percentage in all variants. The variant with 5% concentration, 4 hours of action, induced the maximum rate (0.94%) of fragments.

The associations between bridges and fragments were met only in variants with 5% concentration, time of action 48 hours, 24 hours and 2 hours. In all these cases, the values were subunitary (0.06-0.23%)

Micronuclei were induced only by the variants with 5% concentration, time of action 48 hours, 24 hours, 4 hours and 2 hours.

Retardatary chromosomes met in metaphases and aberrant ana-telophases were present in all variants, in subunitary proportions (0.12-0.37%), the maximum proportion being produced by the variant with 5% concentration, 4 hours.

Multipolar ana-telophases were registered in all variants, in subunitary proportions (0.12-0.89%). The mast efficient variants which induced high proportions of mulipolar ana-telophases were those with 5% concentration, 4 hours, 1%, 4 hours, 0.1%, 4 hours and 0.1%, 2 hours.

Chromosomial aberrations spontaneously produced in the control plot were: bridges (0.02%), fragments (0.03%), micronuclei (0.07%), retardatary chromosomes (0.06%).

Besides the types of chromosomial aberrations, acetate of lead has induced the formation of long nuclei and picnotic nuclei in proportion dependent directly proportional on concentration and time of agent action, in all the variants under study (5.7-12.11%).

Different aspects of chromosomial aberrations induced by acetate of lead are presented in figures 4-7.

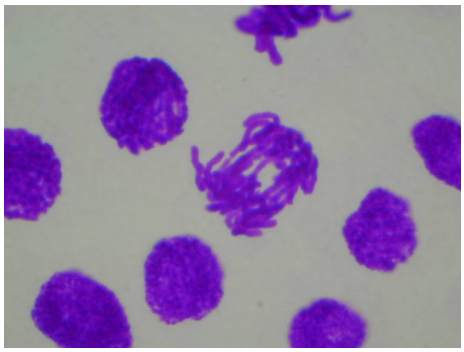

Fig. 4 Ana-telophase with multiple bridges in root meristem at *Allium cepa* L. treated with acetate of lead, 5%, 2 hours (1000 X)

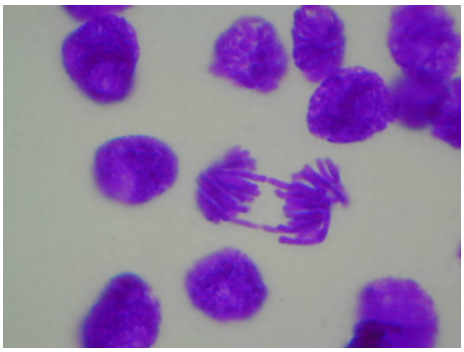

Fig. 5 Ana-telopfase with two bridges in root meristem at *Allium cepa* L. treated with acetate of lead, 0.1%, 4 hours (1000 X)

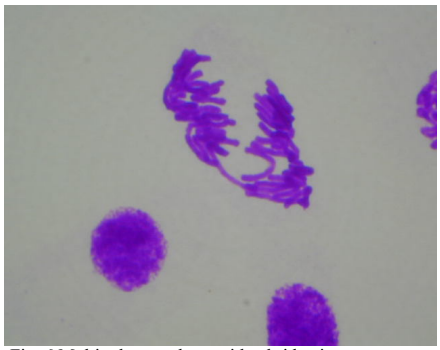

Fig. 6 Multipolar anaphase with a bridge in root meristem at *Allium cepa* L., treated with acetate of lead 5%, 4 hours (1000 X)

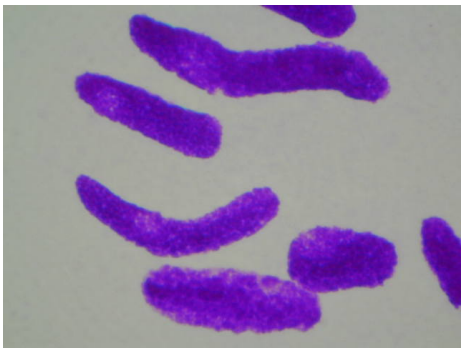

Fig. 7 Long nuclei in root meristem at *Allium cepa* L., treated with acetate of lead, 5%, 2 hours (1000 X)

CONCLUSIONS

Acetate of lead, known as a polluting agent has a strong inhibitory effect on mitotic division of *Allium cepa* L. The proportion of the cells in division is in inverse correlation to the increase of concentration and the time of action of the agent.

In each phase of mitotic division, the cells reacted differently to the action of the polluting agent.

Acetate of lead has a real mutagenic potential; this fact being confirmed by the aberrations induced on mitotic chromosomes in *Allium cepa* L. It acted at chromatidic level causing the occurrence of the fragments and bridges; at centromere level, whose activity it disturbes, acetate of lead determines the occurrence of retardatary chromosomes, which in the end will be changed into micronuclei acting at the level of the dividing spindle, it results spindles with 3, 4 or even more poles, determining the

appearance of multipolar ana-telophases, when the cromatides distribution among daughter cells is not equal.

Long nuclei and picnotic nuclei represents a characteristic of the acetate of lead effect.

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Table 1

Distribution of cells in different phases of mitotic division in root meristem at *Allium cepa* L. after treatment with acetate of lead.

Concentration	Time of action	Total analised cells	Cells in interphase		Cells in division		Cells in prophase		Cells in metaphase		Cells in anaphase		Cells in telophase	
			No.	%	No.	%	No.	%	No.	%	No.	%	No.	%
Control	0	11500	6220	54.09	5280	45.91	324	28.17	63	5.48	47	4.09	94	8.17
5%	48	8100	7865	97.10	235	2.90	130	1.60	14	0.17	12	0.15	79	0.98
5%	24	8023	7551	94.12	472	5.88	290	3.61	55	0.69	45	0.56	82	1.02
5%	4	8070	7240	89.71	830	10.29	280	3.47	220	2.73	270	3.35	60	0.74
5%	2	8560	7490	87.50	1070	12.50	448	5.23	229	2.68	296	3.46	97	1.13
1%	4	8004	6840	85.46	1164	14.54	402	5.02	258	3.22	300	3.75	204	2.55
1%	2	8848	7432	84.00	1416	16.00	560	6.33	275	3.11	316	3.57	265	3.00
0.1%	4	7938	6525	82.20	1413	17.80	494	6.22	327	4.12	321	4.05	271	3.41
0.1%	2	8001	6377	79.70	1624	20.30	535	6.69	352	4.40	329	4.11	408	5.10