CYTOGENETIC EFFECTS INDUCED BY HEXANITROGEN-COBALTIAT III OF SODIUM ON MITOTIC DIVISION AT *TRITICUM AESTIVUM* L.

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The paper presents the influence of hexanitrogen-cobaltiat III of sodium upon the cellular division at Triticum aestivum L. The treatment with hexanitrogen-cobaltiat III of sodium in three concentrations: 5%, 1%, 0.1% for 4 hours and 2 hours of on wheat radicular meristems were expressed by chromosomial mutations, especially in ana-telophase: chromosomal bridges, little chromosomal fragments and retardatary chromosomes, multi-polar ana-telophases, many micronuclei, whose rate was differentiated depending on the concentration function and time of action of respective chemical agent. Frequently chromosomial aberrations induced by hexanitrogen-cobaltiat III of sodium are bridges very thick who determine citomixia phenomenon. Also, hexanitrogen-cobaltiat III of sodium induced many micronuclei and picnotic nuclei non-functional. Hexanitrogen-cobaltiat III of sodium has a strong inhibitory effect on mitotic division of Triticum aestivum L. Particularly metaphase and anaphase are inhibition. The experiment proved that hexanitrogen-cobaltiat III of sodium, known as a polluting agent has a mutagenic potential on the plants.

Keywords*: root meristem, cells in mitotic division, chromosomial aberrations.*

Cobalt plays an important part in producing red globules in animals because of situated in the centre of molecules of B12 vitamin. In plants, cobalt as microelement, has a good effect on their life. Cobalt salts, such as hexanitrogen – cobaltiat III of sodium are toxic for plant metabolism [1, 3, 4].

Cobalt bio-accumulations was a study object for amphipod *Hyalella azteca* in Burlington City tap (Ontario Lake) water [5].

At plants, action of the polluting (for example salts cobalt etc.) demonstrated on various chromosomial aberrations [2, 3, 4]. 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 METHOD

The biological material used in the experiment, was represented by seeds of *Triticum aestivum* L., *Rubin* variety.

The seeds were put to germination in lab conditions. When the roots reached 15 – 17 mm in length, they were treated with hexanitrogen-cobaltiat III of sodium.

Hexanitrogen-cobaltiat III of sodium 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%, 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 6 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 Hund Wetzlar. The microphotographies were made with the camera from the endowment of the microscope.

RESULTS AND DISCUSSIONS

Analysis of the mitotic index

The treatments made with hexanitrogen-cobaltiat III of sodium have induced quite a strong inhibition upon the mitotic division *(fig. 1, 2).*

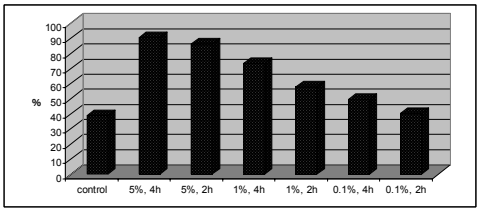

Fig. 1. **Proportion of cells in interphase**

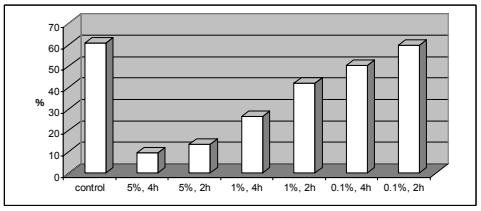

Fig. 2. **Proportion of cells in division**

In prophase, the cells percentage is much lower in variant with 5% concentrations and 1% (7.54–33.51% cells) if compared to the control (54.22% cells) *(fig. 3).*

In metaphase, in variant with 5% concentration (4 and 2 hours) and 1% (4 hours) on constate very small the proportion of cells by comparison with the control *(fig. 4).*

In anaphase the cells in variant with 5% concentration are to be found even in a more reduced number (0.30-0.33%) if compared to the control *(fig. 5).*

In telophase, in variant with 1% concentration (2 hours) and 0.1% (4 and 2 hours), the proportion of cells exceed the control *(fig. 6).*

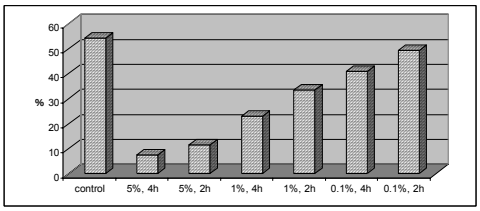

Fig. 3. **Proportion of cells in prophase**

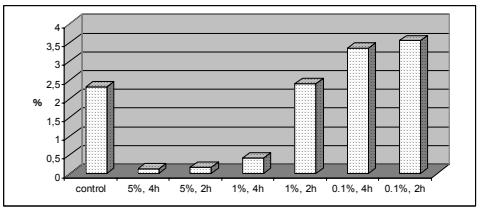

Fig. 4. **Proportion of cells in metaphase**

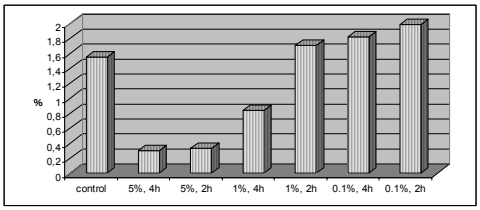

Fig. 5. **Proportion of cells in anaphase**

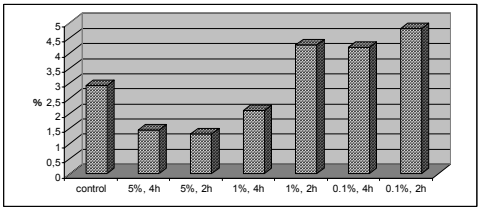

Fig. 6. **Proportion of cells in telophase**

Analysis of cells proportion in aberrant metaphase and ana-telophase

The mutagen effect of the polluant used in this experiment had been demonstrated by aberrant metaphases (0.08-0.27%) and aberrant ana-telophases (0.28-2.38%) inducement *(fig. 7, 8).* Aberrant metaphases consist in picnotic chromosomes.

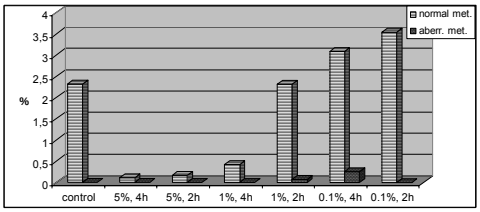

Fig. 7. **Proportion of cells in normal and aberrant metaphase**

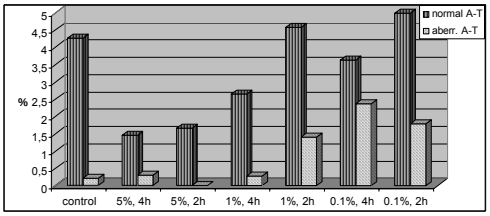

Fig. 8. **Proportion of cells in normal and aberrant ana-telophases**

Analysis of chromosomal aberration types

Figure 9 represents the proportion of aberration types in the wheat's meristem treated with hexanitrogen-cobaltiat III of sodium.

The chromosomal bridges from ana-telophases appeared in four variants, in over-unitary proportion in case of 0.1% concentration, (4 and 2 hours) and in sub-unitary proportion in variants with 5% concentration, 4 hours and 1%, 2 hours. The bridges were various types: entire, ragged, single, multiple, thick, thin, indifferent by variant. To predominate the bridges very thick who determine citomixia phenomenon, indifferent by variant.

The chromosomal fragments from ana-telophases appeared in case of 5% concentration, 4 hours and 1% concentration, 2 hours only, in sub-unitary proportion.

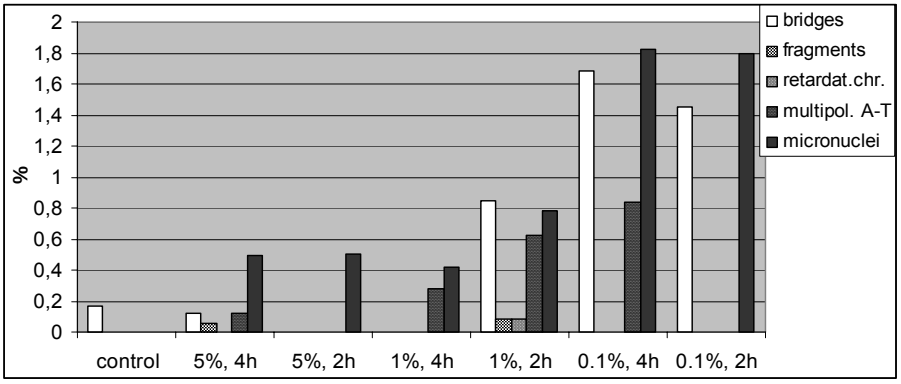

Fig. 9. **Proportion of chromosomial aberration types**

Retardatary chromosomes appeared in case of 1% concentration 2 hours only.

Multi-polar ana-telophases had been produced only at the variants with 5% concentration, 4 hours, 1% (4 and 2 hours) and 0.1%, 4 hours.

Micronuclei from interphase have been produced to all variants. Micronuclei appeared in metaphase in variant with 0.1% concentration, 4 hours.

Along with the presented chromosomal aberrations, the pollutive agent used in the experiment has also induced the formation of picnotic nuclei in over-unitary proportions at variant 5% concentration, 4 hours and sub-unitary proportion in variants 5%, 2 hours and 1%, 4 and 2 hours, in connection with direct proportionality with both polluant's concentration and time of action *(fig. 10).*

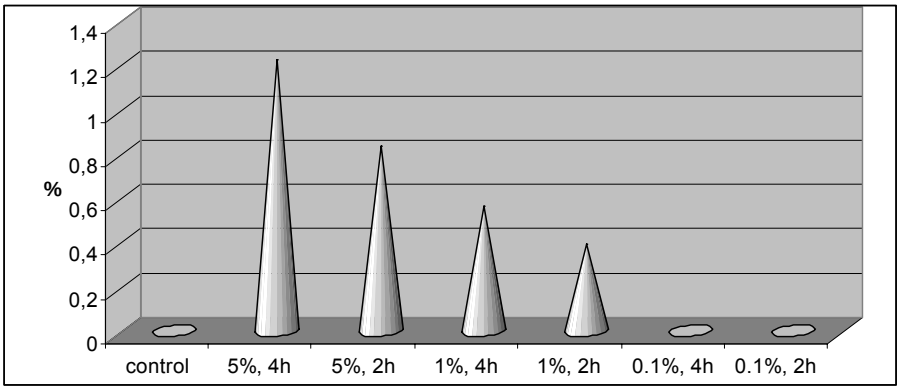

Fig. 10. Proportion of picnotic nuclei in root meristem at wheat, treated with hexanitrogen-cobaltiat III of sodium

Chromosomial aberrations spontaneously produced in the control plot were bridges (0.17%).

The limit differences of effects of the pollutive agent is represented in *table 1*.

Table 1

The influence of hexanitrogen-cobaltiat III of sodium upon mitotic division in root meristems at *Triticum aestivum* L.

Variant	Aberrant metaphases		Aberrant ana-telophases	
	Average value (%)	Significance of difference	Average value (%)	Significance of difference
Control	0	-	0.2	-
5%, 4 hours	0	-	0.30	-
5%, 2 hours	0	-	0	-
1%, 4 hours	0	-	0.28	-
1%, 2 hours	0.08	***	1.40	***
0.1%, 4 hours	0.27	***	2.38	***
0.1%, 2 hours	0	-	1.80	***
DL 5% = 0.018 DL 1% = 0.026 DL 0.1% = 0.036			DL 5% = 0.150 DL 1% = 0.210 DL 0.1% = 0.298	

Different aspects of chromosomial aberrations induced by hexanitrogen-cobaltiat III of sodium are presented in *figures 11 – 16.*

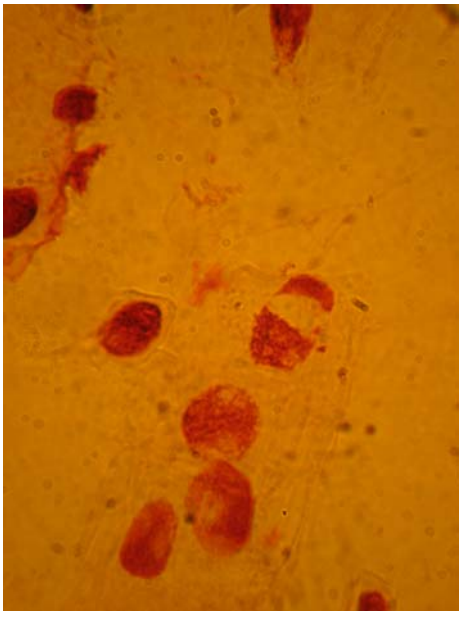

Fig. 11. **Telophase with a two ragged bridges in root meristem at wheat treated with hexanitrogen-cobaltiat III of sodium, 5%, 4 hours (100X)**

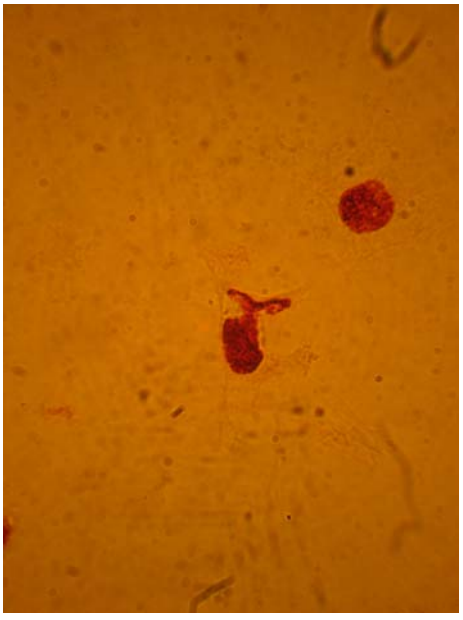

Fig. 12. **Telophase with a thick bridge in root meristem at wheat treated with hexanitrogen-cobaltiat III of sodium, 5%, 4 hours (100X)**

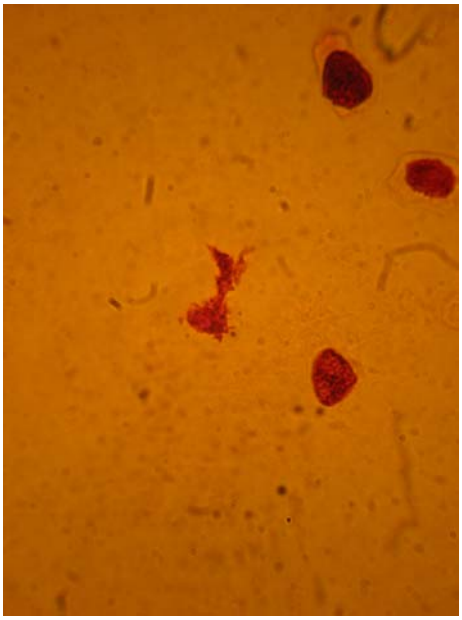

Fig. 13. **Telophase with a thick bridge in root meristem at wheat treated with hexanitrogen-cobaltiat III of sodium, 1%, 2 hours (100X)**

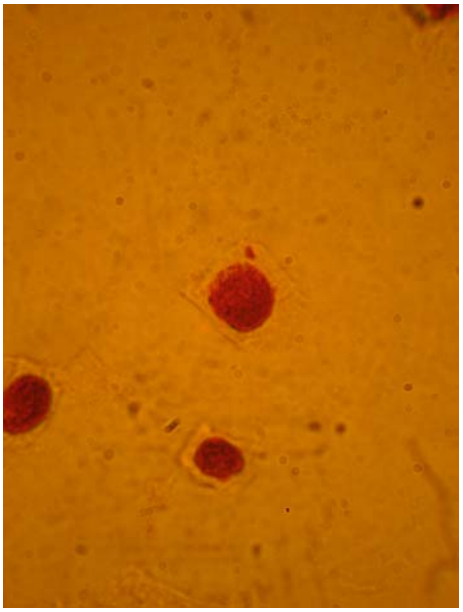

Fig. 14. **Interphase with micronucleus in root meristem at wheat treated with hexanitrogen-cobaltiat III of sodium, 5%, 2 hours (100X)**

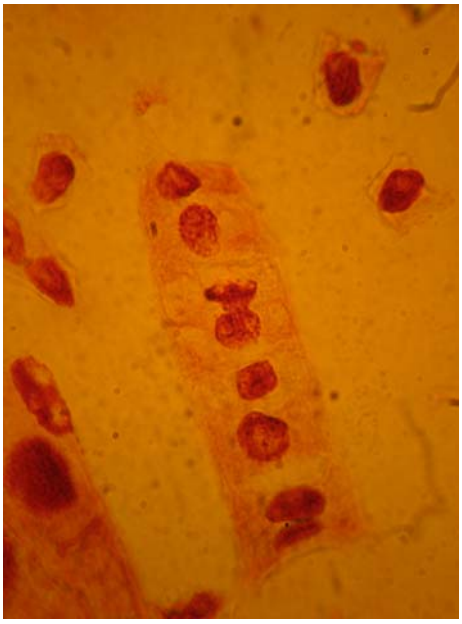

Fig. 15. **Telophase with a thick bridge in root meristem at wheat treated with hexanitrogen-cobaltiat III of sodium, 1%, 2 hours (100X)**

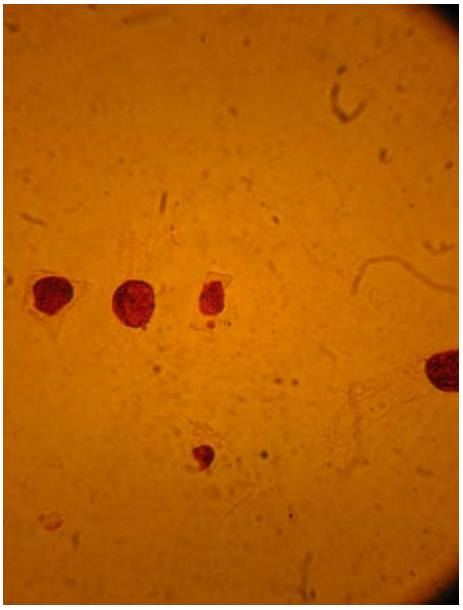

Fig. 16. **Interphase with micronucleus in root meristem at wheat treated with hexanitrogen-cobaltiat III of sodium, 0.1%, 2 hours (100X)**

CONCLUSIONS

1. Hexanitrogen-cobaltiat III of sodium, known as a polluting agent has a strong inhibitory effect on mitotic division of *Triticum aestivum* L.

2. The cells reacted differently in each phase of mitotic division to the action of the polluting agent. Particularly metaphase and anaphase are inhibition.

3. Hexanitrogen-cobaltiat III of sodium has a real mutagenic potential, confirmed by diversity chromosomial aberrations induced: chromosomal bridges thick particularly, little chromosomal fragments and retardatary chromosomes, multi-polar ana-telophases, many micronuclei.

4. Picnotic nuclei non-functional represent a characteristics of the hexanitrogen-cobaltiat III of sodium effect.

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