EFFECT OF 2,4-DINITROPHENOL AND POTASSIUM IODATE ON WHEAT GERMINATION

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Abstract - The dinitrophenol is an agent which enters frequently in the pesticides’ component, used largely in the practice of maintaining the agricultural cultures and management. The toxicity mechanism of these pesticides, acknowledged metabolic modulators, was discussed strongly correlated with the germinative faculty of wheat seeds, the Secuieni genre, but also with the level of photosynthetic and carotenoids pigments, respectively, the enzymes’ activity of oxidative stress (the peroxidase, the superoxide-dismutase and the catalase). The analyze of experimental results signalize the negative impact of dinitrophenol, but also of KIO₃ (used like chemical agent of comparison for testing the toxicity degree of dinitrophenol) on seeds, respectively on Triticum aestivum seedlings, using like reference swatches, plots treated with distilled water.

Key words - wheat, dinitrophenol, KIO₃, germination, chlorophyllien and carotenoid pigments, oxidative stress enzymes

I. INTRODUCTION

The cereals represent the most important foodstuff for human being very rich in proteins, lipids and glucids, totally 50-55% from the consumed calories in the whole world derived from cereals. No food doesn’t satisfy so economically the requirements in nutritive and active principles like the bread from wheat (1; 2). The wheat culture knew a large amplitude on worldwide and along with this were developed, in the last years, new techniques and practices of maintaining the agricultural lands (3) which lead to using, on more largely scale of different chemical substances (4) of organic fertilizer type and inorganic or of pesticides type with the aim of increasing the wheat production or of forbidding the diseases’ apparition and of specificpests (5; 6).

Besides, the agriculture is, in a way, an intervention against the nature, which, when it is practiced irrational, leads to the fertility’s decreasing, to biocenoses and ecosystems’ modification, this intervention of the human becoming sometimes extremely aggressive.

The literature data highlights the fact that, besides the benefic effect that these substances have on the growing and development of plants and implicit on the agricultural production (7 - 10), their biggest majority present also more negative effects than positive due, mainly, to their log remanence (some of them persist in the soil lots of years from their apply), the soil actioning like a receiver and reservoir for pesticides, from where, either are dispersed in the environment, either translocated in plants and, through the agency of these ones, in human and animal organism (11 - 13).

It is known the high toxicity of dinitrophenol and its derivates (14), substances that are refund in the pesticides’ component (15 - 17), producing disequilibrium of the metabolic reactions with repercussions at the level of different tissues and organs, translated through the generation of the reactive oxygen species (ROS). Alongside to other biotic and abiotic factors like the hypoxia, the light, the drought, the high salt, the col, the metallic ions, the xenobiotics, the toxins, the reoxygenation after anoxia, the experimental manipulation, the pathogen infections and the organs’ aging from plants, the oxidative stress, like a glitch of the equilibrium between the prooxidative reactions and those antioxidant, can appear under the pollutants’ influence of dinitrophenol type (18). To counteract the oxidative stress, the plants developed itself in time strategies of intracellular defense which are represented by the enzymatic antioxidant systems (the catalase, the peroxidase, the superoxide-dismutase, the ascorbat-oxidase, the glutation-reductase and the polyphenol-oxidase) and non-enzymatic (the ascorbic acid, the α-tocoferole, the caroten, the theopilphenols, the flavons) (19 - 21).

This study aim to evaluate the influence that the dinitrophenol and the potassium iodate exert, on the one hand on wheat seeds’ germination and on the growing and the development of the seedlings, and on the other hand on the level of oxidative stress through view of the antioxidant enzymes’ activity of catalase, peroxidase and superoxide-dismutase, in parallel with the concentration of chlorophyllien and carotenoid pigments.

II. MATERIAL AND METHODS

It was worked on wheat caryopses from 2013 crop, derived from the Secuieni Research and Agricultural Development Resort, which were put to germination in lab conditions, in Petri boxes, after a preliminary treatment, for an hour, with 2,4 - dinitrophenol (DNP) and potassium iodate (KIO₃) solutions in 10⁻³, 10⁻⁴ and 10⁻⁵M concentrations.
The samples were daily watered with a constant volume of distilled water, for ten days, after which it was evaluated the physiological and biochemical analyze (through the determination of chlorophyll and carotenoids concentration, respectively to the activity of some enzymes of oxidative stress and to the concentration of total soluble proteins) of seedlings.

Hereby, for determining the content of assimilatory pigments and provitamin A, after the pigments’ extraction with acetone, was used the spectrophotometric method, the reading of samples being done at 472, 645 and 663 nm wavelength (22). For the dosage of peroxidase’s activity (POX, EC 1.11.1.X.) was used the Gudkova and Degtiari method, 1968 (23) which is based on the measuring of color’s intensity of o-dianisidine oxidation product with peroxide’s help, for catalase’s activity (CAT, EC 1.11.1.6) was used the Sinha method, 1972 cited by Cojocaru, 2009 (23), while the superoxid-dismutase’s activity (SOD, EC 1.15.1.1) was evaluated through Winterbourn method et al. (1975) adapted by Cojocaru, 2009 (23), the method consisting in the enzyme’s capacity of inhibiting the Nitro Blue Tetrazolium reduction by the superoxid radicals generated in the reaction environment through riboflavine’s photoreduction. The concentration of total soluble proteins, needful for calculating the enzymes’ specific activity, was dosed through Bradford reactive method (24).

For each type of treatment in part were realized three parallel determinations, the obtained experimental results being statistically processed with Microsoft’s Excel (Descriptive statistics, t test-Student) help, being considered significant at p’ values less than 0.05.

We mention that all the used reactives were of analytic purity, the solutions being prepared in peroxide obtained in a MilliQ (18,2 Ω) system, and the UV - VIS measurements were realized with a ShimadzuUV-VIS 1700 spectrophotometer, in quartz tanks of 1 cm from a reactives’ control.

III. RESULTS AND DISCUSSIONS

To obtain some maximized wheat productions, in the last years was recourse to the using more frequently of different pesticides, although it is well known the harmful effect of these ones on human organism and animal, from this reason trying the limitation of using these substances in the near future, even to the detriment of agricultural production diminution (25; 10).

In a first series of experiments we had an eye to the influence that DNP and KIO₃ exert in different concentrations (10⁻³, 10⁻⁴ and 10⁻⁵ M) on germination degree of *Triticum aestivum* seeds, Secuieni genre. With this purpose in view, the pretreated seeds with the respective solutions were layed out in Petri boxes padded with paper filter and watered daily with a constant volume of distilled water. After ten days from the germinative process’ debut we recourse to count the seedlings from each box, calculating the germinative faculty and the number of non-viable caryopses, respectively sprouted, but not grew. Subsequent, the seedlings were subjected to metric and gravimetric study through the measurement and weighting of each seedling in part.

As it can be observed from Table I-II the apply of DNP and KIO₃ solutions influenced significantly the rate of wheat caryopses’ germination. If in the DNP’s case, can be remarked significant decreases of germinative faculties comparatively with the control for each three concentrations used, the 10⁻⁵ M solution having the most striking effect (48.666 ±2.905% comparatively with 74±1.154% in the reference plot), in the KIO₃’s case the situation is inverse, the most significant decreasing being signalized at the 10⁻⁵ M concentration (51.333±1.763%, the limits of confidence intervals of the average being, with a probability of 95%, of 47.876 - 54.79%). It must be remarked, in the same time, the decreased germination rate of wheat seeds (74% in the reference plot), which can be put, besides the species specificity and the different climatic factors - humidity, temperature etc. - (26 - 29) and on account of the fact that the seeds derive from the 2013 year culture, and the process of seeds’ frazzle begins immediately after the harvest through the corroboration of different intrinsic and extrinsic factors (30).

The markant influence of types of treatments can be correlated with other data from specialty literature, which denote the fact that the substances of dinitrophenol type, azides and fluorides can be notorious inhibitors in certain concentrations, their interventions in the metabolic processes being different (31).

<table>
<thead>
<tr>
<th>Control</th>
<th>DNP</th>
<th>KIO₃</th>
</tr>
</thead>
<tbody>
<tr>
<td>M (%)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>S (p)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>S (o)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Conf.</td>
<td></td>
<td></td>
</tr>
<tr>
<td>SL</td>
<td></td>
<td></td>
</tr>
<tr>
<td>IL</td>
<td></td>
<td></td>
</tr>
<tr>
<td>VC%</td>
<td></td>
<td></td>
</tr>
<tr>
<td>m%</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Table I. The germinative faculty of wheat seedlings pretreated with DNP and their main statistical indices

<table>
<thead>
<tr>
<th>Control</th>
<th>KIO₃</th>
</tr>
</thead>
<tbody>
<tr>
<td>M (%)</td>
<td></td>
</tr>
<tr>
<td>S (p)</td>
<td></td>
</tr>
<tr>
<td>S (o)</td>
<td></td>
</tr>
<tr>
<td>Conf.</td>
<td></td>
</tr>
<tr>
<td>SL</td>
<td></td>
</tr>
<tr>
<td>IL</td>
<td></td>
</tr>
<tr>
<td>VC%</td>
<td></td>
</tr>
<tr>
<td>m%</td>
<td></td>
</tr>
</tbody>
</table>

M = average, S (p) = standard error, S (o) = standard deviation, Conf. = confidence level, SL = superior limit, IL = inferior limit, VC% = average variation coefficient, m% = average precision coefficient

Table II. The germinative faculty of wheat seedlings pretreated with KIO₃ and their main statistical indices
In what concerns the wheat seedlings’ weight (Tables III-IV), it is remarked again the same tendency of valoric decreasing towards the control, in strong connection with the concentration degree of respective agent. If at the control, the wheat seedlings’ weight at the age of ten days was between 1.806 - 2.194 g/board, with an average of 2.0003±0.098g/board, at samples treated with DNP 10⁻⁴M is of 1.44±0.121g/board, at those with DNP 10⁻³M is of 1.377±0.104g/board, while, at the 10⁻²M solution the medium average goes up to approximately 50% from that of reference seedlings (1.073±0.093g/board).

By comparison, the apply of KIO₃ treatment induced different answers, in the sense that, at the most high concentration it is remarked a stimulation of seedlings’ elongating, the medium averages highlighted overtaking the marked out limits in the reference samples (2.0826±0.023g/board towards the 2.0003±0.098g/board). At the other two concentrations used, was remarked an inhibitor influence, but less egregious towards the DNP (1.763±0.0977g/board at the 10⁻³M solution and 1.557±0.0688g/board at 10⁻²M solution).

The specialty literature signalizes (32 - 34), otherwise, the apparition of some significant changes in the habitual development of the plant on the score of the different substances intervention in the habitual deployment of seed’s metabolism in the middle of the germination process, in strong connection with the type of substances, their concentration and their toxicity.

**Table III. The DNP influence on wheat seedlings’ weight and its main statistical indices**

<table>
<thead>
<tr>
<th></th>
<th>Control</th>
<th>DNP 10⁻⁴M</th>
<th>DNP 10⁻³M</th>
<th>DNP 10⁻²M</th>
</tr>
</thead>
<tbody>
<tr>
<td>M (g/board)</td>
<td>2.0003</td>
<td>1.073</td>
<td>1.3773</td>
<td>1.4406</td>
</tr>
<tr>
<td>S (σ)</td>
<td>0.0989</td>
<td>0.093</td>
<td>0.1049</td>
<td>0.1215</td>
</tr>
<tr>
<td>Conf.</td>
<td>0.1714</td>
<td>0.1611</td>
<td>0.1817</td>
<td>0.2105</td>
</tr>
<tr>
<td>SL</td>
<td>2.1943</td>
<td>1.2553</td>
<td>1.5829</td>
<td>1.6789</td>
</tr>
<tr>
<td>IL</td>
<td>1.8063</td>
<td>0.8906</td>
<td>1.1717</td>
<td>1.2023</td>
</tr>
<tr>
<td>VC%</td>
<td>8.5696</td>
<td>15.014</td>
<td>13.1924</td>
<td>14.6162</td>
</tr>
<tr>
<td>m%</td>
<td>4.9476</td>
<td>8.668</td>
<td>7.6166</td>
<td>8.4386</td>
</tr>
</tbody>
</table>

**Table IV. The KIO₃’s influence on wheat seedlings’ weight and its main statistical indices**

<table>
<thead>
<tr>
<th></th>
<th>Control</th>
<th>KIO₃ 10⁻⁴M</th>
<th>KIO₃ 10⁻³M</th>
<th>KIO₃ 10⁻²M</th>
</tr>
</thead>
<tbody>
<tr>
<td>M (g/board)</td>
<td>2.0003</td>
<td>2.0826</td>
<td>1.763</td>
<td>1.5576</td>
</tr>
<tr>
<td>S (σ)</td>
<td>0.0989</td>
<td>0.0238</td>
<td>0.0977</td>
<td>0.0688</td>
</tr>
<tr>
<td>Conf.</td>
<td>0.1714</td>
<td>0.0412</td>
<td>0.1693</td>
<td>0.1192</td>
</tr>
<tr>
<td>SL</td>
<td>1.9133</td>
<td>2.1293</td>
<td>1.9545</td>
<td>1.6926</td>
</tr>
<tr>
<td>IL</td>
<td>2.1806</td>
<td>2.0359</td>
<td>1.5714</td>
<td>1.4226</td>
</tr>
<tr>
<td>VC%</td>
<td>8.5696</td>
<td>1.9828</td>
<td>9.6038</td>
<td>7.6572</td>
</tr>
<tr>
<td>m%</td>
<td>4.9476</td>
<td>1.1447</td>
<td>5.5447</td>
<td>4.4209</td>
</tr>
</tbody>
</table>

**Table V. The influence of DNP on wheat seedlings’ highness and their main statistical indices**

<table>
<thead>
<tr>
<th></th>
<th>Control</th>
<th>DNP 10⁻⁴M</th>
<th>DNP 10⁻³M</th>
<th>DNP 10⁻²M</th>
</tr>
</thead>
<tbody>
<tr>
<td>M (cm/board)</td>
<td>267</td>
<td>151.133</td>
<td>191.266</td>
<td>192.433</td>
</tr>
<tr>
<td>S (σ)</td>
<td>10.4</td>
<td>14.039</td>
<td>12.717</td>
<td>13.072</td>
</tr>
<tr>
<td>Conf.</td>
<td>18.013</td>
<td>24.317</td>
<td>22.027</td>
<td>22.642</td>
</tr>
<tr>
<td>SL</td>
<td>20.383</td>
<td>27.517</td>
<td>24.9258</td>
<td>25.622</td>
</tr>
<tr>
<td>IL</td>
<td>287.383</td>
<td>178.651</td>
<td>216.192</td>
<td>218.055</td>
</tr>
<tr>
<td>VC%</td>
<td>6.7465</td>
<td>16.091</td>
<td>11.516</td>
<td>11.766</td>
</tr>
<tr>
<td>m%</td>
<td>3.8951</td>
<td>9.289</td>
<td>6.649</td>
<td>6.793</td>
</tr>
</tbody>
</table>

M = average, S X̄ = standard error, S (σ) = standard deviation, Conf. = confidence level, SL = superior limit, IL = inferior limit, VC% = average variation coefficient, m% = average precision coefficient
Table VI. The influence of KIO₃ on wheat seedlings’ highness and their main statistical indices

<table>
<thead>
<tr>
<th></th>
<th>Control</th>
<th>10⁻³M</th>
<th>10⁻⁴M</th>
<th>10⁻⁵M</th>
</tr>
</thead>
<tbody>
<tr>
<td>M (cm/board)</td>
<td>267</td>
<td>323.466</td>
<td>270.37</td>
<td>237.746</td>
</tr>
<tr>
<td>S ±x</td>
<td>10.4</td>
<td>6.4307</td>
<td>15.106</td>
<td>13.234</td>
</tr>
<tr>
<td>S (σ)</td>
<td>18.013</td>
<td>11.138</td>
<td>26.165</td>
<td>22.9235</td>
</tr>
<tr>
<td>Conf.</td>
<td>20.383</td>
<td>12.604</td>
<td>29.608</td>
<td>25.939</td>
</tr>
<tr>
<td>SL</td>
<td>287.383</td>
<td>336.070</td>
<td>299.978</td>
<td>263.686</td>
</tr>
<tr>
<td>IL</td>
<td>246.616</td>
<td>310.862</td>
<td>240.761</td>
<td>211.806</td>
</tr>
<tr>
<td>VC%</td>
<td>6.7465</td>
<td>3.4434</td>
<td>9.6779</td>
<td>9.642</td>
</tr>
<tr>
<td>m%</td>
<td>3.8951</td>
<td>1.988</td>
<td>5.5874</td>
<td>5.5668</td>
</tr>
</tbody>
</table>

M = average, $S \bar{x}$ = standard error,
S (σ) = standard deviation, Conf. = confidence level,
SL = superior limit, IL = inferior limit,
VC% = average variation coefficient,
m% = average precision coefficient.

It is known the fact that the pollutants, no matter their nature (including the pesticides) exert phytotoxic effects for all the plants’ organs, the specialty literature highlighting the negative impact that these substances exert on vegetable organisms, mostly in what concerns the biochemical, physiological, morphological and citogenetical transformations (35 - 40).

The chlorophyll is a green photosynthetic pigment which helps plants to procure energy from the light, the photosynthesis could be affected by a series of factors like the bright intensity, the concentration of carbon dioxide and the temperature (41).

In what concerns the chlorophyll’s a concentration in *Triticum aestivum* seedlings, the Secuienti variety (Fig.1), obtained from caryopses treated with DNP and KIO₃ solutions, it is ascertainment a fluctuation of the medium level of this pigment, on the one hand in function of the type of treatment, but also by the concentration used. Hereby, the DNP 10⁻³ and KIO₃ 10⁻³M solution exerted significant strong influences - $p < 0.001$ (0.596±0.022 mg/g fresh tissue and 0.555±0.011 mg/g fresh tissue), stimulating the chlorophyll a synthesis by comparison with the control (0.464±0.019 mg/g fresh tissue). To remark the fact that KIO₃, in concentration of 10⁻⁴M doesn’t determine major changes in what concerns the content of chlorophyll a (0.469±0.024 mg/g fresh tissue), while the KIO₃ 10⁻⁵M solution exerted even a slight inhibitory influence (0.441±0.015 mg/g fresh tissue).

Fig.1. The content of chlorophyll a in wheat seedlings pretreated with DNP and KIO₃ of different concentrations

(*P < 0.05 – less significant; **P < 0.01 – significant; ***P < 0.001 – very significant)

To realize the photosynthesis process, the green plants are based on the interactive cooperation of two photosystems (42; 43), the primary cofactor for the photochemical reactions from these complexes being chlorophyll a, needful also for the assembly of these complexes. Also, additionally, the plants contain also the chlorophyllb, an accessory of the chlorophyll, found only in the periphery complexes of light absorption. For maximum efficiency, each of the two photosystems must absorb an equal quantity of light, having for this aim a mobile basin of chlorophyll a/ chlorophyllb to maintain an optimum equilibrium of exertion (44 - 46). The statistical analyze of obtained results (Fig.2), emphasized influences less markant of treatments applied to caryopses in what concerns the concentration of chlorophyllb, significant differences ($p < 0.01$) and less significant ($p < 0.05$) giving points to KIO₃ 10⁻³M solutions (0.156±0.007 mg/g fresh tissue), respectively DNP 10⁻³M (0.152±0.005) and 10⁻⁴M (0.257±0.004 mg/g fresh tissue) comparison with the control plot (0.225±0.009 mg/g fresh tissue).

Fig.2. The content of chlorophyllb in wheat seedlings pretreated with DNP and KIO₃ of different concentrations

(*P < 0.05 – less significant; **P < 0.01 – significant; ***P < 0.001 – very significant)

The majority of economic activities represent the primary source of progress and development of human society, but this development led to a strong impact on all ecosystems, through overtaking the limits of ecologic equilibrium, giving rise to prejudices on the environment, and also in the plants’ life. Hereby, was showed that the pollutants affect a series of metabolic functions like transpiration, respiration and the plants’ photosynthesis (47). Data from literature signalize that the carotenoid pigments are the most important photosynthetic pigments, forasmuch protect the tilacoids’ membranes and the chlorophylls against the damages caused by the energy absorbed through photooxidation (48). If in the reference samples, the medium content of provitamin A is of 0.0828±0.004 mg/g fresh tissue, in the case of tests treated with DNP 10⁻³ and 10⁻⁴M it is ascertainment an intensification of the carotenoid fraction’s synthesis, the concentration getting to medium threshold of 0.097±0.004 mg/g fresh tissue and 0.107±0.011 mg/g fresh tissue, this last value being comparable with that highlighted in the seedlings treated with KIO₃ 10⁻³M (0.101±0.009 mg/g fresh tissue). However, from statistically point of view, aren’t remarked
differences strongly significantly in the case of any treatment applied to caryopses, significant results (p=0.0056) showing only the DNP 10^{-5}M solution and less significant (p< 0.05) towards the variety (in increasing or decreasing sense) the DNP and KIO_3 solutions in concentration of 10^{-4} and respectively 10^{-3}M (Fig.3).

Our data concord with those from the literature data (39) which show that the foliar answer induced by the atmospheric pollutants at Aesculus hippocastanum L. and Tiliatomentosa L. species it is translated by a different content of assimilator pigments in the sense that both chlorophyll _a_ and _b_, as also the carotenoidic pigments have different values, bigger or smaller, comparatively with the reference value.

**Fig.3.** The content of carotensin wheat seedlings pretreated with DNP and KIO_3 of different concentrations

(*P < 0.05 – less significant;**P < 0.01 – significant; ***P < 0.001 – very significant)

It is known the fact that the use on scale larger and larger of the pesticides in agriculture, determines the storage of some high quantities of these substances in the soil (49; 50) and, there from, in the tissues and in the plants’ organs (51; 52) with direct role in the apparition of the habitual metabolism’s perturbations, defined, among other things, by the producing of free oxygen radicals.

The action of biotic and abiotic factors on plants has as result the increasing of the antioxidant enzymes’ activity with the aim of protecting the cell by the negative effect of xenobiotics (53). To metabolize the excess of free radicals, formed on the score of the pesticides action on vegetable tissues, in non-toxic products for the cell (water and oxygen), interfere a series of oxidoreductases like CAT, POX, SOD etc., the adjustment of these protective enzymes being essential for keeping the optimum level of reactive compounds (54 - 56).

The following stage of our study consisted in the determination of POX activity in wheat seedlings of ten days aged, hemoprotein with role in oxidation of some extensive categories of organic substratum and anorganic (57), but also in lignonification and suberization, the cellular wall’s metabolism, the plants’ resistance, the auxines’ metabolism, the elongation of cells, the phenolic oxidation etc. (58 -60).

From Figure 4, it is ascertained that the agents with which were treated the wheat seeds, no matter the concentration used, produced a certain level of oxidative stress, differences strongly significant (p< 0.001), through comparison with the reference sample (17.317±0.651 POXU/µg protein, the confidence interval’s limits being between 16.222 - 18.412 POXU/µg protein), laying out in the DNP’s case in concentrations of 10^{-5} and 10^{-4}M (24.528±0.0542 POXU/µg protein, respectively 23.648±0.745 POXU/µg protein). At variants with KIO_3 it is ascertained significant differences (p< 0.01) in the case of 10^{-3}M solution (21.461±0.713 POXU/µg protein, the confidence interval’s limits being between 20.011 - 22.912 POXU/µg protein) and less significant (p< 0.05) in the case of 10^{-4} and 10^{-5}M solutions (approximately 20 POXU/µg protein).

**Fig.4.** The peroxidase’s activity in wheat seedlings pretreated with DNP and KIO_3 of different concentrations

(*P < 0.05 – less significant;**P < 0.01 – significant; ***P < 0.001 – very significant)

At cell’s level, SOD constitute the first line of defense against the defensive of reactive oxygen species, the enzyme converting the superoxide radicals in oxygen and peroxide (61; 62). This metalo-enzyme is present in all the aerobe organisms where it plays a primordial role in the defensive against the ROS generated like products of primary biologic oxidations (63; 64). The sources of superoxide radical generation may be natural (products of metabolic activities) or induced by external agents (ozone, gamma rays, water deficiency, high temperature, light-induced photoinhibitory conditions or chemicals like pesticides) (65).

The obtained experimental results (Fig.5) show that all the types of treatment applied on caryopses determined a stimulation of this enzyme’s activity, registering significant differences (p< 0.01) between the control plot (8.121±0.221 SODU/µg protein) and the variants treated with DNP (10^{-3}M - 10.061±0.301 SODU/µg protein; 10^{-4}M - 9.977±0.231 SODU/µg protein; 10^{-5}M - 9.272±0.154 SODU/µg protein). In what concerns the KIO_3 effect, we remark an impact extremely strong on the SOD activity reflected also in the value of statistical indicator _p_ (< 0.001) which indicates differences strongly significant (10.495±0.293 SODU/µg protein in the case of the solution of 10^{-3}M concentration and 9.977±0.162 SODU/µg protein in the case of those of 10^{-5}M).

**Fig.5.** The superoxide-dismutase’s activity in wheat seedlings pretreated with DNP and KIO_3 of different concentrations
A last purpose of our study was to determine the influence that DNP and KIO₃ exert on CAT activity, metalo-enzyme that has in quality of prosthetic group the hemoporfirine, with role in removal of the hydrogen peroxide from the vegetable and animal cells (66). Significant amount of ROS, synthesized in the photosynthetic tissues of plants, under the influence of different pathogen agents, may be removed by CAT, which represents the first line of defense of the cell in the battle against the harmful molecules of hydrogen peroxide (67; 56).

Data from specialty literature highlight the apparition of some changes of the cellular redox status at the treated wheat, existing direct correlations between the germination process and the antioxidant capacity of the future seedlings (68 - 70). In the same time, were signalized striking modifications of the metabolic activity of the seed and of the new seedling, the hydrogen peroxide intensifying the germination rate of the wheat caryopses and being eliminated, when it is in excess (71), by CAT which interferes actively in facilitating the redox changes from seeds during the germination (72 - 74).

The differences registered between the medium activity of CAT from the reference samples and all the variants of treatment in part, are extremely marked, the values of statistical indicators denoting the influences strongly significant of the chemic agents used. As we can ascertained from the Figure 6, CAT mobilizes quantities very high of hydrogen peroxide, the activity of this enzyme being, in some cases, even three times bigger comparatively with the control. If in the reference sample the medium activity of CAT reached the valoric threshold of 22.661±1.672 CATU/µg protein, with variations between 19.261-26.062 CATU/µg protein, in the case of experimental variant DNP 10⁻⁴ M the enzyme reach to maximum quotes of activity (60.884±2.112 CATU/µg protein), results approached to those highlighted in the case of KIO₃ of maximum concentration (56.181±3.017 CATU/µg protein).

![Fig.6. The catalaseˈs activity in wheat seedlings pretreated with DNP and KIO₃ of different concentrations](image)

(*P < 0.05 – less significant; **P < 0.01 – significant; ***P < 0.001 – very significant)

IV. CONCLUSIONS

- The DNP and KIO₃ solutions in different concentrations, used for prior treatment of caryopses submitted to germination, determined a significant influence on the germination rate of the wheat, the most decreasing germinative faculty being registered in the case of DNP 10⁻⁵ M solution, in strong connection with the toxicity extremely high of this agent.
- Although the germination process was influenced negatively at all the experimental variants, there werenˈt marked out significant differences in what concerns the growing in highness and weight of viable seedlings, the only registered difference, by comparison with the control, being signalized at KIO₃ 10⁻⁴ M batches, which stimulated the increase in highness of the seedlings.
- In what concerns the content of chlorophyllien and carotenoidic pigments, there are ascertained influences more or less markant of treatment used, in direct relation with the concentration degree of the applied agent, the highest concentrations manifesting a light inhibitor effect on the synthesis of these physiological parameters.
- The apply of treatments with DNP and KIO₃ solutions on Triticum aestivum caryopses, the Secuieni variety, determined the storage of some sizeable quantities of reactive oxygen species, the antioxidant enzyˈmeˈs activity being net superior to control, the catalase laying out, in some cases, activitieseven three times bigger than the reference plot.

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