

Ali-zadeh Gafar Islam oglu,  
candidate of biological sciences, associate professor,  
Jalilova Ainura Rafik kyzy, junior researcher,  
Baku State University,  
Suleymanova Leyla Maharram kizi,  
candidate of biological sciences, associate professor,  
Azerbaijan Medical University

### **THE STABILITY OF FUNCTIONAL ACTIVITY AND ANTIOXIDANT SYSTEM OF DUNALIELLA SALINA IPPAS D-294 MODIFIED BY (IONOL) BHT IN OPTIMAL AND HIGH SALINITY**

*The article discusses the effect of various concentrations of ionols on growth, the activity of the endogenous antioxidant system of the alga Dunaliella salina IPPAS D-294 and their UV-protective activity in the cell under conditions of optimal (1.5 M NaCl) and a mineral environment of high salinity (3.0 M NaCl).*

*Key words: Dunaliella, salinity, synthetic antioxidants, UV-B radiation, LPO, catalase activity, carotenoids, functional stability.*

Али-заде Гафар Ислам оглы, к.б.н., доцент,  
Джалилова Айнура Рафик кызы, м.н.с.,  
Бакинский государственный университет,  
Сулейманова Лейла Магеррам кызы, к.б.н., доцент,  
Азербайджанский медицинский университет

### **УСТОЙЧИВОСТИ ФУНКЦИОНАЛЬНОЙ АКТИВНОСТИ И АНТИОКСИДАНТНОЙ СИСТЕМЫ КЛЕТОК DUNALIELLA SALINA IPPAS D-294 МОДИФИЦИРОВАННЫХ ИОНОЛОМ В ОПТИМАЛЬНЫХ И УСЛОВИЯХ ВЫСОКОЙ СОЛЕННОСТИ**

*В статье рассматриваются влияние различных концентраций ионолов на рост, активность эндогенной антиоксидантной системы водоросли Dunaliella salina IPPAS D-294 и их УФ-защитную активность в клетке в условиях оптимального (1,5 M NaCl) и минеральная среда высокой солености (3,0 M NaCl).*

*Ключевые слова: Dunaliella, соленость, синтетические антиоксиданты, УФ-В излучение, ПОЛ, активность каталазы, каротиноиды, функциональная стабильность.*

Али-заде Гафар Ислам оглу, б.и.к., доцент,  
Жалилова Айнура Рафик кызы, кенже илимий  
кызматкер, Баку мамлекеттик университети,  
Сулейманова Лейла Махаррам кызы, б.и.к., доцент,  
Азербайжан медициналык университети

### **DUNALIELLA SALINA IPPAS D-294 КЛЕТКАЛАРДЫН ОПТИМАЛДУУ ЖАНА ТУЗДУК ШАРТТАРДА ӨЗГӨРТҮЛГӨН ФУНКЦИОНАЛДУУ ИШТҮҮЛҮГҮНҮН ЖАНА АНТИОКСИДАНТТАРЫНЫН СТАБИЛДҮҮЛҮГҮ**

*Макалада ионолдордун ар кандай концентрациясынын өсүүгө таасири, Dunaliella salina IPPAS D-294 балырынын эндогендик антиоксидант системасынын активдүүлүгү жана алардын оптималдуу (1,5 М NaCl) жана минералдык шарттарда клеткадагы УКтан коргоочу активдүүлүгү талкууланат. туздуу чөйрө (3.0 М NaCl)*

*Негизги сөздөр: Дуналиелла, туздуулук, синтетикалык антиоксиданттар, UV-B нурлануусу, LPO, каталазанын активдүүлүгү, каротиноиддер, функционалдык стабилдүүлүк.*

**Introduction** It is known, that the wedge influence physiological and biochemical characteristics of plants. Sustainability mechanisms can be existed through changes in ratio of ions, synthesis of osmolytics, of organic medium nature activation of work of antioxidant system and so on. Genetic mechanisms of saline sustainability are identified by great number of genes which work is necessary for responsiveness of plants to saline stress [1,13].

There are a large number of synthetic compounds, which regulate plant growth in exogenously applied. Compounds which inhibit plant growth by inhibition cell stretching and their division, are called growth retardants which exited growth-stimulation [2,8]. Significant interest performs investigation of influence of synthetic antioxidants like ionol and 2,6 di-tert-butyl phenol, which belong to the class of spatially difficult phenols [4]. For more efficient use of antioxidants, it is urgent to link chemistry and biology by antioxidants, i.e. the dependence of the biological activity of antioxidants. Such dependence of biological activity of antioxidants features. As inhibitors of radical reactions also their efficient concentrations [2,7]. Actively was investigated the possibility of antioxidant use in plant growth as growth stimulators [5]. It is also known that, synthetic antioxidants in high, concentrations begin to act in the opposite direction and not broke; on the contrary, accelerate free radical reactions [9].

Very little information about synthetic antioxidants and their antiradical features in green microalgae is very little [9]. Due to that, the purpose of our work was to investigation of influence of various concentrations of antioxidant 2,6 di-tert-butyl cresol (ionol - a classic synthetic antioxidant) on growth, activity of endogen antioxidant system of Dunaliella algae and their UV protection activity in cell, under low temperature stress and high salinity.

**Materials and methods:** As investigation object was used green algae Dunaliella Salina IPPAS-294, taken from the saline lake Masazir located on the north eastern part of Baku. The algae were grown at 27<sup>0</sup>C temperature in glass photo reactors (250ml), in the installation for growing unicellular algae. Mineral medium contained (g\l): NaCl-87, 5 (1,5M) and NaCl-175, 5 (3,0M), KNO<sub>3</sub> - 5,0; KH<sub>2</sub>PO<sub>4</sub> -1,25; MgSO<sub>4</sub>-50; FeSO<sub>4</sub> - 0,009 and microelement solutions (1mg\l) – Ca(NO<sub>3</sub>)<sub>2</sub>·H<sub>2</sub>O-735; H<sub>3</sub>BO<sub>3</sub>-735; ZnSO<sub>4</sub>·7H<sub>2</sub>O-615; (NH<sub>4</sub>) MoO<sub>4</sub>-100; MnCl<sub>2</sub>·4H<sub>2</sub>O-180. The cell suspension in photo reactors was irradiated by white light (16Wt\m<sup>2</sup>) within 24 hours and permanently purged with mixture (air+1,5% CO<sub>2</sub>) at 25<sup>0</sup> C temperature. The source of UV irradiation was mercury lamp SVD-120. The cells were cultivated within 24 hours, in intensive – accumulated cultivation regime and was irradiated day and high. The culture growth rate was determined by periodically counting cell number in Qoryayev chamber under the microscope or by nephelometric measurement of optic suspension density in photo electro colorimeter.

In this work were used various concentrations of classic antioxidant ionol (2,6 di-tert-butyl cresol).

The content of pigments in cellular extracts (100% acetone) was measured in spectrophotometer and counted on base of Wettshain coefficient [9]. To measure the photosynthetic cell activity of grown algae were precipitated by centrifugation 30000 rev\min. within 10 minutes at room temperature and transferred into newly made mineral medium. The suspension density in cells was led to 10<sup>6</sup> cells\ml (optic density OD=0, 8). The speed of

oxygen evolution in cells was measured in polarographic installation, using platinum electrode Clark lighting the suspension in thermostated volume, saturating the intensity by white light ( $100 \text{ Wt/m}^2$ ).

In order to measure the catalase activity in cells, the suspension precipitated by centrifugation (30000 rev/min). The sediment was transferred into a mortar with 0,5 g  $\text{CaCO}_3$  was added 5 ml distilled water and triturated into homogenous mass. Then gained mass quantitatively transferred into a glass with 50 ml capacity till the monk and infused with periodic shaking 3-4 hours ( $4^\circ\text{C}$ ). Within that time happens enzyme extraction in plant material. After infusion the suspension was filtered in dry glass catalase activity was measured by gasometric method, which based on determination of volume after adding into the aqueous extract of plants containing catalase, hydrogen peroxide [9].

The evaluation degree of lipid peroxidation (POL) was carried out by the method of determining MDA content in *Dunaliella salina* cells – method based on the reactions with triobarbituric acids.

The cell suspension (35ml) was centrifuged 30000 rev/min. within 10 minutes. The resulting sediment was homogenized in 20 ml 0, 1% TCA. Homogenate was centrifuged at 30000 rev/min. within 10 minutes. To the 1 ml supernatant was added 4 ml 20% TCA, containing 0, 5% TBA. The mixture was heated in water bath at  $95^\circ\text{C}$  within 30 minutes and immediately cooled in running water. After centrifugation of mixture at 30000 rev/min. within 10 minutes was determined optic density of supernatant at 532 nm and 600 nm. MDA content was counted after subtraction of non-specific absorption at 600nm [1].

**Result and discussions:** In figure 1, has been presented quantitative indicators of 24 hourly growth of *Dunaliella salina* IPPAS D-294 cells biomass in mineral medium of 1.5M NaCl (1) and 3.0M NaCl (2). As seen in the figure, control suspension of cells in optimal conditions (temperature  $27^\circ\text{C}$ , light intensity  $16 \text{ W / m}^2$ ) in 250 ml in glass photoreactors by air mixture with  $25^\circ\text{C}$  in a periodically accumulative cultivation regime within 24 hours showed that, optic density of cell solution increases 3.5–4 times (1). Growth dynamics of *Dunaliella* microalgae culture in such conditions, but at high salinity of mineral medium (3.0 M NaCl), optical density of cell suspension increases only 3 times (2).

Such trend of population growth continues and also at following repeated cultivation variants of these suspensions.

On the base of conducted investigations has been established that *Dunaliella* cells have lower growth rate and perform less productivity by increasing NaCl concentrations in mineral medium. Decrease of growth rate at the same time increases carotenoid content and naturally glycerin in cells. It is known that, under influence of high concentration NaCl, glycerin content in *Dunaliella salina* IPPAS D-294 cells increases and, at the same time, as show in obtained results, decreases growth rate 25%. The decrease of productivity in *Dunaliella* cells at salinity associated with glycerin biosynthesis and change of carotenoid quantity. In optimal and high salinity of mineral culture medium of cultivation it was interesting investigate pigment formatting indicators of *Dunaliella salina* IPPAS D-294 label cells. *Dunaliella* algae cells respond to factor change of mineral medium of cultivation, which leads to irregular synthesis of definite metabolites. It is known that, also high content of NaCl in mineral medium significantly influences on biosynthesis indicators of chlorophyll and carotenoids in *Dunaliella* cells [7,13].

In table 1 has been presented pigment content indicators in *Dunaliella salina* IPPAS D-294, grown in mineral medium with 1,5M NaCl (1) and 3,0M NaCl (2). As seen in the table, in cells grown in optimal regime of cultivation (1,5M NaCl), chlorophylls biosynthesis and carotenoids amounts have certain indicators: so ratio of “a” and “b”, contains classical indicators (2: 1), in fact. Ratio parameters, of chlorophyll and carotenoid amounts, characterizing of energization of photosynthetic membrane, contain 5.5.

This indicates a high indicator of photosynthetic activity of *Dunaliella salina* IPPAS D-294, grown in given conditions.

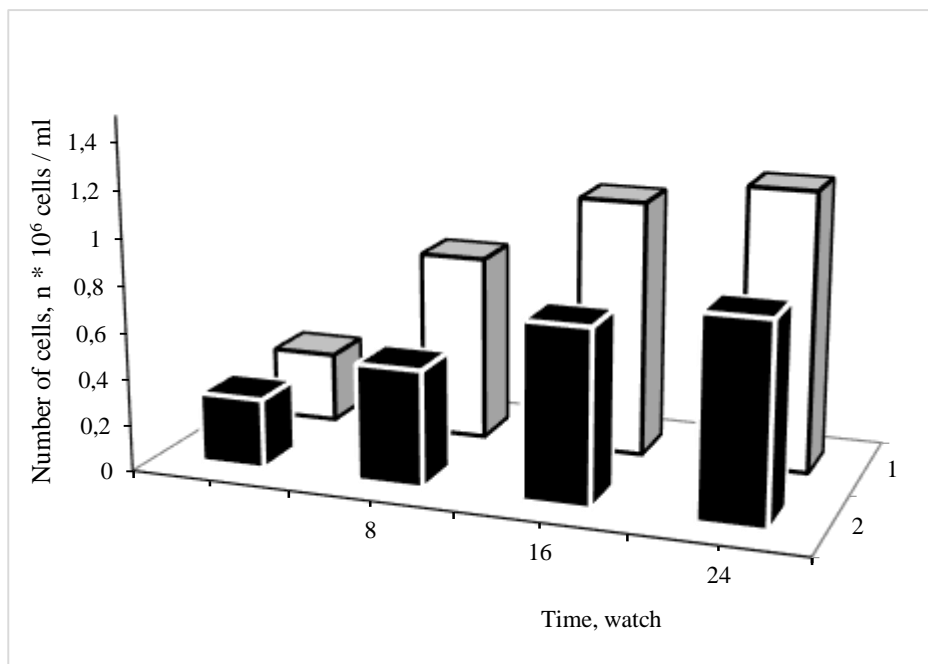


Fig. 1. The dependence of population growth in *Dunaliella salina* IPPAS D-294 in mineral medium with 1,5M NaCl (1) and 3,0M NaCl (2). Temperature 27<sup>0</sup>C, light intensity 16 Wt/m<sup>2</sup>

Table 1

Pigment content in *Dunaliella salina* IPPAS D-294 cells, grown in mineral medium with 1,5M NaCl (1) and 3,0M NaCl (2).

1,5M	3,27± 0,05	1,68±0,05	0,9±0,01	5,5
3,0M	3,13± 0,05	1,67±0,05	1,06±0,01	4,5

Note: optical density OD-0,8; temperature 27<sup>0</sup>C, light intensity 16Wt/m<sup>2</sup>.

Increase of medium salinity (3,0M NaCl) lead to minor decrease 4,3% of chlorophyll biosynthesis “a” and 0,5% “b”, and in the end, their amount, but carotenoid amounts increase 12%. Chlorophyll / carotenoid ratio, being one of the indicators of photosynthetic activity of *Dunaliella* cells in this case by increasing salinity from optimal value (1,5M NaCl) decreases and effects on photosynthetic activity and final bioproductivity (table 1).

Carotenoid accumulation in *Dunaliella* cells NaCl showed that, also in these conditions, from cells save (typical for them) their responsiveness, and by increasing sodium chloride concentration in medium carotenoid content in them increases. Parameter of chlorophylls and carotenoid amount ratio characterizing energization of photosynthetic membrane contains at optimal salinity (1,5 M NaCl) of cultivation regime, and 4,5 at high salinity (3,0 M NaCl).

In figure 2, have been presented the data of photosynthetic oxygen evolution in *Dunaliella salina* IPPAS D-294 cells grown in optimal and high content of NaCl in mineral medium of cultivation. As been in the figure, functional activity of cells, grown in optimal and high content of NaCl in mineral medium, in then 40<sup>0</sup>C temperature and light intensity 100 Wt/m<sup>2</sup> significantly differ. Comparison of the data of photosynthetic oxygen evolution indicates an significant difference, containing about 17%. As seen in the figure of cell photosynthetic activity, grown at high salinity (3,0 M NaCl) in mineral medium, somewhat lower (17%) than in cells, grown in optimal conditions. This is probably, related to various pigment compounds.

In was interesting to observe how various concentrations of synthetic antioxidants would influence on bioproductivity, pigment composition, and functional activity of

Dunaliella cells. In this work, have been used synthetic antioxidants in range of concentrations (25-500 mkM).

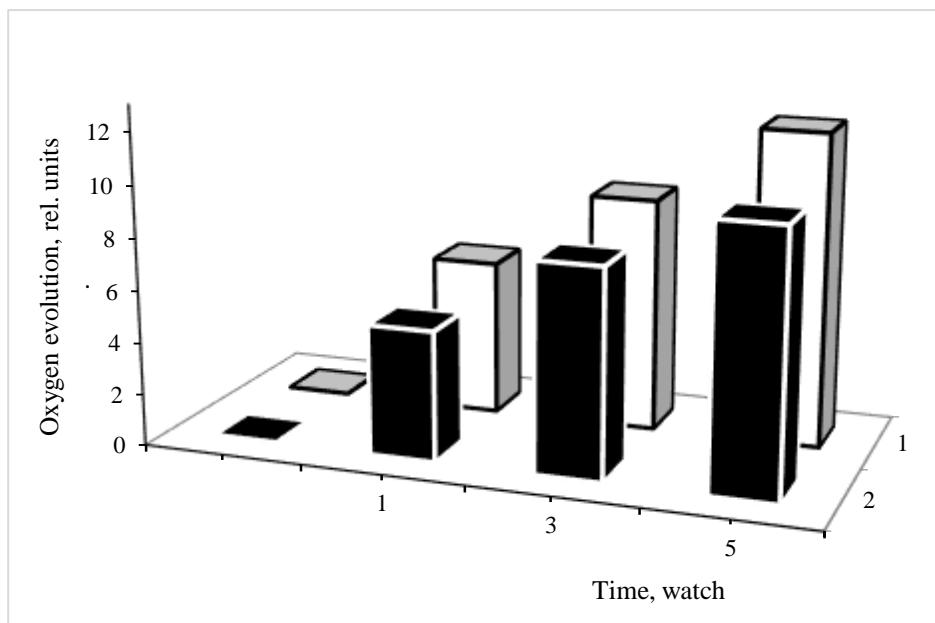


Fig. 2. Photosynthetic oxygen evolution of *Dunaliella salina* IPPAS D-294 cells, in mineral medium with 1,5M NaCl (1) and 3,0M NaCl (2). Temperature 40<sup>0</sup>C, light intensity 100 Wt/m<sup>2</sup>

Figure 3 has been presented the growth dependence of *Dunaliella salina* IPPAS D-294 cells in intensive accumulative regimes of cultivation in mineral medium with 1,5M NaCl (1) and 3,0M NaCl (2) on various concentrations of ionol in mineral medium.

As seen in the figure, ionol presence in mineral medium significantly influences on growth of Dunaliella cells. So, in concentrations 25 mkM and 50 mkM of ionol in mineral medium in optimal regime of cultivation (1,5M NaCl) was observed stimulation of cell culture growth 2,5% and 4,2% accordingly, compared to control suspensions.

So, ionol at low concentrations 25 mkM and 50 mkM was compared to activity of common photo hormones. At concentrations 150-250 mkM in mineral medium its growth stimulating action notably decreases (100-96%). By increasing the content of BHT about in orderly (350-500 mkM) it gets reverse sign, was observed the reduction to (13-15%) accordingly of culture growth within 24 hourly cultivation in intensive- accumulative regime. Under influence of this antioxidant maximal differentiation was observed at concentrations 50 mkM (4,2%) compared to control cells. The dependence of population growth in *Dunaliella salina* IPPAS D-294 cells in intensive – accumulating regime of cultivation in mineral medium with high content of NaCl (3,0 M) on various concentrations of BHT in mineral medium was shown at table 2. As seen figure presence of ionol in mineral medium of cultivation at high salinity visibly affects culture growth. So, at concentrations 25 mkM and 50 mkM in mineral medium of ionol (2) was observed dynamic stimulation of cell culture growth 4% and 6% accordingly, compared to control suspensions. At concentrations (150; 250; 350 mkM) in mineral medium growth stimulation remains at high level (107; 106; 102%) (table 2). By increasing the content of 2,6 di-tret- butyl cresol in mineral medium (500 mkM) it gets reverse sign was observed suppression till (8-9%) accordingly culture growth within 24 hourly cultivations in intensive- accumulative regime. Under influence of this synthetic antioxidant maximal differentiation was observed at concentrations 150 mkM (7%) compared to control cells.

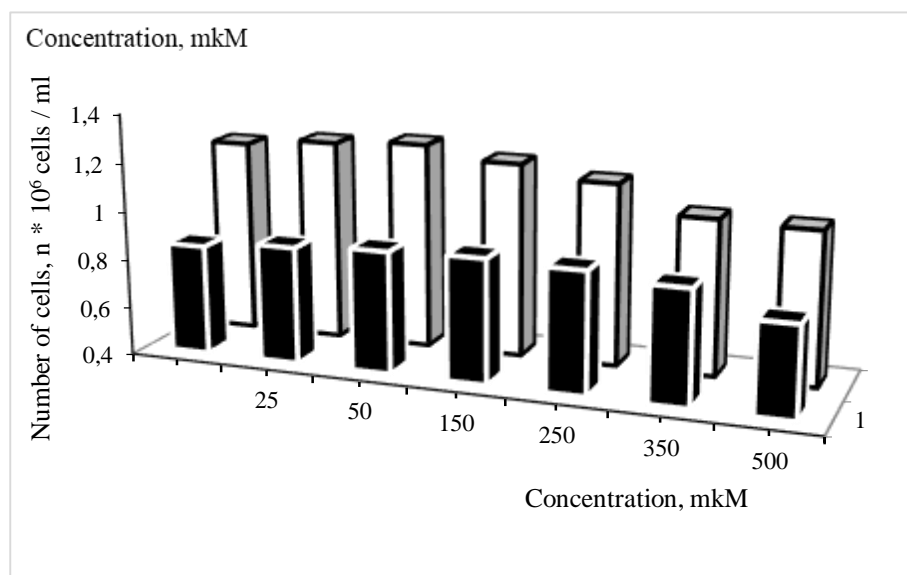


Fig. 3. The dependence of population growth in *Dunaliella salina* IPPAS D-294, grown in mineral medium with 1,5M NaCl (1) and 3,0M NaCl (2); at various concentrations of 2,6 di-tert-butyl cresol.

Temperature 27<sup>0</sup>C, light intensity 16 Wt/m<sup>2</sup>

In this case, increases cell tolerance to the antioxidant compared to cells, grown in optimal and high content of NaCl in mineral medium, ....., related to work of cell endogen antioxidant and ionol expressed growth stimulating BHT activity at its low concentrations 25-100 mkM in mineral medium in optimal regime of cultivation at concentration range of 25-100 mkM at high salinity of mineral medium makes, this antioxidant perspective and effective means of available and reliable regulation of cell culture growth of *Dunaliella salina* IPPAS D-294.

In figure 4, has been shown dependence results of biosynthesis pigments in *Dunaliella salina* IPPAS D-294 cells on various concentrations of ionol in mineral medium grown in optimal salinity (1,5 M NaCl).

As seen in figure, growth stimulating concentrations 250-500 mkM and subsequent high BHT concentrations decrease biosynthesis of whole chlorophyll of quality (till 69% chlorophyll "a", 63% chlorophyll "b") and also amount of carotenoid biosynthesis till 44%. The concentration increase of synthetic antioxidant in mineral medium leads to 150-250 mkM decrease and then 350-500 mkM increase in ratio of chlorophyll / carotenoids.

So, concentration increase of synthetic antioxidant – ionol in mineral medium optimal conditions of *Dunaliella salina* IPPAS D-294 cell cultivation leads to (150-250 mkM) and then the increase (350-500 mkM) in ratio of chlorophyll "a"/ chlorophyll "b".

In figure have been present dependence results of biosynthesis pigments in *Dunaliella salina* IPPAS D-294 cells on various concentrations of BHT in mineral medium, grown at high salinity in mineral medium (3,0 M NaCl). As seen in the figure growth regulating concentrations 25-350 mkM and consequent high concentration 500 mkM of ionol decrease biosynthesis of whole chlorophyll amount (till 82% chlorophyll "a"); 85% chlorophyll "b") synthesis of carotenoid amount in this case, increases till 6%, compared to control cells. It is typical for *Dunaliella* algae where clearly was said that, the change of existence conditions, absolute content and ratio of pigments change. Concentrations of synthetic antioxidant in mineral medium do not affect the ratio of chlorophyll "a"/ chlorophyll "b". So, in this ratio chlorophyll / carotenoids decreases by increasing ionol concentrations in mineral medium.

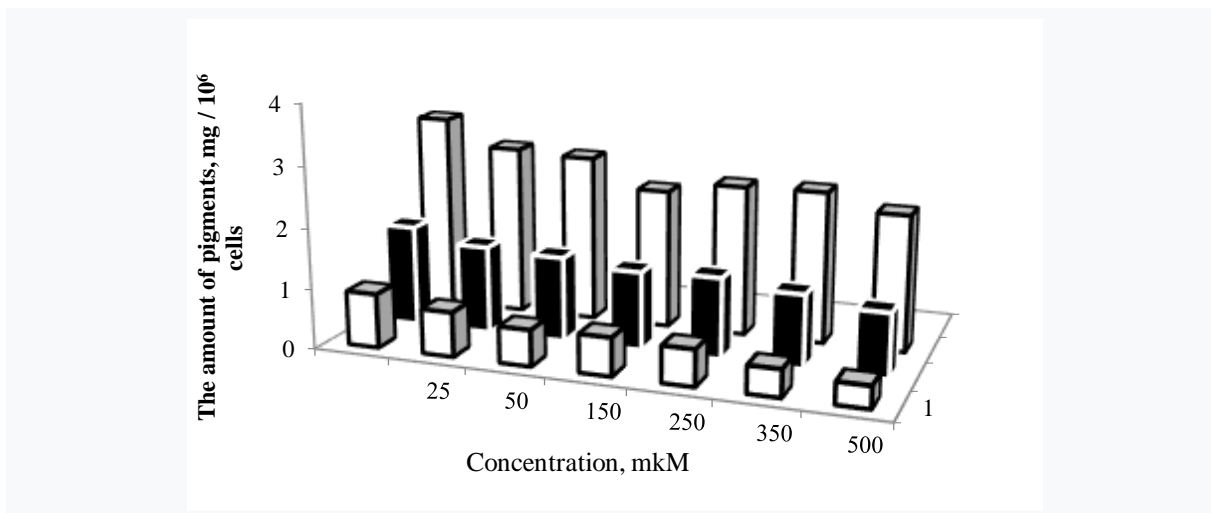


Fig.4. The dependence of pigment biosynthesis in *Dunaliella salina* IPPAS D-294 cells on various concentrations of 2,6 di-tret-butyl cresol (ionol) in mineral medium (1,5M NaCl).

1 – chlorophyll “a” biosynthesis;  
 2 – chlorophyll “b” biosynthesis;  
 3 – biosynthesis of carotenoid amount  
 Temperature 27<sup>0</sup>C, light intensity 16 Wt/m<sup>2</sup>

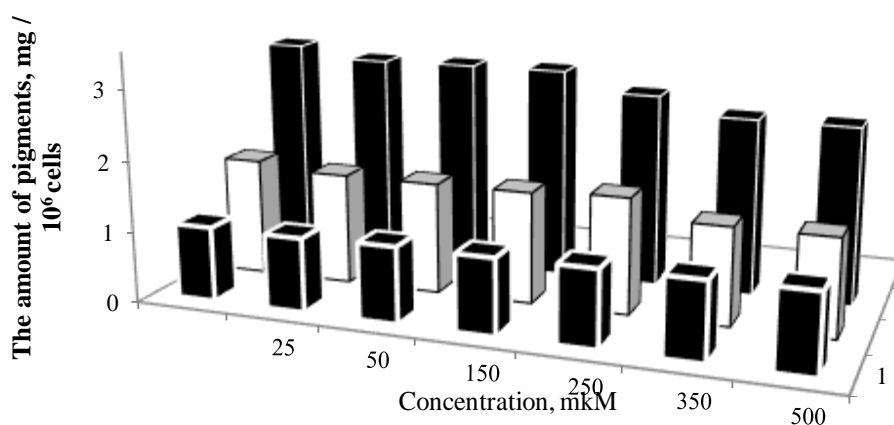


Fig.5. The dependence of pigment biosynthesis in *Dunaliella salina* IPPAS D-294 cells on various concentrations of 2,6 di-tret-butyl cresol (ionol) in mineral medium (3,0M NaCl). 1 – chlorophyll “a” biosynthesis; 2 – chlorophyll “b” biosynthesis; 3 – biosynthesis of carotenoid amount. Temperature 27<sup>0</sup>C, light intensity 16 Wt/m<sup>2</sup>

In the picture 6, have been presented the data of photosynthetic oxygen evolution by *Dunaliella salina* IPPAS D-294 cells, grown at various concentrations of BHT in optimal (1) and at high salinity (2) of mineral medium. As seen in figure, oxygen evolution by *Dunaliella salina* IPPAS D-294 cells, by modification various concentrations of BHT (25-500mkM) in optimal salinity (1,5M NaCl) despite of growth stimulation at low concentrations 25 mkM and 50 mkM is greatly suppressed already at concentrations 25 mkM (20%) (table 1). The cell modification by BHT at high concentrations 50-150mkM suppressed cell functional activity 30%-32%, and at 350-500mkM till up to 35-37%.

Photosynthetic oxygen evolution of *Dunaliella salina* IPPAS D-294 cells by modification with various ionol concentrations (25-500mkM) at high salinity of mineral



medium despite of growth stimulation at concentrations 25-350mkM (fig. 6,2), is greatly suppressed already at concentrations 25 mkM (5%). The cell modification by BHT at high concentrations 50-150mkM suppressed cell function 12-13%, but at 350-500mkM up to till 20-28%.

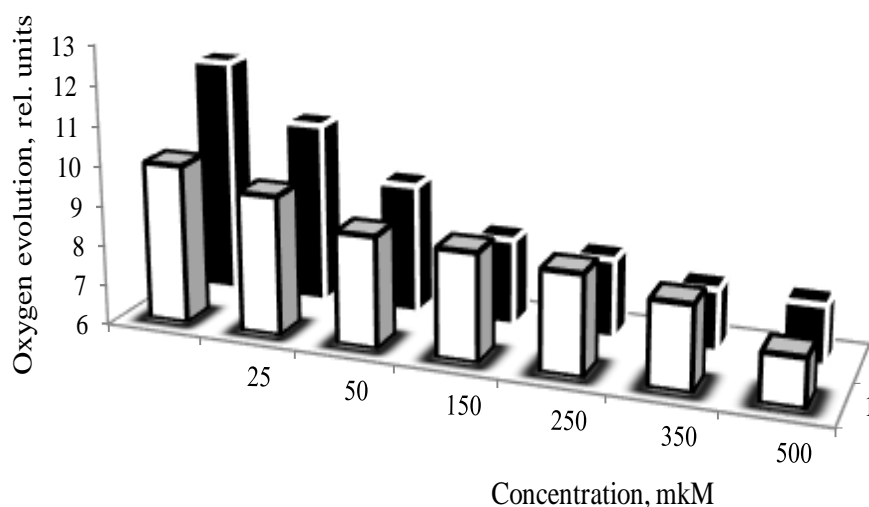


Fig. 6. Photosynthetic oxygen evolution of *Dunaliella salina* IPPAS D-294 cells, grown at various concentrations of 2,6 di-tert-butyl cresol (ionol) with presence of 1,5M NaCl (1) and 3,0M NaCl (2) in mineral medium. Temperature 40<sup>0</sup>C, light intensity 100 Wt/m<sup>2</sup>

In the picture7, have been presented data dependence of catalase activity in *Dunaliella salina* IPPAS D-294 cells on various ionol concentrations, grown in optimal (1) and high salinity (2) in mineral medium. As seen in the picture, various concentration of synthetic antioxidant ionol strongly affect catalase activity in cells, so in optimal salinity (2) in mineral medium where was observed the increase up to 55-65% the range of ionol concentrations 25-50 mkM within 24 hourly cultivation in fallowing increase of concentrations leads to stasionar level installation of catalase activity in Dunaliella cells.

Catalase activity in *Dunaliella salina* IPPAS D-294 cells at various concentrations of ionol in mineral medium with high content of NaCl (3,0M NaCl) increases up to 30-35% in the range of ionol concentration 25-350 mkM within 24 hourly cultivations (table 2). Fallowing increase of concentrations 500mkM leads to level suppression of catalase activity in Dunaliella cells (86%).

So, 24 hourly modifications of Dunaliella cells with ionol significantly decreases the quality of reactive oxygen species which causes the increase of catalase activity finally algae bioproductivity. The increase of catalase activity can be related to modification of general nonspecific protective reaction of cells against ionol presence in optimal conditions of cultivation and at high salinity with a significant increase of oxygen species.

In picture 8, have been presented the dependence of quantitative indicators of malonic dialdehyde in cells on various ionol concentrations in mineral medium with 1,5M NaCl (1) and 3,0M NaCl (2). As seen in the picture various concentrations of synthetic antioxidant ionol strongly affects malonic dialdehyde content in cells. So, in optimal regime of cultivation (table 1), was observed decrease of malonic dialdehyde content down to 65-60% in the range of ionol concentrations 25-150 mkM in 24 hourly cultivations. Fallowing increase of concentrations leads to more decrease of MDA and more effective suppression of POL process (tree time – decrease of malonic dialdehyde content) in cells.



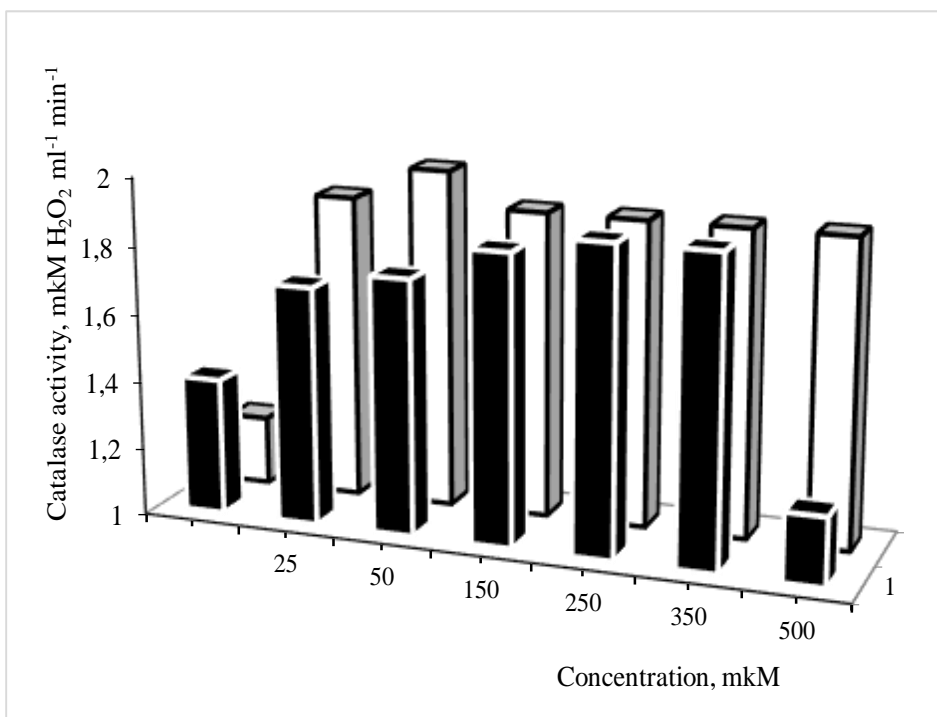


Fig 7. The dependence of catalase activity in *Dunaliella salina* IPPAS D-294 on various concentrations of 2,6 di-tert-butyl cresol grown in mineral medium with 1,5M NaCl (1) and (2) 3,0M NaCl. Temperature 27<sup>0</sup>C, light intensity 16 Wt/m<sup>2</sup>

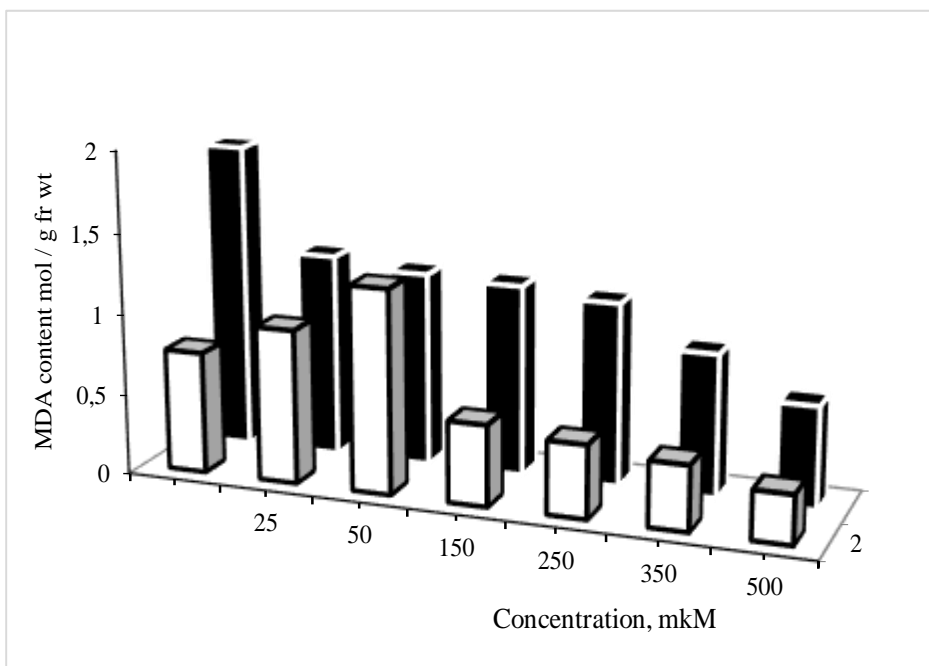


Fig 8. The dependence of MDA content in *Dunaliella salina* IPPAS D-294 on various concentrations of 2,6 di-tert-butyl cresol grown in mineral medium with 1,5M NaCl (1) and (2) 3,0M NaCl. Temperature 27<sup>0</sup>C, light intensity 16 Wt/m<sup>2</sup>

So, 24 hourly modification of *Dunaliella* cells by ionol significantly decreases amount of active oxygen enzymes, which in POL process and finally in amount of algae productivity.

In picture 9, have been presented results of photosynthetic oxygen evolution grown at optimal salinity (1,5M NaCl) and irritated by various acute doses of UV-B light on control *Dunaliella salina* IPPAS D-294 and in cells modified within 24 hourly intensive cultivation with 25 mkM and 50 mkM of 2,6 di-tret-*butyl cresol*. As seen in the picture, in controle cells, irritated by acute doses  $2,2 \cdot 10^3$  Erq/mm<sup>2</sup> functional activity strongly suppressed 30-32%.

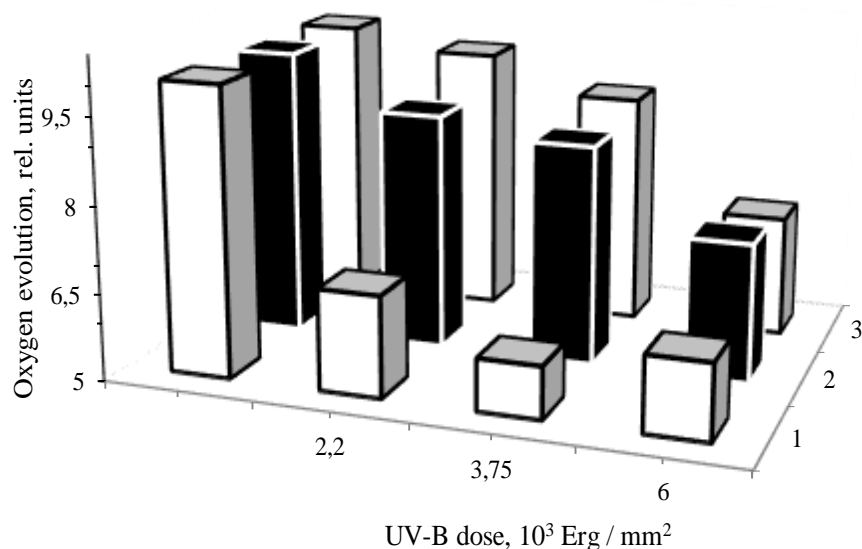


Fig. 9. Photosynthetic oxygen evolution in *Dunaliella salina* IPPAS D-294, grown optimal salinity (1,5 M NaCl) and irritated by various acute doses of UV-B light;

1 – control cells; 2 – cells modified by 2,6 di- tret-*butyl cresol* at concentration 25 mkM; 3 – cells modified by 2,6 di- tret-*butyl cresol* at concentration 50 mkM. Temperature 40<sup>0</sup>C, light intensity 100 Wt/m<sup>2</sup>

Following increase of acute UV-B radiation  $3,75 \cdot 10^3$  Erq/mm<sup>2</sup> leads to deeper suppression (40%) of cell (photo synthetic oxygen evolution) function (table 1). Acute doses  $6 \cdot 10^3$  Erq/mm<sup>2</sup> significantly did not increase the suppression of photo synthetic oxygen evolution, compored to doses  $3,75 \cdot 10^3$  Erq/mm<sup>2</sup>, manifested some stability.

The cells, modified by 2,6 di- tret-*butyl cresol* in concentration 25 mkM under the influence of acute doses of UV-B radiation  $2,2 \cdot 10^3$  Erq/mm<sup>2</sup> show high functional stability 95-96%. The increase of acute doses to  $3,75 \cdot 10^3$  Erq/mm<sup>2</sup> didn't affect on functional activity of modified cells. Acute doses of UV-B radiation  $6 \cdot 10^3$  Erq/mm<sup>2</sup> greatly reduced (77%) photo synthetic oxygen evolution of modified cells (table 2).

The increase of synthetic antioxidant concentration (50 mkM) BHT during cell modification showed that the stability of functional activity is saved on high level at low acute doses  $2,2 \cdot 10^3$  Erq/mm<sup>2</sup> UV-B radiation then suppression character is saved, as in concentration 25 mkM.

In picture 10, have been presented the results of photo synthetic oxygen evolution by control *Dunaliella salina* IPPAS D-294 cells grown at high salinity (3,0M NaCl) and irritated by various doses of UV -B light and the cells modified by 24 hourly intensive cultivation with 25 mkM and 50 mkM concentrations of 2,6 di- tret-*butyl cresol*. As seen in picture, in control cells irritated by acute doses  $15 \cdot 10^3$  Erq/mm<sup>2</sup> functional activity suppressed up to 94%.

Following increase of acute doses  $18 \cdot 10^3$  Erq/mm<sup>2</sup> significantly decreases (74%) cellfunction (table 1). Acute doses  $21 \cdot 10^3$  Erq/mm<sup>2</sup> lead to deeper suppressed 35% of photosynthetic oxygen evolution.

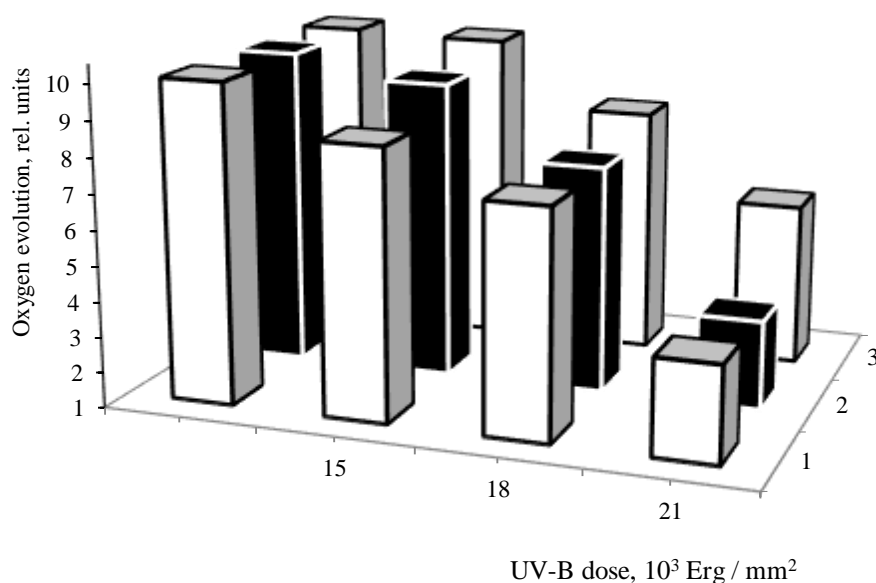


Fig. 10. Photosynthetic oxygen evolution in *Dunaliella salina* IPPAS D-294, grown optimal salinity (3,0 M NaCl) and irritated by various acute doses of UV-B light;

1 – control cells; 2 – cells modified by 2,6 di- *tret-butyl* cresol at concentration 25 mkM; 3 – cells modified by 2,6 di- *tret-butyl* cresol at concentration 50 mkM, Temperature 40°C, light intensity 100 Wt/m<sup>2</sup>

The cells modified by 2,6 di- *tret-butyl* cresol with concentration 25 mkM irritated by acute doses 15\*10<sup>3</sup> Erq/mm<sup>2</sup> of UV-B radiation didn't show functional stability 86% compared to control cells.

Increase of acute doses up to 18\*10<sup>3</sup> Erq/mm<sup>2</sup> suppressed functional activity of cell modified cell (74%). But acute doses 21\*10<sup>3</sup> Erq/mm<sup>2</sup> of UV-B radiation decrease photosynthetic oxygen evolution (37%) cells modified by ionol (table 2).

The increase of ionol concentration (50 mkM) during cell modification showed that stability of functional activity remains at high level (99%) at high doses 15\*10<sup>3</sup> Erq/mm<sup>2</sup> of UV-B radiation (table 3). Acute doses 18\*10<sup>3</sup> Erq/mm<sup>2</sup> of UV-B suppressed functional activity (80%) of cells which differs from control cells where suppressed is 74%

The increase of acute doses 21\*10<sup>3</sup> Erq/mm<sup>2</sup> of UV-B radiation decreases functional activity of cells modified by ionol conditions of high salinity compared to control cells (35%).

### Conclusions:

1. It has been showed that during intensive cultivation of *Dunaliella salina* IPPAS D-294 cells in optimal mineral medium (1,5M NaCl) within 24 hours optic density of cell suspension increases 3-4 times, but in high salinity of mineral medium (3,0M NaCl), bioproductivity decreases 25%. So, data of photosynthetic oxygen evolution points out at significant difference, decrease is about 17%.

2. It was indicated that synthetic antioxidant ionol at low concentrations 25 mkM and 50 mkM in optimal and high salinity of mineral medium stimulated culture growth.

3. Concentrations 25 - 50 mkM of ionol decrease biosynthesis of common chlorophyll amount, also carotenoid amount in cells, grown in optimal and high salinity in mineral medium.

4. The cell modification by various concentrations of ionol in optimal (1,5M NaCl) and high (3,0M NaCl) salinity in mineral medium suppresses functional activity of cells.

5. The cell modification by various concentrations of ionol in optimal (1,5M NaCl) and high (3,0M NaCl) salinity in mineral medium increases catalase activity in cells.

6. The cell modification by various concentrations of ionol in optimal (1,5M NaCl) and high (3,0M NaCl) salinity in mineral medium decreases content MDA and POL process is suppresses in cells.

7. It was established that, the cell modification by various concentrations of ionol in optimal (1,5M NaCl) and high (3,0M NaCl) salinity in mineral medium resistance of *Dunaliella salina* IPPAS D-294 to various acute doses of UV-B light increases.

#### Literature:

1. **Abdullayeva, T.M.** Investigations of scientists on study influence of phytohormones and antioxidants on apoptos at the plants analysis was make up [*Sostavleny issledovaniia uchenykh po izucheniiu vliianiia fitogormonov i antioksidantov Na apoptoz pri analize rastenii.*] [Text] / M.A. Magomedova // South of Russia: ecology, development, 2007, vol. 2, No. 4, p. 44-47.
2. **Alizade, G.I.** Functional activity and UV B tolerance of *Dunaliella* cells modified with synthetic antioxidants under low temperature stress // *Funktsional'naia aktivnost' i ustoichivost' k ul'trafioletovomu izlucheniuiu B kletok Dunaliella, modifitsirovannykh sinteticheskimi antioksidantami, pri nizkotemperaturnom stresse.* [Text] / A.R. Jalilova, I. I. Aliyev, H.H. Magerramova // News of ANAS, Biology and Medical Sciences, 2017, vol. 72, No. 2, p. 106-113.
3. **Alizadeh, G.I.** Lipid peroxidation in *Dunaliella salina* IPPAS D294 cells modified by ionol at high salinity in optimal and low temperature cultivation conditions. [Text] / A.R. Jalilova, I.B. Aliyeva, Kh.Kh. Maharramova, I.I. Aliyev // *European Journal of Biotechnology and Bioscience*, 2020, vol. 1, issue 1, p. 1-3.
4. **Borovkov, A.B.** Semi commercial cultivation of *Dunaliella salina* teod. (Chlorophyta) in the eastern part of Ukraine [Goodwillovich I. N.] // *Algology*, 2014, vol. 24, No. 3, p. 421-425.
5. **Bailly, Ch.** Active oxygen species and antioxidants in seed biology. [Text] // *Seed Science Research*, 2004, vol. 14, issue 2, p. 93-107.
6. **Gate, L.** Oxidative stress induced in pathologies: The role of antioxidants // *Okislitel'nyi stress, indutsirovannyi pri patologiiakh: rol' antioksidantov.* [Text] / G.N. Ba, K.D. Tew, H. Tapiero // *Biomedicine& Pharmacotherapy*, 1999, vol. 53, issue 4, p. 169-180.
7. **Hamid, S.** Antioxidant System Response in Green Microalga *Chlorococcopsis minuta* Against Nutrient Stress in Growth Media. [Text] / G. Sibi // *Asian Journal of Biological Sciences*, 2018, vol. 11, issue 4, p. 210-216.
8. **Ivanishchev, V.V.** Manifestations of oxidative stress in triticale seedlings under short term exposure to sodium chloride [Text] / N.N. Zhukov // *Butlerov Communications*. 2017, vol. 52, No. 11, p. 123-130.
9. **Kolupaev, Yu.E.** Mechanisms of plant adaptation to hypothermia: the role of the antioxidant system [Text] / E.I. Gorelova, T.O. Iastreba // *Bulletin of Kharkiv national agrarian university. Series: Biology*, 2018, issue 1 (43), p. 6-33.
10. **Matés, J.M.** Effects of antioxidant enzymes in the molecular control of reactive oxygen species. [Text] // *Toxicology*, 2000, vol. 153, issue 1-3, p. 83-104.
11. **Namiki, M.** Antioxidants, antimutagens in food. *Critical Reviews in Food Science and Nutrition*, 1990, vol. 29, issue 4, p. 273-300.
12. **Riaz, A.** Oxidative Stress and Antioxidant Defense Mechanisms in Plants Under Salt Stress. In *Plant Abiotic Stress Tolerance: Agronomic, Molecular and Biotechnological Approaches*. Eds. Mirza Hasanuzzaman et al./ H. Sajjad, M.A. Anjum, M.F. Khalid et al. // Springer Nature Switzerland AG, 2019, p. 191-205.
13. **Trunova, T.I.** Plants and low temperature stress [*Rasteniia i nizkotemperaturnyi stress*]. Reported at the 64th annual Timiryazev reading on June 3, 2003. Moscow, 2007, 54 p.

