herbicides in the mixture with biological preparations, the coefficient of morphostructure of epidermis was 0,80 – 0,90 and this corresponds to mesomorphic type of leaf area and conforms with the highest indices of plant productivity.

Generelized data of the research into morphostructure of leaf epidermis when applying herbicides of different chemical classes and their combinations with biologically active substances show that the leaf apparatus of spring barley with optimal structure is formed when the index of morphostructure is 0,7-0,9, its value is 0, 9 -1,0 and higher. In this case leaf apparatus with xerophytic properties is formed which leads to the decrease of leaf area and its productivity.

Thus anatomical changes in the epidermis structure of the leaf apparatus under the application of phisiologically active substances is a direct reflection of the level of the preparations influence on metabolic processes in plants and may serve for disclosing of mode of action of investigated preparation on the plant body at different stages of its growth.

EFFECT SIZE OF SOME FACTORS INFLUENCING PRODUCTIVITY INDEXES IN *DUNALIELLA SALINA* TEOD. CULTURE

Mil'ko S.I. [1], **Komaristaya V.P.** [1], **Rudas A.N.** [2]
[1] *V.N. Karazin Kharkov national university, Kharkov*
[2] *"Betacar-X" LLC, Kharkov*

The number of environmental factors induces b-carotene accumulation in the cells of microalga Dunaliella salina, which serves as a source of its industrial manufacturing. To so called carotenogenesis factors there are attributed elevated medium concentrations of osmotically active salts (medium density, salinity), elevated illuminance, elevated or reduced temperature, addition of carbon sources, deprivation of nutrients (nitrogen and phosphorus). b-carotene accumulation is thought to occur in the cu ltures, the growth in which is inhibited by unfavorable values of these factors. Therefore, culturing most often is performed in two stages: at the first stage cell concentration is grown, then the culture is exposed to carotenogenesis factors. Maximum intensity of carotenogenesis is thought to be achieved at simultaneous effect of all the factors [1]. Nevetherless, most researchers use in their work only some factors and their combinations, e.g. elevated salinity and illuminance, sometimes in combination with nitrogen deprivation. However, the input of each certain factor as well as their interactions into culture growth inhibition and carotenogenesis remains not investigated. And the two stage culturing scheme of D. salina is rather time and labor consuming.

The goal of present research is to elucidate factors having the maximum effect on b-carotene accumulation in D. salina cells at the minimal culture growth inhibition for possible use in single stage culturing of D. salina. For that, in multiple-factor experiment design the effects of medium density, illuminance, carbon, nitrogen and phosphorus sources availability on the yield of cells and b-carotene content in them were determined and the effect sizes of each factor on culture growth and carotenogenesis were calculated [2].

There were investigated 5 factors taken in 2 gradations selected on the basis of reference data and preliminary field and laboratory research, so that the culture preserved the ability to grow: medium density 1,10 and 1,15 g/cm^3, illuminance 2 and 5 klux, carbon source being dissolved atmospheric CO_2 only and with the addition of 100 mg/L $NaHCO_3$, nitrogen and phosphorus deprived and minimal concentrations for growth support added. The peculiarity of this experimental design was that nitrogen and phosphorus concentrations in the medium were kept at relatively constant level by the periodical additions of suitable quantities of nutrient salts [3]. Full experimental design consisted of 2^5=32 variants. For statistical processing of the results nonparametric tests were used, as preliminary research had shown that cell concentrations and b-carotene content in *D. salina* did not fit normal distribution [3].

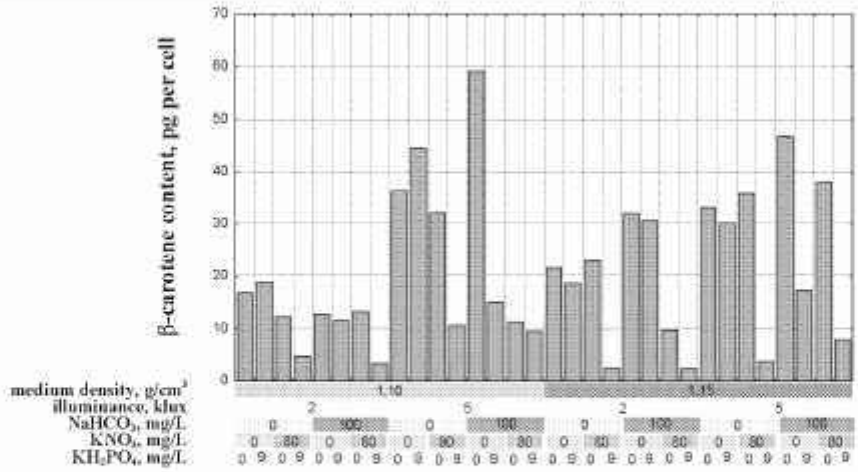

Fig. 1. Effect of carotenogenesis factors on β-carotene content in *D. salina* cells on 28th day of culture growth

Fig. 1 and 2 represent respectively the values of b-carotene content in the algal cells and cell concentrations on 28th day of growth (stationary phase) in all 32 variants of the experiment. The maximum b-carotene accumulation was observed not at simultaneous effect of all the factor values, which were anticipated to promote carotenogenesis (medium density 1,15 g/cm³, illuminance 5 klux, 100 mg/L $NaHCO_3$, nitrogen and phosphorus deprivation), but at lower medium density (1,10 g/cm³) (fig. 1). Stimulating effect of nutrient deprivation on carotenogenesis not always occurred at $NaHCO_3$ additions (fig. 1), though concentrated stock solution of $NaHCO_3$ was shown not to contain nitrogen or phosphorus traces.

Nitrogen deprivation substantially inhibited culture growth (fig. 2). Growth inhibition by deprivation of phosphorus manifested to greater extent at the higher medium density (fig. 2). It is likely that high salinity of medium solution decreased solubility and availability of phosphate traces to the cells. The higher illuminance promoted both more intensive culture growth and b-carotene accumulation in the cells (fig. 1 and 2).

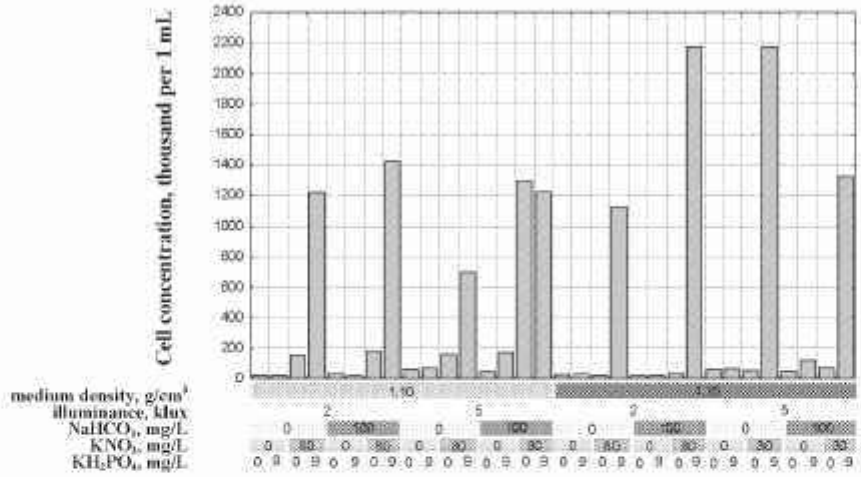

Fig. 2. Effect of carotenogenesis factors on concentration of *D. salina* cells in the culture on 28th day of growth

Kruskal-Wallis test showed that only illuminance, nitrogen and phosphorus availability influenced statistically significantly both cell concentration in stationary phase and cell b-carotene content (table 1). The effect of culture medium density and carbon source in the range of the factors values studied appeared insignificant.

Table 1. Values of Kruskal-Wallis test H for investigated factors and variables (df=1; $p \leq 0,05$; $\chi^2_{tabl} = 3,84$)

Variable Factor	Cell concentration	Cell b-carotene content

Medium density	0,51	0,32
Illuminance	4,07*	4,14*
$NaHCO_3$ addition	0,26	0,57
Nitrogen availability	13,09*	9,32*
Phosphorus availability	4,30*	7,36*

Note: * - H^3 χ^2_{tabl}

For studied factor value gradations the maximum effect size on culture growth and carotenogenesis was caused by nitrogen availability, and the minimum – by illuminance (table 2). Phosphorus deprivation caused almost the same little effect on culture growth as lowered illuminance and practically the same effect as nitrogen deprivation on carotenogenesis (table 2).

Table 2. Influence power η^2 of carotenogenesis factors on cell concentration and b-carotene content in D. salina Teod. culture (df=1; $p \leq 0,05$; $\chi^2_{tabl}=3,84$)

Variable Factor		Cell concentration	Cell b-carotene content
Medium density	$\eta^2 \pm s_{\eta 2}$	0,02±0,03	0,01±0,03
	χ^2	0,62	0,31
Illuminance	$\eta^2 \pm s_{\eta 2}$	0,13±0,03	0,13±0,03
	χ^2	4,03*	4,03*
$NaHCO_3$ addition	$\eta^2 \pm s_{\eta 2}$	0,01±0,03	0,02±0,03
	χ^2	0,31	0,62
Nitrogen availability	$\eta^2 \pm s_{\eta 2}$	0,42±0,03	0,30±0,03
	χ^2	13,02*	9,3*
Phosphorus availability	$\eta^2 \pm s_{\eta 2}$	0,14±0,03	0,24±0,03
	χ^2	4,34*	7,44*

Note: * - χ^2 > χ^2_{tabl}

That means, that phosphorus deprivation to lesser extent than nitrogen deprivation inhibited culture growth, but to greater extent stimulated carotenogenesis, and therefore phosphorus deprivation can be considered to be promising method for carotenogenesis induction in single-stage *D. salina* culture.

1. Масюк Н.П. Морфология, систематика, экология, географическое распространение рода Dunaliella Teod. и перспективы его практического использования. – Киев: Наук. думка, 1973. – 244 с.

2. Атраментова Л.О., Утєвська О.М. Статистичні методи в біології: підручник для вузів.- Х.: Вид-во ХНУ, 2007. - 286 с.

3. Комаристая В.П., Антоненко С.П., Рудась А.Н. Культивирование Dunaliella salina Teod. при субоптимальных концентрациях азота и фосфора и исключении их из среды // Альгология. – 2010. – Т. 20, № 1. – С. 42-55.

THE INTERACTIONS OF LEAD AND SALICYLIC ACID ON GROWTH AND AMOUNTS OF CHLOROPHYLL IN ROOT AND SHOOT OF TWO CULTIVARS OF *BRASSICA NAPUS* L. UNDER HYDROPONIC CULTURE

***Ranjbar M.[1], Boroumand-Jazi Sh.[2]**

[1,2] Department of Biology, Islamic Azad University, Falavarjan Branch, Esfahan, Iran.

**Corresponding Author: ranjbar@iaufala.ac.ir*

Lead is a danger heavy metal, which plotted the environment. Toxicity of lead on plant caused inhibition of root growth and the amount of chlorophyll (Mathe-Gaspar, 2002). It has been proposed that salicylic acid acts as an endogenous signal molecule responsible for inducing abiotic stress tolerance in plants (Gunes, 2007). In this research the effect of lead poison on some parameters of growth and amount of chlorophyll on 20 day old seedlings