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COMPLEXES OF BISCITRATOGERMANATES AND BISCITRATOSTANATES WITH METALS ARE MODIFIERS OF *Bacillus thuringiensis* var. *israelensis* PEPTIDASES AND *Penicillium canescens*, *Cladosporium cladosporioides* AND *Aspergillus niger* α- GALACTOSIDASES ACTIVITIES

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The aim of the research was to study the effect of a number of germanium and stanum coordination compounds (compounds 1–8) as modifiers of peptidases and α-galactosidases activity. The coordination compounds of the same type of structure based on chelating ligand that is biologically activities citric acid were investigated as enzymes effectors. Two types of complexes: 1) $[M(H_2O)_6][Ge(HCitr)_2]\cdot 4H_2O$ (M = Mg(1), Mn(2), Co(3), Ni(4), Zn(5)), containing biscitrate-germanate anion ($[Ge(HCitr)_2]^{2-}$), and 2) $[M(H_2O)_6][Sn(HCitr)_2]\cdot 4H_2O$ (M = Mg(6), Co(7), Ni(8)), containing biscitrate-stanate anion and various hexaaquacations ($[M(H_2O)_6]^{2+}$, M= Mg, Mn, Co, Ni, Zn) were studied. It is shown that the compound 6, which is biscitrate-stanate complex containing magnesium ions as metal, can be used for the stimulation of *B. thuringiensis* var. *israelensis* IMV B-7465 peptidase 1 and peptidase 2 collagenase activity by 20–25%. Compounds 1 (biscitrate-germanate complex containing magnesium ions as metal) and 7 (biscitrate-stanate complex containing cobalt ions as metal), and also compound 6 in a concentration of 0.001% are able to increase elastolytic activity of peptidase 1 by 55–58%. However, compound 7 has shown the greatest activating effect. It increased the elastolytic activity of peptidase 2 by 100–140% (at both tested concentrations). This goes to prove that the compound 7 can be used henceforth as effector of peptidase 2 elastolytic activity. While investigating the effect of the considered coordination compounds on the *Penicillium canescens*, *Cladosporium cladosporioides* and *Aspergillus niger* α-galactosidase activity, it has been found that, when using a number of complexes (1–2 and 4–8), there is a slight increase (by 12–20%) of *P. canescens* enzyme activity, and the maximal effect (~20%, concentration 0.01%) was provided by complex 6.

Key words: *B. thuringiensis* var. *israelensis* IMV B-7465 peptidases, with collagenase, elastolytic and fibrinolytic activity, α-galactosidases, biscitrate-germanate and biscitrate-stanate complexes.

Despite the fact that enzymes are present in animals, plants and microorganisms, the most technologically advanced sources of these biopolymers obtaining are microorganisms since they multiply rapidly and perform synthesis under man-controlled conditions. The following enzymes were taken for the study: 1) *Bacillus thuringiensis* peptidases, exhibiting a wide range of activities towards insoluble protein substrates such as collagen, elastin and fibrin; 2) *Penicillium canescens, Aspergillus niger, Cladosporium cladosporioides* α-galactosidases. Peptidases are classified as hydrolases which action is aimed at accelerating the hydrolysis of peptide bonds in proteins and peptides. Their

important properties are high catalytic activity and selective action. Peptidases are widely used in various industries: in leather industry — for hides softening, in film industry — to dissolve the gelatin layer at films regeneration, in the production of detergents, as additives to remove the contaminations of protein nature, in medicine — for the treatment of inflammation, thrombosis and other complications.

α-Galactosidases are glycosidases capable of cleaving, usually preserving their optical configuration, the terminal residues of non-reducing α-D-galactose from α-D-galactosides, including galactooligosaccharides, galactomannans, galactolipids. This allows the use of α-galactosidase in food biotechnology, in particular in sugar industry to improve its quality and yield increase. Since in sugar production from sugar beets the concentration of raffinose, stachyose and verbascose increases, complicating the crystallization of the sugar, the use of α-galactosidase not only eliminates hindrance for crystallization caused by a high concentration of mentioned oligosaccharides, but also promotes the formation of crystals.

For the successful application of these enzymes in various biotechnological processes, important aspect is to provide methods to enhance their activity. The main factors influencing the growth and metabolism of microorganisms, as well as enzymes synthesis, include physicochemical conditions of cultivation, composition of culture medium, and the introduction of substances promoting the increase in the yield of the enzyme that appears as its activity increase. At the same time, the compounds capable of enzymes modifying with both stimulatory and inhibitory effects on their activity can be used.

In this regard, the aim of the work was to study the effect of a number of coordination compounds of germanium and stanum as modifiers of peptidases and galactosidases activity.

Materials and Methods

The objects of investigations were *Bacillus thuringiensis* var. *israelensis* IMV B-7465 extracellular peptidases with collagenase, elastolytic and fibrinolytic activity, as well as *Penicillium canescens* 239, *Cladosporium cladosporioides* 189 and *Aspergillus niger* 185sh α-galactosidases.

B. thuringiensis var. *israelensis* IMV B-7465 is isolated from the waters of Snake Island (Black Sea) and is in the collection of live cultures of the Department of Microbiology, Virology and Biotechnology of Mechnikov Odesa National University. *P. canescens, C. cladosporioides* and *A. niger* are obtained from the collection of the Department of Physiology and Systematics of Micromycetes of Zabolotny Institute of Microbiology and Virology of the National Academy of Sciences of Ukraine.

For peptidases synthesis, *B. thuringiensis* var. *israelensis* IMV B-7465 was cultured in a liquid medium of the following composition (g/l): KH_2PO_4 — 1.6; $MgSO_4 \cdot 7H_2O$ — 0.75; $ZnSO_4 \cdot 7H_2O$ — 0.25; $(NH_4)_2SO_4$ — 0.5; maltose — 1.0; gelatin — 10.0; yeast autolysate — 0.15; pH — 6.5–6.7 [1]. The strain was grown for 24 h in shaker flasks (200 ml of medium, 28 °C, 250 rev/min). The inoculum was prepared by culturing the strain in a suitable medium for 24 h and seeded into the flasks in an amount of 10^5–10^6 CFU/ml.

For the synthesis of α-galactosidases, *A. niger, C. cladosporioides, P. canescens* were grown in the medium of the following composition (g/l): soybean meal — 15.0; urea — 0.3; $(NH_4)_2SO_4$ — 1.4; KH_2PO_4 — 2.0; $MgSO_4 \cdot 7H_2O$ — 0.3; $CaCl_2$ — 0.3; yeast autolysate — 0.5; pH 5.2. Submerged cultivation was performed in Erlenmeyer flasks (500 ml) for 96 h at 25 °C and stirring speed of 220 rev/min.

Preparations of peptidases and α-galactosidases are isolated from culture supernatants of producers *B. thuringiensis* var. *israelensis* and *A. niger, C. cladosporioides, P. canescens*, respectively, purified to homogeneity by column with neutral and charged TSK-DEAE-650 (M) and Toyopearl HW-55 (Toyosoda, Japan) gels as described previously [2–4]. As a result of *B. thuringiensis* var. *israelensis* complex enzyme preparation purification, peptidase 1 was obtained with specificity to collagen and elastin, and peptidase 2 with specificity to elastin, collagen and fibrin.

The protein content is evaluated by the Lowry method [5], using bovine serum albumin as a standard.

α-Galactosidase activity was determined using *p*-nitrophenyl-α-D-galactopyranoside [6]. The degree of the substrate hydrolysis was determined by colorimetric method. One unit of activity was defined as the amount of enzyme catalyzing the hydrolysis of 1 μM of the substrate per 1 min under standard test conditions (temperature 37 °C, pH 5.2).

The specific activity of *A. niger, C. cladosporioides, P. canescens* α-galactosidases was 12, 32 and 50 U/mg of protein.

Elastolytic activity was determined colorimetrically by the color intensity of the solution upon the enzymatic hydrolysis of elastin stained with Congo red using Trombridg et al. method [7] in Bondarchyuk et al. modification [8]. Incubation mixture contained 5 mg of elastin, 2.0 ml of 0.01 M Tris-HCl buffer (pH 7.5) supplemented with 0.005 M $CaCl_2$ and 1 ml of the tested preparation solution. The mixture was incubated for 5 h at 37 °C. Non-hydrolyzed elastin was separated by centrifugation at 8 000 g, 10 min. The color intensity was measured using the SF-26 spectrophotometer at 515 nm. Activity was calculated according to the standard curve, which was obtained during supernatant color measurement at full enzymatic hydrolysis of known amounts of elastin stained with Congo red. One unit of activity was defined as the amount of enzyme catalyzing the hydrolysis of 1 mg of substrate per 1 min under standard conditions.

Collagenase activity was measured by ninhydrin colorimetric method [9]. In order to get that done, incubation mixture, which contained 10 mg of collagen, 2.5 ml of 0,01 M Tris-HCl buffer (pH 9.0–10.0), and 1 ml of the tested preparation was heated on a steam bath for 3 h at 37 °C. Thereafter, 0.1 ml of the reaction mixture was transferred into test tubes that contained 0.5 ml of 4% solution of ninhydrin in a mixture with 0.2 M citrate buffer. Incubation was carried out for 20 min in boiling water bath, thereafter into the cooled mixture 5 ml of *n*-propanol 50% solution was added and allowed to stand for 15 min at room temperature. The digestion products were determined using the SF-26 spectrophotometer at wavelength of 600 nm. From the standard curve constructed for free L-leucine, an equivalent amount of micromoles of amino acids released during hydrolysis was determined. One unit of activity is equivalent to 1 μmol of L-leucine, released from the substrate for 3 h of hydrolysis at 37 °C.

Fibrinolytic activity was measured by Masada method [10], fibrin obtained from human blood plasma was used as a substrate. The reaction mixture contained 1 mg of fibrin, 1.8 ml of 0.01 M Tris-HCl buffer (pH 7.5); 0.005 M $CaCl_2$ and 0.2 ml of the tested preparation were added. The incubation mixture was kept for 30 min at 37 °C. The digestion products were determined using the SF-26 spectrophotometer at 275 nm. One unit of activity was defined as the amount of the enzyme that increases the extinction of the reaction mixture by 0.01 for 1 min.

The specific activity of peptidase 1 was 442 U/mg of protein (elastolytic activity) and 212.7 U/mg of protein (collagenase activity), while the specific activity of peptidase 2 was 289.5 U/mg of protein (elastolytic activity), 345.8 U/mg of protein (collagenase activity) and 250.5 U/mg of protein (fibrinolytic activity). Protein content was 0.1 mg/ml.

Coordination compounds synthesized by developed techniques [11] and described by the set of physical and chemical methods of research were tested at concentrations of 0.01 and 0.001% as enzyme effectors [12–14]. Complexes $[M(H_2O)_6][Ge(HCitr)_2]\cdot 4H_2O$ (M = Mg(**1**), Mn(**2**), Co(**3**), Ni(**4**), Zn(**5**)) contain biscitrate-germanate anion ($[Ge(HCitr)_2]^{2-}$), and complexes $[M(H_2O)_6][Sn(HCitr)_2]\cdot 4H_2O$ (M = Mg(**6**), Co(**7**), Ni(**8**)) — biscitrate-stanate anion and various hexaaquacations ($[M(H_2O)_6]^{2+}$, M = Mg, Mn, Co, Ni, Zn) (Fig. 1). Coordination compounds were dissolved in distilled water.

All experiments were performed at least 3–5 repetitions. Statistical processing was carried out by standard method using Student's t-test at 5% significance level [15]. The results presented graphically were obtained using Microsoft Excel 2003 program. The values at $P < 0{,}05$ were regarded as credible.

Results and Discussion

The results are a continuation of previous studies [16] concerning the effect of various, including bivalent, metal cations, several anions and chemicals, including chelating, capable of binding with metal ions (1,10-phenanthroline, ethylenediaminetetraacetic acid), on the activity of a number of enzymes. Since it is known [17] that the combined presence of several metal ions in the solution can have a variety of effects: stimulation (synergism), attenuation of one metal ion action on enzymes activity, so for the first time coordination compounds of the same type of structure based on chelating ligand that is biologically active citric acid were taken as enzymes effectors. It is known that the ligand choice has a significant impact on the ability of coordination compounds exhibit both inhibitory and activating effect on the enzyme.

Two types of complexes have been investigated: 1) $[M(H_2O)_6][Ge(HCitr)_2]\cdot 4H_2O$ (M = Mg(**1**), Mn(**2**), Co(**3**), Ni(**4**), Zn(**5**)), containing biscitrate-germanate anion ($[Ge(HCitr)_2]^{2-}$); 2) $[M(H_2O)_6][Sn(HCitr)_2]\cdot 4H_2O$

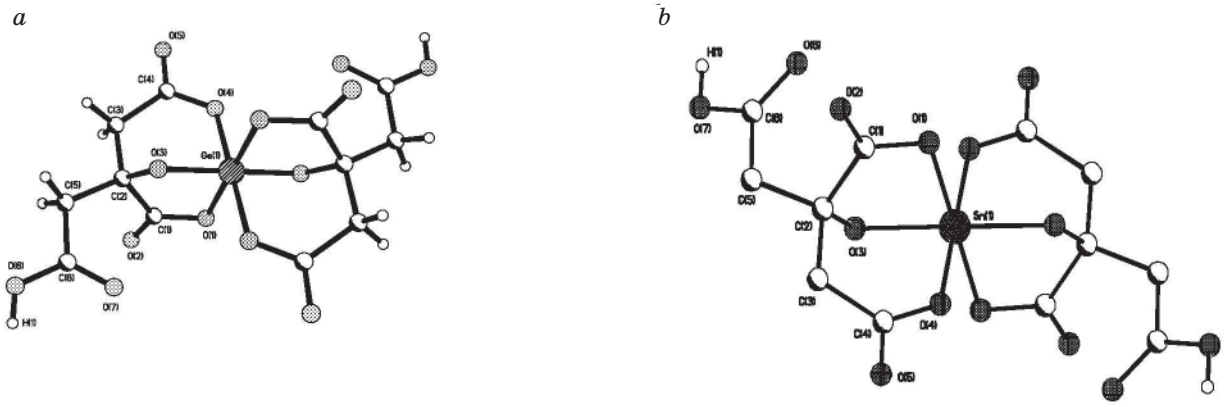

Fig. 1. **The structures of complex biscitrate-germanate (*a*) and biscitrate-stanate various hexaaquacations (*b*) anions in complexes 1–8**

(M = Mg(**6**), Co(**7**), Ni(**8**)) containing biscitrate-stanate anion and various hexaaquacations ($[M(H_2O)_6]^{2+}$, M = Mg, Mn, Co, Ni, Zn).

The peculiarity of such compounds is that in one molecule there are two different metal ions and their chelating agent. Study of a number of effectors makes it possible to trace the influence of individual metal ions and combination thereof, as well as the molecule as a whole on the investigated enzymes activity.

Study of the effect on *B. thuringiensis* var. *israelensis* peptidase 1 and peptidase 2 collagenase activity have shown that all compounds except 6 (Mg-Sn) inhibit the activity (Fig. 2), at that for the peptidase 1 there is an inverse dependence on the concentration of effector used: more marked inhibition when used its lower concentration (0.001%). This suggests a complex dependence nature of enzymes inhibition in these cases, which is a consequence of not only enzymes-binding capacity of these effectors, but also their substrates-binding capacity. In solution these compounds dissociate:

$$[M(H_2O)_6][Ge(HCitr)_2] \leftrightarrow [M(H_2O)_6]^{2+}{}_{sol} + [Ge(HCitr)_2]^{2-}{}_{sol};$$

$$[M(H_2O)_6][Sn(HCitr)_2] \leftrightarrow [M(H_2O)_6]^{2+}{}_{sol} + [Sn(HCitr)_2]^{2-}{}_{sol}.$$

In this case, complex anion is stable and is not destroyed in the pH range from 5.0 to 7.5 and a temperature of 37 °C that are corresponding to the experimental conditions.

M^{2+}sol reacts with both the enzyme and the substrate, i.e. there is a competition between them, which in many cases is culminated in formation of the ternary complex, as determined by X-ray crystallographic analysis of carboxypeptidase [18].

The process becomes even more complicated when you consider that biscitrate-germanate (stanate) anion is also able to bind to the enzyme, substrate by electrostatic interaction, the presence of active carboxyl groups, and complexing ability of germanium and stanum. By comparison the action of effectors **1** and **6** with the same cations $[M(H_2O)_6]^{2+}$ on peptidase 1 collagenase activity it has been found (Fig. 2, *a*) that at higher concentration, a slight inhibition of ~ 5% for the **1** and activation by ~ 20% for the **6** is observed, which can only be explained by the specificity of stanum interaction compared with germanium, due to differences in their radii ($r(Ge^{4+})$ = 0,053 nm, $r(Sn^{4+})$ = 0,069 nm) and, respectively, in the electron density.

The consistent pattern of influence of the studied effectors on enzymes collagenase activity is maintained when replacing peptidase **1** to **2**. Effector **6** stands apart — only it increases the activity up to 120% at a concentration of 0.001% and somewhat smaller (110%) — at a concentration of 0.01%. All other effectors to a far lesser extent affect the activity of peptidase 2 (Fig. 2, *b*), indicating that different functional groups participate in the processes of effectors and enzymes binding.

A somewhat different picture is observed when considering the impact of the tested effectors on the peptidase 1 elastolytic activity (Fig. 3, *a*). According to the degree of inhibition (concentration of 0.01%), the effectors form a series: **1** — no effect, **8** <= **4**, **5**, **6** <**7**, and activation at the same concentration is observed in two cases: **2** and **3** in the range of 140%. At a concentration of

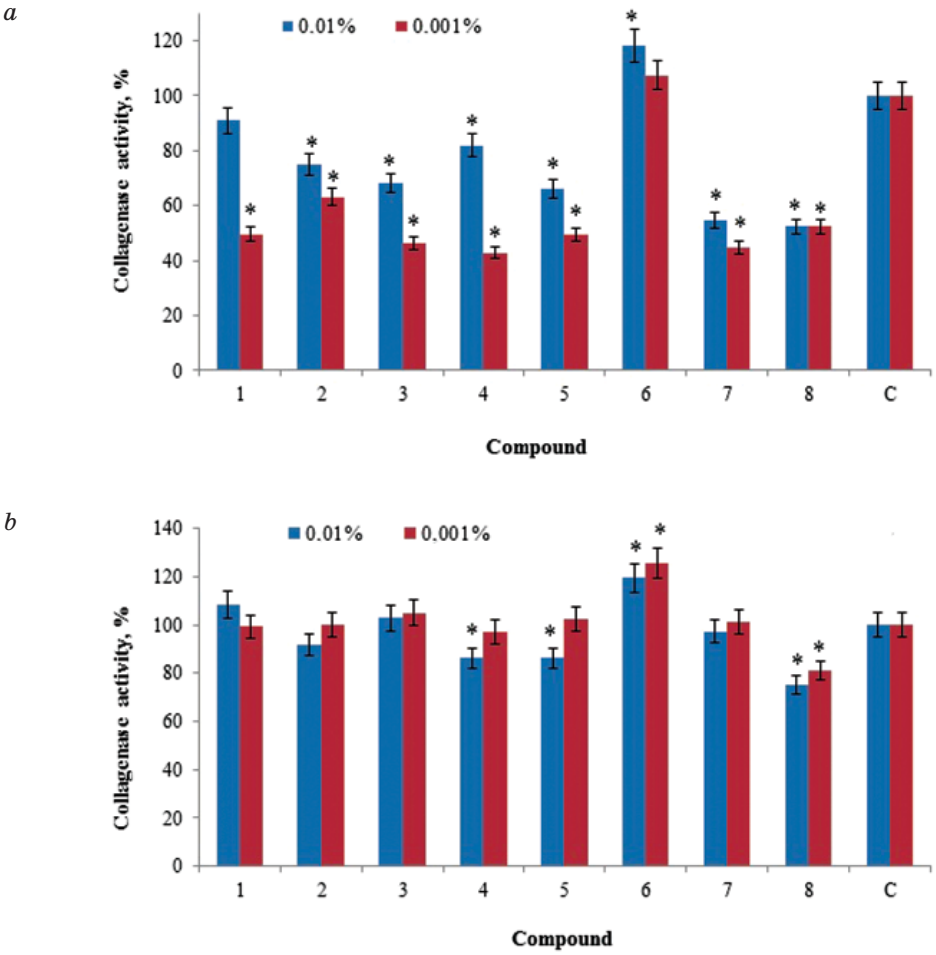

Fig. 2. **Coordination compounds effect on *B. thuringiensis* var. *israelensis* IMV B-7465 peptidase 1 (*a*) and peptidase 2 (*b*) collagenase activity**

Hereinafter: * — P <0,05; C — control

0.001% activation is observed in three cases, **1** = **7**> **6**.

Consequently, the mechanism of effectors with the enzyme and the substrate interaction is specific and varies depending on the type of activity studied. The inhibition of peptidase 1 elastolytic activity by the compounds **8, 4, 5, 6, 7** can be explained by their preferential binding to the substrate, which prevents further adsorption and enzyme action [16]. These processes are significantly affected by the nature of the effector, so for **2** and **3** similar regularity is not observed. Elastolytic activity increase at low concentrations of **1, 7, 6** probably can be explained by the formation of ternary complex "effector-enzyme-substrate", in which an effector acts as link and participates in the formation of catalytically active conformation of the enzyme.

It is characteristic that, when replacing peptidase 1 to peptidase 2, the overall picture of effectors influence on elastolytic activity has changed. This once again confirms the position that the nature of the enzyme contributes to the change in the mechanism of its interaction with the substrates and effectors. Of all the examined compounds, the **7** only significantly increases elastolytic activity (~240% — at 0,001% and ~ 200% — at 0.01%) (Fig. 3, *b*.). It should be noted that the same complex **7**, in the case of peptidase 1, at a concentration of 0.001% increased the activity (Fig. 3, *a*), but in a less degree (160%), at that the increase in the concentration up to 0.01% caused the opposite effect (inhibition reached 90%).

The specificity of each effector interaction with peptidase 2 is seen when analyzing their effect on the enzyme fibrinolytic activity (Fig. 4): the complexes **3–5** and **8** exert slight impact, inhibition by the **1** and **6** is similar due to the presence in their composition of germanium (**1**) and stanum (**6**), at that the **6** is more active, as noted above with respect to collagenase activity. The most active (131%) at

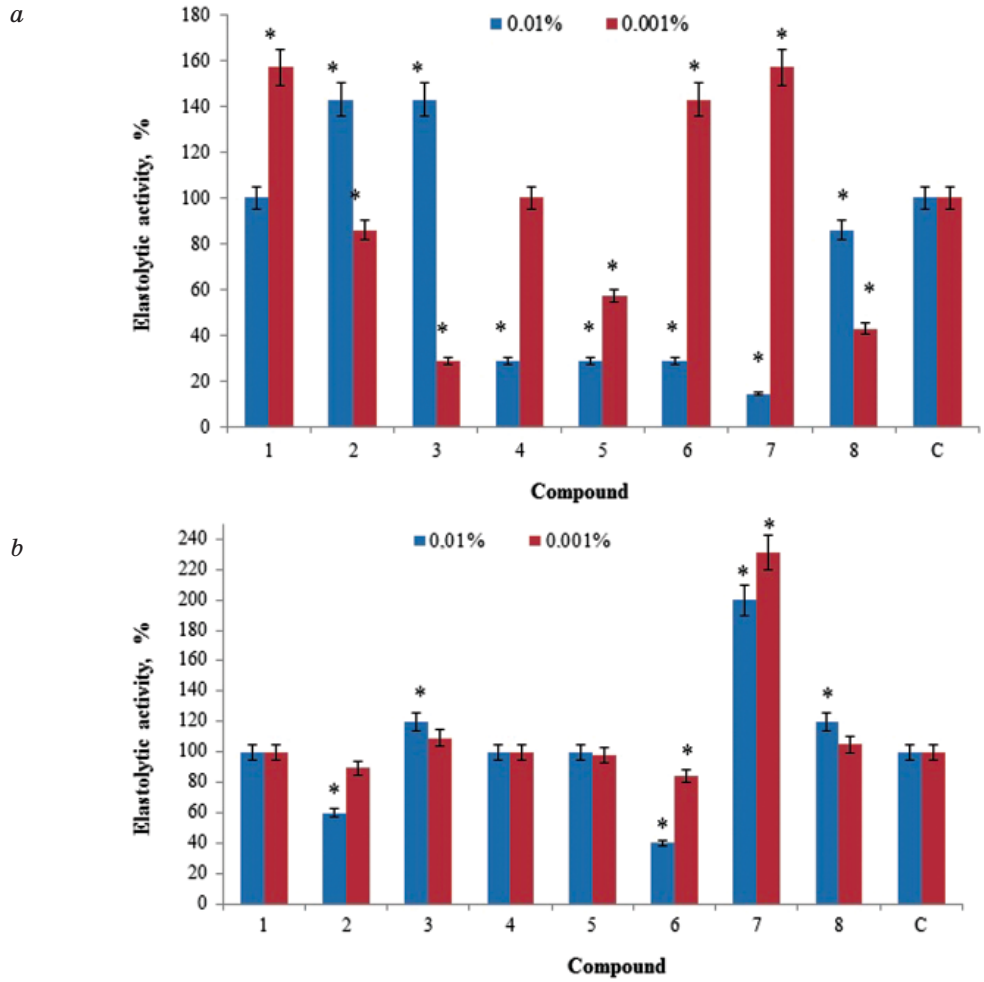

Fig. 3. **Coordination compounds effect on *B. thuringiensis* var. *israelensis* IMV B-7465 peptidase 1 (*a*) and peptidase 2 (*b*) elastolytic activity**

a concentration of 0.01% is the effector **7** with maximal elastolytic activity of peptidase **2**.

In studying of the tested coordination compounds **1–8** influence on *P. canescens* (Fig. 5, *a*), *C. cladosporioides* (Fig. 5, *b*) and *A. niger* (Fig. 5, *c*) α-galactosidase activity, it has been found that when using the complexes **1–2** and **4–8**, a slight (12–20%) increase in *P. canescens* enzyme activity is registered, and the maximal effect (~20%, the concentration of 0.01%) the **6** complex exerted (Fig. 5, *a*).

In other cases, the observed changes in the enzyme activity are negligible and come in a range of experimental error. This difference in their behavior compared with peptidase 1 and 2 is explained by the different nature of the enzymes studied. Catalysis by α-galactosidase does not include metal ions, microbial α-galactosidase usually are independent of metals enzymes that are not sensitive to the action of chelating agents [19]. Apparently, the coordination compounds **1–8** do not bind to catalytically active groups of α-galactosidase. Because of this, there is no formation of more accessible to the substrate its molecule conformation and the substrate at the active center orientation [20].

Based on the evidence found it can be assumed that the considered effectors compete with the substrate for binding sites in the enzyme active center. At the same time, they can interact with various groups of protein molecule, regulatory regions located outside of the active center, but influence on its catalytic functions. As a result of ternary complex enzyme-substrate-effector formation a conformational rearrangement ceases, the molecule is surrounded by other functional groups which provide appropriate catalytic conversions [21]. It should be noted that enzymes catalytic activity may vary substantially depending on the type of effectors interaction. The study of these processes is of great theoretical and practical importance.

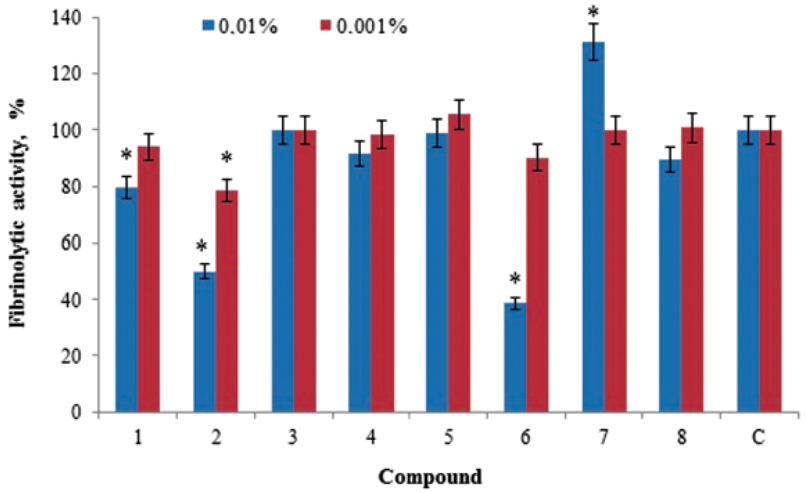

Fig. 4. **Coordination compounds effect on *B. thuringiensis* var. *israelensis* IMV B-7465 peptidase 2 fibrinolytic activity**

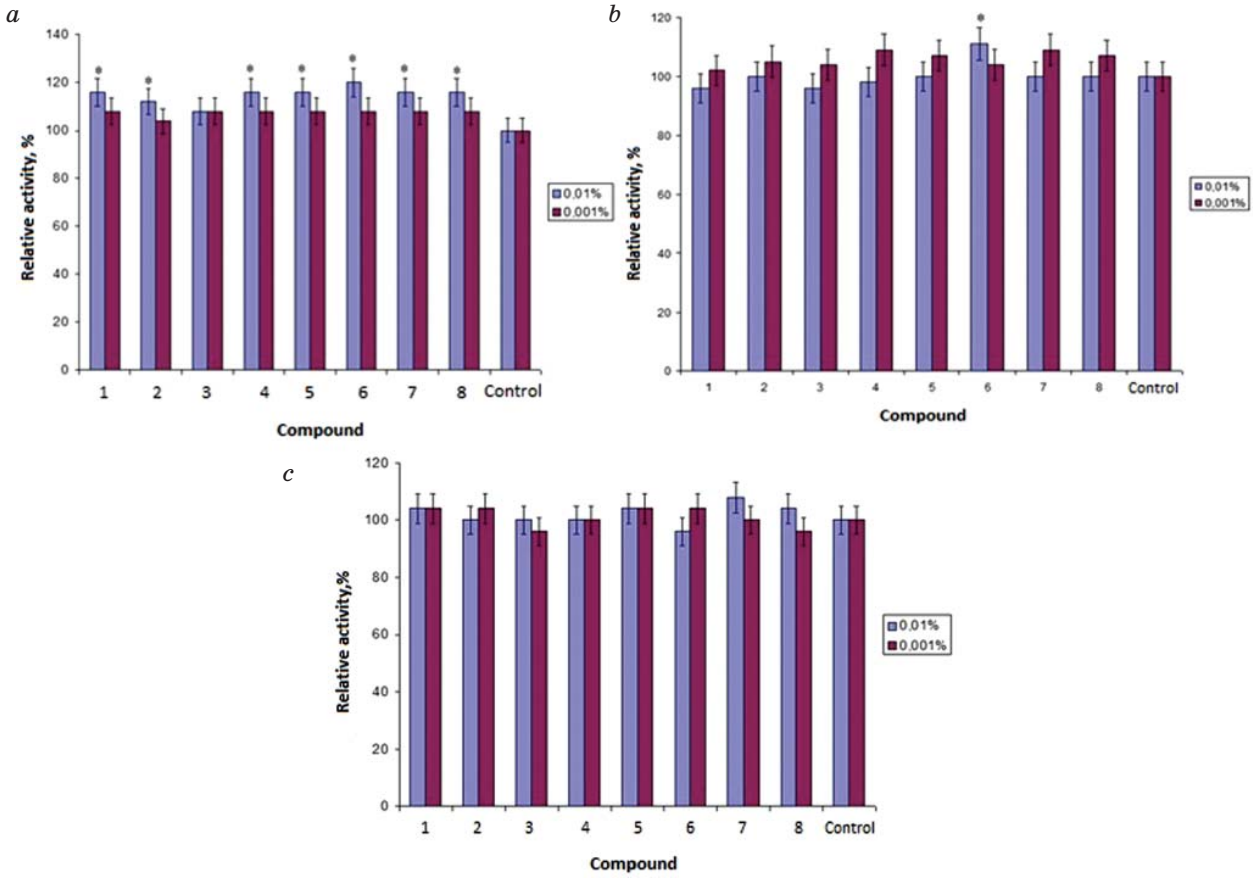

Fig. 5. **Various compounds effect on *Penicillium canescens* (*a*), *Cladosporium cladosporioides* (*b*), *Aspergillus niger* (*c*) α-galactosidase activity**

Thus, the compound **6**, which is biscitrate-stanate complex containing magnesium ions as metal, can be used to stimulate collagenase activity of *B. thuringiensis* var. *israelensis* IMV B-7465 both peptidases by 20–25%. Compounds **1** (biscitrate-germanate complex containing magnesium ions as metal) and **7** (biscitrate-stanate complex containing cobalt ions as metal), as well as **6** at a concentration of 0.001% can increase elastolytic activity of peptidase 1 by 55–58%. However, the greatest activating effect the compound **7** has shown, that at both tested concentrations increased the elastolytic activity of peptidase 2 by 100–140%. This goes to prove that the substance **7**, which is biscitrate-stanate

complex containing cobalt ions as metal, can henceforth be used as effector of peptidase 2 elastolytic activity.

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КОМПЛЕКСИ БІСЦИТРАТОГЕРМАНАТІВ ТА БІСЦИТРАТОСТАНАТІВ З МЕТАЛАМИ — МОДИФІКАТОРИ АКТИВНОСТІ ПЕПТИДАЗ *Bacillus thuringiensis* var. *israelensis* ТА α-ГАЛАКТОЗИДАЗ *Penicillium canescens, Cladosporium cladosporioides* І *Aspergillus niger*

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Метою роботи було вивчити вплив низки координаційних сполук германію і стануму (сполуки 1–8) як модифікаторів активності пептидаз та галактозидаз. Як ефектори ензимів застосовували однотипні за структурою координаційні сполуки на основі хелатуючого ліганда — біологічно активної лімонної кислоти. Досліджували 2 типи комплексів: 1) $[M(H_2O)_6][Ge(HCitr)_2]\cdot 4H_2O$ (M = Mg(1), Mn(2), Co(3), Ni(4), Zn(5)), що містять бісцитратогерманатний аніон ($[Ge(HCitr)_2]^{2-}$); 2) $[M(H_2O)_6][Sn(HCitr)_2]\cdot 4H_2O$ (M = Mg(6), Co(7), Ni(8)) зі вмістом бісцитратостанатного аніона та різних гексааквакатіонів ($[M(H_2O)_6]^{2+}$, M = Mg, Mn, Co, Ni, Zn). Показано, що сполуку 6, яка становить бісцитратостанатний комплекс, що містить як метал іони магнію, можна використовувати для стимуляції на 20–25% колагеназної активності пептидази 1 і пептидази 2 *B. thuringiensis* var. *israelensis* IMB B-7465. Сполуки 1 (бісцитратогерманатний комплекс, що містить як метал іони магнію) і 7 (бісцитратостанатний комплекс, що містить як метал іони кобальту), а також сполука 6 в концентрації 0,001% здатні на 55–58% підвищувати еластолітичну активність пептидази 1. Однак найбільшу активувальну дію виявила сполука 7, яка на 100–140% (в обох досліджуваних концентраціях) підвищувала еластолітичну активність пептидази 2. Це свідчить про те, що сполуку 7 можна надалі використовувати як ефектор еластолітичної активності пептидази 2. Під час досліджень впливу розглянутих координаційних сполук на активність α-галактозидаз *Penicillium canescens, Cladosporium cladosporioides* та *Aspergillus niger* встановлено, що в разі використання низки комплексів (1–2 та 4–8) відзначається неістотне (на 12–20%) підвищення активності ензиму *P. canescens*, а максимальний ефект (~20%, концентрація 0,01%) виявляє комплекс 6.

Ключові слова: пептидази *B. thuringiensis* var. *israelensis* IMB B-7465, α-галактозидази, бісцитратогерманатні та бісцитратостанатні комплекси.

КОМПЛЕКСЫ БИСЦИТРАТОГЕРМАНАТОВ И БИСЦИТРАТОСТАНАТОВ С МЕТАЛЛАМИ — МОДИФИКАТОРЫ АКТИВНОСТИ ПЕПТИДАЗ *Bacillus thuringiensis* var. *israelensis* И α-ГАЛАКТОЗИДАЗ *Penicillium canescens, Cladosporium cladosporioides* И *Aspergillus niger*

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Целью работы было изучить влияние ряда координационных соединений германия и станума (соединения 1–8) в качестве модификаторов активности пептидаз и α-галактозидаз. Как эффекторы энзимов были исследованы однотипные по структуре координационные соединения на основе хелатирующего лиганда – биологически активной лимонной кислоты. Изучали 2 типа комплексов: 1) $[M(H_2O)_6][Ge(HCitr)_2]\cdot 4H_2O$ (M = Mg(1), Mn(2), Co(3), Ni(4), Zn(5)), содержащие бисцитратогерманатный анион ($[Ge(HCitr)_2]^{2-}$); 2) $[M(H_2O)_6][Sn(HCitr)_2]\cdot 4H_2O$ (M = Mg(6), Co(7), Ni(8)) с бисцитратостанатным анионом и различными гексааквакатионами ($[M(H_2O)_6]^{2+}$, M= Mg, Mn, Co, Ni, Zn). Показано, что соединение 6, которое представляет собой бисцитратостанатный комплекс, содержащий в качестве металла ионы магния, можно использовать для стимуляции на 20–25% коллагеназной активности пептидазы 1 и пептидазы 2 *B. thuringiensis* var. *israelensis* ИМВ B-7465. Соединения 1 (бисцитратогерманатный комплекс, содержащий в качестве металла ионы магния) и 7 (бисцитратостанатный комплекс с содержанием в качестве металла ионов кобальта), а также соединение 6 в концентрации 0,001% способны на 55–58% увеличивать эластолитическую активность пептидазы 1. Однако наибольшее активирующее действие оказывало соединение 7, которое на 100–140% (в обеих исследованных концентрациях) повышало эластолитическую активность пептидазы 2. Это свидетельствует о том, что вещество 7 в дальнейшем может быть использовано как эффектор эластолитической активности пептидазы 2. При исследовании влияния рассмотренных координационных соединений на активность α-галактозидаз *Penicillium canescens, Cladosporium cladosporioides* и *Aspergillus niger* обнаружено, что при использовании ряда комплексов (1–2 и 4–8) отмечается незначительное (на 12–20%) увеличение активности энзима *P. canescens*, а максимальный эффект (~20%, концентрация 0,01%) оказывал комплекс 6.

Ключевые слова: пептидазы *B. thuringiensis* var. *israelensis* ИМВ B-7465, α-галактозидазы, бисцитратогерманатные и бисцитратостанатные комплексы.