

FILAMENTOUS CYANOBACTERIA FROM VOJVODINA REGION AS SOURCE OF PHYCOBILIPROTEIN PIGMENTS AS POTENTIAL NATURAL COLORANTS

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ABSTRACT: Phycobiliproteins are a group of colored proteins present in cyanobacteria (blue-green algae). They are extensively commercially used in foods, cosmetics, biotechnology, pharmacology and medicine. In order to determine production of phycobiliproteins in cyanobacterial strains, the quantity of these pigments in 10 filamentous cyanobacteria was investigated. The study was conducted with terrestrial cyanobacterial strains isolated from different soil types in Vojvodina region (Serbia) which belong to *Nostoc*, *Anabaena* and *Spirulina* genera. The obtained results showed that the qualitative and quantitative contents of phycobiliproteins depended not only on cyanobacterial strain, but also on the composition of growth media. In case of strains which belong to *Nostoc* and *Anabaena* genera higher pigment content was found in case of strains cultivated in nitrogen free medium compared to strains grown in medium with nitrogen. The highest content of phycocyanin was found in *Anabaena* strain C2 (22.62 µg/ml) grown in nitrogen free medium, while *Nostoc* strain S1 contained the highest PC amount of 18.37 µg/ml growing in presence of nitrogen. The *Nostoc* strain S1 had the highest content of allophycocyanin growing in both type of nutritious media (25.11 µg/ml and 13.88 µg/ml, respectively). The highest phycoerythrin concentration was characterized the strain *Anabaena* LC1B in nitrogen free medium (24.87 µg/ml) and in presence of nitrogen (20.85 µg/ml). The results of total phycobiliprotein content of tested cyanobacteria showed that the highest content in nitrogen free condition characterized *Anabaena* strains C2 (57.70 µg/ml) and LC1B (58.75 µg/ml) and *Nostoc* strain S1 (57.26 µg/ml). Compared to these *Nostoc* and *Anabaena* strains the contents of all three pigments as well as total pigment content were much lower in both tested *Spirulina* strains. Therefore, some studied strains of *Anabaena* and *Nostoc* genera represent excellent sources of one or more phycobiliproteins.

Key words: cyanobacteria, microalgae, natural colors, pigments, phycobiliproteins

INTRODUCTION

Natural colorants for food are made from renewable sources. Most often, the colorants are extracted from plant material, but other sources such as insects, algae, cyanobacteria (blue-green algae) and fungi are used as well. Legislation restricts which colorants are allowed, what sources may be used for that particular colorant, what solvents may be used to extract it, and the purity of the pigment (Mortensen

A., 2006; Eriksen, 2008). Natural colorants such as phycobiliproteins are gaining importance over synthetic ones, as they are nontoxic and non-carcinogenic. The natural colorants allowed in the EU and the USA are also allowed in most parts of the world (Mortensen A., 2006).

Cyanobacteria (blue-green algae) as specific group of microorganisms represent a potential source of commercially important

chemicals and pharmaceutical products. Among them, phycobiliproteins are very interesting cell constituents with high commercial value. Because of their protein nature, unique color, fluorescence and other properties a wide range of promising applications of phycobiliproteins are possible (Zhao et al., 1995; Rossano et al., 2003, Sekar and Chandramohan, 2008). Due to the toxic and possible carcinogenic effects of several synthetic dyes, there is an increasing preference to use natural colors such as phycobiliproteins (Sekar and Chandramohan, 2008). These pigments can be used as natural colorants in food and drug industry and in cosmetic preparation replacing the synthetic dyes (Cohen, 1986; Soni et al., 2006). Phycocyanin isolated from cyanobacterial species *Spirulina platensis* is widely used as a natural pigment in food, such as dairy products and jellies (Santiago-Santos et al., 2004), coated soft candies (Lone et al., 2005), fermented milk products, ice creams, deserts, milk shakes and sweet cake decoration (Sekar and Chandramohan, 2008). Use of phycobiliproteins for that purposes has to be supported with toxicity testing. Also, phycobiliproteins are applied as fluorescent markers in immunoassays, in biomedical research for cancer diagnostics and as therapeutics (Glazer, 1994; Soni et al., 2006). Moreover, recent studies have shown their immunomodulating and anticarcinogenic activities (Rossano et al., 2003) and their neuroprotective and hepatoprotective properties (Spolaore et al. 2006; Sekar and Chandramohan, 2008). In global patent databases there are 297 patents of phycobiliproteins, and the majority of them are from USA, Japan and Europe (Sekar and Chandramohan, 2008). Patents from USA are mostly related to application of pigments as fluorescent dye while in Japan phycobiliprotein investigations and patents are focused on production, purification and application for therapeutic and diagnostic purposes (Sekar and Chandramohan, 2008). The greatest part of more than 3000 tons of *Spirulina platensis* dry weight is annually produced worldwide because of phycobiliproteins, which are used for

health food products and animal feed additives (Spolaore et al., 2006).

Phycobiliproteins are the major photosynthetic accessory pigments in cyanobacteria which are brilliantly colored, water-soluble proteins, bearing covalently attached open chain tetrapyrroles (Patel et al., 2005). According to the color and absorption ability, phycobilins are divided in 4 main groups: phycoerythrin (PE)-red pigment, allophycocyanin (APC)-bluish green, phycocyanin (PC)-blue, and phycoerythrocyanin (PEC)-orange pigment (Cohen-Bazire, Bryant, 1982). Phycocyanin is a phycobiliprotein that has been recently reported to exhibit a variety of pharmacological properties. In this regard, antioxidant, anti-inflammatory, neuroprotective and hepatoprotective effects have been experimentally attributed to PC (Romay et al., 2003). Phycoerythrin can be used to mark antibodies and other biological elements in laboratory testing.

Phycobiliproteins are variably distributed in the representatives of the division of *Cyanobacteria*. Blue pigments, phycocyanin and allophycocyanin are present in all cyanobacteria, red pigment, phycoerythrin, is widely spread but it is not found in all cyanobacteria, while phycoerythrocyanin usually found in filamentous species (Hoffmann et al., 1990). The ratio of these pigments can be environmentally altered. Filamentous cyanobacteria are particularly attractive for the photoproduction of phycobiliproteins and other chemicals (Borowitzka, 1995). Since that it is of great significance to investigate phycobiliprotein production of especially filamentous cyanobacteria originating from different habitats.

In Serbia research in the field of cyanobacterial products such as phycobiliproteins and their commercial exploitation is very nascent and need adequate attention.

In this study ten strains of terrestrial filamentous cyanobacteria, isolated from different soil types in Vojvodina region, were screened for their potential as producers of phycobiliprotein pigments.

MATERIAL AND METHODS

Analysis of the phycobiliprotein contents was performed on nine cyanobacterial strains originating from various terrestrial environments (solonetz, meadow black soil, chernozem, sand from the banks of the Danube river) in the Vojvodina region and one *Spirulina* strain SJ originating from Algal Culture Collection of Tokyo (Japan) (Table 1).

All tested cyanobacterial strains belong to 3 different genera, *Nostoc*, *Anabaena* and *Spirulina*. Determination of phycobiliproteins: phycocyanin (PC), allophycocyanin (APC) and phycoerythrin (PE) in cyanobacterial strains was carried out during their growth in the laboratory, under certain controlled conditions of temperature, light, mineral content. Pigment content was determined after 21th days (during the stationary phase of growth). Cyanobacterial strains were grown photoautotrophically in BG 11 medium with and without nitrogen (Rippka, et al., 1979), while *Spirulina* strains were cultivated in SOT medium (Soong, 1980). Cultures were maintained at 22-24 °C under illumination by cool white fluorescent light (50 $\mu\text{mol m}^{-2}\text{s}^{-1}$). In terms of duration of light and dark period is most commonly used mode 12 hours light and 12 hours of darkness.

Content of phycobiliproteins (PC, APC and PE) was determined using spectrophotometric method (Bennett, Bogorod 1973). Strains were grown in Erlen-Meyer vessels

where the inocula (1 ml) were streaked in the medium (80ml). After 21 days of incubation, 10 ml of samples (cultures) were taken for the determination of phycobiliproteins and then each sample was centrifuged at 3000 rpm for 5 minutes. The collected cell mass was then washed with buffer 1 M Tris-Cl (pH 8.1). One volume of cell mass was then resuspended in five times of the volume of the same buffer. To extract pigments, it was necessary to make splitting the cell wall of cyanobacterial strains. The procedure of continuous freezing at -20 ° C and thawing at +4 ° C, and sonication of samples (10 minutes with cycles of 30 seconds), allowed the destruction of the cell wall of the strains. After that the separation of cell fragments by centrifugation at 12,000 rpm for 10 minutes was done.

All supernatants of tested cyanobacteria were then separated and concentration of pigments was determined spectrophotometrically at wavelengths of $\lambda = 562 \text{ nm}$ for phycoerythrin (PE), $\lambda = 615 \text{ nm}$ for phycocyanin (PC) and $\lambda = 652 \text{ nm}$ for allophycocyanin (APC). Absorbance measurement was performed on the spectrophotometer "NICOLET Evolution 100" (Thermo Electron Corporation). Concentration of pigments was determined using the following formula:

$$\begin{aligned} \text{PE [mg/ml]} &= (\text{A}_{562} - 2.41 \times \text{PC} - 0.849 \times \text{APC}) / 9.62 \\ \text{PC [mg/ml]} &= (\text{A}_{615} - 0.474 \times \text{A}_{652}) / 5.34 \\ \text{APC [mg/ml]} &= (\text{A}_{652} - 0.208 \times \text{A}_{615}) / 5.09 \end{aligned}$$

Each sample was analyzed in duplicate and buffer was used as a blank.

Table 1
Tested cyanobacterial strains and their origin

Cyanobacterial strains	Genus	Origin
C2	<i>Anabaena</i>	chernozem
C5	<i>Anabaena</i>	chernozem
LC1B	<i>Anabaena</i>	meadow black soil
2S7B	<i>Anabaena</i>	solonetz
2S9B	<i>Nostoc</i>	solonetz
S1	<i>Nostoc</i>	solonetz
S7B	<i>Nostoc</i>	solonetz
2C1	<i>Nostoc</i>	chernozem
SS	<i>Spirulina</i>	Danube bank (Serbia)
SJ	<i>Spirulina</i>	Japanese Culture Collection

RESULTS AND DISSCUSION

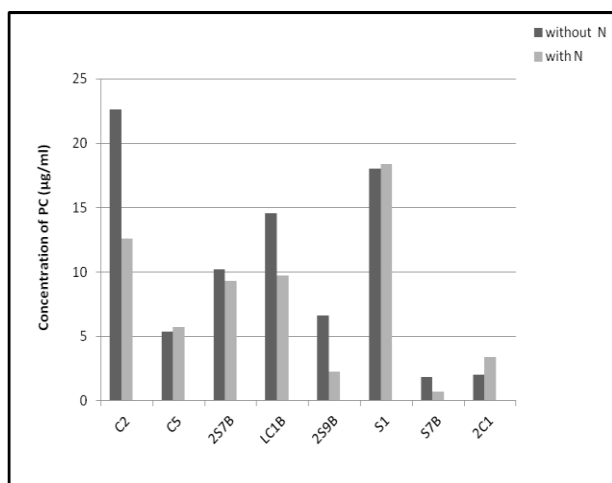


Fig. 1 Content of phycocyanin in investigated cyanobacterial strains of *Nostoc* and *Anabaena* genera

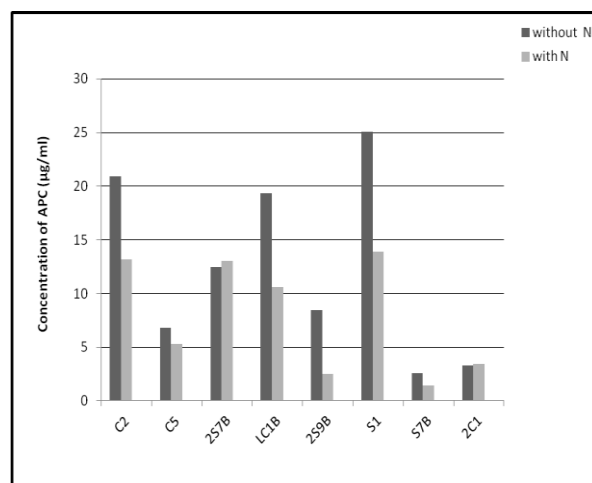


Fig 2. Content of allophycocyanin in cyanobacterial strains of *Nostoc* and *Anabaena* genera

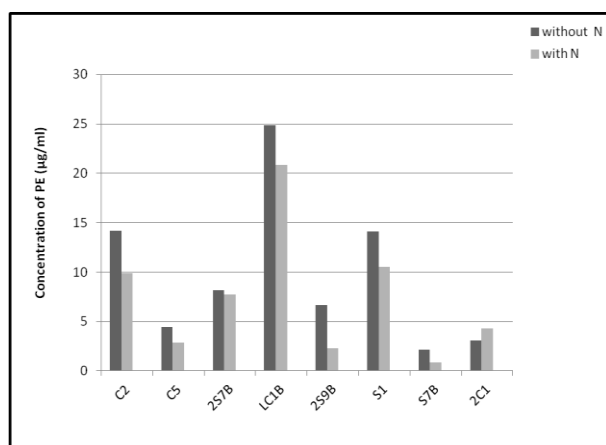


Fig. 3. Content of phycoerythrin in cyanobacterial strains of *Nostoc* and *Anabaena* genera

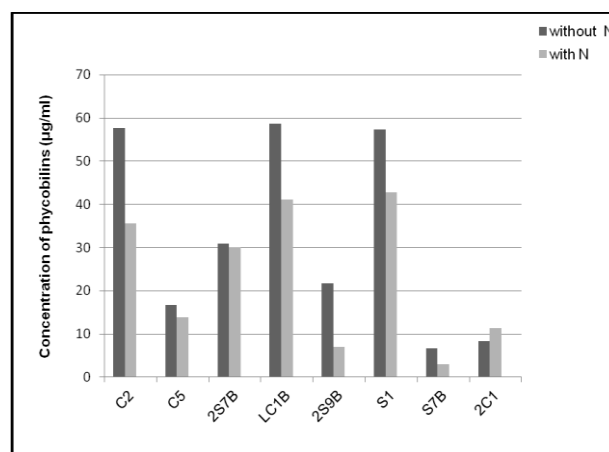


Fig. 4. Content of total phycobiliproteins in cyanobacterial strains of *Nostoc* and *Anabaena* genera

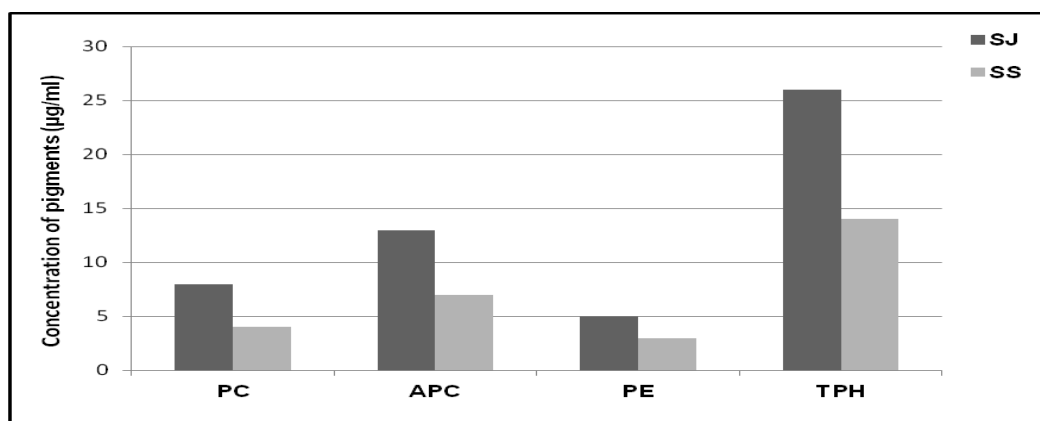


Fig 5. Content of phycobiliproteins in cyanobacterial strains of *Spirulina* genus

The results of PC concentrations in investigated cyanobacteria showed that higher contents of this pigment were found in most strains during the growth in nitrogen free medium, compared to presence of nitrogen (Fig 1). The lowest content was characterized *Nostoc* strain S7B (1.86 µg/ml), while the highest content of PC was found in *Anabaena* strain C2 (22.62 µg/ml). Also very high production of PC in nitrogen free condition was detected in *Anabaena* strain LC1B and *Nostoc* strain S1 with values of 14.57 and 18.03 µg/ml, respectively. Growing in presence of nitrogen, *Nostoc* strain S1 contained the highest PC amount of 18.37 µg/ml (Fig 1). Other tested strains had smaller contents of PC which were in the range of 0.70 (strain S7B) -12.61 µg/ml (strain C2).

The content of pigment allophycocyanin in almost all strains growing in nitrogen free medium was higher than during their growth in medium with nitrogen (Fig 2). Only two strains, 2S7B and 2C1 had higher content of this pigment when they grown in presence of nitrogen. The *Nostoc* strain S1 had the highest amount of APC (25.11 µg/ml). Also very high content was found in *Anabaena* strains such as LC1B (19.31 µg/ml) and C2 (20.92 µg/ml). In the presence of nitrogen the highest concentration of APC was characterized *Nostoc* strain S1 (13.88 µg/ml). Very high content was also found in *Anabaena* strains C2, 2S7B and LC1B (13.18 µg/ml, 13.02 µg/ml, 10.63 µg/ml, respectively) (Fig 2). Other tested strains had content of APC less than 10 µg/ml.

The content of red pigment phycoerythrin was higher in almost all tested strains during their growth in nitrogen free medium (Fig 3). Only strain 2C1 grown in presence of nitrogen had higher amount of this pigment compare to condition without nitrogen. Concentration of PE of strains grown in nitrogen free medium varied from 2.15 µg/ml (strain S7B) to 24.87 µg/ml (strain LC1B). Strains grown in condition with nitrogen showed lower production of phycoerythrin with concentrations in the range of 0.89 µg/ml (strain S7B) – 20.85 µg/ml (strain LC1B) (Fig 3).

The results of total phycobiliprotein content of tested cyanobacteria showed that the highest contents in nitrogen free medium characterized *Anabaena* strains C2 (57.70 µg/ml), LC1B (58.75 µg/ml) and *Nostoc* strain S1 (57.26 µg/ml). The same strains had the highest content of phycobiliproteins during their growth in the medium with nitrogen compared to other tested strains (Fig 4). The lowest concentration of phycobiliproteins was found in *Nostoc* strain S7B, both during the growth in nitrogen free medium and in presence of this element.

The results showed that qualitative and quantitative content of total and individual phycobilin pigments was different, which clearly shows the existence of specific features in the pigment composition of every examined strain. The analysis of the distribution of phycobiliproteins in all strains, showed the presence of all three types of phycobilin pigments – PC, APC, PE in different proportions. Moreno et al. (1995) found that in some strains of *Anabaena* and *Nostoc* genera the prevalent type of phycobiliproteins was C-phyco-cyanin, followed by allophycocyanin, with levels of 17 and 11% d.wt, respectively, while C-phycoerythrin was the major pigment in several *Nostoc* strains, reaching 10% d.wt. In the present study most tested strains of *Anabaena* and *Nostoc* genera had the highest content of allophycocyanin (6 in nitrogen free medium and 4 in the presence in nitrogen), while the highest content of phycocyanin was found in one *Anabaena* strain in nitrogen free medium (C2) and in two strains in the presence of nitrogen (C5 and S1). The highest content of phycoerythrin was characterized in *Anabaena* strain LC1B in both condition and *Nostoc* strain 2C1 in the presence of nitrogen.

There is a lot of information about some average contents of phycocyanin, allophycocyanin and phycoerythrin which are found for some representatives of cyanobacteria. Metabolic examinations showed that an extract of cyanobacteria *Fremyella diplosiphon* contains from 35-40 µgml⁻¹ of phycoerythrin, while an extract of *Calothrix* sp. biomass had 84 µgml⁻¹ of phycocyanin

(Santiago-Santos et al. 2004). Colyer et al. (2005) in the examinations of pigment and protein contents of cyanobacteria detected various concentrations of 3 phycobilins. The values varied in the range of 0.20-4.92 μgml^{-1} for allophycocyanin, 0.73-18.24 μgml^{-1} for phycocyanin and 0.77-19.30 μgml^{-1} for phycoerythrin. In the present study the results of phycobiliprotein content for the examined terrestrial cyanobacterial strains are very similar to the values which were found in these studies. But, some strains tested in our investigation, such as *Anabaena* strains C2 and LC1B and *Nostoc* strain S1 had higher contents of investigated pigments. Therefore, these filamentous strains represent promising sources of one or more phycobiliproteins. However, the detected concentrations of all examined pigments are significantly different from the values found by Soni et al. (2006) in the examination of phycobilin content of the species *Oscillatoria quadripunctulata*, which varied from 27.43 mgml^{-1} for phycocyanin, 15.80 mgml^{-1} for allophycocyanin and 0.45 mgml^{-1} for phycoerythrin.

The results of phycobiliprotein content in tested *Spirulina* strains showed that higher content of all investigated pigments had *Spirulina* strain SJ (Fig 5). The concentration of PC in *Spirulina* SJ strain was 8 $\mu\text{g/ml}$, while *Spirulina* SS strain had content of 4 $\mu\text{g/ml}$. The concentrations of APC and PE in the case of strain SJ were 13 $\mu\text{g/ml}$ and 5 $\mu\text{g/ml}$, respectively. Strain SS had smaller content of both pigments with values of 7 $\mu\text{g/ml}$ for APC and 3 $\mu\text{g/ml}$ for PE (Fig 5). Total pigment content was 26 $\mu\text{g/ml}$ in *Spirulina* SJ strain, while lower pigment content of 14 $\mu\text{g/ml}$ was found in *Spirulina* strain SS (Fig 5).

PC is one of the major pigment constituent of *Spirulina*, a microalga used in many countries as dietary supplement whose nutritional and therapeutic values have been very well documented (Eriksen, 2008). Because of that *Spirulina* is one of the economically important cyanobacterial genus. Species of *Spirulina* have now become the wellknown and the most broadly cultivated microalgae in the world (Vonshak, 1997). Nagaoka et al. (2005) showed that phycocyanin from *Spirulina pla-*

tensis strongly influenced serum cholesterol concentrations and imparted a stronger hypocholesterolemic activity. In this respect *Spirulina* is one of the most promising micro-alga. In the present study we tested two strains of *Spirulina* genera for phycobiliprotein production. *Spirulina* strain SJ contained higher amount of all three pigments (PC-8 $\mu\text{g/ml}$, APC-13 $\mu\text{g/ml}$ and PE-5 $\mu\text{g/ml}$) compared to *Spirulina* strain SS (PC-4 $\mu\text{g/ml}$, APC-7 $\mu\text{g/ml}$ and PE-3 $\mu\text{g/ml}$) (Fig 5). Pigment composition of both strains was very similar. Among phycobilins the highest amount was found for allophycocyanin, then for phycocyanin. The lowest content of phycoerythrin compared to other two types of phycobilins, characterized both strains. This analysis demonstrates that *Spirulina* strains SJ and SS did not show the highest content of tested phycobiliproteins. Strain SJ had higher content of total phycobiliproteins compared to 50% of other tested strains which belong to *Nostoc* or *Anabaena* strains (C5, 2S9B, S7B and 2C1).

Patel et al., (2005) investigated the quantitative of phycobiliprotein (C-PC, APC, and PE) content in three different cyanobacterial species, i.e., *Spirulina* sp., *Phormidium* sp., and *Lyngbya* sp. Among all the three cyanobacterial species, *Spirulina* sp. contains maximum phycobiliprotein content, i.e., 22.5% (w/w) of total freeze-dried cell mass, while *Phormidium* sp. and *Lyngbya* sp. contain 5.4% (w/w) and 5.8% (w/w), respectively. Based on these results the maximum quantity of C-PC exists in *Spirulina* sp. (17.5% (w/w)) as compared to *Phormidium* sp. (4.1% (w/w)) and *Lyngbya* sp. [3.9% (w/w)], while APC and PE are present at lower quantities (Patel et al., 2005). The results of the present study suggest that some *Nostoc* and *Anabaena* strains had much higher content of phycobiliproteins compared to both *Spirulina* strains SJ and SS.

The obtained results clearly prove the fact that the composition and content of phycobilin pigments are specific characteristics of every individual cyanobacterial strain which is very dependent on growing conditions. The medium composition influences the normal growth of cyanobacteria

and normal development of physiological processes. The conditions of cultivation, particularly nitrogen and carbon sources, determines the content of phycobiliproteins in cyanobacteria (cit. in Seker and Chandramohan, 2008). In our study the greatest content of total phycobiliproteins was present in the most of the tested strains which were cultivated in the conditions without nitrogen. Kaushik (2000) obtained similar results by examining the content of phycobiliproteins of 41 strains of cyanobacteria. Results showed that single-celled and colonial cyanobacteria had the lowest content of phycobiliproteins (less than 2.93% of dry mass), non-heterocistous cyanobacteria also had low level of pigment content, while heterocistous nitrogen fixing cyanobacteria had the greatest production of phycobilin which varied from 14.72 to 17.52% of dry mass. Also, Hemlata (2009) found that *Anabaena* NCCU-9 produces the largest amount of phycobilins in the condition without nitrogen. Loreto et al. (2003) showed that the strain *Anabaena* 7120 produces more phycobiliproteins if it is cultivated in the medium without nitrogen, in comparison to the growth in presence of nitrogen. Patel et al. (2005) also noticed the importance of growing conditions on the pigment composition and production of phycobilins for cyanobacterial species. Likewise, Prassana et al. (2004) pointed out the fact that cyanobacteria can regulate their composition and content of basic unit of phycobilin, tetrapyrrole depending on the conditions or signals from the surroundings, such as the availability of nutrients, intensity and quality of light and temperature. In order to find out optimum culture condition for algal growth, Kumar et al. (2011) investigated the effect of light irradiance and temperature on growth rate, biomass composition and pigment production of *Spirulina platensis*. Maximum contents of phycobiliproteins were found in cultures grown at 35 °C i.e. 7.73 % phycocyanin (PC), 3.46% allophycocyanin (APC) and 1.80 % phycoerythrin (PE) and minimum was observed at 20 °C (5.39 % PC, 2.59% APC and 0.64 % PE). But the phycobiliprotein accumulation (except PE) did not show any significant difference at

temperatures 30 °C and 35 °C (Kumar et al., 2011).

It is significant to point out that the content of phycobiliproteins of examined cyanobacterial strains from Vojvodina region represent the pigment content characteristic for every strain during the stationary phase of growth and in constant and usual conditions of cultivation. There was no additional stimulation of production, and changes of the environmental factors (intensity and quality of light, temperature and concentration of nutrients) could influence pigment production in all strains and combining appropriate conditions a high amount of phycobiliproteins can be synthesized. Improvement in the phycobilin content with changes of environmental factors such as light intensity and quality could be a good basis for the exploitation of studied microalgae as a source of biopigments. Still, it is possible that the production of phycobilins would have been increased if other factors had been used, which requires an additional research.

CONCLUSION

The phycobiliproteins have good potential and diverse applications. There is increasing interest in the production of these pigments primary because of their food application (for coloring purposes in foods, dairy products, ice creams, soft drinks, beverages and cake icing). The results of the present study showed that some autochthonous terrestrial filamentous cyanobacterial strains from Vojvodina region had good potential for phycobiliprotein production.

Strains such as *Anabaena* C2, *Anabaena* LC1B and *Nostoc* S1 are very promising producers since they showed the highest content of all three phycobiliproteins among other tested strains. In that respect, it is significant to point out that these strains had much higher phycobiliprotein content compared to both tested *Spirulina* strains.

Further investigations are needed to improve their productivity in suit conditions in mass cultivation. Also, their toxicity should be investigated.

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ФИЛАМЕНТОЗНЕ ЦИЈАНОБАКТЕРИЈЕ ПОРЕКЛОМ СА ПОДРУЧЈА ВОЈВОДИНЕ КАО ИЗВОР ФИКОБИЛИНСКИХ ПИГМЕНАТА КАО ПОТЕНЦИЈАЛНИХ ПРИРОДНИХ КОЛОРАНАТА

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Сажетак: Фикобилипротеини представљају специфичну групу пигмената, који су карактеристични за групу микроалги, познатих под називом цијанобактерије (модрозелене алге). На основу боје и максимума апсорпције светлости фикобилипротеини су подељени у 4 основна типа: фикоцијанин-плави, алофикоцијанин-плави, фикоеритрин-црвени и фикоеритроцијанин-наранџасти пигмент. Ови пигменти се интензивно комерцијално примењују у прехранбеној и козметичкој индустрији, као и у биотехнологији, фармацији и медицини. С обзиром на то да нису токсични нити канцерогени, често се користе као замена многих синтетичких боја. Управо због таквог изузетног значаја фикобилипротеина и њиховог великог биотехнолошког потенцијала, вршена су испитивања продукције 3 фикобилинска пигмента (фикоцијанина, алофикоцијанина и фикоеритрина) код 10 различитих земљишних сојева цијанобактерија. Испитивања су вршена на аутохтоним сојевима пореклом из различитих типова земљишта у Војводини, који припадају цијанобактеријским филаментозним родовима *Nostoc*, *Anabaena* и *Spirulina*. Осим тога, у испитивањима је коришћен и један сој рода *Spirulina* пореклом из колекције култура алги из Јапана. Добијени резултати су показали да количина и квалитативни састав фикобилипротеина не зависи само од цијанобактеријског соја већ пре свега од састава подлоге и услова гајења сојева. У случају сојева који припадају родовима *Nostoc* и *Anabaena*, већи садржај пигмената детектован је у условима њиховог раста у безазотној подлози у поређењу са растом у присуству азота. Највећи садржај плавог пигмента фикоцијанина забележен је код соја *Anabaena* Ч2 (22.62 µg/ml) током гајења у одсуству азота, док је у случају раста у присуству азота највећи садржај овог пигмента (18.37 µg/ml) констатован код соја *Nostoc* С1. Исти сој *Nostoc* С1, у поређењу са осталим тестираним сојевима, одликовао се највећим садржајем пигмента алофикоцијанина растом у оба типа подлоге, при чему је садржај у одсуству азота био значајно већи (25.11 µg/ml) од садржаја регистрованог током раста у присуству азота (13.88 µg/ml). Највећи садржај црвеног пигмента фикоеритрина био је карактеристичан за сој *Anabaena* ЛЦ1Б у условима одсуства азота (24.87 µg/ml), као и током раста у присуству овог елемента (20.85 µg/ml). Резултати садржаја укупних фикобилинских пигмената су указали на то да су растом у безазотним условима највећу продукцију остварили *Anabaena* сојеви Ч2 (57.70 µg/ml) и ЛЦ1Б (58.75 µg/ml), као и *Nostoc* сој С1 (57.26 µg/ml). У поређењу са тим сојевима код оба соја рода *Spirulina* забележен је значајно нижи садржај како појединачних, тако и укупних фикобилина. Садржај укупних фикобилина код соја *Spirulina* СЈ износио је 26 µg/ml, док је у случају соја *Spirulina* СС био значајно нижи, 14 µg/ml. Због тога се може закључити да поједини тестирани филаментозни сојеви родова *Anabaena* и *Nostoc* представљају врло значајан извор фикобилинских пигмената, који могу бити примењени као природне боје. С обзиром на то да на продукцију ових пигмената значајно утичу фактори средине, неопходна су даља истраживања у циљу оптимизације и стимулације продукције и добијања већих количина фикобилина код испитиваних сојева.

Кључне речи: цијанобактерије, фикобилипротеини, микроалге, пигменти, природне боје

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