



## Article Effects of Silver Nanoparticles on the Red Microalga Porphyridium purpureum CNMN-AR-02, Cultivated on Two Nutrient Media

Ludmila Rudi \*D, Liliana Cepoi D, Tatiana Chiriac, Svetlana Djur, Ana Valuta and Vera Miscu

Institute of Microbiology and Biotechnology, Technical University of Moldova, 2028 Chisinau, Moldova; liliana.cepoi@imb.utm.md (L.C.); tatiana.chiriac@imb.utm.md (T.C.); svetlana.djur@imb.utm.md (S.D.); ana.valuta@imb.utm.md (A.V.); vera.miscu@imb.utm.md (V.M.)

\* Correspondence: ludmila.rudi@imb.utm.md

**Abstract:** The purpose of this study was to examine the influence of 10 and 20 nm nanoparticles (AgNPs) on the growth and biochemical composition of microalga *Porphyridium purpureum* CNMN-AR-02 in two media which differ by the total amount of mineral salts (MM1 with 33.02 g/L and MM2 with 21.65 g/L). Spectrophotometric methods were used to estimate the amount of biomass and its biochemical composition. This study provides evidence of both stimulatory and inhibitory effects of AgNPs on different parameters depending on the concentration, size, and composition of the nutrient medium. In relation to the mineral medium, AgNPs exhibited various effects on the content of proteins (an increase up to 20.5% in MM2 and a decrease up to 36.8% in MM1), carbohydrates (a decrease up to 35.8% in MM1 and 39.6% in MM2), phycobiliproteins (an increase up to 15.7% in MM2 and 56.8% in MM1), lipids (an increase up to 197% in MM1 and no changes found in MM2), antioxidant activity (a decrease in both media). The composition of the cultivation medium has been revealed as one of the factors influencing the involvement of nanoparticles in the biosynthetic activity of microalgae.

**Keywords:** *Porphyridium purpureum;* silver nanoparticles; nutrient media; biomass; biochemical composition; antioxidant activity; malondialdehyde content

## 1. Introduction

Interest in the use of microalga *Porphyridium purpureum* (formerly *Porphyridium cru-entum*) (Rhodophyta) as a biotechnological object is steadily increasing, as it represents a valuable source of substances with pronounced biological effects, such as phycobiliproteins, carbohydrates, especially sulfated polysaccharides and lipids, which contain appreciable quantities of polyunsaturated fatty acids [1–5]. Being a marine species, microalga is cultivated under industrial conditions in media with high mineral salt content, and this involves both high production costs and negative environmental impacts due to the excessive discharge of saline wastewater. At the same time, it has been shown that under the conditions of a short biotechnological cycle, *Porphyridium purpureum* culture can provide the same technological advantages in terms of the quantity and quality of biomass, using various media with different mineral nutrient content, with a significantly lower mineral salt content [6,7].

Another aspect of improving *Porphyridium purpureum* cultivation technologies lies in the application of various procedures to stimulate biomass growth and direct biomass composition. In the context of applying stimulants to produce higher amounts of biomass with a valuable and predictable biochemical content, nanoparticles of various natures are increasingly being explored from this perspective [8,9]. For example, nanoparticles of Mg, Al, Zn, Cu, Pb, Ag, Fe, Fe<sub>3</sub>O<sub>4</sub> are used as effective stimulants to enhance lipid synthesis in microalgae for biofuel production purposes [10–12]. However, this effect is more often



Citation: Rudi, L.; Cepoi, L.; Chiriac, T.; Djur, S.; Valuta, A.; Miscu, V. Effects of Silver Nanoparticles on the Red Microalga *Porphyridium purpureum* CNMN-AR-02, Cultivated on Two Nutrient Media. *Mar. Drugs* 2024, 22, 208. https://doi.org/ 10.3390/md22050208

Academic Editors: Celine Laroche and Leonel Pereira

Received: 26 March 2024 Revised: 23 April 2024 Accepted: 23 April 2024 Published: 1 May 2024



**Copyright:** © 2024 by the authors. Licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (https:// creativecommons.org/licenses/by/ 4.0/).

ff

a result of the stress caused by high concentrations of these nanoparticles. At the current stage, the greatest successes in the study of the interactions between nanoparticles and microalgae pertain to elucidating these entities' toxicity mechanisms, with the metabolic pathways involved in the response reactions being highlighted [13]. At the same time, by applying low concentrations of nanoparticles, it is possible to accelerate the growth rate of microalgae, improve nutrient absorption, stimulate biomass production, and optimize the synthesis of bio-products of interest, such as proteins, carbohydrates, pigments, and lipids [8,14,15]. The remarkable bioavailability of nanoparticles supports the idea that they could represent more efficient sources of trace elements compared to their macroscopic forms, thereby contributing to improve vital functions [16].

Silver nanoparticles (AgNPs) are among the most actively used in various fields, such as the food industry, agriculture, textile industry, medical industry, and phycobiotechnologies, including for the valorization of *Porphyridium purpureum* biomass [8,11,17–19]. Thus, AgNPs are used as stimulants for biosynthetic activity, primarily lipids, the content of which can increase in microalgal biomass by 3–8 times [15,18]. Studies have shown the stimulating action of AgNPs on other biosynthetic processes such as carbohydrate production [8].

Modeling the cultivation conditions of microalgae growth in relation to the characteristics of nanoparticles, such as type, size, coating, and concentration, creates new opportunities to enhance the yield of biologically active substances [18,20]. In this context, the study of the influence characteristics of silver nanoparticles on microalgff *Porphyridium purpureum* CNMN-AR-02 is of particular interest. The potential positive effects of silver nanoparticles can be amplified through the additional benefits offered by the possibility of applying a medium with a reduced content of mineral salts. Moreover, under such nutritional conditions, the effects of the nanopfarticles can be significantly altered both in terms of magnitude and the direction of this effect.

Thus, the purpose of this study was to highlight the effects of two-dimensional nanoparticles—10 and 20 nm—applied in concentrations from 0.01 to 10.0  $\mu$ M toff the culture of *Porphyridium purpureum* CNMN-AR-02 on two mineral media characterized by different total salt content.

## References

- 1. Liberti, D.; Imbimbo, P.; Giustino, E.; D'Elia, L.; Silva, M.; Barreira, L.; Monti, D.M. Shedding Light on the Hidden Benefit of *Porphyridium cruentum* Culture. *Antioxidants* **2023**, *12*, 337. [CrossRef] [PubMed]
- Tsvetanova, F.; Yankov, D. Bioactive Compounds from Red Microalgae with Therapeutic and Nutritional Value. *Microorganisms* 2022, 10, 2290. [CrossRef] [PubMed]
- 3. Yin, H.C.; Sui, J.K.; Han, T.L.; Liu, T.Z.; Wang, H. Integration bioprocess of B-phycoerythrin and exopolysaccharides production from photosynthetic microalga *Porphyridium cruentum*. *Front. Mar. Sci.* **2022**, *8*, 836370. [CrossRef]
- Casas-Arrojo, V.; Decara, J.; de los Ángeles Arrojo-Agudo, M.; Pérez-Manríquez, C.; Abdala-Díaz, R.T. Immunomodulatory, antioxidant activity and cytotoxic effect of sulfated polysaccharides from *Porphyridium cruentum*. (S.F. Gray) Nägeli. *Biomolecules* 2021, 11, 488. [CrossRef] [PubMed]
- Kiran, B.R.; Venkata Mohan, S. Microalgal Cell Biofactory—Therapeutic, Nutraceutical and Functional Food Applications. *Plants* 2021, 10, 836. [CrossRef]
- 6. Mehariya, S.; Goswami, R.K.; Karthikeysan, O.P.; Verma, P. Microalgae for high-value products: A way towards green nutraceutical and pharmaceutical compounds. *Chemosphere* **2021**, *280*, 130553. [CrossRef] [PubMed]
- 7. Li, T.; Xu, J.; Wu, H.; Jiang, P.; Chen, Z.; Xiang, W. Growth and Biochemical Composition of *Porphyridium purpureum* SCS-02 under Different Nitrogen Concentrations. *Mar. Drugs* **2019**, *17*, 124. [CrossRef]
- Cepoi, L.; Rudi, L.; Chiriac, T.; Valuta, A.; Zinicovscaia, I.; Miscu, V.; Rudic, V. Silver Nanoparticles as Stimulators in Biotechnology of Porphyridium cruentum. In Proceedings of the 5th International Conference on Nanotechnologies and Biomedical Engineering. Proceedings of ICNBME-2021, Chisinau, Moldova, 3–5 November 2021; Springer Nature: Cham, Switzerland, 2022; Volume 87, pp. 530–536. [CrossRef]
- 9. Huang, Y.; Gao, M.; Wang, W.; Liu, Z.; Qian, W.; Chun Chen, C.; Zhu, X.; Cai, Z. Effects of manufactured nanomaterials on algae: Implications and applications. *Front. Environ. Sci. Eng.* **2022**, *16*, 122. [CrossRef]
- 10. Hasnain, M.; Munir, N.; Abideen, Z.; Dias, D.A.; Aslam, F.; Mancinelli, R. Applying silver nanoparticles to enhance metabolite accumulation and biodiesel production in new algal resources. *Agriculture* **2023**, *13*, 73. [CrossRef]
- 11. Rana, A.; Parmar, A.S. Re-exploring silver nanoparticles and its potential applications. *Nanotechnol. Environ. Eng.* **2023**, *8*, 789–804. [CrossRef]
- 12. Dev Sarkar, R.; Singh, H.B.; Chandra Kalita, M. Enhanced lipid accumulation in microalgae through nanoparticle-mediated approach, for biodiesel production: A mini-review. *Heliyon* **2021**, *7*, e08057. [CrossRef] [PubMed]
- 13. Cao, M.; Wang, F.; Zhou, B.; Chen, H.; Yuan, R.; Ma, S.; Geng, H.; Xing, B. Mechanisms of photoinduced toxicity of AgNPs to the microalgae *Chlorella pyrenoidosa* in the presence of hematite nanoparticles: Insights from transcriptomics, metabolomics and the photochemical index. *Environ. Sci. Nano* **2020**, *9*, 3525–3537. [CrossRef]
- Yap, J.K.; Sankaran, R.; Chew, K.W.; Halimatul Munawaroh, H.S.; Ho, S.H.; Rajesh Banu, J.; Show, P.L. Advancement of green technologies: A comprehensive review on the potential application of microalgae biomass. *Chemosphere* 2021, 281, 130886. [CrossRef]
- 15. Pham, L. Effect of Silver Nanoparticles on Tropical Freshwater and Marine Microalgae. J. Chem. 2019, 2019, 9658386. [CrossRef]
- Komazec, B.; Cvjetko, P.; Balen, B.; Letofsky-Papst, I.; Lyons, D.M.; Peharec Štefanić, P. The Occurrence of Oxidative Stress Induced by Silver Nanoparticles in *Chlorella vulgaris* Depends on the Surface-Stabilizing Agent. *Nanomaterials* 2023, 13, 1967. [CrossRef] [PubMed]
- 17. Nie, P.; Yu, Z.; Xu, H. Synthesis, applications, toxicity and toxicity mechanisms of silver nanoparticles: A review. *Ecotoxicol. Environ. Saf.* **2023**, 253, 114636. [CrossRef]
- 18. Rudi, L.; Cepoi, L.; Chiriac, T.; Miscu, V.; Valuta, A.; Djur, S. Effects of citrate-stabilized gold and silver nanoparticles on some safety parameters of *Porphyridium cruentum* biomass. *Front. Bioeng. Biotechnol.* **2023**, *11*, 1224945. [CrossRef]

- 19. Dawadi, S.; Katuwal, S.; Gupta, A.; Lamichhane, U.; Thapa, R.; Jaisi, S.; Lamichhane, G.; Bhattarai, D.; Parajuli, N. Current Research on Silver Nanoparticles: Synthesis, Characterization, and Applications. J. Nanomater. **2021**, 2021, 6687290. [CrossRef]
- Wang, F.; Liu, T.; Guan, W.; Xu, L.; Huo, S.; Ma, A.; Zhuang, G.; Terry, N. Development of a Strategy for Enhancing the Biomass Growth and Lipid Accumulation of *Chlorella* sp. UJ-3 Using Magnetic Fe<sub>3</sub>O<sub>4</sub> Nanoparticles. *Nanomaterials* 2021, 11, 2802. [CrossRef]
- 21. Decamp, A.; Martineau, E.; Grizeau, D.; Pruvost, J.; Gonçalves, O. Effects of the salinity on the biosynthesis of the polysaccharides of the marine microalgae *Porphyridium cruentum*. *Algal Res.* **2023**, *71*, 103089. [CrossRef]
- 22. Vargas-Estrada, L.; Torres-Arellano, S.; Longoria, A.; Arias, D.M.; Okoye, P.U.; Sebastian, P.J. Role of nanoparticles on microalgal cultivation: A review. *Fuel* **2020**, *280*, 118598. [CrossRef]
- 23. Sun, X.M.; Ren, L.J.; Zhao, Q.Y.; Ji, X.Y.; Huang, H. Microalgae for the production of lipid and carotenoids: A review with focus on stress regulation and adaptation. *Biotechnol. Biofuels* **2018**, *11*, 272. [CrossRef] [PubMed]
- 24. Fazelian, N.; Movafeghi, A.; Yousefzadi, M.; Rahimzadeh, M.; Zarei, M. Impact of silver nanoparticles on the growth, fatty acid profile, and antioxidative response of *Nannochloropsis oculata*. *Acta Physiol. Plant.* **2020**, *42*, 126. [CrossRef]
- 25. He, M.; Yan, Y.; Pei, F.; Wu, M.; Gebreluel, T.; Zou, S.; Wang, C. Improvement on lipid production by *Scenedesmus obliquus* triggered by low dose exposure to nanoparticles. *Sci. Rep.* **2021**, *7*, 15526. [CrossRef]
- Luo, S.W.; Alimujiang, A.; Cui, J.; Chen, T.T.; Balamurugan, S.; Zheng, J.W.; Wang, X.; Yang, W.D.; Li, H.Y. Molybdenum disulfide nanoparticles concurrently stimulated biomass and β-carotene accumulation in *Dunaliella salina*. *Bioresour. Technol.* 2021, 320, 124391. [CrossRef] [PubMed]
- 27. Rana, M.S.; Bhushan, S.; Sudhakar, D.R.; Prajapati, S.K. Effect of iron oxide nanoparticles on growth and biofuel potential of *Chlorella* spp. *Algal Res.* **2020**, *49*, 101942. [CrossRef]
- Yuan, X.; Gao, X.; Liu, C.; Liang, W.; Xue, H.; Li, Z.; Jin, H. Application of Nanomaterials in the Production of Biomolecules in Microalgae: A Review. *Mar. Drugs* 2023, 21, 594. [CrossRef] [PubMed]
- 29. Dey, N.; Vickram, S.; Thanigaivel, S.; Manikandan, S.; Subbaiya, R.; Karmegam, N.; Kim, W.; Govarthanan, M. Aftermath of nanomaterials on lipid profile of microalgae as a radical fuel supplement—A review. *Fuel* **2023**, 333, 126444. [CrossRef]
- Tzanakis, N.; Aravantinou, A.F.; Manariotis, I.D. Short-Term Toxicity of ZnO Nanoparticles on Microalgae at Different Initial Nutrient Concentrations. *Sustainability* 2023, 15, 7853. [CrossRef]
- 31. Johari, S.A.; Sarkheil, M.; Tayemeh, M.B.; Veisi, S. Influence of salinity on the toxicity of silver nanoparticles (AgNPs) and silver nitrate (AgNO<sub>3</sub>) in halophilic microalgae, *Dunaliella salina*. *Chemosphere* **2018**, 209, 156–162. [CrossRef]
- 32. Huang, Z.; Zhong, C.; Dai, J.; Li, S.; Zheng, M.; He, Y.; Wang, M.; Chen, B. Simultaneous enhancement on renewable bioactive compounds from *Porphyridium cruentum* via a novel two-stage cultivation. *Algal Res.* **2021**, *55*, 102270. [CrossRef]
- Lichtenthaler, H.K. Chlorophylls and Carotenoids: Pigments of Photosynthetic Biomembranes. *Methods Enzymol.* 1987, 148, 350–382. [CrossRef]

**Disclaimer/Publisher's Note:** The statements, opinions and data contained in all publications are solely those of the individual author(s) and contributor(s) and not of MDPI and/or the editor(s). MDPI and/or the editor(s) disclaim responsibility for any injury to people or property resulting from any ideas, methods, instructions or products referred to in the content.