The influence of nanomaterials on pyocyanin production by *Pseudomonas aeruginosa*

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Introduction

Pseudomonas aeruginosa is a Gram-negative bacterium producing various industrially relevant metabolites, i.e. pigments. This group includes phenazines, with **pyocyanin** as the most studied example. Pyocyanin is also a well-known virulence factor. However, it also shows the potential for **application** in many branches of technology (Fig. 1).

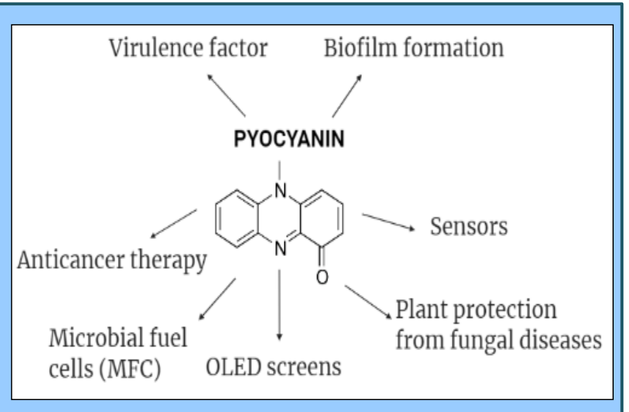


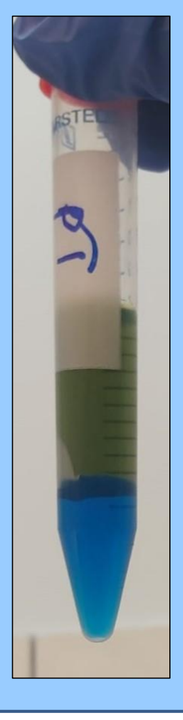
Fig. 1 The applications of pyocyanin

Therefore, concerning both potential **inhibitors and stimulants** of production of pyocyanin by *P. aeruginosa* are widely studied. In the area of interest are various chemical substances, including **nanomaterials**.

THE AIM OF THE STUDY: The study aimed to examine the influence of nanomaterials such as zinc oxide (ZnO) and multi-walled carbon nanotubes (MWCNT) on the pyocyanin yield of *P. aeruginosa* ATCC®27853™.

Materials and methods

1. The **optimization of pyocyanin production** concerning the **volume of culture, mixing and interface.**
2. **Examination** of the influence of different concentrations of nanomaterials (1000.00-0.06 µg/mL) on bacteria growth and fluorescence values.
3. The assessment of **pyocyanin yield** after incubation with the chosen concentrations of **ZnO and MWCNT** using chloroform-hydrochloric acid extraction and absorbance measurements. (λ=520 nm).
4. The assessment of the influence of nanomaterials on bacterial cells by **SEM** analysis.



Results

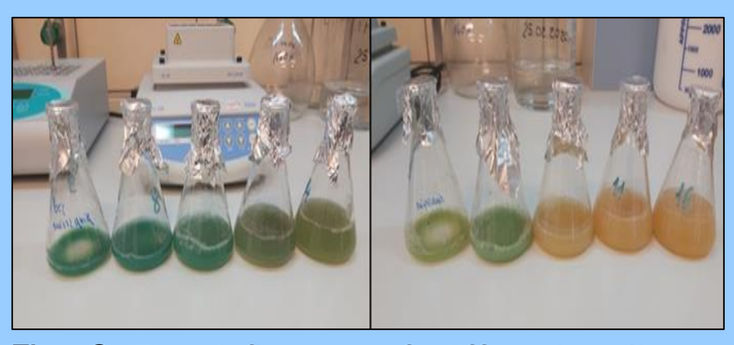


Fig. 2 The influence of different culture volume and mixing on pyocyanin production

The optimization of pyocyanin production revealed that the **highest concentration** of pigment was obtained when *P. aeruginosa* was cultivated **without mixing, in the lowest culture volume (10 mL) and with the biggest interface area**. Exemplary results are shown in Fig. 2. Based on these findings, further experiments were conducted in stationary liquid culture in Petri dishes.

The results show the **diversified influence** of the tested nanomaterials on pyocyanin yield. **MWCNT exhibited a stimulative effect on pigment production when applied in high concentrations**. On the other hand, **higher concentrations** of nanometric **ZnO decreased pyocyanin production**, while **smaller dosages increased** the concentration of the bioproduct. The results are presented in Fig. 3.

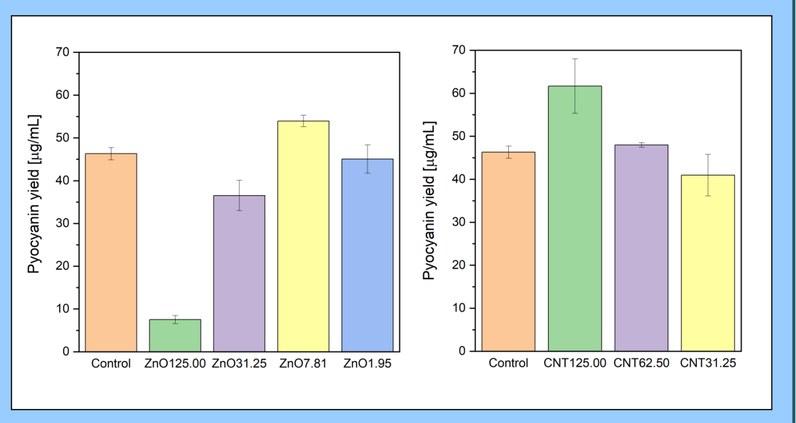


Fig. 3 The influence of ZnO (a) and MWCNT (b) on pyocyanin production

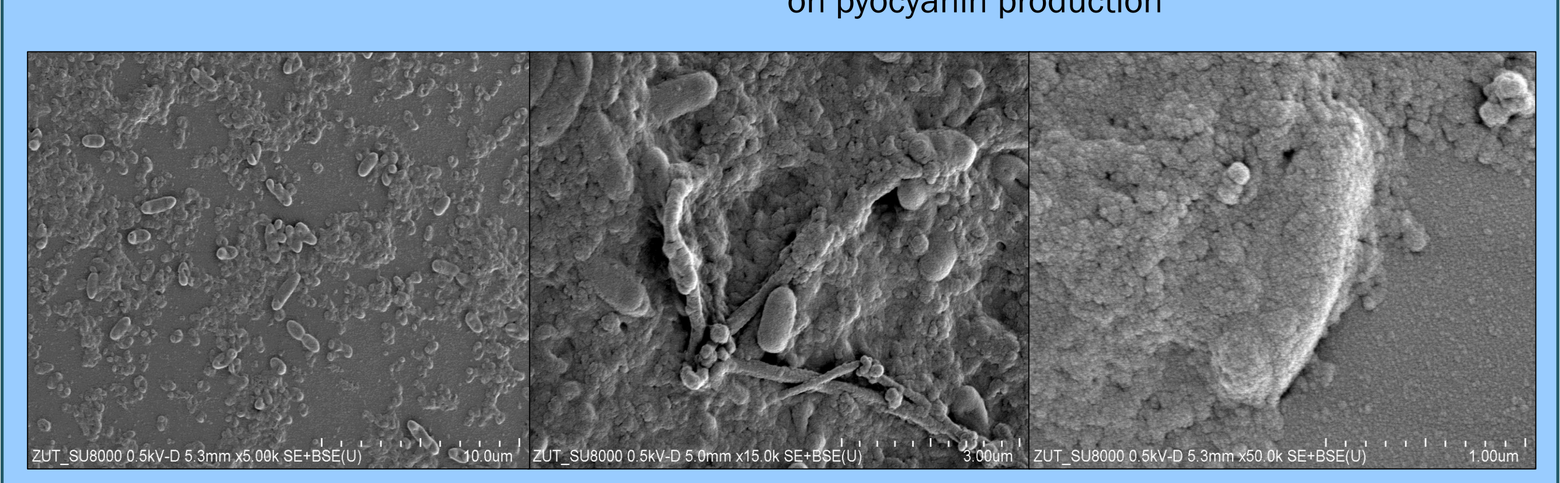


Fig. 4 SEM micrographs of *P. aeruginosa* a) control culture, b) incubated with CNT, c) incubated with ZnO

SEM analyses showed that MWCNT incubated with *P. aeruginosa* was covered with exocellular substance and bacterial cells (Fig 4.). In case of ZnO nanoparticles, the identification of the nanomaterial was not possible due to its size and the need of sputtering of the biological samples. However, the morphology of the cells incubated with nanomaterials **did not differ from the control culture**.

Conclusions

1. Pyocyanin production depends on the **culturing parameters** such as mixing, interface and volume of the culture.
2. Incubation of *P. aeruginosa* with nanomaterials **did not influence the morphology** of cells.
3. **Nanomaterials,** i.e. ZnO and MWCNT, exhibit the potential to be applied as the **modulators** of pyocyanin production, when used in specific concentrations.
4. The influence of nanomaterials on pyocyanin production should be **further explored** to understand the mode of action.

Literature

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