BIOINFOLET 18 (1B) : 199 - 200, 2021

OPTIMIZATION OF MEDIA COMPOSITION AND PHYSICO-CHEMICAL CONDITIONSFOR MAXIMUM PHENAZINE-1-CARBOXYLIC ACID (PCA) PRODUCTION BY *PSEUDOMONAS* RSML35.

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ABSTRACT

Phenazine-1-carboxylic acid (PCA) is a phenazine compound produced by *Pseudomonas* species. Present study deals with optimization of medium composition and physico-chemical conditions for increased production of PCA by Pseudomonas RSML 35. Maximum yield of PCA (965.5 mg/l) was obtained with 30 g/l glucose as a source of carbon, 40 g/l Soya peptone as nitrogen source, at pH 7.0 and 28°C.

Key words: PCA, Optimization, *Pseudomonas* RSML35

Introduction

Pseudomonas, a ubiquitous bacterial genus with biocontrol aptitude, is of immense importance as number of rhizosphere pseudomonad species are termed as plant growth-promoting rhizosphere (PGPR).This bacterium improve plant growth, indirectly by producing antifungal secondary metabolites, such as phenazine-l-carboxylic acid (PCA), Pyrrolnitrin (PRN), pyoluteorin (Plt) and the c-acetyl-phloroglucinols (David and O'Gara, 1994). Present study was undertaken on optimizing carbon (C) and nitrogen (N) in the medium on which *Pseudomonas* is grown, improving PCA production by *Pseudomonas sp.* RSM35.

Material and Methods

The RSM 35 strain of *Pseudomonas* sp. was obtained from Pomegranate rhizospheric soil and used during present study. The organism was grown on fresh King's Medium B (Shtark et al. 2003) for 10 h and a loop of single colony culture was used for inoculation, aseptically, into a 250-ml Erlenmeyer flask containing 100ml KMB broth. The flasks were kept on a rotary shaker at 190 rpm for 24hr at 28°C.

The PCA production medium in the experimental designs composed of carbon source, nitrogen source, $MgSO_4$, NaCl, K_2HPO_4. For batch culture, the 100 mL of the production medium inoculated with 5% of inoculums in 250-mL flasks then flasks were kept on a rotary incubator shaker at 190 rpm at 28^0C(Li He,2008).

The yield of PCA from *Pseudomonas* sp. *RSM35* was investigated using different carbon sources: glucose, fructose, lactose, sucrose, maltose and glycerol, using PCA production medium containing 30 g carbon source (Li He, 2008). The effect of nitrogen source was studied using peptone, soy peptone, beef extract, and yeast extract. The fermentation medium contained 40 g nitrogen source per liter of the broth.

The supernatant was collected after centrifuging at 8000g for 7 min, For PCA extraction, supernatant was acidified to pH 4 with HCl and then the addition of chloroform (50 ml) for 100 ml, finally dried in a vacuum. PCA concentration was determined by specrtrophotometric assay by using different concentration of Standard phenazine (0-1000µg/ml) were employed for recording absorbance at 248 nm on UV-VIS

spectrophotometer (Systronics 2202) (Yihe Ge, 2004).

Results and Discussion

The maximum synthesis of PCA (965.5 mg / l) was observed when glucose was used as a source of carbon, followed by that with fructose (854 mg/l). Minimum PCA production was observed with Maltose as a source carbon (Table 1). Data presented in Tables 2 and 3 indicated thatSoya Peptone (Hi-Media, India) proved to be the best nitrogen source for maximum PCA production (965.5 mg/L), while optimum pH fr maximum PCA production was found to be 7.0 (965.5 mg /l). On the other hand the optimum temperature was found to be 28 °C, as shown in able 4 with the yield of 965.5 mg / l PCA.

Table 1: Optimization of Carbon sources for PCA Production

Carbon Source	Yield of PCA (mg/L)
Sucrose	560.2
Maltose	452.1
Glycerol	665.2
Glucose	965.5
Fructose	854.0
Lactose	675.0

Table 2: Optimization of Nitrogen sources for PCA Production

Nitrogen Source	Yield of PCA (mg/L)
Peptone	653.2
Soya Peptone	965.5
Beef Extract	530.8
Yeast Extract	620.2

Table 3: Optimization of pH for PCA Production

pH	Yield of PCA (mg/L)
4.0	73.2
6.0	540.2
7.0	965.5
9.0	430.8

Table 4: Effect of temperature on PCA Production

Temperature(^{0}C)	Yield of PCA (mg/L)
20^0 C	304.0
28^0 C	965.5
37^0 C	665.2
45^0 C	80.2

Acknowledgements:

The authors acknowledge UGC, New Delhi for financial support for Junior Research Fellowship. They also wish to extend deep sense of gratitude towards Shri Vyankatesh College, Deulgaon Raja and Rajarshi Shahu Mahavidyalaya (Autonomous), Latur for providing laboratory facilities and continuous encouragement.

References

David, N. Dowling and Fergal O'Gara (1994), *TIBTECH* **12**: 133

Shtark O, Shaposhnikov AI, Kravchenko LV (2003) *Mikrobiologiia* **72**:645

Yihe Ge, Xianqing Huang, Sulian Wang, Xuehong Zhang, Yuquan Xu (2004) *FEMS Microbiology Letters :* 237-41

Li He, Yu-Quan Xu, Xue-Hong Zhang (2008). *Biotechnology and Bioengineering*, **100 (2)** : 250