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Explore Pyocyanin Pigment extracted from *Pseudomonas* spp as Plant Growth Promoter

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ABSTRACT: Agriculture is a source of income for over 55% of India's population. Chemical fertilizer and pesticides are used by farmers to increase productivity and profits. Increased use of chemical fertilizers and pesticides results in recalcitrant problems and harm to the texture of the soil. The use of biological agents to boost productivity and manage pests has been extensively researched as a solution. When utilized as bio-fertilizers or plant growth promoters, biological agents had stability and soil condition issues that prevented them from producing the best results. The most prevalent bacteria in the rhizosphere are *Pseudomonas* spp. It produces several metabolites in the soil, one of which is pyocyanin. It is a water-soluble blue-green phenazine pigment. In present studies *Pseudomonas* spp was isolated from a petrol pump soil sample near Jalgaon city. Pyocyanin pigment was extracted from the isolated organism using chloroform and HCl System. Pycocyanin's potential as a plant growth stimulant has been investigated. Extracted pigment was analysed and confirmed by UV visible Spectrophotometer, FTIR and HPLC. In UV- Visible spectrophotometer analysis absorption was observed at 520nm. FTIR analysis revealed different functional groups (-OH, C=N, -CH_3, etc.) which belong to the aromatic structure of pyocyanin. HPLC analysis revealed three peaks with retention durations of 4.572, 5.009, and 5.295 with acetonitrile as mobile phase.

Keywords: Pyocyanin, Phenazine, pigment, *Pseudomonas*, plant growth promoter.

INTRODUCTION

Pseudomonas spp. is the most prevalent species in the rhizosphere, and these strains have long been known for their PGPR activity. The majority of PGPR activities/metabolites include the production of ACC deaminase, phosphate solubilization, phytohormone production, siderophore production (including the production of pyoverdin and pyochelin), antibiotic production (including the production of phenazines and pyrrolnitrin, etc.), and the production of lytic enzymes (cellulase, glucanase, chitinase and protease, etc.). Since PGPR are the most popular substitute for conventional agrochemicals, their survival, shelf life, and productivity are a matter of worry. Consequently, the PGPR who excel under laboratory conditions do not do so in the field since modern agriculture produces (Shaikh *et al.*, 2018; Jha, 2015; Saosoong *et al.*, 2007). Another application of pycocyanin which is widely explored is as one of the good antimicrobial agent, which can inhibit both Gram positive and Gram negative bacteria Such as *Staphylococcus aureus*, *Bacillus cereus*, *Escherichia coli* etc. The problem of antimicrobial resistance was also resolved as a result. (Popy *et al.*, 2017). The pigment's anti-bacterial and antifungal qualities make it a desirable technique for the topical treatment of wound infection (EI-Shouny *et al.*, 2011). The pigment was discovered to have siderophore-like antibacterial properties. Because of its characteristics, pigment is a vital bioactive chemical that can stop bacterial electron transport chains and exhibit antibacterial activity against *E. coli*, *Proteus* species, *S. aureus*, and Klebsiella (Sudhakar *et al*., 2015; Abdul-Hussein and Atia 2016).

As a natural colourant for materials used in the production of fabrics and carpets, paper, and possibly inks, pyocyanin also has broader industrial applications. Additionally, it has the potential to be used as a colouring agent in puddings, cakes, and other baked goods. The food item may also be displayed and decorated using the colour (Alzahrani and Alqahtani 2016; Yogananth *et al*., 2016). Hamad *et al*. (2020), reported that, pyocyanin can be used as a low-cost and secure source in the food and pharmaceutical industries. Pyocyanin modulated root system architecture by inhibiting primary mot growth and increasing the creation of lateral roots and root hairs without compromising meristem viability or triggering cell death in Arabidopis seedlings (Ortiz-Castro *et al.*, 2014). According to Singh *et al.* (2022) microbial inoculants along with various combinations of chemical fertilizers result in enhanced yield as well as luxurious plant growth characters of cabbage (*Brassica oleracea* L. var. *capitata*) in comparison to sole application. Kirubashre *et al.* (2023) also reported, for maintaining productivity and enhancing crop quality, integrated use

of liquid bio-fertilizers, chemical fertilisers, and organic manures can be good choice. But the continued use of chemical fertilisers reduces soil organic matter (SOM) content and agricultural soil quality while weakening microbial activity in the cropping system (Pahalvi *et al.,* 2021; Bhetwal *et al.,* 2021). Muthusamy *et al.* (2023) noted that, the various factor responsible for restricted use bio-fertilizer by farmer such as competition for niches between the bioinoculant and the natural soil flora, poor soil qualities, environmental and soil pollution, extreme climatic conditions, lack of an appropriate strain and an appropriate carrier material in the production unit, lack of resources from public and commercial organisations, as well as lack of infrastructure for storage and transportation. Pyocyanin, which promotes plant growth, could be used as a low-cost source of fertiliser for crop plants. Their ability to produce may be enhanced further with optimisation and familiarisation in accordance with the state of the soil; in the future, they are projected to replace chemical fertiliser and support appropriate agriculture (Baeshen, 2016). According to Farrukh *et al*. (2007), pyocyanin was employed as fertiliser for a variety of crops and demonstrated protective effects against root rot. *Pseudomonas* spp. in soil serves as a model organism for regulating and promoting plant growth, as reported by Anuroopa *et al*. (2022). Pyocyanin has potential uses in a variety of fields, including the production of green energy in microbial fuel cells, biocontrol in agriculture, therapy in medicine, and environmental protection, as shown by Jablonska *et al.* (2023); Jha (2015). In a recent study, pyocyanin, chemical fertiliser substitute that is safe for the environment, was extracted from isolated *Pseudomonas* spp. and tested for its effect on plant growth.

MATERIAL AND METHODS

Sample Collection. Using a sterile spatula and a depth of 5–10 cm, soil samples were taken from the Gujral Petrol Pump area in Jalgaon and aseptically transferred into sterile polythene bags as described by Gahlout *et al.* (2017).

Isolation and Identification of bacteria. The soil sample was diluted with sterile distilled water up to a factor of 10-6. Pigmented bacteria were isolated from diluted samples. A loopful of the diluted sample was removed from the 10-6 tube using a nichrome wireloop, streaked onto a sterile Pseudomonas agar plate, and then cultured for 48 hours at 37°C. The colony that produced the most strong and vibrant pigments were chosen for the remaining research studies. Morphological, biochemical, cultural, and physiological traits were used to identify the organism as suggested by (Schaad *et al.,* 2001), and Bergey's Manual of Determinative Bacteriology was used to cross-check the results (Buchannan and Gibson 1974).

Production of pyocyanin. Pyocyanin was produced in accordance with the method by El-Foulty *et al.* (2015). The chosen isolated colony was injected in 100 ml of nutrition broth supplemented with 1% glycerol and incubated at 37 °C for 48 hours in static condition.

4) Extraction of pyocyanin pigment. Nutrient broth supplemented with glycerol was used for pigment production and was then centrifuged at 10000 rpm for 15 min and the supernatant was collected. It was then filtered through 0.45um pore size membrane filter and used as a crude extract. The pigment extraction process was followed as described by Popy *et al*. (2017). A blue solvent layer was produced after adding chloroform to the culture broth in a ratio of 2:1 and vortexing the mixture. The blue layer was collected, and 0.1 N HCL solution was added and vortexed to create an acidified red layer (20% of the volume of the blue layer). To make it pure, the entire process was performed numerous times.

Concentration of Pyocyanin (ug/ml). A UV-Visible spectrophotometer was used to test the solution's absorbance at 520 nm (Shimadzu). The concentration was given as the number of micrograms of pyocyanin produced per millilitre of culture supernatant (Popy *et al.,* 2017). Concentration of Pyocyanin (µg/ml) = O.D (520) × 17.072

Pyocyanin as a Biofertilizer: The capacity of pyocyanin to stimulate plant development was evaluated using the method recommended by Gupta *et al.* (2015); Suresh *et al*. (2021). Pyocyanin's effectiveness on the growth of wheat and groundnut seeds was examined. Ground nut and wheat seeds in the test tray received 1ml of pyocyanin solution. The pyocyanin was not added to the control tray. Solution of Azotobacter and Rhizobium was added to another set of trays to test the effectiveness of the pyocyanin. Various factors, such as leaf area, root size, and shoot length, have been estimated after 10 days (Baeshen, 2016).

Analysis of pyocyanin. Pyocanin pigment was analysed using UV Visible spectrophotometer, FTIR and HPLC.

UV-Visible Spectrophotometer: Chloroform was used to extract the pigment from the supernatant for two hours at a 1:2 (v/v) ratio. The blue pigment from the chloroform extraction was then treated with acidified water (0.1 N HCL) while being stirred continuously to extract the entire pyocyanin. This process turned the blue pigment into red (acidic form). The absorption of this solution was measured at 520nm. The concentration of pyocyanin produced per mililiter of culture supernatant, is measured in micrograms (Popy *et al.,* 2017).

FTIR: The pigment was further examined using Shimadzu IR Affinity-I Fourier transferred infrared (FTIR) Spectrophotometer, with potassium bromide (KBr) serving as the window material (Popy *et al.,* 2017).

HPLC: Cyberlab Liquid Chromatography with RI detector was used for the HPLC analysis. The column used C18. Acetone nitrile made up the entire mobile phase. The flow rate was 0.5ml.min^{-1}. The column temperature was maintained at 30^{o}C. The peaks obtained were compared with the standard (Al-Qasi *et al.,* 2012).

RESULTS AND DISCUSSION

It was determined that the isolated organism was *Pseudomonas* spp. based on the colony appearance and culture characteristics. (Fig.1). Gram-negative, short-

rod-shaped bacteria are seen to be present singly and to have active movement when the Gram staining and motility was examined. From the various biochemicals performed, the obtained result was compared with Bergey's manual. The isolated organism was identified as *Pseudomonas* spp (Fig. 1 and 2 and Table 1) in accordance with (Schaad *et al.,* 2001) and Bergey's Manual of Determinative Bacteriology.

Production of pyocyanin. The isolate started producing pigment after 24 hours. Due to the presence of soluble pyocyanin pigments, the colour of the solid medium was altered. In a liquid medium, pyocyanin synthesis was evident as a bluish-green tint result obtained was like as described by Alzahrani and Alqahtani, (2016); Popy *et al.* (2017. Day wise production of pycocyanin was monitor. 4th day showed maximum production 2.23 ug/ml (Fig. 6).

Extraction of pyocyanin. Chloroform was used to extract the pigment, producing a blue-coloured product that was later confirmed by adding 0.1 N HCL. Colour shift from blue to red which indicated only the presence of pyocyanin pigment (Fig. 3 and 4). Pyocyanin produces a blue tint when it dissolves in chloroform. as suggested by Alzahrani and Alqahtani (2016); Popy *et al.* (2017).

Pyocyanin as a Biofertilizer. Pot assay was used to determine whether pyocyanin had the ability to promote plant development. The Fig. 9 and Table 5 shows that, in comparison to control plants, plants treated with pyocyanin has increased shoot and root length and produce more no of leaves, and branches in 10 days. Thus, it is evident that the pyocyanin pigment stimulates plant growth. KiKi (2022) investigated the treatment of soil with pyocyanin resulted in considerable increases in some nutrients and minerals including magnesium, chlorine, and iron, whose levels steadily increased with rising pyocyanin concentration in the soil from both plants. Additionally, his research demonstrates a beneficial interaction between pyocyanin and environmental elements that influence plant development.

Analysis of pyocyanin

(a) UV-Visible Spectrophotometer: The absorbance spectrum of pyocyanin was monitored from 200 to 700 nm using UV-Vis spectrophotometer. Pyocyanin dissolved in 0.2 N HCl exhibited the maxima at 270, 414, 400 and 525nm (Fig. 5). According to DeBritto *et al.* (2020) the absorbance peaks of standard pyocyanin were eluted at 382 and 521 nm against 0.2 M HCL These results were near about similar with previous findings. Also, El-Fouly *et al.* (2015) studied the UV-Vis spectrum of pyocyanin and found its characteristics peaks at 300, 388 and 518 nm when dissolved in 0.1 M HCl. The minor difference in the UV Visible spectrum may be due to the media used to produce pyocyanin. Similar type of result is also reported by DeBritto *et al.* (2020); Uzair *et al.* (2018).

(b) FTIR: The spectra, as in Fig. 7, of the extracted compound shows bands between 3400-3300 cm which indicate the presence of –OH group and the appearance of bands between 3000-2900 is an indication of C-H stretch for aromatic compound. As an Aromatic hydrocarbon the compound shows absorptions in the regions 1500-1400cm and 1600-1585 cm^{-1} due to vibrations of carbon-carbon stretching in the aromatic ring. Absorption between 1590-1600 cm^{-1} and 280-1250 cm^{-1} are indicative of C=N bounds and C-N bonds respectively in aromatic stretching. In case of PP_3 sample a hump appeared at 1630 cm^{-1} which can be assigned for C=N bond. The presence of $–CH_3$ group is confirmed with the –CH stretches of the alkyl (methyl) group in the 1380-1400 cm^{-1} range. It shows no peak at 1690-1760 cm^{-1} that means the compound has no chance to have a C=C group in its structure. By comparing the spectra, it was concluded that the purified pigment was pyocyanin because it contained most of the functional groups found in the pyocyanin structure slightly similar which were reported by Devnath *et al.* (2017).

(c) HPLC: Pyocyanin was characterised by HPLC and revealed to include three peaks with retention durations of 4.572, 5.009, and 5.295 with acetonitrile as mobile phase as show in Fig. 8. According to Bacame-Valenzuela *et al.* (2020) HPLC was used to characterize the pyocyanin that was generated and noticed with optimized production of a redox metabolite (Pyocyanin) by *Pseudomonas aeruginosa* nej01r using a maize by-product with C18 column for reverse-phase chromatography was used for the chromatographic separation, The mobile phase was water-adjusted pH to 2.5 by HCl: Acetonitrile (15:85%). The extracted pyocyanin was proven by HPLC followed by Abdelaziz *et al.* (2022) standard pyocyanin which was comparable to that of the extracted purified and lyophilized pyocyanin from *Pseudomonas aeruginosa* derived pyocyanin.

Fig. 1. Pseudomonas agar plate

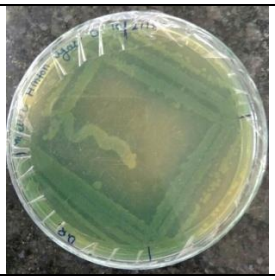

Fig. 2. Growth on Muller Hinton Agar

Fig. 3. Production of pyocyanin

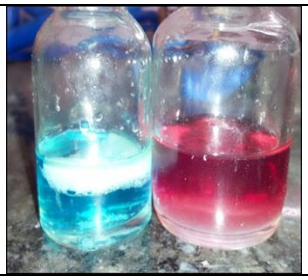

Fig. 4. Extraction of pyocyanin pigment

Fig. 1-4. Physiological and Biochemical Characteristics of *Pseudomonas* Spp Colony characters.

Table 1: Colony characteristics of isolated organism on different media.

Biochemical test:

Media	Size	Shape	Margin	Elevation	Consistency	Opacity	Pigment	Gram reaction	Motility
Nutrient agar plate with glycerol	1 mm	Circular	irregular	Raised	Smooth	Translucent	Greenish	Gram Negative small rods, mostly single	Motile
Pseudomonas agar plate	2 mm	Circular	irregular	Raised	Smooth	Opaque	Diffusible Green pigment	Gram Negative small rods, mostly single	Motile
Muller Hinton Agar	2 mm	Circular	irregular	Raised	Smooth	Translucent	Diffusible Green pigment	Gram Negative small rods, mostly single	Motile

Table 2: Biochemical test result of an isolated organism.

Sugar fermentation:

Indole production	Methyl red test	Vogas Proskauer test	Citrate utilisation test	Catalase	Starch utilization	Gelatin Liquification	Lipid hydrolysis	Nitrate Test
Negative	Negative	Negative	Positive	Positive	Negative	Positive	Positive	Positive

Table 3: Sugar fermentation and TSI test for an isolated organism.

Glucose fermentation	Mannitol fermentation	Sucrose fermentation	Fructose fermentation	TSI agar slant with butt
Acid	Acid	No acid, No gas	Acid	Alkali/Alkali (Red/Red)

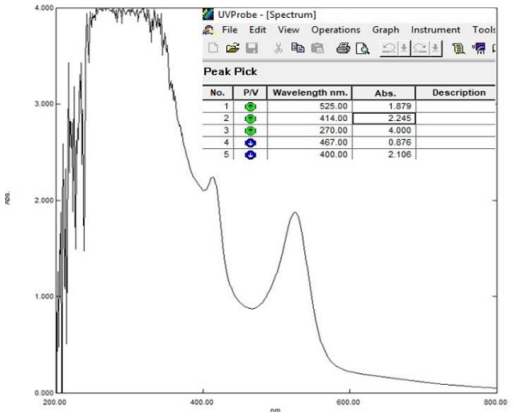

Fig. 5. Absorbance spectrum of pyocyanin using UV-Vis spectrophotometer.

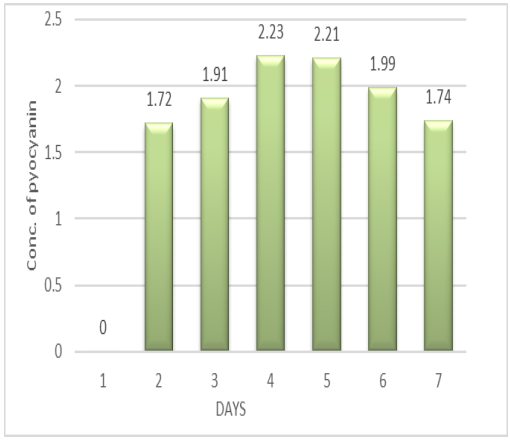

Fig. 6. Day wise production of Pycocyanin

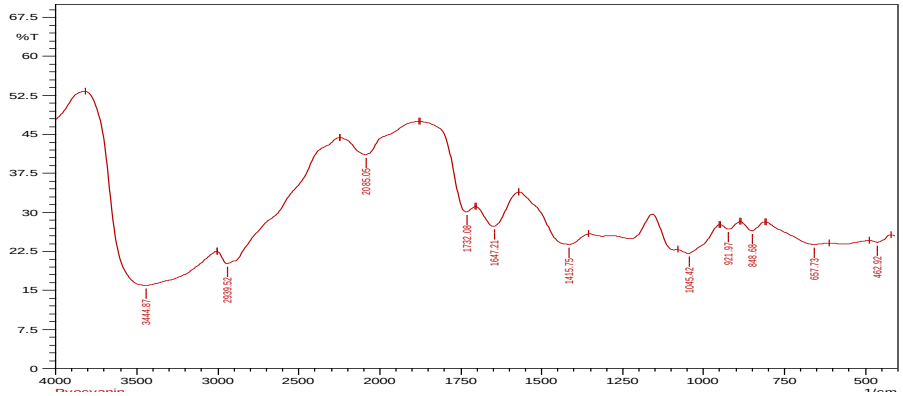

Fig. 7. Absorption bands of Infra-Red (IR) for pyocyanin produced by *Pseudomonas* spp. isolates).

Table 4: FT-IR profile analysis of pyocyanin functional groups extracted from *Pseudomonas* spp.

Compounds Phenazine	H-O	C-H aromatic	C=N	C=C aromatic	C=O	C-O	C-N	ip O-H	ip C-H aromatic	oop C-H aromatic
Standard Phenazine (Aziz et al., 2012)	3475.73	3059.1	1627.9	1554.63	1469.76	1323.17	1284.59	1207.44	856.39	748.38
Extracted Pyocyanin	3444.87	2939.52	1647.21	-	1415.75	-	1045.42	1045.52	848.68	657.73

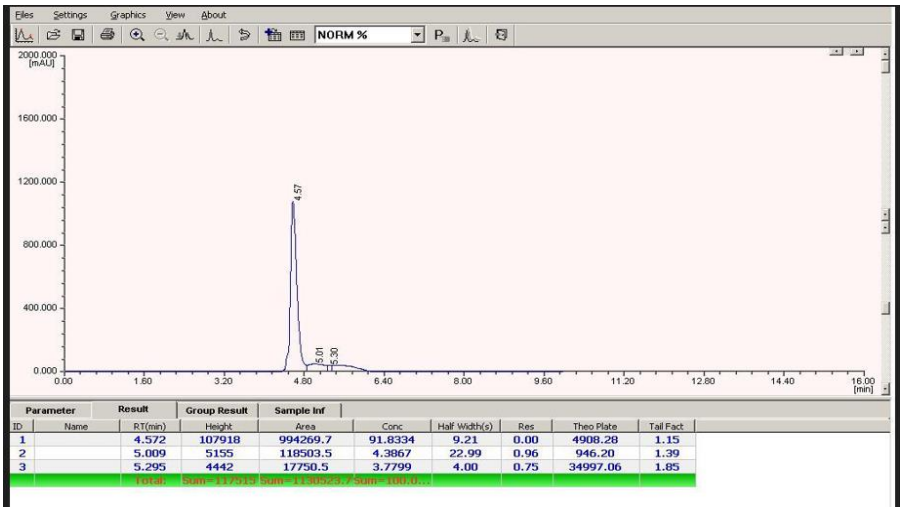

Fig 8. HPLC Scan of extracted Pycocyanin extracted from *Pseudomonas* spp.

Table 5: Effect of pyocyanin on root and shoot of Wheat and ground nut seeds.

Effect of pyocyanin on plant

	With Pyocyanin				Without Pyocyanin			
	Root length	Average root length	Shoot length	Average Shoot length	Root length	Average root length	Shoot length	Average Shoot length
	15cm		10 cm		10cm		8cm	
Ground Nut	22cm	17.3	11cm	10	7cm	9.33	9cm	8.3
	16cm		9cm		11cm		8cm	
	25cm		17cm		7cm		11cm	
Wheat	19cm	21.3	12cm	14.6	12cm	9	9cm	9.83
	20cm		15cm		8cm		9.5cm	

Seed	With Pyocyanin	Without Pyocyanin
Wheat		
Ground nut		

Fig. 9. Effect of pyocyanin on root and shoot of Wheat and ground nut seeds.

CONCLUSIONS

The current study found that *Pseudomonas* spp. pigment is a naturally occurring material with the potential of function as a bio-fertilizer, contributing in accelerating the development of an environmentally friendly alternative to chemical fertilizers. For the benefit of society, its use as a fertiliser should be further investigated, along with toxicity testing**.**

FUTURE SCOPE

Pyocyanin from *Pseudomonas* spp. has been studied for its potential as an antibacterial, antifungal, biosignaling, agent etc. It is also being investigated as a colourant in the textile and food industries. However, pycocyanin has not been investigated as a plant growth promoter. Recent research has produced remarkable results as a plant growth promoter. More research should be done in this area to develop an environmentally friendly product that would increase agricultural productivity. *Pseudomonas* spp. pigment has the potential to serve as a bio-fertilizer and contribute to the creation of an environmentally friendly substitute for chemical fertilizers. Its application as a fertilizer should be researched further, as should toxicity testing, for the benefit of society. It has no side effect of biomagnifications.

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Conflict of Interest. None.

REFERENCES

Abdelaziz, A. A., Kamer, A. M. A., Al-Monofy, K. B. and Al-Madboly, L. A. (2022). A purified and lyophilized *Pseudomonas aeruginosa* derived pyocyanin induces promising apoptotic and necrotic activities against MCF-7 human breast adenocarcinoma. *Microbial Cell Factories, 21*(1), 262.

Abdul-Hussein, Z. R. and Atia, S. S. (2016). Antimicrobial effect of pyocyanin extracted from *Pseudomonas aeroginosa*. *European Journal of Experimental Biology, 6*(3), 1-4.

Al-Qasi, L. M. (2012). Purification, characterization and genetic evaluation of Phenazine produced by *Pseudomonas aeruginosa* local isolates. Degree of Doctorate. College of Science, University of Baghdad.

Alzahrani, S. H. and Alqahtani, F. S. (2016). Pyocyanin pigment extracted from *Pseudomonas aeruginosa* isolate as antimicrobial agent and textile colorant. *Int J Sci Res., 5*(9), 467-470.

Anuroopa, N., Ram, A. B., Ranadev, P., Ashwin, R. and Bagyaraj, D. J. (2022). *Pseudomonas* species in Soil as a Natural Resource for Plant Growth Promotion and Biocontrol Characteristics-An Overview. *Madras Agricultural Journal, 108* (december (10-12), 1.

Aziz, L. M., Hamza, S. J. and Abdul-Rahman, I. A. (2012). Isolation and characterization of phenazine produced from mutant *Pseudomonas aeruginosa*. *Al-Anbar J Vet Sci., 5*(1), 42-53.

Bacame-Valenzuela, F. J., Perez-Garcia, J. A., Figueroa-Magallon, M. L., Espejel-Ayala, F., Ortiz-Frade, L. A. and Reyes-Vidal, Y. (2020). Optimized production of a redox metabolite (*Pyocyanin*) by *Pseudomonas aeruginosa* NEJ01R using a maize by-product. *Microorganisms, 8*(10), 1559.

Baeshen, A. A. (2016). Use of *Pseudomonas aeruginosa* as fertilizer in Eruca sativa. *IJCAMS, 5*, 301-12.

Bhetwal, S., Rijal, R., Das, S., Sharma, A., Pooja, A., & Malannavar, A. B. (2021). *Pseudomonas fluorescens*: Biological Control Aid for Managing Various Plant Diseases: A Review. *Biological Forum–An International Journal 13*(1), 484-494.

Buchannan, R. E. and Gibson N. E. (1974). Bergey's manual of Determinative Bacteriology. Eighth Edition. Williams and Wilkins Co., 217-236.

DeBritto, S., Gajbar, T. D., Satapute, P., Sundaram, L., Lakshmikantha, R. Y., Jogaiah, S., & Ito, S. I. (2020). Isolation and characterization of nutrient dependent pyocyanin from *Pseudomonas aeruginosa* and its dye and agrochemical properties. *Scientific reports, 10*(1), 1542.

Devnath, P., Uddin, M. K., and Aha, F. (2017). Extraction, purification and charact *Pseudomonas aeruginosa. Extraction, 6*(5), 1-9.

El-Fouly, M. Z., Sharaf, A. M., Shahin, A. A. M., El-Bialy, H. A. and Omara, A. M. A. (2015). Biosynthesis of pyocyanin pigment by *Pseudomonas aeruginosa*. *Journal of Radiation Research and Applied Sciences, 8*(1), 36-48.

El-Shouny, W. A., Al-Baidani, A. R. H. and Hamza, W. T. (2011). Antimicrobial activity of pyocyanin produced by *Pseudomonas aeruginosa* isolated from surgical wound-infections. *Int. J. Pharm. Med. Sci., 1*(1), 1-7.

Farrukh, Mansoor., Sultana, V. and Ehteshamul-Haque, S. (2007). Enhancement of biocontrol potential of *Pseudomonas aeruginosa* and *Paecilomyces lilacinus* against root rot of mung bean by a medicinal plant *Launaea nudicaulis* L. *Pak J. Bot., 39*(6), 2113-2119.

Gahlout, M., Prajapati, H., Chauhan, P., Patel, N. and Solanki, D. (2017). Isolation and screening of pyocyanin producing *Pseudomonas* spp. from soil. *Int J Adv Res Biol Sci., 4*(4), 147-152.

Gupta, G., and Nath, J. P. (2015). Screening of potential PGPR candidates as future biofertilizers-a strategic approach from lab to field. *Res J Biotechnol, 10*(11), 48-62.

Hamad, M. N. Marrez, D. A. and El-Sherbieny, S. M. (2020). Toxicity evaluation and antimicrobial activity of purified pyocyanin from *Pseudomonas aeruginosa*. *Biointerface Res Appl Chem., 10*, 6974–6990.

Jablonska, J., Augustyniak, A., Dubrowska, K., and Rakoczy, R. (2023). The two faces of pyocyanin-why and how to steer its production? *World Journal of Microbiology and Biotechnology, 39*(4), 103.

Jha, P. N. (2015). Biocontrolling and colonization potential of *Pseudomonas aeruginosa* PM389 Isolated from *Pennisetum glaucum* under field conditions. *International Journal for Research in Emerging Science and Technology, 2*(3), 104-116.

Kiki, M. (2022). Effects of pyocyanin pigment on the chemical and physical characteristics of agricultural soils. *Applied ecology and environmental research, 20*(4), 3117-3127.

Kirubashre, K., Shanthi A. and Manikandan M. (2023). Influence of Liquid Bio-fertilizers Along with Inorganic and Organic Sources of Nutrients on Quality Parameters of Cluster Bean (*Cyamopsis tetragonoloba* (L.) Taub.). *Biological Forum – An International Journal, 15*(4), 598-602.

Muthusamy, Y., Sengodan, K., Arthanari, M., Kandhasamy, R. and Gobianand, K. (2023). Biofertilizer and Consortium Development: An Updated Review. *Curr. Agri. Res., 11*(1), 01-17.

Ortiz-Castro, R., Pelagio-Flores, R., Mendez-Bravo, A., Ruiz-Herrera, L. F., Campos-García, J. and Lopez-Bucio, J. (2014). Pyocyanin, a virulence factor produced by *Pseudomonas aeruginosa*, alters root development through reactive oxygen species and ethylene signaling in Arabidopsis. *Molecular Plant-Microbe Interactions*, *27*(4), 364-378.

Pahalvi, H. N., Rafiya, L., Rashid, S., Nisar, B. and Kamili, A. N. (2021). Chemical fertilizers and their impact on soil health. Microbiota and Biofertilizers, Vol 2: Ecofriendly Tools for Reclamation of Degraded Soil Environs, 1-20.

Popy, D. M., Kamal, U., Forkan, A. and Mohammed, A. (2017). Extraction, purification and characterization of pyocyanin produced by *Pseudomonas aeruginosa* and evaluation for its antimicrobial activity. *Int J Biol Res.*, *6*, 230-250.

Saosoong, K., Tongtumma, S., Chanthai, S., Wongphathanakul, W., Bunyatratchata, W. and Rungviriyachai, C. (2007). Isolation and study of chemical properties of pyocyanin produced from *Pseudomonas aeruginosa* TISTR 781 (ATCC 9027)(Thai). *Asia-Pacific Journal of Science and Technology*, *12*(1), 24-32.

Schaad, N. W., Jones, J. B., and Chun, W. (2001). Laboratory guide for the identification of plant pathogenic bacteria (No. Ed. 3). American Phytopathological society (APS press).

Shaikh, S. S., Wani, S. J., & Sayyed, R. Z. (2018). Impact of interactions between rhizosphere and rhizobacteria: a review. *J Bacteriol. Mycol.*, *5*(1), 1058.

Singh, K., Jeyakumar, E., Singh, J. P. and Jaiswal, D. K. (2022). Combined Effect of *Pseudomonas aeruginosa* MCCB 0035 with NPK on Growth and Yield Attributes of Cabbage (*Brassica oleracea* L. var. capitata). *Biological Forum–An International Journal 14*(3), 1252-1258.

Sudhakar, T., Karpagam, S. and Premkumar, J. (2015). Biosynthesis, antibacterial activity of pyocyanin pigment produced by *Pseudomonas aeruginosa* SU1. *Journal of Chemical and Pharmaceutical Research*, *7*(3), 921-924.

Suresh, P., Vellasamy, S., Almaary, K. S., Dawoud, T. M. and Elbadawi, Y. B. (2021). Fluorescent pseudomonads (FPs) as a potential biocontrol and plant growth promoting agent associated with tomato rhizosphere. *Journal of King Saud University-Science*, *33*(4), 101423.

Uzair, B., Kausar, R., Bano, S. A., Fatima, S., Badshah, M., Habiba, U. and Fasim, F. (2018). Research Article Isolation and Molecular Characterization of a Model Antagonistic *Pseudomonas aeruginosa* Divulging In Vitro Plant Growth Promoting Characteristics.

Yogananth, N., Kalidoss, A., Syed, A. M. and Anuradha, V. (2016). Production of food grade pigments from *Pseudomonas aeruginosa*. *Int J. Curr. Res. Biol. Med.*, *1*, 14-18.

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