RESEARCH PAPER | Science | Volume : 5 | Issue : 10 | October 2015 | ISSN - 2249-555X

Pyocyanin Exerts Antimicrobial Activity on Staphylococci

KEYWORDS: hypoxia, psychophysiology, memory, altitude.

DR ARIJIT CHATTERJEE

ABSTRACT *Different isolates of Pseudomonas aeruginosa were obtained from wound swab, pus and urine specimens. The isolates were cultured for the production of the blue pigment; pyocyanin. Considerable amounts of blue pigment were produced by*

P. aeruginosa isolates when grown on nutrient agar media. Müller-Hinton agar was

further used for growth, pigment production and sensitivity tests.

P.aeruginosa isolates were grown in peptone water liquid medium by incubating at 370C. for 24hrs. Then, culture tubes were shaken gently and centrifuged 10000rpm for 30 minutes to extract the supernatant. Using 5mL of the bacterial supernatant, 1mL of chloroform was added and shaken vigorously. The chloroform layer was removed (which should be on the bottom and turn blue to contain the pyocyanin. 0.5mL of 0.01M HCl was added and the aqueous layer was removed which becomes red in colour. 5µL of 10nM NaOH was added again to revert the colour to blue and thus purified. The growth of Staphylococcus aureus was completely inhibited when cultivated on the agar plates containing the blue pigment.

INTRODUCTION

Pyocyanin is a water soluble bluish-green phenazine pigment produced by active culture of Pseudomonas aeruginosa. Pyocyanin (N-methyl-1-hydroxyphenazine) has antibiotic activity against many microorganisms. The nature of bacterial inhibition by the phenazine is not well understood. It was proposed that, during aerobic respiration or oxidative utilization of sugar, pyocyanin becomes reduced and reduces O_2 to the toxic superoxide radical. The resistance of different microorganisms to pyocyanin may therefore be dependent upon the levels of superoxide dismutase and catalase possessed by the organism and on the presence of oxygen.

MATERIALS AND METHODS

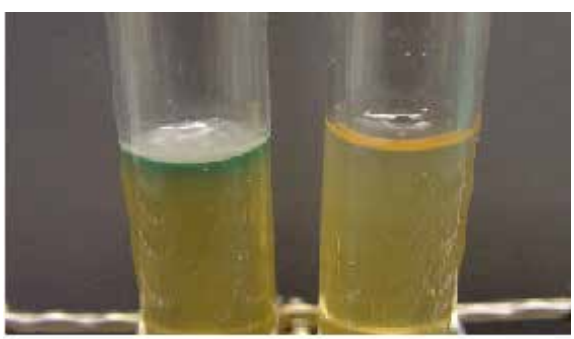
Different isolates of *Pseudomonas aeruginosa* were obtained from wound swab, pus and urine specimens. The isolates were cultured for the production of the blue pigment; pyocyanin and were subjected to standard microbiological and biochemical techniques for identification in SRL Reference Laboratory, Kolkata.

MEDIA

Nutrient agar, Müller Hinton agar and peptone water liquid medium were used for the growth of *P. aeruginosa* and detection of pyocyanin.

PROCEDURE

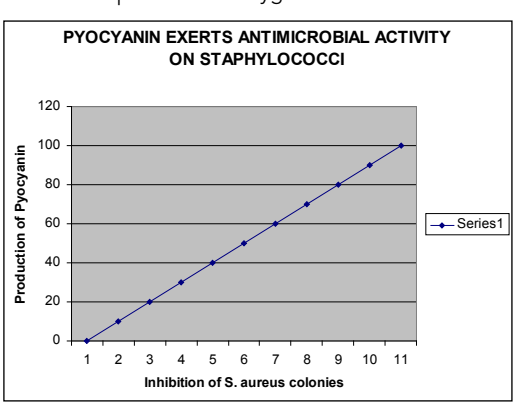
P.aeruginosa isolates were grown in peptone water liquid medium by incubating at 37^{0}C. for 24hrs. Then, culture tubes were shaken gently and centrifuged 10000rpm for 30 minutes to extract the supernatant. Using 5mL of the bacterial supernatant, 1mL of chloroform was added and shaken vigorously. The chloroform layer was removed (which should be on the bottom and turn blue to contain the pyocyanin. 0.5mL of 0.01M HCl was added and the aqueous layer was removed which becomes red in colour. 5µL of 10nM NaOH was added again to revert the colour to blue and thus purified. The growth of Staphylococcus aureus was completely inhibited when cultivated on the agar plates containing the blue pigment.



PYOCYANIN EXTRACTION

RESULT AND DISCUSSION

The nature of bacterial inhibition by the phenazine is not well understood. It was proposed that, during aerobic respiration or oxidative utilization of sugar, pyocyanin becomes reduced and reduces O_2 to the toxic superoxide radical. The resistance of Staphylococcus aureus to pyocyanin may therefore be dependent upon the levels of superoxide dismutase and catalase possessed by the organism and on the presence of oxygen.



Volume : 5 | Issue : 10 | October 2015 | ISSN - 2249-555X

ACKNOWLEDGEMENTS
The technical supervision of Dr. Satadal Das and guidance of Dr. Bhaskar Narayan Chaudhuri was greatly acknowledged in this regard.

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