

INTERNATIONAL JOURNAL OF INSTITUTIONAL PHARMACY AND LIFE SCIENCES

Life Sciences

Research Article.....!!!

Received: 24-02-2015; Revised: 28-02-2015; Accepted: 01-03-2015

GROWTH STUDIES OF DOMINANT BACTERIA ISOLATED FROM MARINE WATER FROM SEASHORE OF NELLORE DISTRICT

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Keywords:

Marine water, Sea shore,
Bay of Bengal, Gram
positive cocci,
Spectrophotometry

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ABSTRACT

The aim of the study is to isolate and characterize by morphological and biochemical methods of dominant bacteria from sea shore water of Nellore district which was bordered by Bay of Bengal. The collected samples were processed for isolation by using spread plate method. Pure colonies were isolated, identified and characterized based on cultural, biochemical characteristics and growth studies. The bacteria isolated from sea shore water sample were identified as Gram positive cocci. Both the spectroscopic and spread plate methods exhibited all growth phases in respect to time period.

1. INTRODUCTION

The Indian marine environment is rich in biodiversity, especially microorganisms. The limited attempts have been made on marine organisms and their metabolites in India^{1,2}. The investigation of marine environment with reference to bioactive molecule production in India is still at its infancy. Therefore, exploration of marine microbes for secondary metabolites production is worthy task. However, the wealth of marine micro-flora has not been fully investigated. Marine microbes flourish not only in the surface water of the sea, but also in the lower and abyssal depths from coastal to the offshore regions³. Marine microbiota are considered as the reservoirs of the beneficial secondary metabolites. Searching of novel antimicrobial secondary metabolites from marine environment is gaining momentum in recent years. 70% of the earth's surface is covered by oceans with rich microbial diversity⁴ which provides the largest biospace for microorganisms, particularly microbes. The bioactive molecules derived from these microbes could be used as therapeutic drugs for the treatments of various ailments in human and animals and as agrochemicals for the management of insect pests, diseases and weeds in agriculture and Microorganisms with their diverse structure, physiology and metabolism survive in different climatic conditions. Studies on marine microorganisms reveals that marine microbial cells exhibit metabolic activity which were yet to be identified and characterized⁵. Cultivation-based studies may provide information on the physiological characteristics of the organisms living in the sediments, and allow the description of new species and the prospecting for microorganisms of biotechnological interest^{6,7}. The aim of this study was to isolate dominant bacteria from the sea shore regions of Nellore district and evaluate their morphological, cultural, biochemical characteristics and growth studies.

2. MATERIALS AND METHODS

Collection of marine water sample

The marine water sample was collected from sea shore of Nellore district, Andhra Pradesh, South India. Samples were collected in sterilized plastic containers at the depth of 10-25 cm and stored in a refrigerator at 4°C till further processing.

Isolation of bacteria from water samples

0.1 ml of marine sample was serially diluted in sterilized distilled water to get a concentration range from 10^{-1} to 10^{-6} . A volume of 0.1 ml of each dilution was transferred aseptically to nutrient agar plates. The sample was spread uniformly and were incubated at 37°C for 24 hours. The bacterial isolates were further sub cultured to obtain pure culture⁸.

Characterization of isolated bacteria

After 24 hours of incubation colonies were grown on the plates and examined for their morphological, cultural and biochemical characterization by using standard Microbiology methods⁹.

Bacterial growth studies

Growth studies by spread plate method

The bacterial concentration of 0.1ml broth suspension was measured by the number of bacteria per milliliter. In this method the suspension was thoroughly shaken and was serially diluted and spread plated¹⁰.

Growth studies by spectrophotometric method

In the Spectrophotometric method, portions of the broth suspension harvested from the plates were pipetted into semi micro cuvettes and the OD of the suspension was measured at 600 nm with a Spectrophotometer. The replicate values were averaged and the OD was plotted against time¹⁰.

3. RESULTS AND DISCUSSION

Isolation & Characterization of Bacteria

The isolated bacteria from marine samples of seashore of Nellore district represents gram positive cocci (Fig 1& 2) under microscopic examination and gram staining and morphological and biochemical tests were tabulated in (table 1).

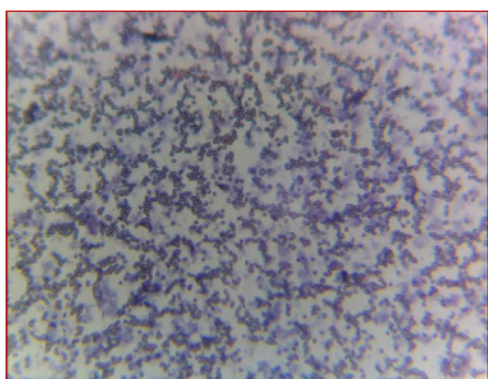


Fig 1: Gram staining of bacteria Fig 2 : Cultural characteristics of Isolated bacteria

Table 1: Morphological, cultural characteristics and biochemical characterization of isolated bacteria

Margin	smooth
Elevation	Raised
Pigment	Cream
opacity	opaque
Gram reaction	positive
Cell shape	cocci
Indole test	Negative
MR Test	Positive
VP Test	Negative
Citrate test	Negative
Urease test	Positive

Growth studies of Bacteria

Growth curves with identifiable lag, log, stationary and decline phases were obtained by both the spread plate and spectroscopic methods performed at different time periods (0-24 hours) were studied (Fig 3 & 4).

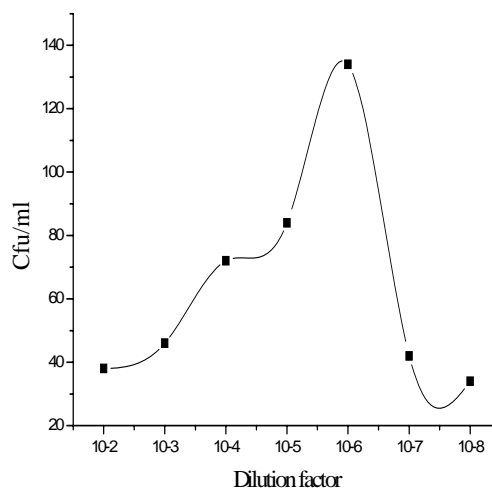
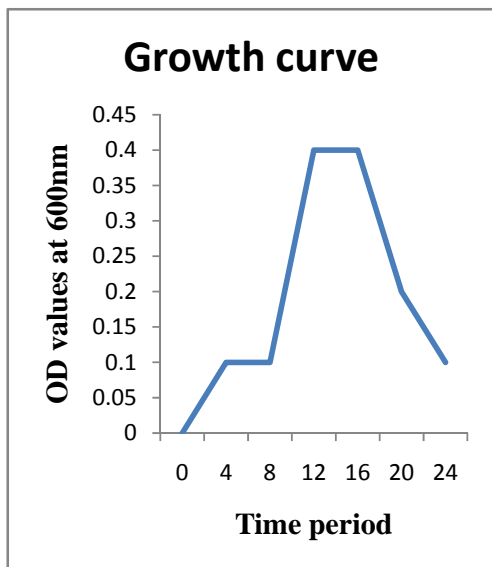


Fig 3 : Optical density Growth curve for Gram positive cocci Fig 4 : Spread plate Growth curve for Gram positive cocci

4. CONCLUSION

The main aim of this present study was to isolate, characterize by morphological, biochemical tests and growth studies of dominant bacteria from sea shore water of Nellore district. The dominant bacterium was identified as gram positive cocci through cultural, biochemical characterization. Growth curves with identifiable lag, log, stationary and decline phases were obtained by both the spread plate and OD methods. The ranges O.D values and colony growth at each time interval is indicated in the graphs. However, data generated from this study clearly revealed the importance of preliminary characterization for further molecular studies.

REFERENCES

1. Ramesh S, Mathivanan N., "Screening of marine actinomycetes isolated from the bay of bengal, india for antimicrobial activity and industrial enzymes", world j. Microbiol. Biotechnol, 2009: doi 10.1007/s11274 - 009 - 0113 - 4.
2. Ramesh S, Rajesh M, Mathivanan N., "Characterization of a thermostable alkaline protease produced by marine *Streptomyces fungicidicus* mml1614", bioprocess biosyst. Eng 2009; 32: 791 - 800.
3. Subhashree parida, Ram chandra jena¹, kailash chandra samal, pradeep kumar chand., 'isolation and identification of pathogenic bacteria from brackish', malaysian journal of microbiology, 2012; vol 8(3): 197-202.

4. Sogin ML, Morrison HG, Huber JA, Welch DM, Huse SM., "Microbial diversity in the deep sea and the Underexplored rarebiosphere.,PNAS.103,12115-12120,doi: 10.1073/pnas.0605127103.
5. Frits schut, Egbert J, De vries jan C, Gotitschal, betsy R., Robertson, Wim harder, Rudolf a, Prins, Don k. button., "Isolation of typical marine bacteria by dilution culture: growth, maintenance, and characteristics of isolates Under laboratory conditions", Applied and environmental microbiology,1993,59 (7): 2150-2160.
6. Zengler K, Toledo G, Rappe M, Elkins J, Mathur EJ, Short JM, Keller M., "Cultivating the uncultured". P natl acad sci , 2002; 99:15681–15686.
7. Pettit RK., "culturability and secondary metabolite diversity of extreme Microbes: expanding contribution of deep sea and deep-sea vent microbes To natural product discovery", Mar biotechnol, 2011;13:1–11.
8. Aneja K., experiments in microbiology plant pathology and biotechnology, fourth edition, new age international (p) ltd., publishers, new Delhi, 2003; pp 320.
9. Panneerselvam A, Arumugam A., "isolation and identification of bacteria from lake water in and around Ranipet area, vellore district", International journal of pharmaceutical & biological archives, 2012; 3(4):1008-1011.
10. Casciato, D A., Stewart PR., Rosenblatt JE., "Growth curves of anaerobic bacteria in solid media", Applied microbiology, 1975 ; 29(5) 610-614.