U of K. J. Vet. Med. & Anim. Prod. Vol. 3, No 1, 2012 (98-106)

ISOLATION OF POTENTIALLY PATHOGENIC BACTERIA FROM THE INTERNAL SUDANESE COASTAL WATERS OF PORT SUDAN

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المستخلص

أجريت هذه الدراسة بهدف عزل بعض أنواع البكتريا التي قد تكون ممرضة في بعض الحالات والتعرف عليها والتي من المحتمل وجودها في مياه البحر الأحمر الداخلية لمدينة بورتسودان (خور سلالاب). جمعت من هذه المنطقة 10 عينات ماء بحر لعزل هذه البكتريا والتي يعزز وجودها من وجود الأنواع الممرضة الصريحة.

تم زرع عينات المياه في وسط مرق السلانايت وبعد فترة تحضين 48 ساعة في درجة حرارة 37ْم تم التخطيط من هذه العينات على وسط أجار الماكونكي ووسط السالمونيلا . شيقلا أجار، بعد ذلك أخذت المستعمرات من الوسطين وتم تنقيتها ومن ثم التعرف عليها.

أظهرت الدراسة وجود عدة أنواع من البكتريا إنتهازية الإمراضية والأنواع التي تم عزلها كانت هي الإشريكية القولونية، الكلبسيلا الرئوية، المتقلبة الرائعة، البكتريا الليمونية، الزائفة الزنجارية، الزائفة القلوية الزائفة المتألقة، العقدية البرازية والضمة الزاوية وهذا يعزز إحتمال وجود الأنواع الممرضة الصريحة سواء كانت بكتريا أو فيروسات أو طفيليات.

أوضحت الدراسة وجوب زيادة الاهتمام بالتلوث البحري وتحسين إدارة المياه ووضع إستراتيجية للارتقاء بالصحة العامة وصحة البيئة في منطقة بورتسودان. وعلى الرغم من عدم وجود

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إحصاءات لعدد الذين يسبحون في تلك المنطقة والأمراض المرتبطة بالسباحة إلا أنه من المؤكد أن ذلك في ازدياد خاصة مع التطور والاهتمام الذي تشهده المنطقة بالسياحة والأنشطة المتعلقة بها.

Abstract

This study was carried out to isolate and identify some species of opportunistic pathogenic bacteria that may be present in the inner Sudanese coastal waters of Port Sudan (Khour Salalab). From this area, 10 sea water samples were collected and used for isolation and identification of possible potentially pathogenic bacteria which increases the chances of the presence of frank pathogens.

Water samples were inoculated in Selenite broth and after an incubation period of 48hrs at 37ºC, streaked onto MacConkey's and Salmonella-Shigella agar then the pure colonies were picked and identified.

A variety of opportunistic pathogens were isolated which were: *Escherichia coli*, *Klebsiella pneumoniae*, *Proteus mirabilis*, *Citrobacter freundii*, *Pseudomonas aeruginosa*, *Pseudomonas alcaligenis*, *Pseudomonas fluoresens*, *Streptococcus faecalis* and *Vibrio anguillarum* and this increases the possibility of the presence of frank pathogens.

The study showed that more efforts should be made to reduce marine pollution and improve water sources management and a need for strategy to improve general and environmental health. there are no records about the number of swimmers and swimming-related diseases in this area. However, due to the develop of tourism and its related activities, chances of swimming related diseases are likely to increase.

Key words: Contamination, Isolation, Bacteria, Coastal water, Red Sea.

Introduction

Human faecal pollution is largely attributed to sewage overflows and constitutes the greatest public health threat because humans are reservoirs for human pathogens including bacteria, protozoa, and viruses (Toranzos and Mcfeters, 1997). Animal faecal waste is also a serious health concern as it may contain pathogens such as *Cryptosporidium*, *E. coli* 0157:H7, or *Salmonella* and may enter recreational water bodies by means of contaminated run–off (Dorfman, 2004). Epidemiological data demonstrate that swimming in sewage–contaminated water is associated with gastroenteritis and inflammation of the eyes, ears, skin, nasal membranes and upper respiratory tract. The likelihood of contracting these symptoms increases with the concentration of pollution and length of exposure to polluted water (Alexander *et al*, 1991).

It is becoming increasingly clear that the concept that enteric pathogens die quickly when exposed to sea water may not be accurate (Charoenca and Fojioka, 1995). Some Gram–negative organisms adapt to low–nutrient environments through reductive division; that is, with no change in total biomass, more organisms develop, but at a greatly reduced metabolic rate (Colwell and Spira, 1992). Additionally, *E. coli*, *Salmonella* spp., *Legionella* spp., *Campylobacter* spp. and *Shigella* spp. demonstrate "viable but nonculturable" (VNC) states (Colwell *et al*., 1985). Despite being undetectable by conventional culture plate methods, VNC organisms have been demonstrated to have clinically virulent potential, and could be present in marine systems (Colwell and Spira, 1992).

Gastrointestinal, respiratory, dermatologic, and ear, nose, and throat infections are not uncommon after recreational or occupational uses of water, but clinicians seldom elicit information regarding potential exposures when interviewing patients with these complaints.

This study was carried out to investigate the quality of Sudanese inner coastal waters of Port Sudan by isolation and identifying the various bacteria which are predominantly excremental in origin. These comprise the normal faecal bacteria that can be found in coastal waters

Materials and Methods

The study area

Water samples were collected from the inner coastal waters of Port Sudan (Khour Salalab, the northern arm of Port Sudan harbour), which showed high counts of indicator bacteria in a previous study (Abdalla, 2006). Ten water samples were collected for the detection of the presence of pathogenic bacteria in Sudanese coastal waters. The samples were collected in one day from one station and were each spaced at distances of about 100 m from the other along the coastal line of the inner water.

Sampling

Sampling was performed according to (Cheesbrough, 2000).

Samples were collected between 8 and 9 AM and were transported as soon as possible to the laboratory and examined within 2 hr or refrigerated (Regional Organization for the Protection of the Marine Environment, 1999).

Isolation and identification of pathogenic bacteria in sea water

For isolation and identification of pathogenic bacteria in sea water, water samples were inoculated in containers of selenite broth medium. After an incubation period of 48 hr, at 37ºC the broth was streaked onto plates of MacConkey's agar. Subcultures were also made at the same time in a new selenite broth medium which was streaked onto Salmonella–Shigella agar (SSA) after an incubation period of 48 hr at 37ºC.

Predominant colonies were frequently picked from both MacConkey's agar and SSA agar and purification was done by repeated subculturing of isolated colonies onto these solid media.

Cultural characteristic, microscopical examinations and biochemical tests were used for identification of isolates which was done in accordance to Barrow and Feltham (1993) procedures.

Results

Pathogenic bacteria in sea water

No primary pathogenic bacteria were isolated from the sea water samples, but some opportunistic pathogens have been isolated and identified as shown in the table (1).

Table 1: Bacterial species isolated from 10 samples of sea water collected from the inner waters of Port Sudan area (khour Salalab).

Bacterial species	No. of isolates	Percentage of total isolates
Escherichia coli	5	22.7 %
Klebsiella pneumoniae	2	9.2 %
Proteus mirabilis	8	36.4 %
Citrobacter freundii	1	4.5 %
Pseudomonas aeruginosa	1	4.5 %
Pseudomonas alcaligenis	1	4.5 %
Pseudomonas fluoresens	1	4.5 %
Streptococcus faecalis	2	9.2 %
Vibrio anguillarum	1	4.5 %

Discussion

Although indicator bacteria are not necessarily pathogenic, their presence in the coastal waters indicates the possibility of the presence of pathogenic organisms such as bacteria, viruses and parasites. Results of isolation and identification tests revealed the presence of a variety of opportunistic bacterial pathogens in the coastal waters which enhances the possibility of the presence of primary pathogenic bacteria and other pathogenic organisms.

The opportunistic pathogens isolated from the inner waters of Port Sudan can cause numerous infections as mentioned by Cheesbrough (2000), but the likelihood of infection also depends on individual susceptibility; for example, acquired immunity may protect an individual from marine–associated infections. Children appear to be at greater risk than adults (Stevenson, 1953), and tourists without prior immunity may be particularly at high risk per exposure (Cabelli, 1983). However, among local populations, the poor are at highest risk because they tend to swim at periurban, polluted beaches and may have both poorer diets and weaker general health. As expected, immunocompromised persons are also a high–risk category for infectious disease (Cabelli *et al*., 1997).

Among the isolated species of bacteria, although most strains of *E. coli* are not regarded as pathogens, they can be opportunistic pathogens that cause infections in immunocompromised hosts. There are also pathogenic strains of *E. coli* which when ingested can cause gastrointestinal illness in healthy human (USFDA, 2002).

Sediments are thought to be reservoirs for certain pathogens. For example, indicator bacteria (faecal coliform and enterococci) and *Vibrio parahaemolyticus* were found to survive in high numbers in sewage–polluted intertidal sediments (Shiaris *et al*., 1987). Bothner *et al*. (1994) isolated faecal coliform from marine sediments beneath a deep ocean dumpsite off New York where sediments could be readily resuspended in high–energy coastal environments. These sediments might be a significant source of pathogens to the water column by the action of waves and the activities of swimmers in the shallow waters as in the case of inner waters of Port Sudan area.

The point of valuable importance that has to be mentioned here is that humid weather of Port Sudan and the Red Sea coast in general, except during summer seasons, can contribute largely to the survival of potentially pathogenic bacteria outside of the water system and increase their chances to reach the sea.

There are no data about the number of swimmers in the recreational sites of the Sudanese Red Sea coast and swimming–related infections. The number of swimmers usually increases during the summer months. However, the high air and seawater temperature and the shortage of rainfall during summer seasons which results in no water run–off from the land, may affect the load, and viability of indicator and pathogenic microorganisms. Consequently, this may affect the rate of marine–associated human infections.

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