Bacterial indicators of pollution of the Douala lagoon, Cameroon: Public health implications

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Abstract

Background: Indiscriminate disposal of untreated wastes which are often heavily laden with sewage microorganisms some of which are pathogenic to humans into aquatic environments near cities could serve as potential dangers to human health**.**

Objective: A prospective study was undertaken to investigate the scope of potential bacterial pathogens and to assess the extent of pollution of the Douala lagoon.

Methods: A total of eighty water samples were collected fortnightly from the lagoon at five stations from March to October 2005 and analysed for heterotrophic bacterial densities, coliform counts, faecal coliform and faecal streptococcal counts. Bacteria were isolated and identified using standard microbiology and biochemical techniques.

Results: High heterotrophic bacterial counts ($33 \times 10^5 - 161 \times 10^5$ CFU/ mL), total coliform counts (1.8×10^2 - 2.4×10^2 CFU/ 100 mL), faecal coliform counts ($2.2 \times 10^2 - 2.4 \times 10^2$ CFU/ 100 mL) and faecal streptococcal counts ($2.1 \times 10^2 - 2.3 \times 10^2$ CFU/ 100 mL) were observed in all sampling stations. Eleven species of bacteria: *Bacteroides fragilis*, *Proteus vulgaris*, *Klebsiella pneumoniae*, *E. coli*, *Enterococcus faecalis*, *Enterobacter aerogenes*, *Citrobacter freundii*, *Aeromonas hydrophila*, *Pseudomonas aeruginosa*, *Bacillus mycoides* and *Serratia marcesens*, were frequently isolated.

Conclusion: The presence of potential bacterial agents such as *Bacteroides fragilis*, *Pseudomonas aeruginosa, Aeromonas hydrophila, Klebsiella pneumoniae* and *E. coli* in the lagoon may pose a serious threat to the health and well being of users of the Lagoon and calls for urgent intervention.

Key words: Bacteria; Pathogenic; Coliform; Heterotrophic; Lagoon; Cameroon.

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Introduction

Aquatic environments near cities are usually prone to overloading with a variety of pollutants either through direct or indirect discharges. This situation may be worsened by the indiscriminate disposal of untreated wastes, which are often heavily laden with sewage into actively used waters. Sewage polluted waters carry sewage microorganisms, some of which are pathogenic to humans[1-4].

Several researchers [5-11] have documented the public health significance of faecal pollution of natural waters. Apart from health concerns, the indiscriminate dumping of untreated wastes into aquatic environments brings about physical, chemical and biological deteriorations of such water bodies when these discharges are beyond their self-purifying capacity[12-14]. This will no doubt endanger the resident aquatic organisms as well as impair the beneficial uses of the water[6, 15, 16].

The Douala lagoon, like many coastal lagoons, serves as a seaport, centre for recreational sailing and a sink for disposal of domestic and industrial wastes. A paucity of information exists on the extent of pollution of the lagoon and hence its public health implication. Ekane and Oben[17] investigated the biochemical characteristics of the Douala lagoon and Limbe estuary and reported a great impact of human activities on these parameters. Oben and Oben[18] in a plankton survey of the lagoon reported a high rate of mortality of some of these organisms and associated their findings to the influence of discharges from food and chemical industry. It is apparent that with the high level of waste disposal and other forms of contamination, the lagoon would likely represent a reservoir of potential bacterial and other pathogens. It is against this background that the present study was initiated since we are not aware of any investigation on the bacteriology of the lagoon. This study therefore appraises the level of bacterial contamination, the sanitary quality and potential health risk of the lagoon to humans.

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Materials and methods

Study Site

The Douala lagoon is part of the coastal plain of Cameroon, which stretches from Rio-Del-Rey through Victoria, Tiko, Douala and Kribi down to Equatorial Guinea. The area experiences an ever-increasing population and industries (chemical, food processing, textile and petroleum) flank the lagoon.

Sampling

Surface water samples (n= 80) were collected fortnightly for a period of eight months (March to October, 2005) using standard methods[19]. Quadruplet composite samples were collected into sterile 300 mL plastic bottles from the following points receiving either domestic or industrial wastes: station 1, behind an engineering company; station 2, after an effluent discharge point from the petroleum depot; station 3, an open space with no trees with recreational activities predominating; station 4, receives waste from a chemical company, also serves as a dump site for domestic wastes and, station 5, after the Wouri bridge, serves as dump site for domestic wastes (Fig.1) .

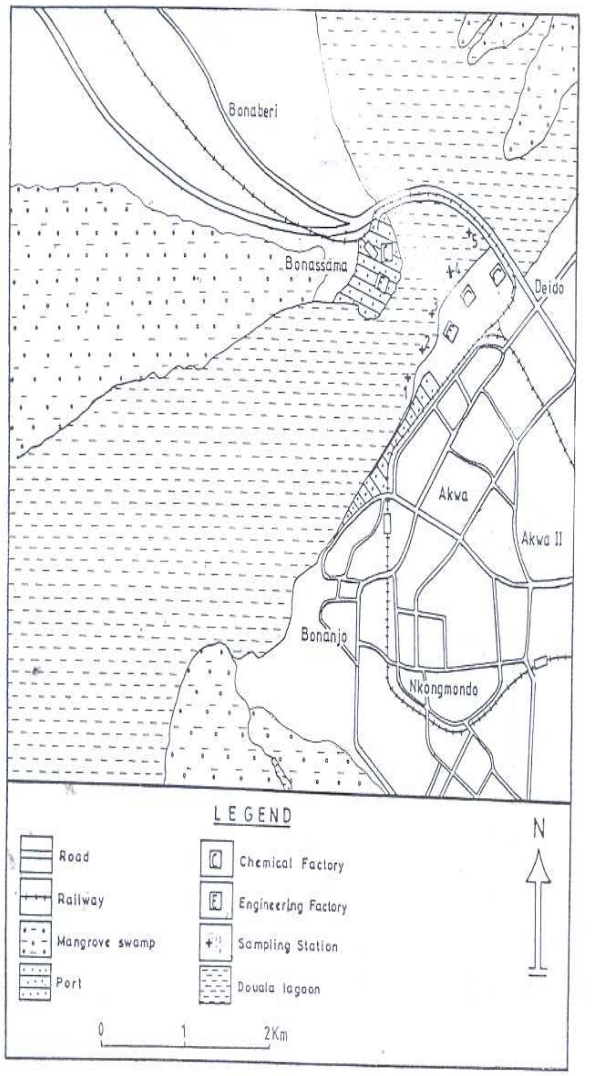

Fig. 1: Map of the Douala Lagoon showing the sampling stations and the sources (Factories) of effluents (pollutants).

Heterotrophic Bacterial Counts

Samples were serially diluted. Total viable heterotrophic bacterial counts were obtained by the standard plate count (SPC) method on nutrient agar. Volumes of 0.1mL were plated in triplicate and incubated at 35°C for 48 hours[19].

Enumeration of Indicator Organisms

Standard methods were employed for the isolation and enumeration of indicator organisms[19]. Three-tube, 10-fold dilutions of McConkey broth were used to determine the most probable number (MPN) of total coliforms. For total coliforms, fermentation tubes (containing Durham tubes) in a 3-3-3-tube regimen were incubated at 37°C for 24 hours. For faecal coliforms, incubation was at 44.5°C for 24 hours. Faecal streptococci counts were obtained by inoculating samples into Hannay and Norton's sodium azide broth in similar regimen as for total and faecal coliforms and incubating at 37 °C for 72 hours[2]. Counts (MPN) were estimated from probability tables[19].

Identification of Isolates

Bacteria were identified based on colonial characteristics on nutrient and McConkey agar, Gram coloration, motility and biochemical reactions using previously established schemes[20-22]. Isolates were confirmed using the API 20E (Biomerieux SA, France) kit according to the manufacturer's instruction.

Statistical analysis

Variations in the parameters analyzed between the sampling stations and sampling period were compared using the two-way analysis of variance test. P values <0.05 were considered significant.

Results

Heterotrophic bacterial counts, ranging from 33×10^5 CFU/mL in station 2 to 161×10^5 CFU/mL in station 3 were recorded in the lagoon (Fig. 2). The counts in station 3 were significantly higher ($P<0.05$) than in other stations. With respect to sampling period, counts were significantly higher ($P<0.05$) in the wet months (May to October) than in the dry months (March to April).

Also, very high coliform counts (1.8×10^2 – 2.4×10^2 CFU/100mL), faecal coliform counts (2.2×10^2 — 2.4×10^2 CFU/100mL) and faecal streptococcal counts (2.1×10^2 –2.3×10^2 CFU/100mL (Fig.3) were observed throughout the study period. Counts though high reflected no significant differences ($P>0.05$) between the stations.

Eleven bacterial species were isolated from the lagoon (Table 1). The majority of these were enteric organisms: *E.coli* (100%), *Klebsiella pneumoniae* (100%), *Proteus vulgaris* (100%) and *Enterobacter aerogenes* (95%) predominating. Other gram-negative rods included *Bacteroides fragilis* (100%), *Aeromonas hydrophila* (50%), *Citrobacter freundii* (50%), *Pseudomonas aeruginosa* (45%) *and Serratia marcescens* (15%). Prominent among the gram-positive genera were *Enterococcus faecalis (*100%) and *Bacillus mycoides* (35%). Of these isolates, *B. fragilis, P. vulgaris, K. pneumoniae, E. coli* and *E. faecalis* were the most predominant, being isolated from all samples.

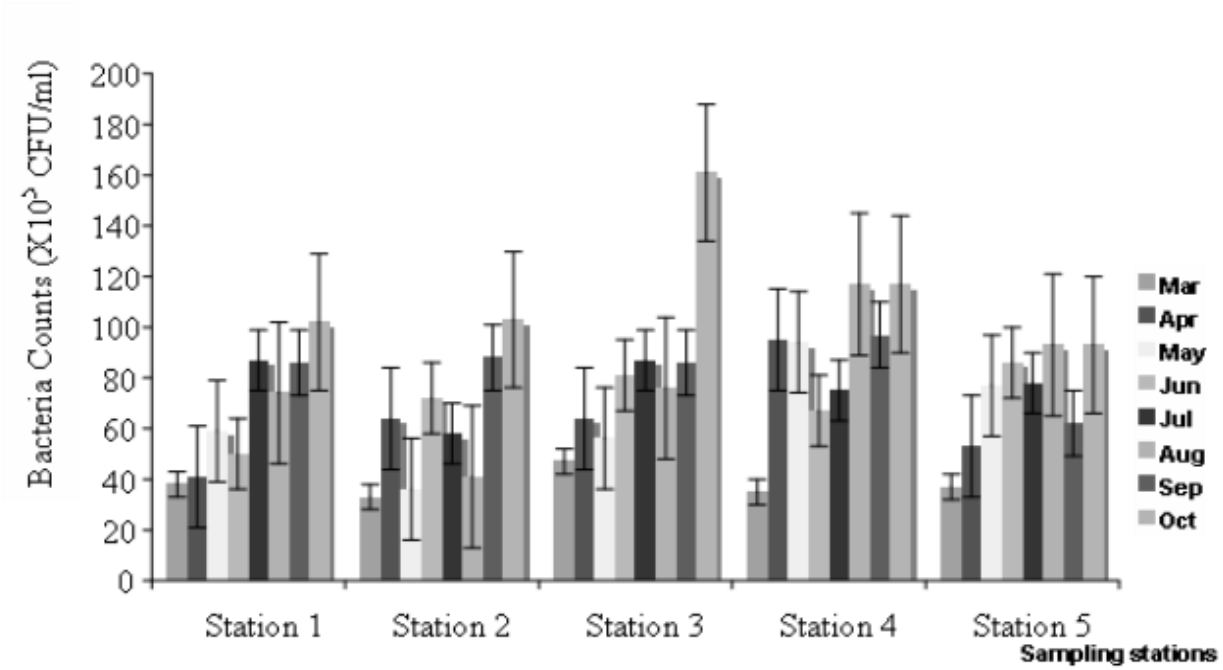

Fig. 2: Monthly Variations in the heterotrophic bacteria counts

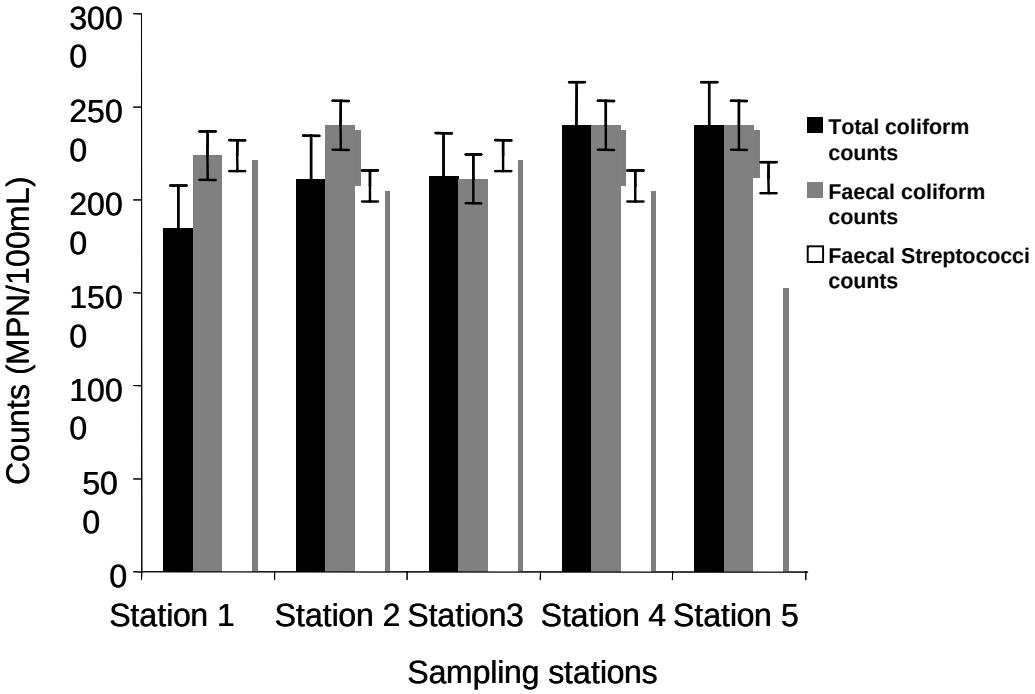

Fig. 3: Fortnightly variations in total coliform, faecal coliform and faecal streptococci counts.

Table 1: Frequency of isolation of bacteria.

Organisms isolated	Samples screened (Number positive)	Frequency of isolation (%)
Bacteriodes fragilis	80 (80)	100
Proteus vulgaris	80 (80)	100
Klebsiella pneumoniae	80 (80)	100
Escherichia coli	80 (80)	100
Enterococcus faecalis	80 (80)	100
Enterobacter aerogenes	80 (40)	95
Citrobacter freundii	80 (76)	50
Aeromonas hydrophila	80 (40)	50
Pseudomonas aeruginosa	80 (36)	45
Bacillus mycoides	80 (28)	35
Serratia marcesens	80 (12)	15

Discussion

Heterotrophic bacterial counts ranging from 33×10^5 CFU/mL to 161×10^5 CFU/mL were recorded in the lagoon. These counts were however significantly higher ($P<0.05$) in the wet months (May to October) than in the dry months (March to April). These high counts could be due to high levels of organic matter present in the lagoon as a result of indiscriminate dumping of wastes, and are in agreement with the findings of Tatah and Ikenebomeh[23] who in a similar study in Nigeria attributed such high counts to the high organic matter content of Ikpoba River. Fleisher *et al* [7] and Sequel *et al*[24] equally reported similar findings. During a rainfall, run off water washes organic matter as well as bacteria[12]. We therefore speculate that this may also explain the significantly higher counts obtained in the wet months during which period run off get into the lagoon. Bacteria form a link between primary producers and consumers; it would therefore appear that pollution affects the aquatic food chain. The high bacterial counts are indicative of the sanitary status of the lagoon. It was also of interest to note the very high total coliform counts ($1.8x10^2 - 2.4x10^2$ CFU/ 100mL), faecal coliform counts ($2.2 x10^2 - 2.4x10^2$ CFU/ 100mL) and faecal streptococcal counts ($2.1x10^2 - 2.3x10^2$ CFU/ 100mL throughout the study period. Such counts are highly undesirable and indicate severe faecal pollution of the lagoon, hence the possible presence of human pathogens.

Eleven bacterial species were isolated from the lagoon. The majority of these were enteric organisms, with members of the Enterobacteriaceae *(E. coli*, *Enterobacter aerogene*s, *Klebsiella pneumoniae, Proteus ulgaris)* predominating. Of these isolates, *B. fragilis, P. vulgaris, K. pneumoniae, E. coli* and *E. faecalis* were the most predominant, being isolated from all samples. This is consistent with the findings of other investigators [1, 2, 12, 25, 26] who isolated these organisms from water bodies heavily contaminated with faecal material. The majority of our isolates are human pathogens. The lagoon is fed by polluted swamps and drainage ditches carrying faecal pollution from neighbouring densely crowded residential areas which have inadequate sanitary facilities. The uncontrolled urbanization around the swampy areas surrounding the lagoon harbours mostly the poor city dwellers who do not have access to clean water or basic sanitary facilities. As waste disposal facilities are grossly inadequate to provide complete evacuation of both solid and liquid wastes, these inhabitants resort to use of the lagoon as an "infinite sink" for their wastes. The high incidence of human pathogenic bacteria in the lagoon may indicate their possible presence in fish and other foods obtained therein. This lays credence to the fact that other researchers[26, 27] have isolated human pathogenic bacteria from aquatic foods from polluted waters. Though the lagoon is not used for drinking, it is of value for fishing and recreation thereby rendering it dangerous to the health of the population. Although cholera is endemic in Douala with some of the contributing factors to its endemicity being the location of the city of Douala at the mouth of the Wouri delta, presence of vast expanses of swamp, streams/drainage ditches infested with algae[28], we did not isolate *Vibrio cholerae* from samples during this study. Recent studies[29-31] have identified conditionally viable environmental cells of pathogenic V*ibrio cholerae* that resist cultivation by conventional techniques existing in surface waters as biofilms of partially dormant cells. This may explain the absence of this organism from our samples.

Conclusion

Based on our findings, it is likely that the lagoon could pose a serious threat to the health and well being of users and calls for urgent intervention. We therefore recommend that adequate waste disposal facilities should be provided to inhabitants of the Douala Municipality to prevent indiscriminate dumping of wastes into the lagoon; the Environmental Protection Agency should ensure that industries treat their wastes adequately before disposal into the lagoon and; the public should be educated on the health risks of indiscriminate disposal of wastes.

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