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**DISTRIBUTION OF HEAVY METALS AND *ESCHERICHIA COLI* IN SABAK BEACH
AND CAHAYA BULAN BEACH, KELANTAN**

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ABSTRACT

The study was carried out for determination of heavy metal and *Escherichia Coli* (*E.Coli*) in coastal water in two popular coasts along Kota Bharu, Kelantan. The samples were collected from July until November 2011. Eight elements of heavy metals which are chromium (*Cr*), manganese (*Mn*), lead (*Pb*), ferum (*Fe*), nickel (*Ni*), zinc (*Zn*), copper (*Cu*) and cadmium (*Cd*) were analyzed using Inductively Coupled Plasma Mass Spectrometry (ICP-MS). The homogenized samples were introduced to ammonium pyrrolidine dithiocarbamate - methyl isobutyl ketone (APDC - MIBK) extraction before it was analyzed by using ICP-MS. The existence of *E.Coli* in coastal water samples was determined by using Multi-tube Fermentation Technique. *E.Coli* used as an indicator for health risk water contact in recreational water. The consequences of heavy metals and microbial contamination in water samples were compared between both coastal. The results show that concentration of heavy metals and *E. Coli* oscillated in both locations of studies. Relative Standard Division (%rsd) of heavy metals Sabak Beach and Cahaya Bulan Beach; *Mn* = 87.49, 125.01; *Pb* = 134.51, 28.09; *Fe* = 112.82, 116.37; *Zn* = 26.19, 48.22; *Cu* = 133.15, 100.35; and *Cr* = 29.29, 39.41. It was observed that Sabak Beach is more polluted compared to Cahaya Bulan Beach this is because the residence area is closely with coastal and it caused coastal erosion. Furthermore monsoon factor also lead to pollution of the beaches because of raining and land and river runoff. Rainfall washed down the elements that contain in atmosphere directly into coastal water or river. However, the concentration of *E.Coli* at Sabak Beach and Cahaya Bulan Beach are 10 and 5 MPN/100ml, respectively. As a conclusion, the concentration metals and *E.Coli* in both beaches were lower than standard suggested by government except for *Fe* and *Cr* but it stay consider safe for any coastal activity.

Keywords: Coastal water, Heavy metals, *Escherichia Coli*, Most Potable Number (MPN), Inductively Coupled Plasma Mass Spectrometry (ICP-MS).

INTRODUCTION

Sabak Beach and Cahaya Bulan Beach are located in Kota Bharu region which is main town of Kelantan. Sabak Beach and Cahaya Bulan Beach located 12 km and 11 km from the middle of Kota Bharu. Sabak Beach near with the airport and industrial area which in Pengkalan Chepa. However, Cahaya Bulan Beach is most popular compared to Sabak Beach because of the traditional industries such as Batik Textile and Handicraft. Other than that, it is also popular among tourists because of the good services of hotels and resorts.

These two coastal strongly influence by a changes of monsoon because Sabak Beach and Cahaya Bulan Beach facing South China Sea. Changes of monsoon depend on vary of wind direction. Normally, Northeast Monsoon occur from late of November until February every year. However, from March to September transpire by Southwest Monsoon; and from September to October is transmission period or inter monsoon season [1].

The most considerable factors that contribute to coastal water pollutions are exhaustive human activity, discharge of municipal waste and effluence from industry near coastal area [2]. Natural sources of heavy metal

pollution in coastal water are through mechanical and chemical wearing rocks and land and river runoff. Rainfall washed down the elements that contain in atmosphere directly into coastal water or river, windblown the element or component from [3-5]. *E. Coli* basically arising from improper treatment of sewage tank, agriculture waste and food waste from household and food industry [6]. Consequently, the distance between coastal area and town, residential area and industry are also influenced the pollution.

The aim of this studies were conducted to determine levels of heavy metals of chromium (*Cr*), magnesium (*Mn*), lead (*Pb*), ferum (*Fe*), nickel (*Ni*), zinc (*Zn*), copper (*Cu*), cadmium (*Cd*) and *E. Coli* in coastal water. Additionally studies were conducted to relate the coastal condition measured to the changes of monsoon physical coastal condition.

MATERIALS AND METHODS

Collection samples and preservation

The experiment was started with collecting sample at three points in two selected coastal along Kota Bharu in July until November 2011. Coastal water samples (500ml) were collected in pre-acid cleaning Polypropylene bottle for heavy metal determination [4]. The sample immediately preserved using 50% HNO₃. However, for *Escherichia Coli* (*E.Coli*) analysis, 125ml coastal water collected using sterilized container without chemically preservation. Both of samples were transferred immediately into ice box to avoid contamination and vaporization during transportation. The analysis of *E.Coli* must begin immediately, but heavy metal analysis within three months after sampling to ensure result is valid [7].

Hydrological variable determination

Hydrological variable such as salinity, temperature, dissolved oxygen, pH, turbidity and conductivity were measured during sampling by using multi-analyzer [7].

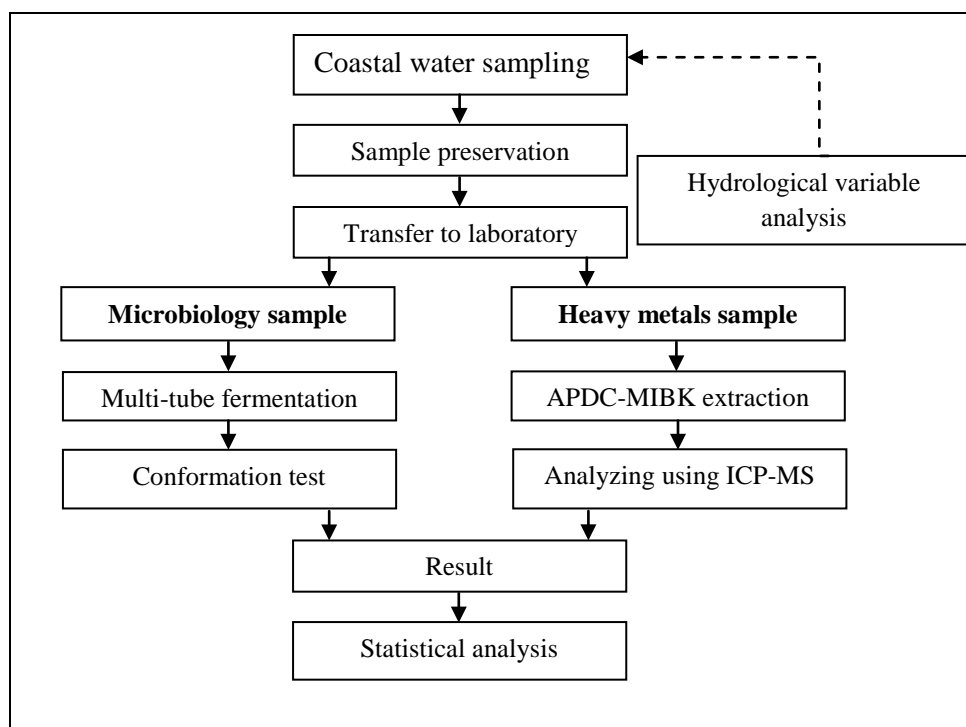


Fig. 1: Flow of research.

Determination of Escherichia Coli (E.Coli)

Multi-tube fermentation technique divided to three stages first presumptive, second confirmed and third completed stage. *Presumptive stage*: fifteen tubes of lactose broth divided into three sets incubated with difference quantity 10, 1.0 and 0.1ml of samples. Each lactose broth tube contains Durham tube inside. Positive presumptive stage was observed after 48 hours at temperature $35\pm 5^{\circ}\text{C}$ where it presents of gas bubble. The coliforms organism was estimated using standard MPN table. *Confirmed stage*: The positive tubes from presumptive stage were continue to confirmed stage by transfer a loopful of each positive presumptive broth to eosin methylene blue (EMB) agar. The *E.Coli* was indicating by presence of dark centers and green metallic sheen after incubated for 24 hours at $35\pm 5^{\circ}\text{C}$. *Completed stage*: Isolated colony was picked from positive confirmatory stage and incubated into lactose broth in test tube contain Durham tube and streaked on nutrient and MacConkey agar slant. The result was observed after 24 hours incubation, colonies on agar slant use to perform gram stain and colonies on MacConkey agar use in BD kit identification [7, 8].

Heavy metals determination

Ammonium pyrrolidine dithiocarbamate - methyl isobutyl ketone (APDC - MIBK) extraction: 100 ml aliquot from homogenized acid preservation samples was transfer to a 250 ml separating funnel. The sample were pre-concentrated and separated from bulk matrix by complexes with 1% APDC and extracted into MIBK. The organic layer containing metals chelates were collect and back extract using 50% nitric acid and diluted with 100 ml deionized water. The resulting solution was aspired to the ICP-MS for determination of *Cr, Mn, Pb, Fe, Ni, Zn, Cu* and *Cd* [1, 3, 7].

Statistical analysis

T test and analysis of variance (ANOVA) were used to explore data and to determine relationships between variables using MINITAB software. Statistical analysis included the determination range, mean, standard deviation and percent relative standard deviation (% rsd) for each of data set.

RESULTS AND DISCUSSION

Total nine coastal water samples were analyzed for determination of eight elements of heavy metals which are chromium (*Cr*), manganese (*Mn*), lead (*Pb*), ferum (*Fe*), nickel (*Ni*), zinc (*Zn*), copper (*Cu*), cadmium (*Cd*) and *Escherichia Coli (E.Coli)* at Sabak Beach (SB) and Cahaya Bualan Beach (CBB).

Table 1: Results of hydrological variables in July, September and November 2011.

Location	Month			Mean	SD	% rsd
	July	Sep	Nov			
Temperature ($^{\circ}\text{C}$)						
SB	31.23	31.10	31.25	31.19	0.08	0.27
CBB	30.26	30.64	30.26	30.39	0.22	0.73
DO (mg/l)						
SB	4.71	4.31	6.00	5.01	0.88	17.64
CBB	5.41	4.23	5.92	5.19	0.87	16.68
pH						
SB	8.15	8.27	8.16	8.19	0.07	0.79
CBB	8.02	8.26	8.21	8.16	0.13	1.57

SD = standard deviation; % rsd = relative standard deviation.

Table 2: Results of hydrological variables in July, September and November 2011.

Location	Month			Mean	SD	% rsd
	July	Sep	Nov			
Conductivity (mS/cm)						
SB	53.71	51.38	50.57	51.89	1.63	3.14
CBB	53.35	51.48	51.44	52.09	1.09	2.09
Salinity (ppt)						
SB	31.20	29.92	29.37	30.16	0.94	3.11
CBB	31.56	29.84	29.60	30.33	1.07	3.53

SD = standard deviation; % rsd = relative standard deviation.

Table 1 and 2 shows, surface water temperature varied from 31.10 to 31.25°C with mean 31.19 ± 0.08 °C at Sabak Beach (SB) and 30.26 to 30.64°C with mean 30.26 ± 0.22 °C at Cahaya Bulan Beach (CBB). Variation of dissolve oxygen (DO) content was from 4.31 to 6.00 mg/l with mean 5.01 ± 0.88 mg/l (SB) and 4.23 to 5.92 mg/l with mean 5.19 ± 0.87 mg/l. pH ranged from 8.15 to 8.27 with mean 8.19 ± 0.07 (SB) and 8.02 to 8.26 with mean 8.16 ± 0.13 (CBB). Conductivity varied from 50.57 to 53.71 mS/cm with mean 51.89 ± 1.63 mS/cm (SB) and 51.44 to 53.35 mS/cm with mean 52.09 ± 1.09 mS/cm (CBB). Salinity ranged from 29.37 to 31.20 ppt with mean 30.16 ± 0.94 ppt (SB) and 29.60 to 31.56 ppt with mean 30.33 ± 3.53 ppt (CBB).

The surface water temperature, pH, conductivity and salinity were higher in non-monsoon season (July) however DO record higher in monsoon season (Nov) [9]. Surface water temperature fluctuates from July until November and influence by sampling time and raining because of solar radiation and water mixing. The salinity was record higher in July compared with during pre-monsoon and monsoon season on September and November at both coastal. The salinity value was decrease in monsoon season because of rainfall and water runoff from land and river was mixed with coastal water and reduced salinity. The concentration of DO was affected by changes of temperature and salinity, DO were recorded increase during monsoon season because of lower utilization by phytoplankton in monsoon season. The values of pH were maximums during post monsoon due to reactions of the bicarbonate degradation and decomposition of organic matter during changes of monsoon [3].

The concentration of cadmium Copper (*Cu*) vary from 0 to 0.021 mg/l with mean 0.008 ± 0.011 mg/l (SB) and 0 to 0.004 mg/l with mean 0.002 ± 0.002 mg/l (CBB). Lead (*Pb*) in coastal water samples ranged from 0.003 to 0.165 mg/l with mean 0.065 ± 0.0087 mg/l (SB) and 0.010 to 0.16 mg/l with mean 0.012 ± 0.003 mg/l (CBB). The value of chromium (*Cr*) were varied from 0.360 to 0.665 mg/l with mean 0.526 ± 0.154 mg/l (SB) and 0.285 to 0.544 mg/l with mean 0.374 ± 0.147 mg/l (CBB). Iron (*Fe*) range from 0.253 to 3.447 mg/l with mean 1.510 ± 1.702 mg/l (SB) and 0.239 to 1.845 mg/l with mean 0.790 ± 0.916 mg/l (CBB) (as shows in Table 2).

Among these two coastal, almost all heavy metal shown higher values in Sabak Beach compared to Cahaya Bulan Beach. It is due to the coastal condition and the location of the coastal itself. From the observation, Sabak Beach located near to the industrial area, agriculture area and traditional village. However Cahaya Bulan Beach is closely with tourism area, for example hotels and resorts, restaurants and outlets for traditional fabric of 'batik'. Mean shows that the most of heavy metals are nested below the Malaysia Interim Marine Water Quality Standard (IMWQS) except *Cr* which is 0.526 mg/l (SB) standard value of 0.500 mg/l and *Fe* which are 1.510 mg/l (SB) and 0.790 mg/l (CBB) with standard 0.100 mg/l (Table 3) (DOE, 2011).

Table 3: Results of heavy metals in Sabak Beach (SB) and Cahaya Bulan Beach (CBB) in July, September and November 2011.

Location	Month			Mean	SD	% rsd
	July	Sep	Nov			
Cadmium, Cd (mg/l)						
SB	0.007	0.001	0.000	0.003	0.004	140.87
CBB	0.001	0.000	0.000	0.001	0.001	86.60
Copper, Cu (mg/l)						
SB	0.004	0.021	0.000	0.008	0.011	133.15
CBB	0.001	0.004	0.000	0.002	0.002	100.35
Lead, Pb (mg/l)						
SB	0.165	0.027	0.003	0.065	0.087	134.51
CBB	0.010	0.016	0.010	0.012	0.003	28.09
Chromium, Cr (mg/l)						
SB	0.552	0.360	0.665	0.526	0.154	29.29
CBB	0.285	0.292	0.544	0.374	0.147	39.41
Nickel, Ni (mg/l)						
SB	0.001	0.005	0.002	0.003	0.002	83.86
CBB	0.001	0.002	0.001	0.002	0.000	24.74
Manganese, Mn (mg/l)						
SB	0.000	0.026	0.034	0.020	0.017	87.49
CBB	0.001	0.046	0.010	0.019	0.024	125.01
Zinc, Zn (mg/l)						
SB	0.067	0.044	0.044	0.051	0.013	26.19
CBB	0.023	0.056	0.068	0.049	0.024	48.22
Ferum, Fe (mg/l)						
SB	0.827	3.447	0.253	1.51	1.702	112.82
CBB	0.278	1.845	0.239	0.79	0.916	116.37

SD = standard deviation; % rsd = relative standard deviation.

Specifically, Figure 2 shows *Cr* only critical at Sabak Beach on July (0.552 mg/l) and November (0.665 mg/l); and Cahaya Bulan Beach on November (0.544 mg/l). *Cr* values was higher than IMWQS standard values of 0.500 mg/l [11]. The results show P value > 0.05 between both coastal. Source of *Cr* in coastal water from textile industries and from natural sources such as leaching from topsoil and rocks [10], Because of that the *Cr* in Sabak Beach is more higher compared in Cahaya Bulan Beach. However on November, *Cr* also shown the highest value at Cahaya Bulan Beach due to raining and land runoff make coastal polluted.

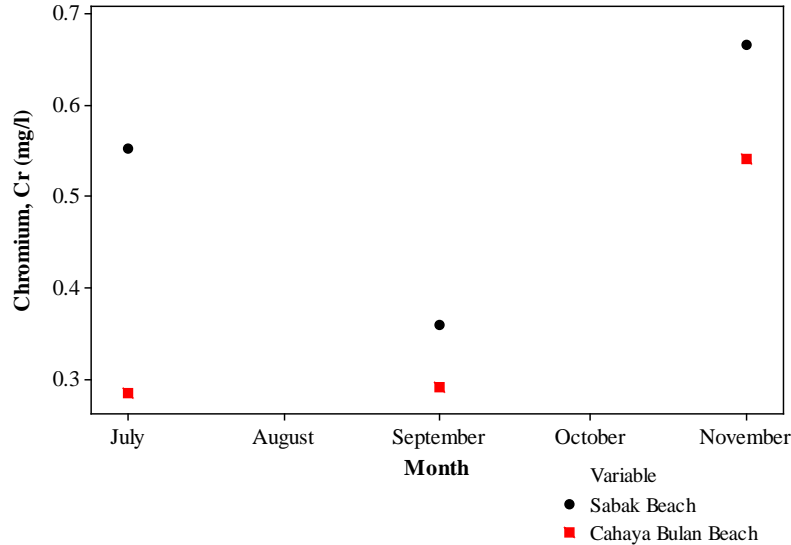


Fig. 2: Variation of chromium concentration in Sabak Beach and Cahaya Bulan Beach.

Figure 3 demonstrates variation of *Fe* concentration in Sabak Beach and Cahaya Bulan Beach on July until November 2011. *Fe* also shows in higher concentration at both coastal with mean values of 1.510 ± 1.702 mg/l (SB) and 0.790 ± 0.916 mg/l (CBB). Mean value of *Fe* were higher than IMWQS standard values of 0.100 mg/l [11]. The results show no significant value between *Fe* in Sabak Beach and Cahaya Bulan Beach.

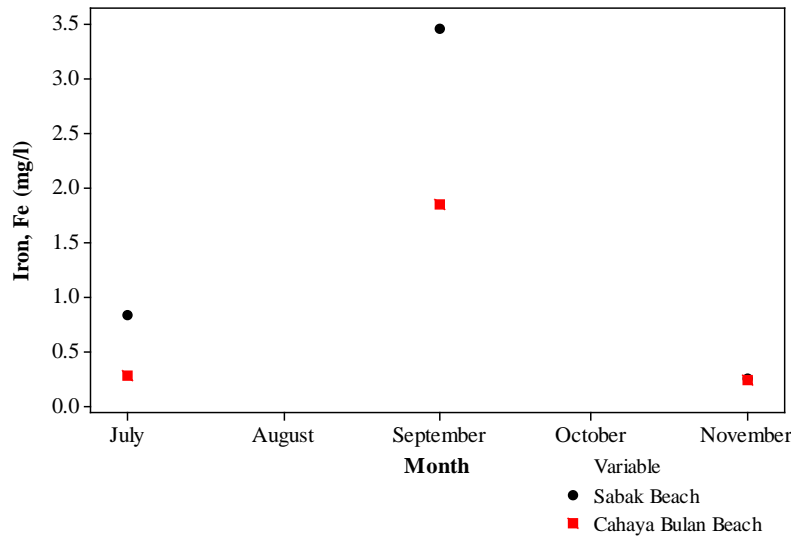


Fig. 3: Variation of Iron concentration in Sabak Beach and Cahaya Bulan Beach.

Concentration of *Escherichia Coli* (*E. Coli*) were relatively low and ranged from 2 to 21 MPN/100ml with mean values of 12 ± 9.512 MPN/100ml (SB); and 8 to 17 MPN/100ml with mean values of 11 ± 5.168 MPN/100ml (CBB) (Table 4). Mean of *E. Coli* were below than IMWQS standard values of 100 MPN/100ml [11]. No significant difference value was observed between both coastal, with variability 82.3 %rsd (SB) and 45.2 %rsd (CBB). Furthermore, no significant difference between *E. Coli* concentration and temperature, DO, salinity and pH in Sabak Beach. Only conductivity shows the significant difference with P- value < 0.05. However, in Cahaya Bulan Beach shows the significant difference between *E. Coli* temperature, turbidity, conductivity and salinity; except DO and pH were no significant difference.

Table 4: Results of *Escherichia Coli* in Sabak Beach (SB) and Cahaya Bulan Beach (CBB) in July, September and November 2011.

Location	Month			Mean	SD	% rsd
	July	Sep	Nov			
Escherichia Coli (E.Coli) (MPN/100ml)						
SB	11	21	2	12	9.512	82.32
CBB	8	17	9	11	5.168	45.15

SD = standard deviation; % rsd = relative standard deviation.

CONCLUSION

In general the present of study clearly shows *Mn*, *Pb*, *Ni*, *Zn*, *Cu*, *Cd* and *E. Coli* concentration were still relatively low compared to IMWQS standard values from July until November 2011 in both Sabak Beach and Cahaya Bulan Beach. However, *Cr* and *Fe* shown the higher concentration due to the location of the coastal near to the traditional industrial of textile which is 'Batik' and others industrial near coastal area [10]. Through the observation monsoon season contribute to changes of coastal condition physically and chemically. All heavy metals and *E. Coli* were increase due to pre-monsoon season on September and monsoon season on November. As a conclusion of this research, it can be conclude that two coastal used in this studied classified as save costal for any coastal activity such as bathing, boating and fishing.

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