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Assessment of Phenolic Compounds in Surface Waters from New Calabar River and Orashi River in the Niger Delta Region of Nigeria

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ABSTRACT: Phenolic compounds are hazardous with diverse health implications. The presence of phenolic compounds in rivers is triggered by several activities including petroleum products. The Niger Delta is one of the world's major hydrocarbon provinces with several health hazards created by oil exploration and exploitation and slow in action to oil spillages. Therefore this paper assessed phenolic compounds in surface waters from New Calabar River and Orashi River in the Niger Delta Region of Nigeria using GC/MS technique (Agilent 6890N Gas agilent 5975 mass selective Detector). Data obtained reveal that the average phenolic compounds recorded during the dry and wet seasons were 37.7156µg/l and 37.5878µg/l for New Calabar River and 94.3078µg/l and 85.5222µg/l for Orashi River, respectively. The level of phenolic compounds in the water samples indicated that both rivers were contaminated and not suitable for domestic use. Therefore, the people located within the riverbank, particularly Orashi River, are advised not to use the river water domestically except after treatment.

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Water covers about 70% of the surface globally but mostly affected due to pollution. Some nations are in situations where the conveying limit of the metropolitan territories is decaying because of population increase that does not match the rate of available assets (Bour, 2003). Most water bodies like rivers are capable of absorbing degradable wastes, but may become saturated at excessive load input into the rivers and this can affect the ecological balance of the rivers (Langat, 2009). Globally, water bodies are recipient of mechanical and municipal wastes, which contain diverse of contaminants that reduce the quality of water. According to *Anku et al. (2017),* there is deficiency of clean water and freshwater, which has led to one-fifth of urban occupants in most developing nations to lack quality and safe water supply. Bad disposal practices of wastes into water bodies and poor guidelines policies are the major reason for surface water pollution. The contamination of water resources by waste from hazardous substances of poorly treated waste treatment plants and contamination from inflowing wastewaters lead to contamination of surface water (*Aniyikaiye et al., 2019*). Polluted surface water is harmful to living organisms, the ecosystems and human health, which can be taken in through bioaccumulation, drinking of contaminated water from supply and recreational systems. Contaminated water also hinders the growth of agricultural products and its accumulation in farm produce can be disastrous to human health (Mainali, 2020).

River pollution by physicochemical and biological parameters has been dominant area of research in Nigeria, but phenolic compounds are also hazardous to aquatic environment, which can results from different sources. According to Michałowicz and Duda (2007), phenolic compounds in rivers occurred due to anthropogenic factor, such as contaminants released from chemical, petrol, and pharmaceutical industries. The compounds penetrate into to surface water or ecosystems through drainage of municipal and

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industrial sewage. Pesticides and phenolic biocides like pentachlorophenol are also sources of phenol pollution in water. Some phenols may form because of natural phenomena like in the case of phenol and p-cresol that formed due to the decomposition of organic matter and synthesis of chlorinated phenols, and they are catalyzed by fungi and plants. Phenolic compounds are widely distributed in the aquatic environment, and they are among the chemicals of major concern because of their persistent tendency in the environment for a long time and toxicity (Ekpete and Horsfall, 2011; Stelmaszewski, 2012).

In fact, they are introduced to the environment in many ways like waste from paper manufacturing, agriculture, pharmaceuticals, petrochemical industry, coal processing or as municipal wastes (*Ekpete et al., 2011;* Sun et al., 2014). They also occur naturally and through interactions with microorganisms, inorganic and some organic compounds in water, producing substituted compounds or other moieties that may be as toxic as phenolic compounds (*Anku et al. (2017).* Discharge of these compounds without treatment may lead to serious health risks to humans, animals, and aquatic systems. Phenolic compounds can inflict both severe and long-lasting effects on both humans and animals, as exposure to phenolic compounds could lead to cancer and damage to the red blood cells and liver, even at low concentrations (*Ekpete et al., 2012*; *Anku et al., 2017*).

Exposure to the compounds can also affect the endocrine system (*Olak et al., 2012).* In a study by Al *Hashemi et al. (2014*), total phenol and its derivatives were found in wastewater from refinery treatment plant discharged into rivers. The detected phenolic compounds, which varied significantly, included straight phenol, combined m- and p-cresols, o-cresol, tri and tetra-chlorophenols, and 4-chloro-3-methylphenol. The total phenol in the refinery effluents stream ranged from 1400-234000µg/l. Also, *Zhou et al. (2017*) recorded a highest average concentration of phenols in water and sediment samples from Yinma River Basin during the wet season at 0.10168µg/l and 0.12776µg/g, respectively. *Sharma et al. (2018*) reported a very high amount of phenolic compounds in Khandari Nala and Narmada Rivers that received wastes from sewage, industries and human activities like bathing and washing with concentrations varying from 450-2560µg/l. Most recent studies have equally reported different concentration levels of phenol and its derivative in surface water. For instance *Dolatto et al. (2020*) recorded 3.3µg/L phenol and 4.7µg/L 2,4-dimethylphenol in river water polluted by shale mining activities and oil shale reserve in São Mateus do Sul, Brazil, while Chen et al. (2021) obtained 0.06 to 14.12µg/L, 0.92 to 34,885µg/g and 3.54 to 34.09µg/g of total phenolic compounds concentration on water surface, suspended particulate matter and the sediment of a river, respectively. The individual phenolic compounds detected include pentachlorophenol, 2,4,6-trichlorophenol and chlorophenols at various degrees of concentrations. Another study by *El-Naggar et al*. (2022) reported a varying range of phenolic compounds in water samples at maximum concentrations of 109.2ng/L for phenol, 117.68ng/L for 2,4-dimethylphenols, 87.09ng/L for 2-nitrophenol, 66.30ng/L for 2,4 dichlorophenol, 60.08ng/L for pentachlorophenol, 91.43ng/L for 2-chlorophenol, 46.04ng/L for 2,4,6-trichlorophenol and 52.3ng/L for 4-chloro-3-methylphenol. Some of the concentration levels are above the recommended limit set by international regulatory bodies.

For example, the EPA set a water purity standard of less than 1 ppb for phenol in surface water (*Kazemi et al., 2014*). The toxicity levels usually ranged from 9 – 25mg/L for both humans and aquatic life (Sharma and Bhattacharya, 2017). Hence, it is necessary to assess the concentration level of phenolic compounds in surface water used for domestic, agricultural and industrial activities for possible treatment before use, as have been highlighted by several researchers (*Ekpete et al., 2010*; Ekpete and Horsfall Jnr, 2011; *Anku et al., 2017; Peters et al., 2018*; Mainali, 2020). Various studies have reported the presence of phenol and its compounds in rivers, but studies on assessment of phenolic compounds in Orash and New Calabar Rivers are still limited or not made available to the public. Therefore, this research assessed the presence of phenolic compounds in surface waters from New Calabar River and Orashi River in the Niger Delta Region of Nigeria

MATERIALS AND METHODS

Description of study areas: Water samples were collected in triplicate at three (3) locations in the rivers. The sampling locations were spaced some kilometres away from one another. In New Calabar River, the water samples were collected in locations within Choba, Ogbogoro and Rumuolumeni Towns in Obio/Akpor Local Government Area, while in Orashi River, the water samples were collected in locations within Mbiama, Odieke and Okarki Towns in Ahoada West Local Government Area. Both Local Governments are located in Rivers State of Nigeria. Table 1 shows the coordinates of the sample collection points.

Table 1: Coordinates of Sample Locations

S/No	River	Sample location	L.G.A.	Coordinates
1	New Calabar	Choba	Obio/Akpor	4.888617N, 6.897029E
2	New Calabar	Ogbogoro	Obio/Akpor	4.845140N, 6.922351E
3	New Calabar	Rumuolumeni	Obio/Akpor	4.811536N, 6.928650E
4	Orashi	Mbiama	Ahoada West	5.061525N, 6.451185E
5	Orashi	Odieke	Ahoada West	5.018959N, 6.432941E
6	Orashi	Okarki	Ahoada West	4.983985N, 6.431361E

Sampling method: The water samples were collected away from the river bank at specific intervals at 15 – 20 cm below the water surface. Few drops of concentrated H_2SO_4 (pH≥2) was added to the water samples to avoid chemisorptions. All the samples were collected into an amber glass bottle and stored in a cooler packed with ice prior to the analysis, which was done almost immediately after the sample collection. Sampling was carried out for a period of one year at one month interval, from December 2020 to April 2021 (dry season) and June to October 2021 (wet season). The water sampling was carried out according to the standard methods for examination of water and wastewater (APHA, 2010).

Determination of phenolic compounds: The phenolic compounds were determined using GC/MS technique (Agilent 6890N Gas agilent 5975 mass selective Detector). First, each cartridge was rinsed with 3ml aliquots of methylene chloride. The cartridge was allowed to drain dry after each flush. Then the cartridge was rinsed again with 3ml aliquots of methanol, but the methanol was not allowed to elute below the top of the cartridge packing nor the cartridge packing allow to go dry. It was further rinsed with 3ml aliquots of 0.05M hydrochloric acid. Meanwhile, before the dilute acid level dropped below the top edge of the packing, the vacuum was turned off. Approximately 3ml additional 0.05M hydrochloric acid was added to the cartridge, the transfer tube was attached, the vacuum turned on, and the sample added to the cartridge.

The inside of each sample bottle was rinsed with 8 – 10ml methylene chloride and vacuum was used to pull the solvent through the transfer tube and through the cartridge, collecting the solvent in a collection tube. The transfer tubing was removed from the top of the cartridge. 2 – 3ml methylene chloride was added to the top of the cartridge with a disposable pipette. This solvent was pulled through the cartridge at low vacuum, such that the solvent exits the cartridge in a dropwise fashion. Small amounts of residual water from the sample container and the SPE cartridge formed an immiscible layer with the eluate. The eluate was passed through the drying column, which is packed with approximately 5 to 7g of anhydrous sodium sulphate, and collected in a clean collection tube. The sodium sulphate was washed with at least 2ml of methylene chloride and collected in the same tube. The extract was concentrated to approximately 0.9ml in a warm (40°C) water bath under a gentle stream of nitrogen. The final volume was adjusted to 1ml using methylene chloride.

An aliquot of the sample extract was analyzed with the GC/MS system under the same conditions used for the initial and continuing calibrations. At the conclusion of data acquisition, the same software that were used in the calibration procedure to identify peaks in predetermined retention time windows of interest was used to examine the abundance of components of the chromatogram. It is important to note that the identification of analytes in the GC/MS system.a sample component was identified by comparing its mass spectrum (after background subtraction) to a reference spectrum in the user-created data base.

RESULTS AND DISCUSSION

The results obtained are presented and discussed in this section. The mean values obtained during the dry season (December 2020, February 2021 and April 2021) and wet season (June, August and October 2021) across the sampling locations in New Calabar and Orashi Rivers are presented in Tables 2 and 3. As shown in Tables 2 and 3, different concentrations of individual phenolic compounds were detected in the water samples, which included phenol, 2-chlorophenol, 3-chlorophenol, 4-chlorophenol, 2-methylphenol, 3-methylphenol, 4-methylphenol, 2-nitrophenol, 3-nitrophenol, 4-nitrophenol, cyclohexyl-4, 6-dinitrophenol, 2,3-dichlorophenol, 2,4-dichlorophenol, 2,5-dichlorophenol, 3,4-dichlorophenol, 3,5-dichlorophenol, 4-chloro-2-methylphenol, 2-chloro-5-methylphenol, 2,3-dimethylphenol, 2,4-dinitrophenol and 2,5-dimethylphenol. The concentrations varied across the sampling locations. Thus, Okarki and Mbiama locations in Orashi River recorded the highest and second highest mean values of the total phenolic compounds. Comparatively, the total phenolic compounds recorded in dry season at the locations in Orashi River were higher than the mean values recorded in wet season. This was also observed in sampling locations in New Calabar River except samples from Ogbogoro location.

Table 2: Phenolic compounds concentration in dry season

	New Calabar River				Orashi River	
Compounds (µg/l)	**Choba**	**Ogbogoro**	**Rumuolumeni**	**Odieke**	**Mbiama**	**Okarki**
Phenol	2.32±0.74	3.11±1.42	3.89±0.42	10.43±2.03	15.77±2.27	24.18±2.47
2-Chlorophenol	1.30±1.05	1.72±0.90	2.37±0.48	1.29±0.90	4.17±1.35	4.79±2.04
3-Chlorophenol	1.46±0.68	2.66±0.41	1.55±0.68	3.26±1.03	3.15±1.63	4.58±1.88
4-Chlorophenol	1.50±1.07	2.08±0.57	2.27±0.58	2.28±0.64	1.06±0.86	7.57±2.53
2-Methylphenol	0.91±0.48	1.36±0.61	1.65±0.70	1.45±0.59	4.29±0.53	2.84±0.90
3-Methylphenol	1.14±0.50	2.23±0.40	1.58±1.03	7.98±2.21	2.30±0.57	4.63±2.02
4-Methylphenol	2.18±1.77	1.95±1.36	2.02±0.58	2.13±1.47	0.72±0.05	3.54±2.53
2-Nitrophenol	1.11±0.39	1.98±1.63	1.25±0.47	4.47±1.20	8.19±2.31	3.76±1.79
3-Nitrophenol	1.52±0.89	1.35±1.09	1.47±1.45	2.40±1.61	1.49±0.58	5.05±2.78
4-Nitrophenol	0.40±0.27	1.29±0.14	1.83±0.39	1.34±0.24	3.09±1.13	4.06±1.33
Cyclohexyl-4,6-dichlorophenol	1.51±1.34	1.78±0.98	1.52±0.82	2.82±0.67	1.29±1.08	4.92±1.18
2,3-Dichlorophenol	2.40±1.73	1.84±1.66	1.00±0.66	6.23±1.83	2.11±1.27	6.74±4.37
2,4-Dichlorophenol	3.22±1.87	2.72±1.01	3.06±1.70	2.24±0.71	2.62±1.45	3.96±1.02
2,5-Dichlorophenol	1.28±0.34	4.34±2.82	4.46±2.61	3.44±0.69	7.40±1.54	10.62±1.31
3,4-Dichlorophenol	0.75±0.14	2.19±0.58	1.02±0.90	1.51±0.62	6.86±4.76	5.41±2.51
3,5-Dichlorophenol	1.06±0.25	2.68±0.48	2.23±0.45	4.70±1.17	6.09±1.25	7.77±1.53
4-Chloro-2-methylphenol	1.41±1.14	1.77±1.17	1.01±0.75	1.24±0.45	3.70±0.89	8.07±3.00
2-Chloro-5-methylphenol	1.81±1.46	2.13±1.29	0.56±0.39	1.59±0.67	1.77±0.91	4.21±2.69
2,3-Dimethylphenol	1.57±0.96	2.10±1.05	1.52±0.74	4.57±1.15	7.12±3.08	3.69±1.13
2,4-Dimethylphenol	1.51±1.17	1.47±1.62	0.77±0.35	1.32±0.73	3.37±1.72	3.75±1.50
2,5-Dimethylphenol	1.26±0.89	1.30±1.45	0.45±0.44	0.46±0.12	2.05±1.22	3.06±1.51

Table 3*:* Phenolic compounds concentrations in wet season

	New Calabar River				Orashi River	
Compounds (µg/l)	**Choba**	**Ogbogoro**	**Rumuolumeni**	**Odieke**	**Mbiama**	**Okarki**
Phenol	3.04±3.81	4.15±4.06	3.34±2.55	7.51±1.73	13.24±4.74	21.98±2.51
2-Chlorophenol	1.06±0.20	3.08±1.94	2.38±1.72	1.24±0.57	3.53±0.17	3.07±0.89
3-Chlorophenol	0.82±0.09	1.17±0.87	0.69±0.22	2.45±0.77	1.40±0.11	2.31±1.04
4-Chlorophenol	3.69±4.32	1.58±0.70	2.08±0.69	1.63±0.76	0.46±0.26	4.07±3.09
2-Methylphenol	0.39±0.10	0.44±0.07	1.41±0.34	0.49±0.23	3.76±0.50	8.77±11.69
3-Methylphenol	0.55±0.17	3.89±4.48	1.79±0.66	6.61±2.68	2.00±1.40	2.1±0.93
4-Methylphenol	6.27±8.53	1.09±0.88	3.04±2.82	1.19±0.55	3.48±5.24	3.64±3.55
2-Nitrophenol	0.44±0.08	1.02±0.60	0.68±0.57	2.64±0.66	7.82±0.70	1.14±0.39
3-Nitrophenol	0.62±0.21	0.60±0.15	2.19±1.17	3.92±2.94	0.70±0.16	7.46±4.54
4-Nitrophenol	0.12±0.07	0.72±0.37	1.24±0.95	1.14±0.06	2.39±1.99	3.24±0.62
Cyclohexyl-4,6-dichlorophenol	1.03±0.22	1.31±0.33	1.63±0.20	1.49±0.27	2.72±4.16	2.73±1.81
2,3-Dichlorophenol	1.75±0.68	1.56±0.49	2.31±2.96	6.99±5.49	1.35±0.28	10.16±9.71
2,4-Dichlorophenol	2.95±0.58	1.99±0.50	1.47±0.18	1.61±0.16	0.59±0.14	0.79±0.73
2,5-Dichlorophenol	0.48±0.19	7.54±5.03	6.19±2.88	2.96±2.98	10.4±10.86	7.83±2.30
3,4-Dichlorophenol	0.40±0.13	0.64±0.30	0.54±0.23	1.74±2.43	3.02±2.51	3.19±1.01
3,5-Dichlorophenol	0.60±0.15	1.95±1.78	1.56±0.54	3.52±3.63	7.69±5.93	10.08±5.30
4-Chloro-2-methylphenol	4.74±5.79	0.92±0.34	0.79±0.63	0.29±0.14	3.46±2.63	3.19±3.49
2-Chloro-5-methylphenol	1.44±0.28	1.35±0.06	1.68±2.39	4.95±5.60	0.80±0.72	7.32±5.43
2,3-Dimethylphenol	0.75±0.54	1.09±0.16	2.45±2.66	1.83±0.49	7.42±1.17	4.17±3.85
2,4-Dimethylphenol	1.23±0.66	2.59±4.11	0.72±0.47	2.62±3.44	5.63±2.89	3.91±2.61
2,5-Dimethylphenol	1.37±0.85	0.55±0.27	2.32±2.41	1.73±1.81	1.52±0.27	5.20±5.72

The average phenolic compounds recorded during the dry and wet seasons were 37.7156µg/l and 37.5878µg/l for New Calabar River and 94.3078µg/l and 85.5222µg/l for Orashi River, respectively. This implied that the average total phenolic compounds concentration recorded during the wet and dry seasons are relatively the same, particularly in New Calabar River. This observation agreed with some studies on seasonal variation of phenolic compounds in rivers (Zhou *et al*., 2017; Sharma *et al*., 2018). The marginal difference in average phenols recorded between the dry and wet seasons in New Calabar River implied that the sources of phenols pollution were relatively constant throughout the period of this study. Thus, artisanal oil refining activities, as potential source of phenol pollution, was continuous within the period of this study, which is justifiably why there was no difference. On the other hand, the level of variation of phenol recorded in the dry and wet seasons in Orashi River can be traced to the higher activities of illegal refining of crude oil and other anthropogenic activities during the wet season. In addition, the level of water in Orashi River was higher during the wet season compared to the dry season. This situation might have led to intrusion of various types of organic matters and runoff from agricultural lands into the river, leading to addition of phenolic compounds in Orashi River.

Conclusion: The total concentration of Phenolic compounds detected in New Calabar and Orashi

Rivers are above the permissible limit of 1.00μg, indicating that the rivers are polluted. Therefore, the rivers are not suitable for domestic use and consumption. Therefore, appropriate treatment is required to reduce the level of Phenolic compounds below the threshold limits before water from the rivers can be applied for domestic and other uses.

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