Effect of Gas Flare on Some Clinical Enzyme Biomarkers of Eleme Residents in Rivers State, Nigeria

Tamuno-boma Odinga\textsuperscript{1*}, Felix U. Igwe\textsuperscript{1}, Christine U. Gabriel-Brisibe\textsuperscript{2} and P. C. Dimkpa\textsuperscript{3}

\textsuperscript{1}Department of Biochemistry, Faculty of Science, Rivers State University, Nkpolu Oroworukwo, Port Harcourt, Nigeria.
\textsuperscript{2}Department of Medical Biochemistry, College of Medical Sciences, Rivers State University, Nkpolu-Oroworukwo, Port Harcourt, Nigeria.
\textsuperscript{3}Department of Chemistry, Faculty of Science, Rivers State University, Nkpolu-Oroworukwo, Port Harcourt, Nigeria.

Authors’ contributions
This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

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(1) Dr. Chunying Li, Georgia State University, USA.
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ABSTRACT

Background: Gas flaring, an environmental harmful practice, is prevalent in Eleme and most communities in the Niger Delta region of Nigeria.
Aim: This study explored the impact of gas flaring on some clinical enzyme biomarkers of Eleme residents who are constantly exposed to the flared gas.
Study Design/Place of Study: Volunteer subjects were randomly selected from representative groups resident in Eleme, an oil and gas producing and refining area, for over 15 years, while similar volunteer subjects resident in Nkpolu-Oroworukwo, a non-gas flaring community, served as the control.
Methodology: Blood samples were collected from the subjects and analyzed for selected clinical enzyme biomarkers including Creatine kinase (CK), Lactate dehydrogenase (LDH) and Aspartate aminotransferase (AST) using standard enzyme activity and Randox test kit methods.

\*Corresponding author: E-mail: Bomaodinga@gmail.com;
1. INTRODUCTION

Gas flaring has been recognized as a major environmental problem, contributing about 150 billion cubic meters of natural gas and contaminating the environment with about 40 metric tonnes of carbon dioxide (CO₂) annually [1,2]. It involves the process of burning-off associated gas from wells, hydrocarbon processing plants or refineries, either as means of disposal or as a safety measure to relieve pressure [3]. Although the flared gases are very similar in composition to natural gas and a cleaner source of energy than other fossil fuels, it has contributed to the increase in methane (CH₄), CO₂ and other greenhouse gas (GHG) emission in the atmosphere [1]. The composition of gas flared depends on the source of the gas going to the flared system. Gas flared from refineries and other process operations commonly contain a mixture of hydrocarbons, particulates and other environmental contaminants [4]. Air is said to be polluted when its natural users are impaired. Air pollution may cause profound undesirable effects on human health and the environment producing negative consequences [5].

Eleme is a Local Government Area (LGA) in Rivers State, (Niger Delta) Nigeria covering an approximately 140 square kilometers of land space. It is bound in the north by Obio/Akpor and Oyigbo LGAs, in the South by Okrika and Ogu Bolo LGAs, in the east by Tai LGA and the west by Okrika and Port Harcourt City LGAs. The Eleme environment host more than six operational gas wells with flare points clustered within the communities. The people of Eleme are traditionally farmers who depend largely on crops they cultivate for their livelihood. Associated gas from oil and gas exploitation is flared at ground levels within very close proximity of cultivated farmlands in the area [6,7]. Eleme LGA is also host to several heavy industries. There are two major petroleum refineries, two fertilizer plants, a petrochemical plant, a sea port, Federal Lighter Terminal (FLT) and Federal Ocean Terminal (FOT) with associated industrial plants that flare gas in the area [7]. Available records shows that most of the industrial wastes and effluents generated in the area are not discharged and disposed in compliance with established standards and Global Best Practices.

Residents of Eleme and other communities in the oil and gas rich Niger Delta region of Nigeria are constantly exposed to air pollution arising from gas flaring and other oil and gas exploration and exploitation activities. To assess the health impact of this exposure, regular epidemiological studies and assay of relevant biochemical parameters are expedient. Biomarkers are biochemical indicators found in the blood, other body fluids or tissues that serve as a sign of a condition or disease [8]. Biomarkers as measurable indicators of the severity or presence of a disease state [9]. It can be specific cells, molecules, genes, gene products, enzymes or hormones. The biomarkers of interest in this study includes; Creatine kinase (CK), Aspartate aminotransferase (AST), and Lactate dehydrogenase (LDH).

| Results: | Results obtained revealed that CK levels for female Eleme subjects increased by 61.20% when compare with the female control subjects. This increase was statistically significant (p<0.05). The CK levels for Eleme male subjects also increased by 37.36% compared to control. However, this increase was not statistically significant (p<0.05). LDH increased by 23.21% in the male subjects and 18.58% in the female subjects compared to control, while AST increased by 42.11% in the male subject and 11.32% in the female subjects compared to control. The results suggest that there could be impending damage to organs for which an increase in the biomarkers-Creatine kinase, Lactate Dehydrogenase and Aspartate Aminotransferase suggest an ongoing pathologic process. |
| Conclusion: | These results therefore suggest that the continuous exposure to gas flare is causing an increase in some critical tissue and organ function enzyme biomarkers in blood. This may eventually affect the health status of the residents, increase the tendency of developing ill health and generally reduce their quality of life. |

Keywords: Gas flaring; clinical enzymes; Eleme communities; creatine kinase; lactate dehydrogenase; aspartate aminotransferase.
high energy phosphate group in a reversible reaction. Hence, it enhances contractility of cardiac, skeletal and smooth muscles as well as being involved in the generation of blood pressure [10]. A small amount of CK in the blood is normal. Higher amounts can mean a health problem. Depending on the type and level of CK found, it can mean a damage or disease of the skeletal muscles, heart, or brain. The CK test can be used to help diagnose a heart attack [11]. CK levels are usually normal or only slightly elevated in all forms of hyperparathyroidism. It is rare for patients with hyperparathyroidism and renal failure to develop ischemic myopathy with elevated serum CK and myoglobinuria [12].

Creatinine Kinase activity is useful in diagnosis of myocardial infarction and cerebral accidents. As the heart muscles become damaged, the CK isoenzymes are released into the bloodstream and may be detected for 6-18 hours after onset of Acute Myocardial Infarction (AMI). The window of detection is quite short, lasting no more than 12-18 hours after heart attack has occurred, because of protein degradation mechanisms that eliminate the CK-MB isoenzyme from the blood. Due to this short time frame, the peak level of CK-MB is often missed, leaving in doubt whether a heart attack has occurred or an indication of mild heart tissue damage or angina. Exposure to Cadmium and/or $^{60}$Co gamma radiation after one day led to an increase in the CK level [13].

Lactate Dehydrogenase (LDH) test measures the level of lactate dehydrogenase (LDH), also known as lactic acid dehydrogenase, in the blood or sometimes in other body fluids. LDH is a type of protein, known as an enzyme. LDH plays an important role in making the body’s energy balance. It catalyzes the inter conversion of lactate and pyruvate with accompanying inter conversion of NAD$^+$ and NADH. It is found in almost all the body’s tissues with high activities in the heart, liver, skeletal muscle, kidney, and erythrocyte and lesser amounts are found in the lung, smooth muscle and brain. When these tissues are damaged, they release LDH into the bloodstream or other body fluids. If the LDH blood or fluid levels are high, it may mean certain tissues in the body have been damaged by disease or injury [14].

An LDH test is most often used to find out if there is tissue damage and/or monitor disorders that cause tissue damage. In conditions such as anemia, liver disease, lung disease, some types of infections and chemotherapy for certain types of cancer, LDH test may be used to show if treatment is effective [15]. Levels reach a maximum approximately 48 hours after the onset of pain and persist for about ten days. The degree of elevation is of value in assessing the extent of damage and in developing a prognosis. The level of isoenzyme LDH$_1$ compared with LDH$_2$ has been used to detect acute myocardial infarction (AMI) because of the high concentration of LDH$_4$ in cardiac muscle fibres. LDH$_1$ isoenzymes begins to leak out of dying heart muscle cells and are detectable in the serum by 12 to 24 hours following heart attack and the inactivation of LDH in the presence of toxicants and reactive chemicals [16].

Aspartate aminotransferase (AST), also called serum glutamic-oxaloacetic transaminase (SGOT), is an enzyme that is found mostly in the liver, but also in muscles. It catalyzes the reversible reaction involving transfer of amino group between aspartate and glutamate in amino acid metabolism. When the liver is damaged, AST is released into the bloodstream. An AST blood test is used in the diagnosis of liver damage or liver disease [17]. Concentration of AST is very high in heart muscles or myocardium. In AMI, serum activity of AST rises sharply within the first 12 hours, with a peak level at 24 hours or over and returns to normal within 3-5 days [18].

High concentrations of AST are found in the liver, muscles, heart, kidney, brain and red blood cells. A small amount of AST is typically present in the bloodstream. Higher-than-normal amounts in blood may be a sign of health problem associated with liver injury [19]. It can also mean damage to another organ like the heart or kidneys [20].

The exposure of wistar albino rats to refinery effluent and Bonny light crude oil significantly increased their AST levels and thus increased the risk of hepatic dysfunction [21, 22]. An increase in AST and ALT levels of humans on exposure to lead and organic solvents within the permissible exposure limit has been reported [23].

The levels of inflammatory mediators and markers of cell damage are used as prognostic and monitoring tools of disease development, activity and progression [24].

This study therefore evaluated the impact of gas flaring on the levels of CK, LDH and AST biomarkers in the residents of Eleme
communities who are constantly exposed to gas flare.

2. MATERIALS AND METHODS

2.1 Exclusion Criteria

The control population were volunteer subjects aged between 18 and 50 years, had no history of any health complications, were not on any special medication for any disease, are not smokers, had no history of alcohol abuse, work or live outside Eleme and have not travelled to Eleme or any other community anywhere in the world where gas flaring and oil and gas exploitation is taking place. Subjects with known illnesses such as cancer, diabetes mellitus and Parkinson's disease as well as tobacco smokers were excluded from this study.

2.2 Inclusion Criteria

The test population includes healthy volunteer subjects within the age range of 18 - 50 years, with no history of recent blood transfusion and no clear diagnosis of other known diseases. They reside, work and farm in the study area (Eleme communities) for over 15 years and have been to Eleme communities or any other community anywhere in the world where gas flaring and oil and gas exploration is taking place.

2.3 Study Area

This study was carried out in two Local Government Areas (LGA) of Rivers State located in the Niger Delta region of Nigeria. Eleme communities, located in Eleme LGA in Rivers East Senatorial District served as the test area while Nkpolu-Oroworukwo community located in Port Harcourt City LGA in Rivers Central Senatorial District served as control.

2.4 Study Population

A total number of 200 male and female volunteer subjects within the ages of 18 and 50 years were randomly selected and recruited for this study. They comprise of 50 males and 50 females (100 subjects) used as test and 50 males and 50 females (100 subjects) used as control. Blood samples were collected from each participant and analyses of the serum levels of CK, LDH and AST of the subjects were conducted individually.

2.5 Specimen Collection and Processing

Blood specimen was collected from a peripheral vein via antecubital venipuncture from the respective volunteer subjects. This was dispensed into serum separator tubes (SST) and then centrifuged at 1000 rpm for 10 minutes. The serum obtained was transferred into plain sample bottles and stored in a freezer at -20°C before analysis. All samples were analyzed within 14 days.

2.6 Determination of Biomarkers

Creatine kinase (CK) was determined in the serum using Creatine Kinase Activity Assay Kit (Colorimetric) (ab155901), USA.

Aspartate Aminotransferase (AST) activity was determined in the serum using the Reitman-Frankel AST/GOT Activity Assay Kit Method [18].

Lactate Dehydrogenase (LDH) was determined in the serum samples using standard “Randox” reagent kit (Randox Laboratories, London, UK).

2.7 Statistical Analysis

All data generated were analyzed using a one way Analysis of Variance (ANOVA) with the aid of Statistical Package for Social Sciences (SPSS) version 20 running on Windows PC. Data for each parameter were expressed as mean value ± standard deviation. The significant differences between the test means and control means were determined at 95% or p<0.05 confidence level.

3. RESULTS

The levels of CK, LDH and AST of Eleme residents (test samples) and

| Volunteer Subjects    | CK (u/l)     | LDH (u/l)    | AST (u/l)     |
|-----------------------|--------------|--------------|---------------|
| Male (Control) n=50   | 8.820±2.30   | 31.760±6.02  | 8.800±1.64    |
| Male (Eleme) n=50     | 14.080±4.01  | 41.360±8.85  | 15.200±4.60   |
| Female (Control) n=50 | 9.680±2.47   | 34.700±4.41  | 9.400±2.51    |
| Female (Eleme) n=50   | 24.740±5.05  | 42.620±4.60  | 10.600±4.93   |

*Values are expressed as mean ± standard deviation. Values with different superscripts show significant difference at p<0.05.*
Nkpou-Oroworukwo residents (control samples) are contained in Table 1. The results show that CK, LDH and AST levels of the exposed subjects (Eleme residents) generally increased for both male and female when compared to the non-exposed subjects (control). While CK increased by 37.36% in the male subjects and 61.20% in the female subjects, LDH increased by 23.21% in the male subjects and 18.58% in the female subjects compared to control. AST levels increased by 42.11% in male subjects and 11.32 in female subjects compared to control.

4. DISCUSSION

Table 1 showed consistent elevation of all clinical enzyme biomarkers assayed in the gas flare exposed (Eleme) resident subjects compared to the non-exposed control subjects. Creatine kinase (CK) levels for the exposed subjects were higher in the females compared to males. CK is often used in the diagnosis of myocardial infarction [25]. An increase in the level of CK is closely associated with cell damage, muscle disruption or diseases. These cellular disruptions can cause the enzyme CK to leak from cells into the bloodstream. The increase in the CK level in all subjects exposed to gas flare could therefore be partly attributed to cell damage arising from consistent exposure to the deleterious atmospheric pollutants emitted from the gas flare. The composition of gas flared depends on the source of the gas [26]. Gas flared from refineries and other process operations commonly contain a mixture of hydrocarbons, heavy metals and particulates which are known pollutants and environmental toxicants.

This comparative elevation in CK levels due to exposure to gas flare environment is consistent with a significant increase in creatine kinase level on exposure to polychlorinated biphenyl, a known environmental pollutant [27]. Consistent elevated blood CK levels in humans may cause hypertension [10], muscle aches and pains, muscle stiffness, weakness or numbness on one side of the body, paralysis, vision changes, confusion or loss of consciousness, etc [11].

An increase in the level of CK is closely associated with cell damage, muscle disruption or diseases. These cellular disruptions can cause the enzyme CK to leak from cells into blood serum. Some of the conditions in which CK is elevated include diseases of muscle and myocardial infarction [25,28].

LDH is generally released during tissue breakdown; therefore its plasma level is often measured as surrogate to monitor tissue damage. It is a common biomarker of disease conditions such as heart failure. The plasma LDH levels in this study were also seen to increase in the exposed (Eleme) subjects compared to the non-exposed (Control) subjects. This finding further strengthens the report of which posited that the presence of toxicants and reactive chemicals led to elevated LDH levels [16]. Residents of Eleme are obviously inundated with reactive environmental toxicants from gas flares and other industrial effluents whose tissue damaging effects can be detected with the elevation in plasma LDH levels.

Since serum LDH activity is a screening test for some tissue damages, this suggests that a body organ such as heart, kidney, liver or skeletal muscle could be compromised in the study participants at Eleme [14].

The AST levels of the exposed (Eleme) subjects were seen to be increased at different levels for both male and female when compared to the unexposed (control) subjects. This could imply that the environmental toxicants prevalent in the area are possibly inducing liver damage. Elevated AST level is associated with liver disease which leads to hypoxic hepatitis caused by anoxic necrosis of the centrilobular liver cell [29]. Refinery effluent has been reported to pose some adverse effect on some biochemical parameters such as an increase in the AST level, attributed to the constituents of the effluent from the refinery [21]. Increase in level of AST in the blood has also been reported when body tissue or organ such as heart or liver is diseased or damaged [30]. The elevation of plasma levels of these enzyme biomarkers in exposed Eleme residents, compared to non-exposed control, is an indication of environmental toxicity in the population exposed to gas flare.

This study is limited in that there is lack of specificity when using total levels of biomarkers like creatinine kinase and lactate dehydrogenase. The diagnosis becomes more specific when the isoenzymes are assayed. Also, other biomarkers may have been beneficial for specific organ injury such as Alanine transaminase (ALT), usually compared with AST for liver damage. And in the case of acute myocardial infarction, levels of biomarkers needed to have been taken within the stipulated
hours after occurrence of the insult to properly make diagnosis.

5. CONCLUSION

This study evaluated the effect of gas flare on some clinical enzyme biomarkers of Eleme residents. The results show that exposure to gas flare led to increased levels of creatine kinase, lactate dehydrogenase and aspartate aminotransferase enzyme biomarkers in the exposed population compared to control. This suggests an increase in the tendency of developing disease conditions such as myocardial infarction, liver dysfunction, kidney dysfunction, tissue damage and a general reduction in health status of the residents.

6. RECOMMENDATION

It is recommended that there should be increased environmental education and public awareness on the adverse effect of consistent exposure to gas flare. Government should take measures to monitor the environment and ensure strict compliance with extant environmental regulations in the oil and gas industries to ensure safe and eco-friendly disposal of gaseous waste.

It is hoped that this study would assist government and the private sector appreciate the health and environmental implications of gas flaring in communities when formulating policies or initiating actions that would impact on the citizenry.

CONSENT AND ETHICAL APPROVAL

Ethical approval for the study was obtained from the Rivers State Ministry of Health (RSMoH), Rivers State Health Research Ethics Committee (RSHREC) and Rivers State Hospital Management Board (RSHMB), Port Harcourt, Nigeria. All test and control subjects for the study duly gave their written consent and filled the requisite questionnaire.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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