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RESEARCH ARTICLE

ASSESSMENT OF SOME CARDIAC ENZYMES AMONG PETROLEUM FILLING STATION ATTENDANTS IN OWERRIIMO STATE

Ukamaka Edward[1], Chiamaka Precious Ezenwa[1] and Emmanuel Ifeanyi Obeagu[2]

1. Department of Medical Laboratory Science, Medical Laboratory Science, Imo State University, Owerri, Imo State, Nigeria.
2. Department of Medical Laboratory Science, Kampala International University, Uganda.

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Abstract

In recent years, occupational health hazards have grown as one of the major public health issues worldwide. The present study was aimed to assess the level of cardiac enzymes among petroleum filling station attendants in Owerri. The study subjects consisted of twenty individuals working as petrol station attendants in Owerri, Imo State for three years and twenty apparently healthy males and females who are not occupationally exposed. Blood samples were collected aseptically by venopuncture, using a 5ml sterile disposable syringe and needle from petrol station attendants and Controls was disposed into a labeled plain dry specimen container. All reagents were commercially purchased and the manufacturer's standard operational procedures were strictly followed. All data obtained in the study were analyzed using the independent student t-test (SPSS.20). The level of significance was set at P=0.05. The mean value of Creatine kinase-MB (CK-MB) was significantly increased in petrol attendants (2.35±0.50)ng/ml when compared to controls (1.17±0.66)ng/ml. The mean value of Lactate dehydrogenase (LDH) was significantly increased in petrol attendants (310.74±20.69)IU/L when compared to controls (282.95±13.76)IU/L. The mean value of Aspartate transaminase(AST) was significantly increased in petrol attendants (22.16±4.39)IU/L when compared to controls (11.05±2.61)IU/L. There was a non-significant positive correlation (r=0.06, p=0.808; r=0.13, p=0.574) between Creatine kinase-MB with Lactate dehydrogenase and Aspartate transaminase in petrol attendants. In conclusion it was observed that the petrol pump attendants are at greater risk of developing biochemical alterations in the cardiac enzymes with time due to the significant increase in the level of Aspartate transaminase, Creatine kinase-MB and Lactate dehydrogenase.

Corresponding Author:- **Emmanuel Ifeanyi Obeagu**

Introduction:-

Petrol (gasoline), diesel and kerosene are frequently used petroleum products for internal combustion of car engine and for operating machines. They are also used in homes as lighting and cooking fuels. These commodities contain predominantly hydrocarbons and have been reported to have significant adverse health effects (Nwanjo and Ojiako, 2007).

Human exposure to petroleum hydrocarbon can occur through ingestion of contaminated food, drinking contaminated water, contact with contaminants (dermal exposure) or inhalation of vapour and air-borne soil (Azeez et al., 2012). Inhalation of petroleum products can be accidental or intentional. Inhalant abuse, the deliberate inhalation of hydrocarbon as a form of recreational drug use has become a significant issue affecting children and adolescents (Steffe et al., 2016). Their low cost, ready availability and ease of use contribute to this problem. Inhalation is most commonly achieved by sniffing, huffing and bagging (Azeez etal., 2012). Sudden death of undiagnosed cause is becoming common in our environment; sudden death was recognised in misuse of volatile substances (Azeez et al., 2012). Pipeline breaks have resulted in exposure of livestock to crude oil or refined petroleum hydrocarbons. Ingestion of petroleum hydrocarbons have resulted in sudden death from per-acute bloat (Edwards, 2019).

Since kerosene, diesel and petrol consist mainly of hydrocarbons, chronic or frequent exposure to them may affect the liver. Humans are exposed to these products especially at the filling station while refilling of vehicle(Obeagu et al., 2021; Obeagu et al., 2018). In Nigeria, due to the unreliable public power supply many homes, offices and business rely greatly on small electric generators and liquid fuel-dependent machines for day to day activities. The fuel for operating these machines are purchased and stored in homes and offices where their fumes constitute environmental health hazards especially during refueling. The use of kerosene in homes as cooking and lighting fuels has further increased the exposure of humans to these products(Obeagu et al., 2018; Emeji et al., 2015).

In Nigeria, the current trend of locating filling station in residential areas exposes the residents of those areas to liquid fuel fumes. It has also been noticed that the owners of filling stations attach well designed and built duplexes or bungalows to their filling stations. This also exposes the occupants of these apartments to petrol fumes. Apart from these, petroleum products have been abused as therapeutics agents for the treatment of snake bites, convulsion, arthritis, gastro-intestinal disorder and a host of other conditions (Patrick et al., 2011).

Individuals who are occupationally exposed like those who work in petroleum industries or petroleum distillates are likely to be more affected than their counterparts who do not work in these industries (Okoro et al., 2016). Automobile mechanics, petrol station pump attendants and petrol tanker drivers belong to this class of people. Unfortunately, they handle these products without proper protection against their possible harmful effects (Udonwa et al., 2019).

Cardiac enzymes have been in use since the mid-20th century in evaluating patients with suspected acute myocardial infarction (MI). The biomarkers used back then are not clinically relevant today as more sensitive and specific biomarkers have replaced them. Some of the biomarkers includes AST, LDH, and creatine kinase.

Creatine is a nitrogenous organic acid that occurs naturally in vertebrates and helps to supply energy to all cells in the body, primarily muscle. Creatine kinase (CK), also known as phosphokinase is a "leakage" enzyme present in high concentration in the cytoplasm of myocytes and is the most widely used enzyme for evaluation of neuromuscular disease (Gasper et al., 2015), unlike other enzymes found in skeletal muscle (e.g. lactate dehydrogenase, aldolase and transaminase). CK has relative predominance in skeletal muscle, is not falsely elevated by hemolysis, and being unbound in cell cytoplasm is readily released in cellular injury (Gasper et al., 2015). Creatine kinase was indicated to be a constituent of the integral proteins of erythrocyte membrane or to be tightly bound to the membrane, and was contrasted to the results obtained with adenylate kinase. CK catalyses the formation of phosphocreatine (PCr) and adenosine phosphate (ADP) from creatine and Adenosine triphosphate (ATP) (Bong et al., 2012). This CK enzyme reaction is reversible such that ATP can be generated from PCr and ADP. There are three isoenzymes: CK-MM, CKBB and CKMB.Lactate dehydrogenase (LDH) is an important enzyme of the anaerobic metabolic pathway. It belongs to the class of oxidoreductases, with an enzyme commission number EC 1.1.1. The function of the enzyme is to catalyze the reversible conversion of lactate to pyruvate with the reduction of NAD+ to NADH and vice versa (Bong et al., 2012). The enzyme is present in a variety of organisms that include plants and animals. Aspartate amino transferase is also known as glutamate oxaloacetate transferase (SGOT). It is a tissue enzymes that catalyses the exchange of amino and keto group between alpha amino acid and alpha keto acid. Aspartate amino transferase is found in a diversity of tissues including liver, heart, muscles, kidney and brain. It is released when any of these tissues is damaged or injured such as during, cardiac injury or myocardial infarction, muscle injury (Nwanjo, 2006).

Materials And Methods:-

Study Area

The study was conducted in Owerri, Imo state.

Advocacy, Mobilization and Pre-survey Contacts

A letter of introduction was obtained from the Head of Department Medical Laboratory Science, Imo State University, Owerri. Approval was obtained from the managers of the filling stations. Written consent of willingness to participate in the study and questionaires were obtained from all participants included in the study. All participants were petrol station attendants who have worked for a particular period of time.

Study Population

Forty subjects who were between the ages of eighteen to forty years were recruited for this study. Twenty were petroleum station attendants who were working and exposed for the past three years. Twenty were apparently healthy individuals who were not occupationally exposed and served as control subjects.

Selection Criteria

a. Inclusion criteria

a) Subjects who signed informed consent to participate in the study.
b) Subjects who were aged 18- 50 years.
c) Subjects who have worked as petroleum attendant for 3 years and above.
d) Subjects with no history of diabetes mellitus, liver disease, malignancy, high blood pressure, renal disorder, HIV and other endocrine disorder.

b. Exclusion criteria

a) Subjects who did not sign informed consent to participate in the study.
b) Subjects who were aged below 18 years and above 50 years.
c) Subjects who had worked for below 3 years.
d) Subjects with medical history of liver diseases, Kidney disease, diabetes, hypertension and other chronic illnesses were excluded from this study.

Sample Collection

Blood samples were collected aseptically by venopuncture, using a 5ml sterile disposable syringe and needle from petrol station attendant and Controls was disposed into a labeled plain dry specimen container. The samples were allowed to clot and centrifuged at 3,000pm for 5 minutes to obtain the sera. The sera were extracted using a pipette and put into another specimen container, and stored at -20^{o}C prior to use.

Laboratory Procedures

All reagents were commercially purchased and the manufacturer's standard operational procedures were strictly followed.

Determination of Creatine Kinase activity (Stolle, 1976) as modified byRandox limited, United Kingdom. Catalog number: BZL-EV4418.

Procedure

The Operation Procedures for programming controls/patients and loading sectors/racks in the Beckman Coulter Synchron UniCel DxC 600/800 was followed. Synchron Systems perform all calculations internally to produce the final reported result.

Determination of Serum Lactate Dehydrogenase (Assay Kitas modified by Randox limited, United Kingdom.Catalog number: KA1653)

Procedure

50 µL samples were transferred into 1-cm cuvettes. 950 µL Working Reagent was pipetted into samples and Mixed briefly. Sample was read at OD565nm shortly after the mixing (ODSO), and again after 25 min (ODS25). OD565nm for 1 mL water (ODH2O) and Calibrator (ODCAL) was read.

A. Serum Aspartate Amino Transferase (AST) (Sridevi, 2020) as modified by Randox laboratory limited, United Kingdom. Catalog number: EV4418.

Procedure

Three test tubes were labeled as test, standard and blank, dinitophynylhydrazine reagent was added to the three test tube. 0.1 ml of serum was added to the test tube labelled test, 0.1ml of standard solution was added to the test tube labelled standard, and while 0.1ml of distilled water was added to the test tube labelled blank and mixed well, 0.5ml of dinitrophenylhydrazine was also added to the tubes. The test was left for 20 minutes at room temperature for colour development.

Five milliliters of 0.4N sodium hydroxide was added, mixed well and left at room temperature for 5 minutes. The absorbance was read at 505nm, and the concentration calculated.

Statistical Analysis

Results were presented as mean ± standard deviation (SD). All data obtained in the study were analyzed using the independent student t-test (SPSS, Version 20). The level of significance was set at 95% confidence limit, and results are presented in tables.

Results:-

Table 1:- Mean Value of Creatine Kinase-MB, Lactate Dehydrogenase and Aspartate Transaminase in Petrol Attendant of the Study Population.

Parameter	Test	Control	t-value	p-value
CK-MB (ng/ml)	2.35±0.50	1.17±0.66	6.28	0.0001
LDH (IU/L)	310.74±20.69	282.95±13.76	4.96	0.0001
AST (IU/L)	22.16±4.39	11.05±2.61	9.67	0.0001

*KEY;
CK-MB: Creatine Kinase-MB
LDH: Lactate Dehydrogenase
AST: Aspartate Transaminase

Table 1 shows the mean values of Creatine Kinase, Aspartate Transaminase and Lactate Dehydrogenase in the study population.

The result showed that mean value of CK-MB was significantly higher (P=0.0001) in petrol attendants (2.35±0.50)ng/ml when compared with the value of control subjects (1.17±0.66)ng/ml.

The mean value of LDH was significantly higher in petrol attendants (310.74±20.69)IU/L when compared with the mean value (0.0001) controlsubjects (282.95±13.76)IU/L.

The mean value of AST was significantly higher in petrol attendants (22.16±4.39)IU/L when compared with the mean value of the controlsubjects (11.05±2.61)IU/L.

Table 2:- Correlation of Creatine Kinase-MB with Lactate Dehydrogenase and Aspartate Transaminase in Petrol Attendant.

Variable	N	R	p-value
LDH	20	0.06	0.808
AST	20	0.13	0.574

*KEY;
LDH: Lactate Dehydrogenase
AST: Aspartate Transaminase

Table 2 shows the Correlation of Creatine Kinase-MB with Lactate Dehydrogenase and Aspartate Transaminase in Petrol Attendant

There was a non-significant positive correlation (r=0.06, p=0.808; r=0.13, p=0.574) between Creatine Kinase-MB with Lactate Dehydrogenase and Aspartate Transaminase in petrol attendant.

Discussion:-

In recent years, occupational health hazards have grown as one of the major public health issues worldwide. In this context, petrol pump workers were studied who by virtue of their occupation is continuously exposed to the various chemical substances and volatile organic compounds liberated in the form of gasoline vapors in their breathing zone (Obeagu, 2018). In this study, the cardiac markers were evaluated since the heart is considered as a vital organ in the human body.

The present study reveals that the mean value of CK-MB was significantly increased ($p<0.05$) in petrol attendant when compared to controls. The significant increase in the mean value of CK-MB in petrol attendants clearly shows that petrol exposure may have resulted to myocardial injury. The exact mechanism is uncertain, but it is thought to be due to increased heart muscle catabolism or membrane permeability. The result of this study is consistent with the report by Azeez et al. (2015). Bartoli et al. (2009) revealed that inhalation of petroleum hydrocarbons leads to increase baroreflex sensitivity. Baroreceptors are sensors in the blood vessels which function in the short term as a part of the baroreflex negative feedback system (Levecchio and Fulton, 2011). These mechanoreceptors reset in the maintenance of a normal arterial pressure. Therefore, the pressor effects of the petrol fumes or particles observed in petrol attendants could explain the increase in CK-MB.

In the current study it was observed that the mean value of LDH was significantly increased in petrol attendant when compared to controls. The reportedly increased LDH as observed in this study clearly shows that petrol has cardiotoxic effect which might have resulted in myocardial injury.

The mean value of AST was significantly increased ($p<0.05$) in petrol attendant when compared to controls. The elevated levels of AST clearly suggest that the petrol filling attendants are at greater risk of developing myocardial injury with time.

Conclusion:-

In conclusion it was observed that the petrol pump attendants are at greater risk of developing biochemical alterations in the cardiac enzymes with time due to the significant increase in the level of AST, CK-MB and LDH.

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