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Serofrequency of Chikungunya Virus Among Blood Donors in Singa City Sinnar State Sudan

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Abstract

Background and Objectives
Chikungunya (CHIK) is a re-emerging disease causing a large negative impact on global health and economics. Clinical manifestations of CHIK are non-specific and difficult to differentiate from dengue hemorrhagic fever or other viral exanthema. A rapid, simple and reliable diagnostic assay is necessary for CHIK outbreak control especially in countries with insufficient access to well-equipped laboratories. This study aimed to detect CHIKV among blood donors in Singa city.
Material and Method
This is descriptive, cross-sectional study, 90 healthy blood donors who attended Singa Hospital blood bank were included in this study, their age ranged from 20 to 50 years with mean 35during period from March toNovember 2017, enzyme linked immunoassay (ELISA) method was used for detection of immunoglobulin antibodies G and M for the CHIK virus from serum samples. Ethical consideration was approved by the research ethical committee of Al Neelain University. Data was collected using direct interviewing questionnaire and analyzed by SPSS.
Result
out of the total 90 blood donors, serofrequency of CHIK was47(52.2%) and5 (5.5%), for IgG and IgM respectively and 12(13.3%) positive for both. Most of seropositivety (46(51.1%)) was observed among 21-30 year
Conclusion
seropositivety rate of CHIKV is fairly common among the blood donors in Singa City (Sudan)

Keywords:-Chikungunya virus, ELISA, Blood Donors ,Singa.

I. INTRODUCTION

Chikungunya virus (CHIKV) is an arbovirus spread predominantly by Aedesaegypti and Ae. Albopictus mosquitoes [(1-2)].Mosquitoes are a significant public health concern due to their ability to transmit a variety of emerging and reemerging arboviruses[(3-4)]..CHIKV belongs to the alphavirus genus within the Togaviridae family. It is a member of the antigenic Semliki Forest Complex[(5-6)]. The alphavirus genome consists of a single-stranded positive-sensed 11.8 kB RNA molecule packaged by the C protein to form the nucleocapsid. This nucleocapsid is surrounded by a host-cell derived lipid bilayer with two inserted transmembrane glycoproteins, E1 and E2[(6)]. The composition of the host-cell derived lipid bilayer strongly resembles the plasma membrane of the infected host cell. For mammalian-derived CHIKV virions, the membrane consists of cholesterol and phospholipids in a ratio of approximately 1:1 [(7-8-9-10)].The virus was first described in 1952 during a febrile illness outbreak in Makonde, a province in southern Tanzania[(11)]. In spite CHIKV has caused millions of human infections in Africa, the Indian Ocean islands, Asia, Europe, and the Americas, it is a neglected disease, because it circulates within these subtropical and tropical regions, has the potential to affect more than 1 billion people, and many at-risk people live in poverty stricken regions[(12-13)], and methods of prevention and treatment are still lacking[(14)]. Mosquito saliva contains a complex repertoire of bioactive factors that are secreted into blood feeding site, the skin. Infected mosquitoes transmit pathogens to the host during feeding via saliva. The bioactive factors in mosquito saliva are responsible for modulating host hemostasis, immune defenses and pain/itch responses, and have been implicated to enhance pathogen infection and establishment in the host[(15)] such as CHIKV infection, which starts when a CHIKV-infected Aemosquito is feeding on a human host[(16)]. During feeding, CHIKV particles are thought to be released within the dermis[(17)], the virus reaches the blood and disseminates to other parts of the body[(18)], include joints, muscle, skin, and less frequently, the liver, kidneys, eye and the central nervous system[(19-20-21)].Symptoms of CHIKV infection start abruptly with fever typically last from several days up to 2 weeks and can be biphasic in nature[(22-23)]. Shortly after the onset of fever, the majority of infected persons develop severe, [(24)].

Blood transfusion saves hundreds of patients' lives every day, but adequate blood supply is needed to continue helping those people who are in need of blood transfusion. Encouraging and

promoting voluntary blood donation is a goal of every country in the world.

Concealing of medical history by captive, paid or professional blood donors, who widely exist in developing countries, also pose a great threat to safe blood supply. There is a long list of viruses, parasites and bacteria, which can be transmitted through blood transfusions. Among them, important transfusion-transmitted viruses are human immunodeficiency virus (HIV-I/II), hepatitis B virus (HBV), hepatitis C virus (HCV), syphilis infection by spirochytes and transfusion associated malaria infection [(25)]. CHIKV transmission through blood transfusion have been reported one case in France, an infection of nurse got infected with CHIKV when had direct contact with blood of patient infected with CHIKV [(26)].

Out of the 270 blood samples investigated for anti-CHIKV IgG, 122 (45%) samples were found positive, while 148 (65%) were found negative in Khartoum State by khirreetal 2017[(27)]

In Saint Martin survey done by Noellie Gay etal 2013 [(28)]to detect seroprevalence of CHIKV they found positive for anti-CHIKVantibodies (19 for IgM, 36 for IgG, and 13 for both).

In Puerto Rico, peaks of CHIKV infection were observed in 2.1% of blood donors. In a study conducted in Sudan, the seroprevalence of CHIKV infection was estimated in 379 patients with fever attending the outpatient clinics of three hospitals in eastern and central Sudan. The seroprevalence rate was found as 1.8%, indicating that CHIKV infections are rare in these parts of Sudan[(29)] This study aimed to detect serofrequency of CHIKV among blood donors in Singa.

II. MATERIAL AND METHOD

This was descriptive- cross sectional study which had been conducted in Sinnar state, Singa city during period from March[20] to November[20] 201, ninety well healthy blood donor males were enrolled, Data was collectedby using direct interviewing questionnaire; ethical clearancewas obtained from research ethical committee of faculty ofgraduate studies and ministry of health Sinnar state, written consent also was obtained from blood donors.

A. Experimental work

a). Samples Collection

Blood samples were collected from 90 blood donors, underdirect medical supervision by medial vein puncture using 5 mlsyringe into plain tube to obtain serum by centrifugation at5000 rpm for 10 min. serums was kept in -20°C till serologicalstudy was performed.Specimens were processed by Enzyme linked immune sorbentassay (ELISA) (3rd generation ELISA) (Euroimmune-Germany) fordetection IgM and IgG Enzyme linked immune sorbent assay for detection anti CHIKVIgM and IgG (the same method for both). All reagents and samples were allowed to reach room temperature for 15minutes before use. Washing buffer was prepared 1:40 from buffer concentrate with distilled water.100µl of sample diluents was added into appropriate wells except the blank well and negative well.10µl from each sample was added to the appropriate wells and mixed by pipette repeatedly until liquids turn blue. 50µl from negative and positive control was dispense and added to the negative and positive wells separately without dispensing liquid into the blank control well.Microtiter wells was flicked for 30 seconds and mixed well, then plate was covered and incubated for 20 minutes at 37°C. Plate was taken out and wash buffer was added to each well(Washing 1) and aspirated off after 20 seconds. This step was repeated for 5 times until each well become dry.50µl of HRP-Conjugate Reagent was added in to each well except the blank, the plate was mixed well and covered with the plate cover and incubated for 20 min at 37°C.The plate cover was removed and discarded. The liquid was aspirated and each well was rinsed in wash buffer (Washing2). This step was repeated for 5 times until each well

+becomedry.50µl of substrate A and 30µl substrate B solution was added into each well including the Blank and mixed by tapping the plate gently. The plate was incubated at 37°C for 10 min.50 µl Stop solution was added into each well and mixed gently.Measuring the absorbance: The plate reader was calibrated with blank well and the absorbance was read at 450nm. The results were calculated by relating each sample optical density(OD) value to the Cut off value of plate.

III. CALCULATION OF RESULTS

The extinction value of the calibrator defines the upper limit of the reference range of non-infected persons (cut-off) recommended by EUROIMMUN. Values above the indicated cut-off are to be considered as positive, those below as negative. Semi quantitative: Results can be evaluated semi quantitatively by calculating a ratio of the extinction value of the control or patient sample over the extinction value of calibrator. Use the following formula to calculate the ratio:

$$\frac{\text{Extinction of the control or patient sample}}{\text{Extinction of calibrator}} = \text{Ratio}$$

EUROIMMUN recommends interpreting results as follows:

Ratio <0.8: negative

Ratio >1.1: positive

A. Interpretation of Results

Negative results: samples giving absorbance less than Cut-off value are negative for this assay. Positive result: sample giving absorbance equal to or greater than Cut-off considered initially reactive. Data was analyzed by SPSS (Statistical Package of Social Science) software program version 21.

B. Result

A total of 90 blood donors who attended Singa Hospital blood bank, their age ranged from 20 to 50with mean 35, the most of them belonged 21-30 years were enrolled in this study table2. The overall result revealed that 05(5.6%),47(52.2%)were positive for CHIKV IgM, IgG respectively while 12 (13.3%)were positive for both and 26(28.9) were negative table 1. Regarding age the highest serofrequency of CHIKV observed among 21-30 age group.

CHIKV	IgM	IgG	IgG/IgM
Positive	05 (5.6%)	47 (52.2%)	12 (13.3%)
Negative	85 (94.4%)	43 (47.8%)	78 (86.7%)
Total	90	90	90

Table1: Frequency of the Positive and Negative Result of Chikvigm and Iggamong Blood Donors.

Age group in years	Number (%)	IgG+ -	IgM+	IgG+/IgM+	IgG-/IgM-	total
21-30	46(51.1%)	19	04	10	13	46
31-40	37(41.1%)	23	01	02	11	37
41-50	07(7.8%)	05	0	0	02	7
Total	90(100%)	47	05	12	26	90

Table 2: Frequency of CHIKV among Blood Donors According To Age Ranges.

IV. DISCUSSION

Chikungunya virus (CHIKV) is an arthropod-borne virus transmitted by Aedesmosquitoes. It may be responsible for acute and chronic articular manifestations. It became a new, unexpected, public health problem in many tropical African and Asian countries within the past decade(1,2).

CHIKV infection was inevitably misdiagnosed as malaria. CHIKV-infected patients may wrongly receive antibacterial or anti-malarial therapy. Such a therapy leads to wastage of health resources and potentially promotes antimicrobial resistance. Unfortunately clinical signs, symptoms, and laboratory findings are unhelpful in distinguishing patients with CHIKV infection from other febrile inpatients[(30)].

In this study 47 (52.2%) anti-CHIKV IgG cases were detected among the blood donors investigated and 05(5.5) positive anti-CHIKV IgMand 12(13.3%) for both This frequency rate was similar to previous studies conducted in Khartoum state-Sudan2017 by khiireetal ([(27)]targeting the blood donors to percentile the CHIKV, it detect positive CHIKV among 45% of involved personal but it revealed the existence of IgG only. Our resultshow frequency rate slightly highly thafrequency reported byNoellie Gay etal 2013 In Saint Martin they found positive for anti-CHIKV antibodies (9.4% for IgM, 17.7 for IgG, and 6.4% for both[(28)] .

And also result is high than result reported by Puerto Rico,in Eastern and centeral of Sudan peaks of CHIKV infection were observed in 2.1% of blood donors. [(29)].

V. CONCLUSION

CHIKV infection was frequent among apparently healthy blood donors in Singa city, Sinnar State-Sudan, so CHIKV screening test is recommended to be set as routine test in blood banking protocol, that would save blood receiver from coming un justified symptoms may present later if given blood was contained CHIK virus.

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