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Prevalence of Dengue and Chikungunya Virus Co-infection in Tertiary Care Hospital in Patiala, Punjab

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Abstract

Background: Arboviruses of public health importance, such as dengue and chikungunya, have caused several epidemics in many tropical and subtropical countries. The clinical manifestations, vector, and pathogenic processes of the dengue and chikungunya viruses are identical.

Aims and objectives: The present study is to study the prevalence of Dengue and Chikungunya co-infection in tertiary care hospital.

Material and Methods: Serum samples were received from RH Patiala and processed in VRDL, Government Medical College, Patiala, between January 2022 to November 2022. The samples were processed depending upon the H/O duration of the fever in days. For fever less than 4 days Dengue NS1Ag detection kits was used and for fever >4 days DENV IgM antibody capture (MAC) ELISA and CHIK IgM ELISA kits were used. The testing was done by using kit protocol. The serotyping was done by using Real time RT PCR kit received from NIV Pune as per kit protocol.

Results: Out of total 2188 samples from suspected patients for dengue infection, 546 samples were positive for DENV while CHIK IgM antibodies were positive in 47 patients out of the total suspected 102 cases and co infection was seen in 2 cases. The results of 43 serotyping revealed that DENV 2 is more common followed by DENV 1 and DENV 3, while no case of DENV 4 serotype was detected. Out of 546 Dengue virus positive cases 321 were male patients and 225 were female patients, whereas, CHIKV positive cases was reported in 21 male and 26 female patients. However, co-infection of DENV and CHIKV was reported in one male and one female patient.

Conclusion: Dengue and Chikungunya coinfection as a global problem worthy of consideration. It is therefore pertinent that both infections be assessed during diagnosis, mosquito vector control practices be implemented, and vaccine development strides be supported globally.

Introduction

Dengue virus (DENV) and Chikungunya virus (CHIKV) are important arboviruses which spread by the bites of female mosquitoes, primarily Aedes aegypti and to a lesser extent Ae. albopictus. Punjab is seeing a resurgence of both illnesses, which pose a serious threat of widespread outbreaks [1]. The rise or re-emergence of vector-borne diseases has been driven by unplanned and rapid urbanization along with alterations in climatic and environmental conditions, greater international travel and trade, and other societal concerns. There are numerous dengue and chikungunya virus (CHIKV) carriers in the tropics, with local variations in risk due to local climate, socioeconomic, and environmental factors [2].

The dengue virus, which is a member of the Flaviviridae family, has four distinct but related serotypes (DENV-1, DENV-2, DENV-3 and DENV-4). Following recovery from infection, it is believed that immunity against that serotype will last a lifetime. Only temporary and limited cross-immunity to the other serotypes exists upon recovery. [3]. The almost identical clinical signs of the two diseases are caused by the same primary pathogenic virus transmission pathways. Several disorders are brought on by dengue. When someone is infected, they may experience anything from minor flu-like symptoms to major sickness. Even though it's less often, dengue can be very serious for certain people, which can lead to a variety of issues include severe bleeding, organ damage, and/or plasma leakage. When severe dengue is improperly treated, a larger risk of death exists. Severe dengue was first discovered during

dengue epidemics in the Philippines and Thailand in the 1950s. Today, severe dengue is a significant contributor to paediatric hospitalisation and mortality, and adults in most Asian and Latin American nations [4].

The beginning of CHIKV sickness in symptomatic patients normally occurs 4–8 days (within a range of 2–12 days) following the bite of an infected mosquito. A quick onset of fever that is often accompanied by acute joint pain sets it apart from other illnesses. The joint pains are frequently excruciating and usually subside after a few days, although it can last for weeks, months, or even years. Joint swelling, muscle distress, headaches, nausea, exhaustion, and rashes are other typical symptoms. Because these symptoms co-exist with those of other diseases, cases may be incorrectly identified, such as those caused by the dengue and Zika viruses. When there is no severe joint pain, an infection may go undetected in an infected person because their symptoms are typically mild [5].

Both Dengue and Chikungunya present substantial health risks with a high fatality rate in the current situation. Patients with symptoms are often only tested for dengue and infrequently for chikungunya virus infection. Because of this, cases of the Chikungunya virus infection in dengue-endemic areas go misdiagnosed, and the real scope of the epidemic is unclear. These infections might not be correctly identified, which would lead to the improper therapy, which would worsen the condition. Therefore, a precise and early identification of co-infection will facilitate effective management and therapy [6,7]. The current study's objective is to determine the prevalence of co-infection with Dengue and Chikungunya in tertiary care facilities.

Materials and Methods:

Serum samples were received from Rajindra Hospital Patiala and were processed in the Virology Research and Diagnostic Laboratory, Government Medical College, Patiala, between January 2022 to November 2022. From suspected patients having fever, serum samples were taken. The samples were processed depending upon the H/O duration of the fever in days. For fever less than 4 days Dengue NS1Ag detection kits was used and for fever >4 days DENV IgM antibody capture (MAC) ELISA and CHIK IgM ELISA kits were used. The testing was done by using kit protocol. All NS1 positive samples were used for DENV serotyping using multiplex real-time PCR assay. The serotyping was done by using Real time RT PCR kit received from NIV Pune. The RT-PCR assay was performed for the detection of four serotype of DENV by the Probe1-Step RT-qPCR System following instructions from the manufacturer. The reaction was carried out using Biorad Real Time PCR system. The processing of all samples included negative and positive controls as well as internal controls.

Results

In the current study, out of total 2188 samples from suspected patients for dengue infection, 546 samples were positive for DENV while CHIK IgM antibodies were positive in 47 patients out of the total suspected 102 cases. In addition to this, 102 samples were tested for both viruses, out of which 2 sera were positive for co-infection of dengue and CHIKV as shown in Table 1. The results of 43 serotyping revealed that DENV 2 is more common followed by DENV 1 and DENV 3, while no case of DENV 4 serotype was detected. Similarly, in gender wise distribution of DENV revealed positivity in 321 male patients and 225 female patients, whereas, positive cases of CHIKV was reported in 21 male and 26 female patients. However, co-infection of DENV and CHIKV was reported in one male and one female patient (Table-2, Figure-1).

Table 1: Samples tested for DENV and CHIKV showing monoinfection and co-infections.

	Total sample tested	Total sample positive	Percentage positive
DENV	2188	546	24%
CHIKV	102	47	46%
COINFECTION	102	2	2%

	Male	**Female**	**Total sample positive**
DENV	321	225	546
CHIKV	21	26	47
Coinfection	1	1	2

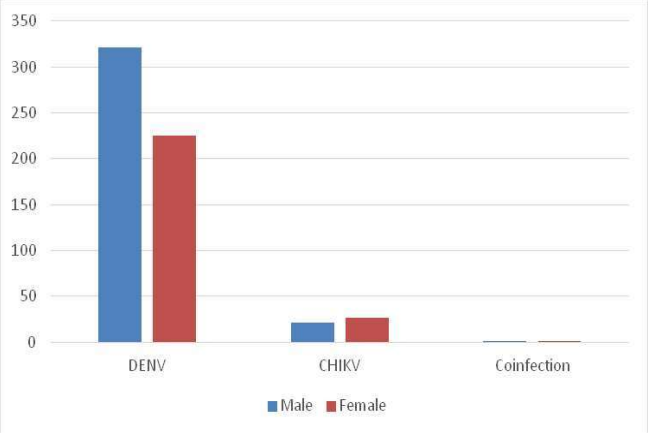


Table 2: Gender wise distribution of positive samples of Dengue and Chikungunya.

Figure 1 Gender wise distribution of DENV and CHIKV positive cases

Discussion:

In this study, we characterized DENV and CHIKV infections and also identified patients having co-infection of these viruses in the state. The frequency of monoinfected with DENV and CHIKV was high and co-infections between them were observed only in two patients.

Numerous research have recently revealed rising rates of Chikungunya co-infections. Since the clinical signs and symptoms of both diseases (chikungunya and dengue) are similar, it can be challenging to distinguish between them clinically. Both infections spread by the bite of infected Aedes species mosquitoes [8,9].

In India, DENV's endemic territories were overlapped by CHIKV's re-emergence in late 2005. Consequently, the incidence of concurrent infection with both viruses has significantly increased in the nation [10]. The first Chikungunya outbreak in India was documented in Calcutta in 1963. There is some indication that the virus may have come from South East Asian nations, the exact source of the pandemic is still unknown [11].Although there is debate, more research tends to support the idea that co-infections did not worsen clinical outcomes [8]. Trends of co-infection of DENV and CHIKV have been shown in Table 2.

Total sample	Positive DENV	Positive CHIK	State	Reference	Co infection	Year
5134	884,	591	Hyderabad	Pathan MM et al 2019 [12]	207	2019
3160	2178	127/373	Amritsar,	Kaur M et al 2018[13]	27/283	2018
326	54%	33%	West Bengal	Mukherjee S et al 2017 [10]	23%	2017
5198	4154	1816	Odisha	Subhadra S et al 2021 [14]		2016-2019
130	48/120	26/65	New Delhi	Hisamuddin M et al 2016 [15]	5	2016
648	141	22	Odisha	SaswatT et al 2019 [16]	4	2015-2016
331	49	55	Tamil Nadu	Venkatasubramani K et al 2015[17]		2012

Table: 2 Trends of co-infection of DENV and CHIKV.

Since then, more dengue outbreak reports have come from various regions of India. The Asian genotype responsible for the concurrent Thai outbreaks was found to be closely linked to the Indian CHIKV (same within-clade cluster), according to a recent phylogenetic analysis based on the NS4 gene peculiar to the Alphavirus genus. A novel strain of chikungunya sprang out in Lamu and later Mombasa on the Kenyan coast in 2004.

This recently emerged strain from the Central/East African lineage reached a relatively high attack rate of 75% in the immunologically undeveloped indigenous human populations in Kenya, where it is typically maintained in a sylvatic cycle. It subsequently spread to islands in the Indian Ocean, India, and South-East Asia through international travel and the transportation of goods [18]. As a result, co-infections with chikungunya and dengue were discovered in 2006 in Malaysia, Madagascar, Sri Lanka, and India.

Numerous parts of the world have records of DENV and CHIKV coexistence. A study observed 321 persons were having DENV2 and CHIKV infections in a febrile sickness during outbreak that hit Gabon in 2007 with 20,000 probable cases; 8 of these patients were found to have co-infections between these viruses [19]. Since 2014, reports of the co-infection and co-circulation of various arboviruses with significant public health implications in Brazil have been made. In Minas Gerais, DENV, CHIKV, and ZIKV cases were recently examined, and a total of 11 individuals had co-infections, five of which had DENV and CHIKV[20].

The reporting of cases of co-infection with Dengue and Chikungunya showed how quickly these viruses had spread across continents and countries. Nimmannitya et al in year 1969 found the first Dengue-Chikungunya co-infection cases in Thailand in 1962, 1963, and 1964. They found four, three and 12 co-infected cases out of 150, 144 and 334 infected patients respectively [21].

A cross-sectional study conducted in Odisha, India, in 2013, we looked into 204 suspected Dengue cases. Forty samples tested positive for DENV solely, 28 for CHIKV exclusively, and intriguingly, 28 patients tested positive for both DENV and CHIKV [22]. This highlights the requirement for a routine CHIKV and DENV diagnosis in febrile patients.

In the study conducted by Singh et al 2018, author reported that all of the instances involved patients with Dengue Fever who most frequently experienced headaches, followed by nausea/vomiting and widespread weakness; whereas those with chikungunya fever were mostly had body pains and joint discomfort [8].

In a study conducted by Savargaonkar D et al 2018, 1536 (27.7%) of the 5536 patients with fever who were screened between 2012 and 2015 had confirmed dengue. The month of September and the month of October saw the highest dengue positivity rates. One of the 60 samples that were examined included all four serotypes of dengue, making up 10 (16.7%) of the samples. Additionally, Dengue, Malaria, and Chikungunya co-infection were also noted in their study [23].

Sagar R et al in 2021 reported a study in which sample included clinically suspected febrile individuals (>7 days) whose sera were obtained in Delhi between 2017 and 2018 (n=87) and between 2008 and 2010 (n=623). During 2017–2018, DENV/CHIKV co-infection was found in 10.34% (n=9/87) cases, which is an interesting finding given that DENV/CHIKV prevalence rates were 41.38% (n=87) and 16.1% (n=87), respectively [24].Another study showed that 648 suspected DENV patients (in 2015 and 2016), 141 (21.7%) tested positive for DENV (serotypes 1-3), 22 (3.4%) tested positive for CHIKV (ECSA), and 4 (2.8%) tested positive for both DENV and CHIKV [16]. Similarly, a study from Colombia revealed that eighty-two individuals tested positive for one or more viruses, including ZIKV, CHIKV, and DENV, with 33 (21.02%, 47, and 29.94% respectively). Patients with ZIKV infections had an average age that was statistically greater than that of those with DENV or CHIKV infections (29.72 years) (21.09 years). There was evidence of the co-circulation and co-infection of these three viruses. Co-infection rates for DENV/CHIKV, DENV/ZIKV, and CHIKV/ZIKV were 7.64%, 6.37%, and 5.10%, respectively, with attack rates of 14.90, 12.42, and 9.93 cases per 100,000 people[25].

The goal of the study was to identify and describe the DENV and CHIKV strains that were in circulation in the DENV endemic area of New Delhi in 2016. By using RT-PCR, CHIKV and DENV were found in the blood samples (n = 130) taken from suspected patients. A total of 26 out of 65 samples (40%) contained CHIKV. DENV was discovered in 48 of 120 samples, or 40% of the total. In five (9%) of the samples, there was co-infection with both viruses [15].

Even though each of these diseases is severe and has health risks, the co-circulation of the viruses has made it possible for concurrent infection to occur in the general population of humans. Studies carried out over the world have recorded numerous instances of this [26,27,28]. More research is necessary to clarify whether or not co-infection by both viruses increases illness severity.

When DENV and CHKV viruses coexist in an area, they might spread simultaneously, and because there isn't concurrent testing, co-infections might go undiagnosed. Although socioeconomic trends like population growth and urbanisation are suspected, the reasons for the sudden rise in dengue and Chikungunya incidence are not well understood.

Conclusion:

Dengue patients' initial serotyping can be used to track the epidemiological trend. Since the vector for both infections is the same, a key public health concern that justifies the installation of strong control measures is the rise in the number of Dengue and Chikungunya infections and their co-circulation.

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