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TO STUDY CLINICAL PROFILE OF SEROLOGICALLY PROVEN DENGUE FEVER

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ABSTRACT

Dengue Fever, known commonly as Break bone fever is the most common Arboviral mosquito borne disease in the world. Many countries especially the countries of the Indian subcontinent have suffered at the hands of this disease. Epidemiology of Dengue Fever in Indian subcontinent is very complex. It has changed over the last few years with regard to the strains, affected regions and disease severity. Dengue has a varied and wide spectrum of clinical presentations, often with unpredictable clinical evolution and outcome. In the Department of Medicine with collaborationof other departments such as Departments of Biochemistry, Microbiology, Pathology, Radiology, Hospital based cross sectional Observational study and approval of the Institutional Ethical Committee with observation of inclusion and exclusion criteria's, complete history, signs,symptoms and laboratory data were taken in to account as per the Proforma in the sample size of 251 cases for a period of one year. The present study is an observational study where we studied the clinical profile of 251 serologically proven dengue patients admitted in the Department of Medicine and Pathology Kasturba Medical College Mangaluru .The study had revealed much observation that concurred with traditional teaching about dengue; there were other findings that were peculiar to this epidemic.

Key words: Dengue Fever, Arboviral Disease, Epidemiology, Clinical Profile, Observational Study

INTRODUCTION

Traditional teaching of saddle Book fever in dengue was not observed during this study.

- Break bone fever (ie) bony pain characteristic of Dengue in surprisingly very low (3-20%)
- Atypical presentation of seizures with GTCS in 1-2% of pts.
- Atypical presentation of myocarditis conformed with
- Peticheae on the palate—unusual site—observerd in some cases.
- WHO recommends IV fluids for 48 hours,but we had to give IVF for an average period of 72 hours according to hematocrit, which was found to be more effective in managing cases in this epidemic.

In this study the clinical profile of serologically proven Dengue fever was studied from April 2012 – March 2013 particularly during the epidemic that stormed Puducherry district. With this clinical profile it is easy to recognize and understand the clinical problem. The application of clinical spectrum of WHO classification system is not as very simple and straightforward as it seems because clinical features may overlap among different categories. The WHO classification system of dengue does not include unusual manifestation like encephalopathy, seizures, myocarditis, etc. which might be life-threatening. Although these manifestations are rare, clinicians should always have a high index of suspicion and knowledge of these atypical manifestations, particularly in view of the increasing burden of dengue in recent years. Dengue Fever, known commonly as Break bone fever is the most common Arboviral mosquito borne disease in the world.

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Dengue is one of the most common mosquito-borne arbo viral diseases. The word DENGUE is derived from the Swahili phrase KA-DINGA PEPO which means "Cramps like seizure". Later Benjamin Rush called it "BREAK BONE FEVER" as it causes severe arthralgia and myalgia [1].

Dengue was first reported in India in 1946 [2]. Then for the next 18 years, there was no further epidemic. In 1963 to 1964, an outbreak was reported in the Eastern coast of our country, which then spread and reached Delhi by 1967 [3].

The Kanpur epidemic [4] in 1967 was due to Dv-4 and in 1969[5] epidemic, both DV-2 and 4 were isolated. the Vellore epidemic [6,7] of 1966 was reported to be due to DV-3 strain DV-3 was also reported from Calcutta[13] in 1983, Gwalior in 2003, 2004 [8,9] and at Tirupur, Tamilnadu in 2010 [10].

The out breaks in Delhi during 1996 reported DV-2 and in 2003 it was due to DV-2 and DV-3 strains. At Delhi till 2003 [11] DV-2 was the predominant serotype reported, which was then replaced by DV-1 strain for about a period of three years from 2007 to 2009 [12]. Concurrent infection by Chikungunya and DV-2 was reported from Vellore [13] and Delhi [14].

The emergence of DV-4 which was related to increased severity was reported in Andra Pradesh in 2007 [15] and in Pune 2009 to 2010 [16]. Phylogenetic analysis revealed that the strains isolated from South India were closely related to Gwalior and Delhi isolates.

Agent:

Dengue virus is a single-stranded RNA virus. it has four serotypes Dengue 1,2,3,4. They belong to the Flavivirus genus of family "Flaviviride" [18]

The spherical shaped virus is of 50nm diameter. It has multiple copies of 3 structural proteins, a bilayered membrane derived from host and a single copy of single stranded RNA.

Vector:

This dengue virus is spread by the bite of infected Aedes mosquito most commonly Ae. aegypti [19,20]. This mosquito is mostly seen in tropical and sub tropical countries. Since it cannot with stand lower temperatures, it is uncommon above 1000 meters [21]. The immature stages inhabit artificial collection of water nearby human dwellings. Dengue epidemics have also been occurred due to Aedespolynesiensis, Aedesalbopictus and Aedescutellaris complex [22]. The eggs can remain viable for long period even without water [23].

Host:

Incubation period of dengue fever is about 4-10 days. The dengue virus enters the host through cutaneous route when an infected mosquito is sucking its blood meal.

Following primary infections, the infected individuals are protected from other serotypes for about 2-3 months but there will be no long-term cross-protect [24, 25]. Monoyte /macrophages play a key role in the pathogenesis of DF and then to the origin of DHF and DSS. These sub neutralizing levels of antibodies enhances the infectivity of virus [26-29].

In the course of secondary infection, antibody-dependent enhancement (ADE) of infection was proposed as a mechanism in the causation of severe dengue [30, 31]. This explains the reason why severe dengue is regularly found during primary infection in infants born to dengue-immune mothers. In this model, non-neutralizing cross-reactive antibodies that are formed during primary infection, or that are acquired passively at birth, bind to the epitopes present on the surface of the heterologous infecting virus and this in turn facilitates virus entry into Fc-receptor-bearing cells [32, 33, 34].

The number of infected cells is therefore increased and this results in a high viral burden and induction of a profound host immune response that includes release of inflammatory mediators, some of which are attributed to cause capillary leakage.

Thrombocytopenia In Dengue Infection

Thrombocytopenia in dengue as a result of

- Alternations in megakaryocytopoiesis when human haematopoletic cells are infected with dengue virus, resulting in impaired progenitor cell growth thereby causing platelet dysfunction.
- Enhanced destruction of platelets.
- Increased consumption (due to peripheral sequestration) of platelets.

Hemorrhage can occur as a consequence of the thrombocytopenic state and associated platelet dysfunction or it can be to disseminated intravascular coagulation [35, 36, 37].

Admission Criteria:

Presence of warning signs related to hypotension, dehydration, profuse sweating, cold extremities, bleeding manifestation or co-morbid condition warrants admission. In these patients, careful and repeated estimation value status and fluid replacement are the cornerstone of management [38]

If the patients have dengue with warning signs, the action plan should be as follows:

Baseline haematocrit should be observed before fluid therapy. Isotonic solutions like 0.9% saline or RL or Hartmann's solution should be preferred.

- To study the clinical profile of serologically proven dengue fever in Kasturba Medical College Mangaluru

- To identify some peculiar features that may help in early recognition and appropriate case management which all together helps in bringing out a good clinical outcome.

Methodology

5-7ml/kg/hr for 1-2 hours,
Assess if vital are stable
3-5ml/kg/hr for 2-4 hours,
Assess if vital are stable
2-3 ml/kg/hr or less depending upon clinical response

Settings:

Department of General Medicine and Pathology Kasturba Medical College Mangaluru

Study Design:

Hospital based cross sectional Observational study

Sample size:

251 cases

Ethical committee approval:

- The present study was approved by the Institutionl Ethical Committee.

Inclusion Creteria:

- Fever with thrombocytopenia with Dengue antigen (NSI) or antibody (IgG or IgM) positivity
- Adults with age more than 13 years

ExcusionCreteria:

- Fever with thrombocytopenia due to other causes.

Materials:

Total of 251 serologically proven dengue cases, who satisfied the inclusion and exclusion criteria were taken up for the study.

Methods:

Complete history, signs, symptoms and laboratory data were recorded as per the performa.

Investigations:

The following investigations were performed in all the patients.

Blood:

TC, Platelet Count, haematocrit, LFT, RFT, Electrolytes.

Radiology:

X-Ray chest,
USG Abdomen

Statistical Analysis:

The data was entered in the Microsoft Excel, spread sheet and analyzed statistically using standard statistical software, SPSS for windows. Chi Square test used for categorical variables. Significance was considered if the 'p' value was below 0.05.

DENGUE Classical Fever (DCF):

Anacute febrile illness with two or more of the following

- Headache
- Retro orbital pain
- Myalgia and Arthralgia
- Skin rash
- Hemorrhagic manifestation
- Nausea, vomiting and
- Supportive serology.

Dengue Hemorrhagic fever (DHF): (all 4 criteria required)

- Fever -- history of fever lasting for 2-7 days.
- Bleeding tendencies indicated by either a positive tourniquet test,petechiae, ecchymoses and purpura
- Bleeding per mucosa, Haematemesis, malena etc.
- Thrombocytopenia less than one lakh
- Plasma leakage evidenced by
- Rise in haematocrit more than 20% and a fall in haematocrit, more than 20% after iv fluids,
- Pleural effusion, ascites, hypoalbuminemia

Dengue Shock Syndrome (DSS):

DSS require all DHF criteria in addition to circulatory failure manifested by

- Rapid and weak pulse
- Narrow pulse pressure<20mm of Hg
- Hypotension

Cold dry skin, restlessness.

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Aim

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RESULTS

The present study is an observational study where we studied the clinical profile of 251 serologically proven dengue patients admitted in the Department of General Medicine and Pathology Kasturba Medical College Mangaluru. The study had revealed much observation that concurred with traditional teaching about dengue; there were other findings that were peculiar to this epidemic. The observations are as follows

Out of 251 cases, 149 patients (59.40%) belonged to DCF, DHF in 76 patients (30.30%), where as 26 patients (10.40%) belonged to more severe variety of DSS. The details are given in the above table.

Majority of cases 47.09 % occurred in young adult <20 years of age. The incidence appeared to reduce with advancing age with least number of cases seen in the age group >60 years of age.

Associations between Dengue type and age

DCF (44.30%) and DSS (42.30%) was more common in the younger age group <20 years DHF (39.50%) was more common in age group of 20-29 years where as DSS is not observed in patients >60 years in this study.

Distribution of dengue in females was slightly higher 51.40% when compared to males 48.60%
DSS was more common in females 65.40%, when compared to males 34.60% where as DHF in men 53.90% when compared to women 46.10%

In this study all patients had fever 100%. Followed by headache 61%, myalgia 54.60%, chills 46.20%, abdominal pain 43.40%, vomiting 42.20%. The characteristic feature of dengue like bone pain and retro-orbital pain was present in only 3.20% and 32.70% respectively. Atypical clinical feature like seizure was present in 1.20%

Even though the bone pain was present in 3.20% it has got statistically significant correlation with Dengue classical fever.

Most common bleeding manifestation encountered was malena 27.50% followed by petechiae 8.40% and gum bleeding in 4.80% less frequent was bleeding manifestation hemoptysis 0.40%

Most common sign observed in this study was conjunctival congestion 23.90%, followed by Hepatomegaly 12%, Ascites 10%, Rashes 10%, Pleural effusion 9.60%, Puffiness of the face & Splenomegaly 8.40% Malena has got statistically significant association with DHF Blood pressure was normal in 76.50%, even Blood pressure was normal in 76.50%, even though 21.50% of patient presented with hypotension, DSS had occurred in 10.40%.

In LFT, All the three Enzymes ALT, AST and ALP were elevated in 47.10%. SGOT more than SGPT in 36.80%; SGPT more than SGOT; both AST and ALP were almost equally elevated in 5.70%, where as SGOT only in 2.30%. The elevation of liver enzymes has no statistical correlation with any particular type of dengue fever. Renal function test was normal in 97.60%, elevated in 2.40%but it has no statistically correlation ECG was normal in 51.40%. Most frequent ECG sign was sinus Tachycardia 33.40%, whereas sinus bradycardia in 11.60%, First degrees AV block in 2.0% Second degrees AV block 0.80%.

CT brain was taken for 4 patients presented with encephalopathy. It was normal in 2 patients, 1 patient with seizure had Intracerebral bleed.

In serology NSI antigen was positive in 61.80%, IgM in 44.60%, IgG in 1.60%.

Approximate number of Dengue cases and death reported during 2012 [17] were

States, Union Territories that were most affected in 2012		
States, Union Territories	**No. of Cases**	**Deaths**
Tamilnadu	12826	66
West Bengal	6456	11
Kerala	4172	15
Karnataka	1924	21
Delhi	2093	04
Puducherry	1506	05

Clinical classification of cases.

Clinical Spectrum of cases	No. of cases	Percent
DSS	**26**	**10.4**
DHF	**76**	**30.3**
DCF	**149**	**59.4**
Total	**251**	**100**

Clinical spectrum of case Distribution of Age

AGE	Frequency	Percent
<20 yrs	**101**	**40.2**
20 – 29	**77**	**30.7**
30 – 39	**27**	**10.8**
40 – 49	**22**	**8.8**
50-59	**19**	**7.6**
>60yrs	**5**	**2.0**
Total	**251**	**100**

Associations between Dengue type and age.

AGE (Code)	Dengue Clinical Type		
	DSS	**DHF**	**DCF**
<20 yrs	**42.30%**	**31.60%**	**44.30%**
20 – 29	**26.90%**	**39.50%**	**26.80%**

30 – 39	**3.80%**	**13.20%**	**10.70%**
40 – 49	**15.40%**	**5.30%**	**9.40%**
50-59	**11.50%**	**6.60%**	**7.40%**
>60yrs	**--**	**3.90%**	**1.30%**

Association of Dengue type and Liver Enzymes

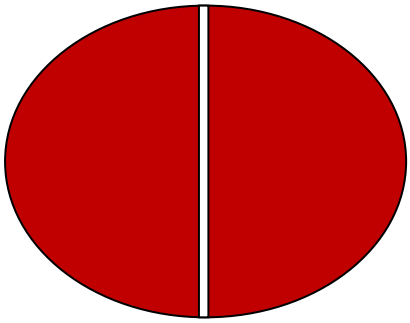
LFT Liver Enzymes	Frequency	Percent
Elevated SGOT, SGPT, Alk, Phosphate	41	47.1
SGOT>SGPT	32	36.8
SGPT=SGOT	5	5.7
SGPT>SGOT	7	8.0
SGOT Only	2	2.3
Total	87	100

In serology NSI antigen was positive in 61.80%, IgM in 44.60%, IgG in 1.60%

Serology	Frequency	Percent
NS 1	155	61.8
IgM	112	44.6
IgG	4	1.6

Gender Distribution

Male 48.60%



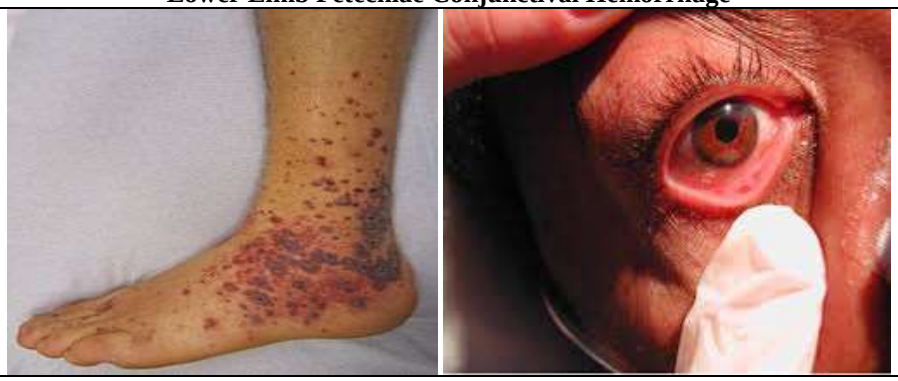
Female 51.40%

(AedesEgypti (Tiger) Mosquito)

Breeding Source of Aedes Mosquito

Conjunctival Suffusion Gum Bleeding **Palatal Petchiae**

Lower Limb Petechiae Conjunctival Hemorrhage



DISCUSSION

This study describes the clinical profile of serologically proven dengue cases in adults aged more than 13 years admitted in our hospital during the current Dengue epidemic of 2012.

A total of 251 cases admitted between April 2012-March 2013 were studied by analyzing the symptoms, signs, hematological and laboratory findings,

Analysis o Blood count among cases

Thrombocytopenia was the most common abnormally observed 167 cases (66.50%) had thrombocytopenia ranging from mild to severe thrombocytopenia.

Haematocrit was elevated in 73 cases studied and they had to be treated with IV fluid therapy in addition to oral fluids. Haematocrit was followed up evey 6 hours and fluids were titrated accordingly. Haematocrit was persistently elevated in 27 cases where the duration of fluid therapy was extended to > 48 hours and even 72 hours to normalize the haematocrit. WHO recommends fluid therapy only for 48 hours but we had to give IVF for an average of 72 hours according to haematocrit. This was a significant change in the fluid management strategy that we followed in this epidemic and was found to be more effective in treating our population with dengue. Further studies dealing with fluid therapy in our population with dengue is needed to validate our finding.

Dengue cases were confirmed by serology. NSI antigen and ELISA was the most important test used for diagnosing dengue cases in our hospital. Dengue IgM and IgGwre performed in cases according to the availability of test kits. NSI was found to be positive in 155 cases (61.80%) and IgM positive in 112 cases (44.60%). IgG was positive only in 4 cases.

Analysis of X-Ray findings:

Chest X-Ray PA view was normal in 230 cases. Right sided pleural effusion was detected in 13 cases and bilateral pleural effusion noted in 6 cases.

CT brain was performed in patients who had presented with encephalopathy and seizures. CT brain was normal in 2 cases. Intracerebral bleed was noted in 1 case and minimal white matter hypo density was noted in the other case.

CONCLUSION

In this study the clinical profile of serologically proven Dengue fever was studied from April 2012– March 2013 particularly during the epidemic that stormed Puducherry district. With this clinical profile it is easy to recognize and understand the clinical problem. The application of clinical spectrum of WHO classification system is not as very simple and straightforward as it seems because clinical features may overlap among different categories. The WHO classification system of dengue does not include unusual manifestation like encephalopathy, seizures, myocarditis, etc. which might be life-threatening. Although these manifestations are rare, clinicians should always have a high index of suspicion

and knowledge of these atypical manifestations, particularly in view of the increasing burden of dengue in recent years. Blood pressure and haematocrit should be monitored for evaluating for the progress of the disease. Bleeding tendency should be closely watched. Management of patient with dengue is mainly supportive simple inexpensive and very effective in saving lives, prophylactic FFP and platelets are not necessary for treating DHF, DSS, but early and meticulous monitoring are the corner stone for positive outcome.

REFERENCE:

1. Dengue in India Indian J Med Res 136, September 2012, pp 373-390.
2. Chaturvedi UC, Mathur A, Kapoor AK, Tandon HO, Mehotra RML, *et al.* Clinicovirological study of the recurrence of dengue epidemic with haemorrhagic manifestation at Kanpur, during 1969. Indian J Med Res, 60, 1972, 329-33.
3. Myers RM, Carey DE, Banerjee K, Reuben R, Ramamurti DV, *et al.* Recovery of dengue type 3 virus from human serum and Aedesaegypti in South India. Indian J Med Res, 56, 1968, 781-7.
4. Myers RM, Carey DE, De Ranitz CM, Reuben R, Bennet B, *et al.* Virological investigations of the 1966 outbreak of Dengue type 3 in Vellore, Southern India. Indian J Med Res,57, 1969,1392-401.
5. Mukherjee KK, Chakravarti SK, Dey PN, Dey S, Chakraborty MS, *et al.* Outbreak of febrile illness due to dengue virus type 3 in Calcutta during 1983. Trans R Soc Trop Med Hyg 81, 1987, 1008-10.
6. Dash PK, Saxena P, AbhyankarA, Bhargava R, Jana AM, *et al.* Reemergence of dengue virus Type-3 (subtype-III) in India implecations for increased incidence of DHF & DSS. Virol J 3, 2006, 55-65.
7. Paramasivan R, Thenmozhi V, Thangaratham PS, Rajendran R, Tewari SC, Dhananjeyan KJ, *et al*. An outbreak of dengue fever in Tirupur, Coimbatore districts, Tamilnadu, Indian J Med Res, 132, 2010, 105-7.
8. Chakravarti A, Kumar A, Matlani M, *et al.* Displacement of dengue virus type 3 and type 2 by dengue virus type 1 in Delhi during 2008. Indian J Med Microbiol 28, 2010, 412-3.
9. Chahar HS, Bharaj P, Dar L, GuleriaR,Kabra SK, Broor S, *et al.* Co-infections with chikungunya virus and dengue virus in Delhi, India Emerge Infect Dis, 15, 2009, 1077-80
10. Dhash Pk, Sharma S, Srivastava A, Santhosh SR, Parida MM, Neeraja M, *et al*. Emergence of dengue virus type 4 (genotypeI) in India.Epidomiol Infect 139, 2011, 857-61.
11. Dayaraj C, Kakade MB, Bhagat AB, Vallentyne J, Singh A, Patil JA, et al. Detection of dengue-4 virus in Pune, Wetern India after an absence of 30years – its association with two severe cases. Virol J, 8, 2011, 46-9.
12. Gubler DJ. The global emergence of arboviral disease as public health problems. Arch Med Res 33, 2002, 330-422.
13. Westaway EG, Blok J. Taxonomy and evolutionary relationships of flaviviruses. In: Gubler DJ, Kuno G, editors. Dengue and dengue hemorrhagic fever. New York: CAB international; 1997.p.147.
14. Leitmeyer KC. Dengue virus structural differences that correlate with pathogenesis. Journal of Virology, 73(6), , 1999, 4738-4747.
15. Messer WB. Emergence and global spread of a dengue serotype 3, subtype III virus. Emerging Infections Diseases, 9(7), 2003,800-809.
16. Wilder-Smith A, Schwartz E. Dengue in travelers. N Engl J Med. 353, 2005, 924-932.
17. Guzman MG, Kouri G. Dengue an update. Lancet-infect Dis. 2, 2002, 33-42.
18. Halstead SB. Etiologies of the experimental dengues of siler and Simmons, American Journal of Tropical Medicine and Hygiene, 23, 1974, 974—982.
19. Trpis M, Hausermann W. Genetic of house – entering behavior in East African populations of Aedesaegypti (L) (Diptera: Culicidae) and its relevance to speciation. Bull Entomol Res, 8, 1978, 521-32.
20. Schexneider, K.I and Reedy, E. A. Thrombocyttopenia in dengur fever. Curr, Hematol. Rep 145-1484
21. Kalayanarooj, S.*et al*. Dengue patients at the children's hospital, Bangkok: 1995-1999. Review. Dengue Bull. 26, 2002, 33-43.
22. Mourao, M. P, *et al*. Thrombocytopenia in patient with dengue virus infection in the Brazilian Amazon. Platelets, 18, 2007, 605-612.
23. Honda, S. et al. Increased Phagocytosis of platelets from patients with secondary dengue virus infection by human macrophages. Am. J. Trop. Med. Hyg, 80, 2009, 841 – 845.
24. Rigau-Perez JG, *et al*. Dengue and dengue hemorrhagic fever Lancet, 352, 1998, 971-97.
25. Kalayanarooj, S.*et al.* Early clinical and Laboratory indicators of acute dengue illness. Journal of Infectious Diseases, 176, 1997, 313-321.
26. Balmaseda A *et al.* Assessment of the World Health Organization scheme for classification of dengue severity in Nicaragua. American Journal of Tropical Medicine and Hygiene, 73, 2005, 1059-1063.
27. Phuong CXT, *et al.* Evaluation of the World Health Organization standard tourniquet test in the diagnosis of dengue infection in Vietnam. Tropical Medicine and International.

28. Srikiatkhachorn A *et al*. Natural history of plasma leakage in dengue hemorrhagic fever: a serial ultrasonic study. Pediatric Infectious Disease Journal, 26(4), 2007, 283-290.
29. Nimmannitya S, *et al.* Dengue and Chikungunya virus infection in man in Thailand, 1962-64. Oservations on hospitalized patients with haemorrhagic fever. American Journal of Tropical Medicine and Hygiene, 18(6), 1969, 954-971.
30. Martinez- Torres E, Polanco-Anaya AC, Pleites – Sandoval EB, *et al.* Why and how children with dengue die? Revistacubana de medicina tropical, 60(1), 2008, 40-47.
31. Wali JP, Biswas A, Chandra S, *et al.* Cardiac involvement in dengue haemorrhagic fever Int J Cardiol. 64(1), 1998, 31-6
32. Hommel D, talarmin A, Reynes JM, *et al*. Acute renal failure associated with dengue fever in French Guiana, Nephron, 1999, 83, 183.
33. Clinical profile of dengue infection at a teaching hospital in North India RituKaroli, Jalees Fatima, ZebaSiddiqi, Kursheed I, Kazmi, Amit R, Sultania Department of Medicine, Era's Lucknow Medical College, Safarazganj, Hardoi Road, Lucknow, Uttar Pradesh, India J Infect DevCtries 6(7), 2012, 551-554. Received 01 April 2011 – Accepted 26 July 2011.
34. Gupta P, Khare V, Tripathi S, Nag VL, Kumar R, Khan MY, Dhole TK, *et al.* Assessment of WHO definition of dengue hemorrhagic fever in North India. J Infect DevCtries, 4, 2010,150-155.
35. Prakash O, Almas A, Jafri Wasmin SM, Hamid S, Akhtar J, Alishah H, *et al*. Severity of acute hepatitis and its outcome in patients with dengue fever ina tertiary care hospital Karachi, Pakistan (South Asia). BMC Gastroenterology, 10, 2010, 43.
36. Manjith Narayanan, Aravind MA, Thilothammal N, PreRna R, Rex CS, Sarguram and Nalini Ramamurthy, *et al.* Dengue fever Epidemic in Chennai – A Study of Clinical Profile and outcome. Indian Pediatrics 39, 2002, 1027-1030B.
37. Ooi ET, Ganesanathan S, Anil R, Kwok FY, Sinniah M, *et al.* Gastrointestinal manifestations of Dengue infections in adults. Med J Malaysia 63, 2008, 401-05.
38. Desouza LJ, Nogueria RM, Dinh The Trung, Le Thi Thu Thao are LC, Soares CE, Ribas BF, Alves FP, Viera FR, Pessanha FE, *et al.* The impact of dengue on liver function as evaluated by aninotransferase levels. Braz J Infect Dis 11, 2007, 407-10.