RESEARCH BRIEF

Profile and Trends of Rotavirus Gastroenteritis in Under-five Children in India (2012-2014): Preliminary Report of the Indian National Rotavirus Surveillance Network

CP GIRISH KUMAR, S VENKATASUBRAMANIAN, *GAGANDEEP KANG, #RASHMI ARORA AND SANJAY MEHENDALE, FOR THE NATIONAL ROTAVIRUS SURVEILLANCE NETWORK

*From National Institute of Epidemiology, Chennai; *Christian Medical College and Hospital, Vellore; and #Indian Council of Medical Research, New Delhi; India*

Correspondence to:
Dr CP Girish Kumar,
National Institute of Epidemiology,
II Main Road, TNHB, Ayapakkam,
Chennai 600 077, India.
girishkumar@nie.gov.in
Received: January 02, 2016;
Initial review: January 04, 2016;
Accepted: May 12, 2016.

Objective: To estimate the burden of rotavirus-associated gastroenteritis in India.

Methods: Hospital-based surveillance network was established, with clinical evaluation and laboratory testing for rotavirus among children aged below 5 years hospitalized with acute gastroenteritis.

Results: Between September 2012 and December 2014, stool samples from 10207 children were tested and rotavirus was detected in 39.6% of cases. Infections were more commonly seen among younger children (<2 years). Detection rates were higher during cooler months of September – February. Among rotavirus infected-children, 64.0% had severe or very severe disease. G1P[8] was the predominant rotavirus genotype (62.7%) observed during the surveillance period.

Conclusions: Surveillance data highlights the high rotavirus disease burden and emphasizes the need for close monitoring to reduce morbidity and mortality associated with rotavirus gastroenteritis in India.

Key words: *Diarrhea, Epidemiology, Prevalence, Trends.*

Rotavirus is the leading cause of severe childhood gastroenteritis/diarrhea worldwide, and is estimated to account for about one-third of deaths attributable to diarrhea in children under five years of age [1,2]. In this report, we present the findings of the rotavirus surveillance carried out as part of the National Rotavirus Surveillance Network (NRSN) established by the Indian Council of Medical Research (ICMR). This preliminary report describes rotavirus burden in children admitted with acute gastroenteritis between September 2012 and December 2014 by age, region, seasons and also the diarrheal disease severity pattern.

METHODS

The NRSN surveillance protocol was developed based on a modification of the WHO generic protocol for rotavirus surveillance [3]. Prospective surveillance for diarrhea-related hospitalizations among children under five years of age was carried out in 28 hospital-based surveillance units spread across 17 states and two union territories (UT) of India (Details of study sites are provided in a companion paper in this issue) [4], and in ***Web Annexure* 1**. The surveillance units or clinical recruitment sites (CRS) were linked with either Referral or Regional Laboratories. These clinical recruitment sites were either governmental (n=16) or private (n=12) health care facilities for pediatric patients. The study was initiated after obtaining approvals by the institutional ethics committees of the National Institute of Epidemiology (NIE), all the participating reference and referral laboratories, and the CRS.

All children aged ≤59 months who presented to a participating CRS with acute gastroenteritis (≥3 loose stools in a 24 hour period for 5 or less days), and required hospitalization for diarrhea management, were enrolled after obtaining informed and written consent from the accompanying parent or guardian. Study medical officers enrolled eligible children and collected clinical and demographic details on standardized clinical recruitment forms (CRF).

Whole stool specimens (~5 mL) were collected and transported within 2 hours to the testing laboratory or stored in a refrigerator at 4^0C until transportation. Samples stored at 4^0C were transported in boxes with ice

packs at weekly or fortnightly intervals to the testing laboratories. All stool samples were subjected to rotavirus screening using commercial enzyme immunoassay (Premier Rotaclone, Meridian Biosciences) kits following the manufacturer's instructions. Rotavirus positive samples were subjected to rotavirus genotyping for VP7 (G-typing) and VP4 (P-typing) by Reverse-transcription polymerase chain reaction (RT-PCR) [5,6]. Initially all positives were genotyped but subsequently it was decided to restrict genotyping to every third positive sample at each testing laboratory. There were no gender, age, region-wise or seasonal considerations for choosing samples for genotyping. Aliquots of all samples were stored at -70^{0}C.

Data captured on the CRF were entered at each of the regional and referral laboratories using the online data capture portal hosted on the NIE web server. Site-specific summary data on diarrheal hospital admissions and rotavirus positivity data from laboratories was received at the Coordination Center (NIE) on a monthly basis.

Statistical analysis: Data were analyzed to assess the proportions of rotavirus-positive cases in terms of demographic factors, symptoms, disease severity (Vesikari score), median duration of hospitalization, genotype status and also by season and regions. Proportion ratios (PR) were calculated to compare the strength of association of severe infection in rotavirus infected and uninfected children with seasonal rotavirus burden and length of hospitalization. Analyses were carried out using MS Excel 2007, SPSS *v.* 17.0 and Stata *v* 10.0.

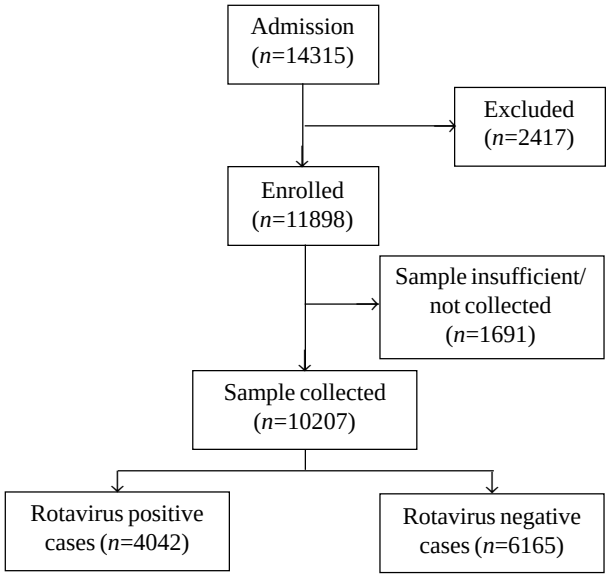

FIG. 1 *Flow diagram summarizing case enrollment and laboratory testing.*

RESULTS

Details of case enrolment and testing are provided in ***Fig.* 1**. Rotavirus was detected in 39.6% (4042/10207) of diarrheal cases. Rotavirus infection was detected throughout the year in all CRS. The detection rates were higher during December- February (56.4%; 1668/2959).

Most cases of rotavirus infection (41.5%; 3404/8206) occurred among children less than 2 years of age (***Table* I**). The highest positivity (46.7%) was observed among children between 12 and 23 months of age. Among infants aged <3 months and <6 months, the proportion of rotavirus positivity was 17.4% (127/729) and 27.4% (497/1814) respectively. There were 118 neonates, and 22.9% (27/118) of them were rotavirus positive. The median (IQR) age for rotavirus infection was 12 (8,18) months, which was not significantly

TABLE I ROTAVIRUS POSITIVITY IN CHILDREN HOSPITALIZED WITH ACUTE GASTROENTERITIS

Variables	*Levels*	*N*	*Rotavirus positivity, n (%)*
Age (mo)	0-2	729	127 (17.4)
	3-5	1085	370 (34.1)
	6-11	3148	1393 (44.3)
	12-23	3244	1514 (46.7)
	≥24	2001	638 (31.9)
	Median (IQR)		12 (8 – 18)
Gender	Male	6363	2520 (39.6)
	Female	3844	1522 (39.6)
Disease	Mild [0-5]	433	123 (28.4)
severity	Moderate [6-10]	3779	1334 (35.3)
	Severe [11-15]	5509	2394 (43.5)
	Very Severe [16-20]	484	191 (39.5)
Treatment	Oral	1987	788 (39.7)
	Intravenous	8220	3254 (39.6)
Season	Dec-Feb	2959	1668 (56.4)
	Mar-May	1838	694 (37.8)
	Jun-Aug	2239	461 (20.6)
	Sep-Nov	3171	1219 (38.4)
Region	East	2032	827 (40.7)
	West	1755	689 (39.3)
	South	4080	1521 (37.3)
	North	2340	1005 (42.9)
Oral RV vaccination	Yes	340	111 (32.6)

different from that seen among cases of non-rotavirus gastroenteritis [median 12 mo. IQR (6,21) mo, *P*=0.51]. Hospital admissions due to rotavirus gastroenteritis among boys (2520/6363) outnumbered that among girls (1522/3844) with no significant difference in rotavirus positivity rates between the two genders (39.6% *vs* 39.6%; *P*=0.992).

Burden of rotavirus varied significantly across regions and seasons (***Table* I**). Rotavirus infections usually occurred more commonly during the cooler months of December - February (56.4%), followed by September -November (38.4%).

Analysis of diarrheal disease severity showed that the proportion rotavirus positive (64%) was greater among children with very severe or severe disease compared to children with mild to moderate infection (36%). A proportion ratio analysis revealed that proportion of severe rotavirus gastroenteritis was more during December-February (ratio: 1.75; CI: 1.65- 1.85), and these children were likely to stay in the hospital for ≥3 days (ratio: 1.72; CI: 1.60-1.85) (***Table* II**).

Only 3.3% (340/10206) of enrolled children had a history of rotavirus vaccination, and among them 111 children (32.6%) were rotavirus positive. Majority of vaccinated children were seen at private hospitals (87.9 %).

TABLE II PROPORTION RATIO ANALYSIS

A. Analysis of prevalence of severe gastroenteritis across seasons

Season	*Rotavirus positive (n=2585)*	*Rotavirus negative (n=3408)*	*PR (95 % CI)*
Dec -Feb	1103	687	1.748 (1.654, 1.846)
Mar-May	400	609	0.904 (0.833, 0.982)
Jun-Aug	282	1029	0.437 (0.393, 0.487)
Sep-Nov	800	1083	0.978 (0.918, 1.042)

B. Analysis of length of hospitalization (≥3 days) among cases of severe gastroenteritis across seasons

Season	*Rotavirus positive (n=1569)*	*Rotavirus negative (n=2121)*	*PR (95 % CI)*
Dec-Feb	651	425	1.723 (1.605, 1.850)
Mar-May	268	396	0.939 (0.848, 1.039)
Jun-Aug	172	647	0.432 (0.376, 0.495)
Sep-Nov	478	653	0.991 (0.914, 1.076)

***Proportion Ratio (PR) = Proportion of RV +ve cases in a particular season / Proportion of RV +ve cases in remaining seasons.*

During the reporting period, 15 (0.15%) enrolled children died during their hospital stay. Rotavirus antigen was detected in stool from four children. Twelve children were below one year of age with a median (IQR) age of 5 (1,7) months. On admission, nine children had severe to very severe diarrhea (median (IQR) Vesikari score 15 (14,15)). Thirteen children with severe diarrheal disease (median (IQR)) Vesikari score 14 (10,15) had received intravenous fluids and the remaining two children with mild to moderate diarrhea had received oral rehydration. Analysis of the cause of death revealed that the children had died due to serious complications *viz*. sepsis and shock (10 cases), sepsis and meningitis (3 cases), bronchopneumonia (1 case), and milk-aspiration (1 case).

Analysis of overall distribution of various rotavirus genotypes showed the preponderance of G1P[8] strains (62.7%) followed by G2P[4] strains (7.6%) (data not shown). Among neonates hospitalized with gastroenteritis, eight genotypes were observed with G1P[8] (45.5%; 10/22) as the commonest strain.

DISCUSSION

Using a standardized approach for patient enrollment and testing in country-wide surveillance, rotavirus was detected in 39.6% of children admitted with diarrhea. This is consistent with findings from the earlier phase of rotavirus surveillance in India [4-6]. Our report highlights the substantial rotavirus disease burden in India. In the present round of surveillance, although most cases of rotavirus gastroenteritis were seen among children between 12 and 23 months of age, it was documented in all age sub-groups, including neonates. These rates are; however, higher than previously reported from India [5-7]. Neonatal infections which are usually mild or asymptomatic are caused by different nursery rotavirus strains, but in this study, nearly half the neonates (10/22 genotyped) had gastroenteritis due to G1P[8], which was also the predominant genotype circulating among older children. These findings are in agreement with data from the previous surveillance that reported early incidence of rotavirus infection in India [6].

Analysis of diarrheal disease severity showed that children with rotavirus infection have severe disease and the occurrence of severe rotavirus gastroenteritis was more commonly observed during December – February period that represents cooler season in India. The seasonal pattern observed in this surveillance period, with more infections occurring during cooler months was similar to the observation during the previous iteration of the NRSN, and is consistent with reports from most parts of the world [6,8,9].

WHAT THIS STUDY ADDS?

- Rotavirus is associated with 40% of diarrheal episodes requiring hospitalization in under-five children.

The reported low mortality among hospitalized children with acute gastroenteritis could not be attributed to rotavirus infection. This probably reflects that an effective diarrheal disease management protocol is practiced in the health care facilities participating in this surveillance. It would be necessary to study the community burden of rotavirus gastroenteritis and related morbidity and mortality outcomes to know the true burden in the community settings and non-hospitalized children. The limitation of the present analysis is that the findings that have been presented in this paper represent interim analysis of data.

The data from the expanded NRSN was already available with policy makers before the recent decision to introduce rotavirus vaccine in the UIP [10]. Continued surveillance and studies among vaccinated children will generate evidence regarding impact of rotavirus vaccine rolled out through the national program on morbidity and mortality due to rotavirus infections in young Indian children.

The successfully implemented NRSN surveillance platform will continue to generate data on trends in rotavirus disease burden and its correlates. It will also contribute significantly in the assessment of the impact of the rotavirus vaccine after implementation.

Acknowledgements: M.Chiranjeevi, Technical Assistant, NRSN project Team at NIE for support in statistical analysis.
Contributors: CPGK, SMM: Network coordination, conceptualization and manuscript writing; SV: Contributed to network coordination, statistical analysis and manuscript writing; GK: Coordinated laboratory activities in the network and provided intellectual inputs for manuscript development; RA: Coordination at ICMR level and provided intellectual inputs to for manuscript development. All authors approved the final manuscript.
Funding: Indian Council of Medical Research; *Competing Interests*: None stated.

REFERENCES

1. World Health Organization. Rotavirus Mortality Estimates. Available from: *http://www.who.int/immunization/monitoring_surveillance/burden/estimates/rotavirus/en*. Accessed March 21, 2016.
2. John J, Sarkar R, Muliyil J, Bhandari N, Bhan MK, Kang G. Rotavirus gastroenteritis in India, 2011-2013: Revised estimates of disease burden and potential impact of vaccines. Vaccine. 2014;32:A5-9.
3. World Health Organization. Generic protocols (i) hospital-based surveillance to estimate the Burden of Rotavirus Gastroenteritis in Children and (ii) a Community Based Survey on Utilization of Health Care Services for Gastroenteritis in Children. Geneva. 2002.
4. Mehendale S, Venkatasubramanian S, Girish Kumar CP, Kang G, Gupte MD, Arora R. Expanded Indian National Rotavirus Surveillance Network in the context of rotavirus vaccine introduction. Indian Pediatr.2016;53:575-81.
5. Kang G, Desai R, Arora R, Chitambar S, Naik TN, Krishnan T, *et al*. Diversity of circulating rotavirus strains in children hospitalized with diarrhea in India, 2005–2009. Vaccine. 2013;31:2879-83.
6. Kang G, Arora R, Chitambar SD, Deshpande J, Gupte MD, Kulkarni M, *et al*. Multicenter, hospital-based surveillance of rotavirus disease and strains among Indian children aged <5 years. J Infect Dis. 2009;200:S147–S53.
7. Saluja T, Sharma SD, Gupta M, Kundu R, Kar S, Dutta A, *et al*. A multicenter prospective hospital-based surveillance to estimate the burden of rotavirus gastroenteritis in children less than five years of age in India. Vaccine. 2014;32:A13-9.
8. Brandt CD, Kim HW, Rodriguez WJ, Arrobio JO, Jeffries BC, Parrott RH. Rotavirus gastroenteritis and weather. J Clin Microbiol. 1982;16:478-82.
9. Jagai JS, Sarkar R, Castronovo D, Kattula D, McEntee J, Ward H, *et al*. Seasonality of rotavirus in South Asia: A meta-analysis approach assessing associations with temperature, precipitation, and vegetation index. PLoS ONE. 2012;7:e38168.
10. Ministry of Health and Family Welfare Notable Achievements and Initiatives- 2015. Press Information Bureau. Available from: *http://pib.nic.in/newsite/PrintRelease.aspx?relid=133853*. Accessed March 21, 2016.

WEB ANNEXURE 1

Other participating investigators of National Rotavirus Surveillance Network [2012 – 2016]

Agarwal A (Netaji SC Bose Medical College & Hospital, Jabalpur); Aneja S (Kalawati Saran Children Hospital (LHMC) New Delhi); Anna Simon (Christian Medical College, Vellore); Aundhakar SC (Krishna Institute of Medical Sciences, Karad); Babji S (Medical College, Vellore and Infant Jesus Hospital, Trichy); Bavdekar A (KEM Hospital, Pune); Baveja. S (Lokmanya Tilak Municipal GH & Medical College, Mumbai); Bhat J (National Institute for Research in Tribal Health, Jabalpur); Biswas D (Assam Medical College & Hospital, Dibrugarh); Bora CJ (Assam Medical College & Hospital, Dibrugarh); Borkakoty B (Regional Medical Research Centre, Dibrugarh); Chatterjee S (Midnapore Medical College, Midnapore); Chaudhary S (Dr. Rajendra Prasad Government Medical College, Tanda); Chawla-Sarkar M (National Institute of Cholera and Enteric Diseases, Kolkata); Chitambar SD (National Institute of Virology, Pune); Das P (Rajendra Memorial Research Institute of Medical Sciences, Patna); Das VNR (Rajendra Memorial Research Institute of Medical Sciences, Patna); Desai K (Surat Municipal Institution of Medical Education & Research, Surat); Dhongade R (Sant Dnyaneshwar Medical Education & Research Centre, Pune); Dwibedi B (Regional Medical Research Centre, Bhubaneswar); Dwivedi R (Kamla Nehru Hospital, Gandhi Medical College, Bhopal); Dzuvichu K (District Hospital, Nagaland); Ganguly N (Institute of Child Health, Kolkata); Gathwala G (Pt. B.D.Sharma PGIMS, Rohtak); Ghosh C (Midnapore Medical College, Midnapore); Gopalkrishna V (National Institute of Virology, Pune); Gupta DS (Surat Municipal Institution of Medical Education & Research, Surat); Jadhav AR (Lokmanya Tilak Municipal GH & Medical College, Mumbai); Jali S (Jawaharlal Nehru Medical College, Belgaum); Kalrao VR (Bharati Vidyapeeth Medical College, Pune); Kar SK (Regional Medical Research Centre, Bhubaneswar); Khuntia HK (Regional Medical Research Centre, Bhubaneswar); Kumar P (Kalawati Saran Children Hospital, New Delhi); Kumar S (National Institute for Research in Tribal Health, Jabalpur); Kumar SS (Pragna Children's Hospital, Hyderabad); Lal BG (BJR Hospital, Port Blair); Manglani M (Lokmanya Tilak Municipal GH & Medical College, Mumbai); Manohar B (Sri Venkateswara Institute of Medical Sciences, Tirupati); Mathew A (St. Stephen's Hospital, Delhi); Mathew MA (MOSC Medical College, Kolenchery); Mehariya KM (BJ Medical College & Civil Hospital, Ahmedabad); Mishra SK (Capital Hospital, Bhubaneswar); Narayanan SA (Institute of Child Health & Hospital for Children, Chennai); Niyogi P (Institute of Child Health, Kolkata); Panda S (National Institute of Cholera and Enteric Diseases, Kolkata); Pandey K (Rajendra Memorial Research Institute of Medical Sciences, Patna); Patankar M (BJ Medical College & Civil Hospital, Ahmedabad); Purani CS (BJ Medical College & Civil Hospital, Ahmedabad); Ray P (Jamia Hamdard, New Delhi); Roy S (Regional Medical Research Centre, Belgaum); Sahoo GC (Rajendra Memorial Research Institute of Medical Sciences, Patna); Singh N (National Institute for Research in Tribal Health, Jabalpur); Singh P (Surat Municipal Institution of Medical Education & Research, Surat); Singh T (Christian Medical College, Ludhiana); Sundari S (Institute of Child Health & Hospital for Children, Chennai); Temsu T (District Hospital, Nagaland); Thakur AK (Nalanda Medical College & Hospital, Nalanda); Topno RK (Rajendra Memorial Research Institute of Medical Sciences, Patna); Upadhyay A (Lala Lajpat Rai Memorial Medical College, Meerut); Utpalkant Singh (Child Care Centre, Patna); Vijayachari P (Regional Medical Research Centre, Port Blair).

WEB ANNEXURE 1

Other participating investigators of National Rotavirus Surveillance Network [2012 – 2016]

Agarwal A (Netaji SC Bose Medical College & Hospital, Jabalpur); Aneja S (Kalawati Saran Children Hospital (LHMC) New Delhi); Anna Simon (Christian Medical College, Vellore); Aundhakar SC (Krishna Institute of Medical Sciences, Karad); Babji S (Medical College, Vellore and Infant Jesus Hospital, Trichy); Bavdekar A (KEM Hospital, Pune); Baveja. S (Lokmanya Tilak Municipal GH & Medical College, Mumbai); Bhat J (National Institute for Research in Tribal Health, Jabalpur); Biswas D (Assam Medical College & Hospital, Dibrugarh); Bora CJ (Assam Medical College & Hospital, Dibrugarh); Borkakoty B (Regional Medical Research Centre, Dibrugarh); Chatterjee S (Midnapore Medical College, Midnapore); Chaudhary S (Dr. Rajendra Prasad Government Medical College, Tanda); Chawla-Sarkar M (National Institute of Cholera and Enteric Diseases, Kolkata); Chitambar SD (National Institute of Virology, Pune); Das P (Rajendra Memorial Research Institute of Medical Sciences, Patna); Das VNR (Rajendra Memorial Research Institute of Medical Sciences, Patna); Desai K (Surat Municipal Institution of Medical Education & Research, Surat); Dhongade R (Sant Dnyaneshwar Medical Education & Research Centre, Pune); Dwibedi B (Regional Medical Research Centre, Bhubaneswar); Dwivedi R (Kamla Nehru Hospital, Gandhi Medical College, Bhopal); Dzuvichu K (District Hospital, Nagaland); Ganguly N (Institute of Child Health, Kolkata); Gathwala G (Pt. B.D.Sharma PGIMS, Rohtak); Ghosh C (Midnapore Medical College, Midnapore); Gopalkrishna V (National Institute of Virology, Pune); Gupta DS (Surat Municipal Institution of Medical Education & Research, Surat); Jadhav AR (Lokmanya Tilak Municipal GH & Medical College, Mumbai); Jali S (Jawaharlal Nehru Medical College, Belgaum); Kalrao VR (Bharati Vidyapeeth Medical College, Pune); Kar SK (Regional Medical Research Centre, Bhubaneswar); Khuntia HK (Regional Medical Research Centre, Bhubaneswar); Kumar P (Kalawati Saran Children Hospital, New Delhi); Kumar S (National Institute for Research in Tribal Health, Jabalpur); Kumar SS (Pragna Children's Hospital, Hyderabad); Lal BG (BJR Hospital, Port Blair); Manglani M (Lokmanya Tilak Municipal GH & Medical College, Mumbai); Manohar B (Sri Venkateswara Institute of Medical Sciences, Tirupati); Mathew A (St. Stephen's Hospital, Delhi); Mathew MA (MOSC Medical College, Kolenchery); Mehariya KM (BJ Medical College & Civil Hospital, Ahmedabad); Mishra SK (Capital Hospital, Bhubaneswar); Narayanan SA (Institute of Child Health & Hospital for Children, Chennai); Niyogi P (Institute of Child Health, Kolkata); Panda S (National Institute of Cholera and Enteric Diseases, Kolkata); Pandey K (Rajendra Memorial Research Institute of Medical Sciences, Patna); Patankar M (BJ Medical College & Civil Hospital, Ahmedabad); Purani CS (BJ Medical College & Civil Hospital, Ahmedabad); Ray P (Jamia Hamdard, New Delhi); Roy S (Regional Medical Research Centre, Belgaum); Sahoo GC (Rajendra Memorial Research Institute of Medical Sciences, Patna); Singh N (National Institute for Research in Tribal Health, Jabalpur); Singh P (Surat Municipal Institution of Medical Education & Research, Surat); Singh T (Christian Medical College, Ludhiana); Sundari S (Institute of Child Health & Hospital for Children, Chennai); Temsu T (District Hospital, Nagaland); Thakur AK (Nalanda Medical College & Hospital, Nalanda); Topno RK (Rajendra Memorial Research Institute of Medical Sciences, Patna); Upadhyay A (Lala Lajpat Rai Memorial Medical College, Meerut); Utpalkant Singh (Child Care Centre, Patna); Vijayachari P (Regional Medical Research Centre, Port Blair).