Disease Specific Documents for XII Plan

Japanese Encephalitis

(JE)/AES

INDIAN COUNCIL OF MEDICAL RESEARCH

Annexure XIV

Disease Specific Documents for XII Plan

Japanese Encephalitis (JE)/AES

High Power Committee to Evaluate the Performance of ICMR, 2012-13

Indian Council of Medical Research, New Delhi

Year of Publication 2014

Dr. V.M. Katoch
Secretary, DHR & DG, ICMR

Coordination, Report Compilation & Editing
Dr. G.S. Toteja, Director, DMRC, Jodhpur & Head, Division of Nutrition, ICMR Hqrs.
Dr. Rajni Kant, Scientist 'E', ICMR, Hqrs.

Technical Support
Dr. Sprhia Rao, Scientist 'B', Division of Nutrition, ICMR Hqrs.

Head P&I
Dr. V.K. Srivastava, Scientist 'G'

Production Controller
JN Mathur, Press Manager

Published by the Division of Publication & Information on behalf of the Secretary DHR & DG, ICMR, Ministry of Health & Family Welfare, New Delhi

Designed & Printed at M/s Aravali Printers & Publishers (P) Ltd., W-30, Okhla Industrial Area, Phase-II, New Delhi-110020 Phone: 47173300, 26388830-32

ICMR INSTITUTES WORKING ON JE/AES

1. National Institute of Virology (NIV), Pune (lead institute)
2. Regional Medical Research Centre (RMRC), Dibrugarh
3. Regional Medical Research Centre (RMRC), Bhubaneswar
4. Desert Medicine Research Centre (DMRC), Jodhpur
5. Vector Control Research Centre (VCRC), Puducherry
6. Centre for Research in Medical Entomology (CRME), Madurai
7. Division of Epidemiology and Communicable Diseases , ICMR Hqrs

1. Current situation of Disease with contribution of ICMR

Japanese encephalitis is the leading viral cause of encephalitis in Southeastern Asian countries. The disease primarily affects children under the age of fifteen years. Over the past 60 years, it has been estimated that JE has infected ~10 million children globally, killing 3 million and causing long-term disability in 4 million. The JE virus (JEV) is a member of the genus Flaviviridae having single stranded RNA genome of about 11 kb. Genome sequence analysis studies, co-circulation of various genotypes of JEV have been documented in different geographic areas. In India, circulation of different genetic variants of genotype III have been documented in various parts of the country and recently introduction of a genotype I strains has been reported. JEV is transmitted by the *Culex* mosquitoes particularly of the *Culex vishnui* group (*Cx. tritaeneorhynchus*). Water birds and pigs play a major role as amplifying hosts. Humans get infected following a bite by an infected mosquito. However, as human are dead end hosts, further spread from human to human does not take place.

The first evidence of JEV transmission in India dates back to 1952 in the Nagpur subdivision of Maharashtra. JE was clinically diagnosed for the first time in 1955 at Vellore in the North Arcot district of Tamil Nadu. In subsequent years, outbreaks have occurred in various parts of the country. The first major JE epidemic was reported from the Burdwan and Bankura districts of West Bengal in 1973 followed by another outbreak in 1976. Outbreaks have been reported from states like Uttar Pradesh, West Bengal, Assam, Andhra Pradesh, Karnataka, Bihar, Tamil Nadu, Haryana and other states through the years. The Case Fatality Rate (CFR) due to AES/ JE in India has been around 17% with wide variations in states. Annual reported cases due to JE range between 1714 and 6727 while deaths due to JE range between 367 and 1684.

Based on the samples referred to various ICMR laboratories from hospitals and public health laboratories from various states, intramural research activities carried out by various ICMR institutes and data generated by NVBDCP on JE, indicates Uttar Pradesh has shown highest JEV

activity followed by Assam, West Bengal, Andhra Pradesh and Tamil Nadu. In contrast Punjab state was free of any JE cases from 2008 to 2012. Continuous but low activity was noted in Goa, Haryana, Jharkhand (except 2011), Karnataka, Kerala, Maharashtra, Manipur and Nagaland. From Bihar no reports of JE activity from the year 2008-2010, but a spur of activity was noted in 2011 with some death cases; which came down in 2012 where no deaths were reported. Overall it appears that there was increase in number of JE cases during the year 2011 during the span of 2008-2012. Number of deaths due to JE cases were 2008 (70/427; 16.39%), 2009 (103/653; 15.77%), 2010 (112/555; 20.18%), 2011 (181/1214; 14.90%), and 2012 (136/726; 19.0%). National Institute of Virology, Pune along with other ICMR centers CRME, Madurai, VCRC Puducherry, RMRC, Dibrugarh, *etc*. investigated all the outbreaks occurring in various parts of the country and also monitoring JEV activities through routine entomological and human sero-surveys to determine the JEV endemicity, epidemiology, prevalence and incidence in various parts of India. These studies suggested the circulation of different genetic variants of genotype III strains in different parts of the country and identified the introduction of genotype I strains.

In 2005, Uttar Pradesh faced a devastating epidemic outbreak of JE mostly confined to Gorakhpur affecting 6061 cases with 1500 deaths followed by another outbreak in 2006 with 2320 cases and 528 deaths. Similarly JE cases in Uttar Pradesh were confined predominantly in Gorakhpur during 2007 reporting 3024 cases and 645 deaths. NIV conducted the clinical, epidemiological and entomological investigation in the Gorakhpur division from 2005 onwards. All the human clinical specimens obtained from the pediatric patients were tested for JEV-IgM antibodies by using MAC ELISA kit and RT-PCR. These findings indicate broadly about 18-20% JE positivity among the hospitalized patients from various parts of the country that varies from year to year.

2. Major achievements with leads emerged during XI Plan

NIV

- An indigenous Tissue culture derived, inactivated Japanese encephalitis vaccine candidate was developed and transferred to Bharat Biotech, Hyderabad. The vaccine is undergoing clinical trials.
- NIV's JE kit was found to be as good as or better than the commercial kits available as certified by the CDC, USA. 504 JE kits supplied for the national program.
- The protective efficacy of the live attenuated JE vaccine SA14-14-2 in healthy adults determined. The vaccine was found safe and immunogenic in healthy adults.

VCRC Puducherry

- DEPA - a synthetic insect repellent.

- Sperifix - a controlled release formulation of *Bacillus sphaericus* for mosquito larval control, assigned to NRDC.
- Deltafix - a controlled release formulation of *Bacillus thuringiensis* var. *israelensis* for mosquito larval control, assigned to NRDC.
- F24 - a mosquito oviposition attractant assigned to NRDC.
- B426 - a mosquito pupicide assigned to NRDC. (6) Controlled release formulation of DPE-28 an IGR for mosquito larval control assigned to NRDC.

CRME, Madurai

- Detection of JE virus antigen in desiccated vector mosquitoes by IFA / ELISA and JEV genome by RT-PCR was established and technology transferred to Department of Public Health, Tamil Nadu.

RMRC, Dibrugarh

- Developed an early warning system for Japanese encephalitis in Dibrugarh district.

3. No. of Publications: 15

4. List of patents

- **NIV, Pune:** Chimeric T helper-B cell peptide as a vaccine for Flaviviruses. WO 02/053182 A1.
- **VCRC, Puducherry:** (1) A mosquito larvicidal preparation of *Bacillus thuringiensis* var *israelensis* Indian Patent Appl. No. 4.22.1.1/94055. (2) A Process for the preparation and formulation of a mosquito larvicide from *Bacillus sphaericus*, Indian Patent Appl.No. 4.22.1.2/94056. (3) Process for the preparation of a controlled release agent for controlling growth of the insecticides. Indian Patent No. 191820 dt. 11.01.2005. (4) F24 - A process for the production of mosquito pupicidal compound. Indian Patent no.199635 dt.22.9.2006. (5) B426 - A process for the production of mosquito oviposition attractant. Indian Patent no.192872 dt.7.4.2006.
- **CRME, Madurai:** An Indian Patent on the invention of "Mechanical Mosquito Sampler", in 2005.

5. Technologies developed/technologies transferred to the industry

Tissue culture derived, inactivated Japanese encephalitis vaccine by NIV: An indigenous JE vaccine candidate strain was developed and transferred to Bharat Biotech India Ltd (BBIL). The formalin-inactivated, Vero cell derived vaccine strain was studied for genetic stability and immunogenicity in lab animals and human subjects. Sera collected from vaccinees enrolled in the Phase I clinical trial after injecting 2-3 doses neutralized all the JEV strains (n=11 tested) co-circulating in India. Phase I, II and III vaccine trials completed using two/three vaccine doses. The

vaccine was safe after one or two doses of vaccination, well tolerated and immunogenic in healthy volunteers in the age group between ≤50 to ≥1. These studies confirmed effectiveness of a single dose of JE vaccine for the immunization (~95% sero-protection and sero-conversion after a single dose) and a two dose schedule can be used for routine immunization (~97% sero-protection and sero-conversion after two doses).

Techniques developed by NIV for research and Industrial purpose: (1) Developed an antigen capture ELISA test of JE. (2) Development and standardization of Qualitative and Quantitative RT-PCR/PCR assay for detection of JEV from clinical specimens and mosquitoes. (3) Developed and validated plaque reduction neutralization test (PRNT) for detection of neutralizing antibody against JEV. (4) Developed a tissue culture derived formalin inactivated JE vaccine and handed over to BBIL for clinical trials and product development.

Techniques/products developed by VCRC Puducherry: (1) DEPA - a synthetic insect repellent. (2) Sperifix - a controlled release formulation of *Bacillus sphaericus* for mosquito larval control, assigned to NRDC. (3) Deltafix - a controlled release formulation of *Bacillus thuringiensis* var. *israelensis* for mosquito larval control, assigned to NRDC. (4) F24 - a mosquito oviposition attractant assigned to NRDC. (5) B426 - a mosquito pupicide assigned to NRDC. (6) Controlled release formulation of DPE-28 an IGR for mosquito larval control assigned to NRDC. (7) IIFT - an insecticide impregnated fabric trap for sampling filariasis vector.

6. Techniques Developed by (ICMR institutes) and used by WHO/Others

- Molecular and serological techniques (JE-MAC ELISA) for diagnosis of JE infection (NIV, Pune).
- Plaque reduction neutralization test (PRNT) for detection of neutralizing antibody against JEV (NIV, Pune).

7. Manpower Trained:

The encephalitis group yearly trains 5 – 6 students from national / international origin for their degree programs. In depth training is given to staff and trainers from different hospitals, research organizations from India and other southeastern countries on JEV MAC ELISA, RT-PCR and diagnostic reagent preparations for JEV and other encephalitic viruses. Additionally, these technologies along with tissue culture, hybridoma, virus neutralization, *etc*. were transferred to different ICMR and non-ICMR research organizations, hospitals and industries.

Table 1. Status of Completed Research Studies undertaken during XI Plan					
Sr. No.	**Thematic area and Title of the Study**	**Objectives**	**Completed with outcome of the study**	**If off-shoot, Refer to XII th plan study**	**Institution**
Basic Research					
1	Understanding pathogenesis of JE virus	To understand pathogenesis and immunoprotective mechanism against various genotypes of JEV virus	Age dependent susceptibility was documented. The protective role CD4 T cells were observed in animal studies. The objectives of proposed study were completed.	To understand disease progression and predicting possible severity of disease with emergency of newer genotypes	NIV, Pune
2	Role of innate immune response in Japanese encephalitis virus infection	To understand the role of various components innate immune system in JEV infection	Different study models were established using cells of human origin	This is necessary to understand the immunological components responsible for the disease progression	NIV, Pune
3	To study the selective expression of Japanese encephalitis envelope protein under different promoters	To study the expression of JEV envelope protein under different promoters to understand its expression levels under different antigen presenting cells	Increase in the humoral and cell mediated immune response was observed under the macrophage specific promoter.	The study is completed	NIV, Pune
4	In-Depth evaluation of JE for NVBDCP	This study was undertaken to understand the ground situation to make progress towards achieving the control of JE.	**Completed.** NVBDCP wanted to achieve the complete roll back of the vector Borne Diseases which are of the major public health concern in India. This study helped to understand the ground situation to make progress towards achieving the targets in Tamil Nadu, AP, UP and Assam	Report submitted to NVBDCP.	CRME, Madurai

5	Silent JE transmission study in Tamil Nadu	To investigate the role of silent JEV transmission in Thanjavur district a non endemic zone Tamil Nadu and its implications of impending JE outbreak in future	ongoing	New research work initiated in the Tanjore district To understand the JEV activity in the pediatric populations	CRME, Madurai
6	Evaluation of Chinese JE vaccination programme in Tamil Nadu	To understand the vaccine coverage in Tamil Nadu	completed	Report submitted to DPH Tamil Nadu	CRME, Madurai
7	Establishment of Field Station in South Arcot District to study Japanese encephalitis epidemiology and control	To predict the JE epidemic in advance and to develop vector control strategies	Most JE studies cumulatively culminated in developing a JEV surveillance system which was handed over to Directorate, Public Health, Tamil Nadu on 25th July, 2010.	Technology was transferred to the Directorate, Public Health, Tamil Nadu on 18th Oct., 2012, to help taxonomic studies and surveillance mechanism. More studies are planned as follows: Determination of critical density of vector of JE. Control of JE vector using neem-coated urea.	CRME, Madurai This is a long term project funded by ICMR from extramural grant.
8	Risk assessment of JE virus emergence in two paddy growing districts of Rajasthan state (Sri Ganganagar and Hanumangarh)	To assess potential emergence and public health risk of JE virus in Sri Ganganagar and Hanumangarh district by carrying out mosquitoes, vertebrate hosts and human surveillance.	Study Completed. It revealed the presence of 6 mosquito vectors of JE, including *Cx. tritaeniorhynchus* with maximum PMHD in August & September month. Prevalence of JE specific IgM antibodies in humans, neutralizing antibodies in pigs and high density of *Cx. tritaeniorhynchus* during paddy season indicates that there is a risk of emergence of JE virus in Sri- Ganganagar and Hanumangarh district	Based on the finding a new project entitled 'Mapping of mosquito breeding habitats and location of vertebrate hosts in North and Southern parts of Rajasthan state prone for emergence of JE virus using space technology (RS &GIS)' has been proposed in 12th plan.	DMRC, Jodhpur

9	Epidemiology and immune response against Japanese encephalitis virus strains at molecular level in North Eastern region of India	To establish the genotypes of JE virus circulating in NE region of India and determine the divergence in nucleotide sequences in genomic region encoding the envelope protein of JE virus isolates from human and mosquitoes.	Genomic characterization of newly isolated JEC strains show circulation of genetically distinct JEV strains in India. Three different strains (GP78-GKP, 1978; 014178-Lakhimpur,2001 and 9117857-GKP, 1991) and their variants have been isolated from UP. Two genetically distinct strains (7812474 and P-20778 like) have been isolated.	No	Extramural grant, ICMR Hqrs
Clinical Research					
1	Improvement of JEV diagnostic assays	Early diagnosis for better clinical management of cases	The JEV diagnostic ELISA and molecular assays have been developed. However, with the emergence of newer genetic variants of JEV, time to time improvements will be necessary.	Early diagnosis for better clinical management of cases	NIV, Pune
2	Evaluation of efficacy of JE virus live, attenuated and killed vaccine	This is mandatory for assessing health ministries' ongoing vaccination program	The JE vaccine SA14-14-2 clinical trials were carried out and reports submitted to ministry	This is mandatory for assessing health ministries' ongoing vaccination program	NIV, Pune
3	Role of innate immune response in Japanese encephalitis virus infection	To understand the role of various components innate immune system in JEV infection	Different study models were established using cells of human origin.	This is necessary to understand the immunological components responsible for the disease progression	NIV, Pune

4	Treatment of Japanese encephalitis: A double blind placebo controlled clinical trial	To study the efficacy of the antiviral agent Ribavirin in reducing mortality, morbidity and sequelae in children with a laboratory confirmed diagnosis of JE.	The study showed that oral ribavirin in the dose used was safe and did not affect the outcome of children with JE or the secondary outcome measures –days to become afebrile, days to start oral feeds and days in hospital.	No	Extramural grant , ICMR Hqrs
Epidemiological/Operational Research					
1	Molecular epidemiology of JEV	Would be main supportive tool in early detection of emergency of newer genotypes	The evolutionary studies on JEV isolated from 1956 onwards suggest multiple introductions of JEV GIII and GI strains in India.	Would provide support to health ministries' ongoing vaccination program	NIV, Pune
2	Epidemiological and entomological studies on JE and WN virus in various parts of the country	Required as proactive research, these studies will directly and indirectly support all the activities listed below	Long term. Till date, the data on prevalence and distribution of various flaviviruses in different geographic regions of India were mapped.	Would strengthen surveillance network and operational research for improvement of JE control strategies and will also generate the baseline data for further control strategies	NIV, Pune
3.	Application of Heuristic Engine: A novel forecasting software tool to predict the outbreak of Japanese encephalitis in Andhra Pradesh.	The specific objectives are to collect entomological and serological data associated with Japanese encephalitis (JE) from various epidemic prone districts of Andhra Pradesh to develop a database for Heuristic Engine.	A novel forecasting software tool to predict the outbreak of Japanese encephalitis in Andhra Pradesh has been developed and field tested at a small scale.	No	Extramural grant, ICMR Hqrs

Translational Research/Techniques Developed					
1	Tissue culture derived, inactivated Japanese encephalitis vaccine	An indigenous JEV vaccine candidate was developed and transferred to Bharat Biotech Hyderabad	This will be used for vaccination in India after the approvals	The vaccine is undergoing clinical trials	NIV, Pune and Bharat Biotech Hyderabad
2	Scaling up of production of diagnostic reagents.	To provide sufficient diagnostic reagents to public health and other laboratories involved in of JE surveillance network for better clinical management of cases	NIV JE kit was found to be as good as or better than the commercial kits available as certified by the CDC, USA. 504 JE kits supplied for the national program.	Continue	NIV, Pune
3	A prospective open-label, uncontrolled, single-centre, evaluation of viraemia in healthy flavivirus seronegative adults after primary vaccination with JE SA14-14-2 live attenuated vaccine	This study was undertaken to determine the protective efficacy of the live attenuated JE vaccine SA14-14-2 in healthy adults.	The vaccine was found safe and immunogenic in healthy adults.	Completed	A collaborative study at NIV, Pune
4	Post marketing study to evaluate the safety and immunogenicity of single dose of JE vaccine Immunogenicity at Burdwan West Bengal	This study was undertaken to study the persistence of anti-JEV neutralizing antibodies against the live attenuated vaccine SA14-14-2	The neutralizing antibody response was observed up to six months of vaccination which was gradually declined thereafter.	Completed	NIV, Pune

Table 2. Important and essential activities which need to be continued in XIIth Plan					
Sr. No.	**Thematic area and title of the study**	**Justification**	**Time frame**	**Deliverable outcome with public impact**	**Institution**
Basic Research					
1	Optimization of new generation sequencing platform for novel virus discovery	Required for early detection of emergency of newer genotypes in an area which may pose problem to current vaccine program	Five years	Would help in formulating better control strategy in the case if any of the control tool become less effective	NIV, Pune
2	Risk assessment of JE virus emergence in two paddy growing districts of Rajasthan state (Sri Ganganagar and Hanumangarh)	To assess potential emergence and public health risk of JE virus in Sri Ganganagar and Hanumangarh district by carrying out mosquitoes, vertebrate hosts and human surveillance. Study Completed. It revealed the presence of 6 mosquito vectors of JE, including *Cx. tritaeniorhynchus* with maximum PMHD in August & September month. Prevalence of JE specific IgM antibodies in humans, neutralizing antibodies in pigs and high density of *Cx. tritaeniorhynchus* during paddy season indicates that there is a risk of emergence of JE virus in Sri - Ganganagar and Hanumangarh district.	Five Years	Based on the finding a new project entitled 'Mapping of mosquito breeding habitats and location of vertebrate hosts in North and Southern parts of Rajasthan state prone for emergence of JE virus using space technology (RS &GIS)' has been proposed in 12th plan.	DMRC, Jodhpur
Clinical Research					
1	Improvement of JEV diagnostic assays	Early diagnosis for better clinical management of cases	Five years	Early diagnosis for better clinical management of cases	NIV, Pune
2	Scaling up of production of diagnostic reagents	To provide sufficient diagnostic reagents to public health and other laboratories involved in JE surveillance network for better clinical management of cases	Five years	Early diagnosis for better clinical management of cases	NIV, Pune

3	Understanding pathogenesis of JE virus	To understand pathogenesis caused by various genotypes of virus	Five years	To understand disease progression and predicting possible severity of disease with emergency of newer genotypes	NIV, Pune
4	Immunological studies on understanding the severity and disease progression	To understand selective expression of JEV envelope proteins in Antigen Presenting Cells	Five years	Would help in understanding immunological components responsible for disease progression which would provide lead for development of antiviral drugs	NIV, Pune
5	Evaluation of efficacy of JE virus live, attenuated and killed vaccine	This is mandatory for assessing health ministries' ongoing vaccination program	Five years	This is mandatory for assessing health ministries' ongoing vaccination program	NIV, Pune
Epidemiological/Operational Research					
1	Molecular epidemiology of JEV	Would be main supportive tool in early detection of emergency of newer genotypes	Five years	Would provide support to health ministries' ongoing vaccination program	NIV, Pune
2	Epidemiological and entomological studies on JE and WN virus in various parts of the country	Required as proactive research, these studies will directly and indirectly support all the activities listed below	Long term	Would strengthen surveillance network and operational research for improvement of JE control strategies	NIV, Pune
3	Development of RS-GIS based model for forecasting JE vector abundance and transmission risk	Early warning system is essential for the programme to plan and implement timely interventions. During the first two years, field data on vectors and environmental conditions including meteorological data are collected. Correlation analysis with the RS data for all seasons will have to be completed and therefore the study needs to be continued and will be completed as originally planned.	2011 -2014	Deliverables: A prediction model for delimitation and monitoring of JE risk areas based on vector abundance and transmission risk for the National programme.	VCRC and CRME in collaboration with ISRO, Bangalore and State Health Dept, Tamil Nadu & Karnataka

4	Establishment of JE surveillance network	For early detection of out breaks	Five years	Would help in saving many human lives	NIV, Pune
5	Establishment of monitoring system for early warning	For early detection of out breaks	Five years	Would help in saving many human lives	NIV, Pune
6	Epidemiology and dynamics of transmission of JE/AES in Eastern part of UP and implement appropriate interventions	Required for developing better tools for effectively breaking the transmission chain	Five years	Would help in controlling epidemics	NIV Field Unit, Gorakhpur , VCRC,Puducherry, CRME, Madurai
7	Sero-prevalence of Japanese encephalitis in Alappuzha district, Kerala	Required to understand the systematic prevalence of JEV in Alappuzha district, Kerala	Five years	Would help to understand the JEV activities in Kerala	NIV Field Unit, Allepy, Kerala
9	Aetiological and epidemiological investigations on arboviral infections and laboratory diagnosis in Karnataka and neighbouring areas with particular reference to JE, DEN and CHIK viruses	To provide early diagnosis to differentiate JEV etiology.	Five years	Would help in saving many human lives	NIV Field Unit, Bangalore, Karnataka
10	In-depth evaluation of JE for NVBDCP	This study was undertaken to understand the ground situation to make progress towards achieving the control of JE.		Completed. NVBDCP wanted to achieve the complete roll back of the vector Borne Diseases which are of the major public health concern in India. This study helped to understand the ground situation to make progress towards achieving the targets in Tamil Nadu, AP, UP and Assam	CRME, Madurai
11	Evaluation of Chinese JE vaccination programme in Tamil Nadu	To understand the vaccine coverage in Tamil Nadu	completed	Report submitted to DPH Tamil Nadu	CRME, Madurai
12	Evaluation of Larvicidal Effect of Actinobacterial Metabolites for Biological Control of Culex Mosquito	To isolate the Acinobacteria from fresh water, to characterize it, to extract the larvicidal compound from it, to evaluate the larvicidal compound for larvicidal activity, to determine the repellency in larvicidal compounds and to evaluate the larvicidal property of potential acinobacterial isolates in experimental aquatic tanks.	Two years	The larvicidal compound of the Acinobacteria will be isolated and tested for its repellency. Also the larvicidal compound will tested for its larvicidal activity against Culex mosquito by larvicidal bio-assays. Also the larvicidal property of acinobacteer will be determined in experimental aquatic tanks	Supported in extramural research mode (Bharathidasan University Triuchirapalli)

13	Mapping of mosquito breeding habitats and location of verterate hosts in North and Southern parts of Rajasthan state prone for emergence of JE virus using space technology (Rs & GIS)	Mapping of mosquito larval habitats, particularly *Cx. tritaeniorhynchus* and *Cx.vishnui* subgroup of mosquitoes and location of vertebrate host using RS and GIS technologies, to identify JE prone areas and to develop early warning systems	Three years	Assesment of potential larval habitats of mosquito vectors of JE and location of vertebrate host in Northern and Southern parts of Rajasthan prone for emergence of JE virus using space technology	Supported in extramural research mode (DMRC, Jodhpur)
Translational Research/Techniques Developed					
1	Field evaluation of Neem (*Azadirachta indica*) leaves and neem cake for the control of Japanese encephalitis vectors in rice agro-ecosystem.	Looking for newer indices for larval control	Five years	These herbal products will be good for safety of environment.	CRME, Madurai
Technologies Developed and Transferred to Industry					
1	Tissue culture derived, inactivated Japanese encephalitis vaccine	An indigenous JEV vaccine candidate was developed and transferred to Bharat Biotech, Hyderabad		The vaccine is undergoing clinical trials	NIV, Pune and Bharat Biotech, Hyderabad
2	1. Sperifix - a controlled release formulation of Bacillus sphaericus for mosquito larval control. 2. Deltafix - a controlled release formulation of Bacillus thuringiensis var. israelensis for mosquito larval control, 3. F24 - a mosquito oviposition attractant 4. B426 - a mosquito pupicide 5. Controlled release formulation of DPE-28 an IGR for mosquito larval control 6. IIFT - an insecticide impregnated fabric trap for sampling filariasis vector.			Assigned to NRDC.	VCRC Puducherry
3	Detection of JE virus antigen in desiccated vector mosquitoes was established.			Transferred to DPH, TN	CRME Madurai
4	Multiangular viewer for JE vector identification			Transferred to DPH and PM, TN	CRME Madurai

Table 3. New proposals to be undertaken during 12th Plan						
Sr. No.	**Thematic area and Title of the study**	**Justification**				
		Off-Shoot of an earlier completed programme	**De Novo idea which is either nationally relevant or it is likely to lead to a new scientific breakthrough.**	**Time frame**	**Deliverables**	**Institution**
Basic Research						
1	Rapid detection of Japanese encephalitis using improved new/ molecular tests	It is observed sensitive tool to detect JE in short duration of the laboratory test compared other existing tests.	It is nationally relevant .	Five years	Early diagnosis for better clinical management of cases	CRME, Madurai
2	Development of a multiplex real time PCR method for the detection of flaviruses (Japanese encephalitis, West Nile and Dengue) and Alpha virus (Chikungunya) in the vector mosquitoes.	Quicker test to differentiate JE from other viruses. Time and reagents can be saved.	Single test would differentiate mentioned viruses. Nationally relevant.	Five years	Early diagnosis for better clinical management of cases	CRME, Madurai
3	Rapid detection of Japanese encephalitis virus in desiccated vector	A newer tools for surveillance of JEV	Nationally relevant	Five years	Early diagnosis for better clinical management of cases	CRME, Madurai
4	Screening of antiviral compounds from different sources	Antivirals against JE may add new dimensions to the research. Some compounds would lead an effective measure.	The origin of this program is based on a need-of-hour requirement is developing an integrated platform using combinatorial approaches, screening and viability testing of potential antiviral compounds of synthetic as well as indigenous origin. This will be of National importance and a multi-institutional collaboration with relevant stakeholders will be set up. The program will be in several operational phases. The major focus will be on medically important viruses of public health importance in India.	Five years	Antiviral agent against JEV	NIV, Pune

5	Role of host and viral factors in different vector borne viral diseases	Needs to understand host and viral factors contributing to the disease	This project is a de novo proposal that will strengthen the knowledge base in understanding in detail vector biology component of arboviruses.	Five years	Basic knowledge for JE virologist	NIV, Pune
6	Role of possible zoonosis in spread of enteroviruses responsible for Acute Encephalitis Syndrome (AES)	AES remained as the complicated issue , need to understand the role of JEV	The etiology of a significant number of AES cases from various parts of India especially Gorakhpur remains incompletely understood. This is a crucial project of National importance as AES has been identified to be a major public health issue. The possible role of zoonotic involvement will be the key focus and findings from this study will be important in further identifying the disease ecobiology of AES.	Five years	JEV contribution to the AES	NIV, Pune
7	Mapping of mosquito breeding habitats and location of vertebrate hosts in North and Southern parts of Rajasthan state prone for emergence of JE virus using space technology (RS &GIS):	Offshoot of earlier study carried out on Japanese encephalitis (JE) in Sri-Ganganagar & Hanumangarh district	During the proposed study, breeding habitats of JE vectors & locations of vertebrate hosts involved in JE epidemiology will be mapped with the help of RS & GIS technology, which may help in forecasting JE outbreak at an early stage in JE prone areas of Rajasthan state. Nationally relevant.	Five years	Forecasting of JE outbreak at an early stage in JE prone areas of Rajasthan state.	DMRC, Jodhpur
8	Silent JE transmission study in Tamil Nadu	Would generate data on silent JE transmission	To investigate the role of silent JEV transmissions in Thanjavur district a non endemic zone Tamil Nadu and its implications of impending JE outbreak in future. Nationally relevant.	Five years	JEV activity in the pediatric populations of Thanjavur district.	CRME, Madurai
9	Establishment of Field Station in South Arcot District to study Japanese encephalitis epidemiology and control	To predict the JE epidemic in advance and to develop vector control strategies.	Most JE studies cumulatively culminated in developing a JEV surveillance system which was handed over to Directorate, Public Health, Tamil Nadu on 25th July, 2010. Nationally relevant	Five years	Generation of Basic data on JEV epidemiology & Control in South Arcot district.	CRME, Madurai

10	Encapsulation of neem oil nanoemulsion laced with urea - A novel nanotechnology to control Japanese encephalitis vector, *Culex tritaeniorhynchus*, larval population in paddy ecosystem of India.	Collaborative research project by VIT, Vellore and CRME to be funded by VSF-ICMR. To synthesize and characterize neem-based nano-emulsion by choosing suitable biodegradable bio-polymers for encapsulation.	Indigenous technology of National relevance	2015	Neem based nanomaterials with biodegradable polymer coatings for sustained release in vast stretches of water bodies breeding major JE vector, *Culex tritaeniorhynchus*.	CRME, Madurai
11	Vector *(Culex tritaeniorhynchus)* - based virological surveillance involving Japanese encephalitis.	To conduct laboratory studies to demonstrate susceptibility or otherwise of the vector mosquito to the "live attenuated vaccine" virus strain (SA-14-14-2). To determine if the "live attenuated vaccine" virus ingested by the vector mosquito is able to multiply (recombination) and mutate in its body.	National relevance	2015	This will bring out the level of susceptibility of the vaccine-virus in the vector mosquitoes.	CRME, Madurai
Clinical Research						
1	Studies on immunological correlates of morbidity, severity, and multiorgan involvement in AES cases.	To understand immunological aspects in pathobiology of JEV.	A component of AES umbrella program that will provide crucial links towards understanding the disease pathogenesis and markers. New knowledge may generate.	Five years	Basic knowledge in understanding immunological components responsible for disease progression	NIV, Pune
2	Pathophysiology of AES and development of management protocols.	Cellular & molecular abnormalities due JE	This is a direct clinical project that will help physicians get a better treatment protocol defined for immediate bedside intervention. Nationally relevant	Five years		NIV, Pune

3	Role of innate immune response in Japanese encephalitis virus infection in humans.	To understand the role of innate immune response and cytokine levels in JEV infected patients.	New knowledge may generate in Indian context.	Five Years	Would help in understanding immunological components responsible for disease progression which would provide lead for development of antiviral drugs	NIV, Pune
4	Studies in AES in Gorakhpur (U.P).	Ongoing programme due to endemicity of AES.	AES has been recognized as a major public health problem and challenge in India. This is a key project that is integrated with other stand-alone and/de-novo proposals for studying different aspects of AES. A very high-priority Project of National importance.	Five years	Contribution of JEV in AES	NIV, Pune
Epidemiological/Operational Research						
1	Surveillance of different viral diseases in Kerala state to determine the dynamics of transition.	To understand JEV status in Kerala	Kerala has multiple vector borne disease. This proposal strengthens the mandate for a separate NIV Kerala to study multidisciplinary aspects of these infections with respect to public health burden.	Five years	Would strengthen surveillance network and operational research for improvement of JE control strategies	NIV, Pune
2	Development of Predictive algorithms for AES using remote sensing and ground level data collection in eastern UP	JEV prediction in eastern UP	An integrated eco-geographical framework development for data collection and analysis to understand better the environmental impact/ factors that might be associated with AES	Five years	Early prediction of JEV in eastern UP	NIV, Pune

3	Environmental monitoring of etiological agents responsible for causation of AES in Eastern Uttar Pradesh	Environmental contribution in JEV	Part of the AES umbrella program that focuses on the possible role of environmental factors in the etiology of AES.	Five years	Role of different environmental factors	NIV, Pune
4	GIS based data acquisition and software development portals for rapid reporting and recording of AES cases	Rapid recording and reporting of cases would saves the lives by treating the patients	A de novo proposal but part of the applied eco-geographical approach studies under the AES umbrella program.	Five years	Rapid recording & reporting system	NIV, Pune
5	Molecular diagnosis and epidemiology of arbovirus infections in Karnataka.	JEV occurrence in Karnataka state – ongoing activity	This is a routine public health diagnostic service activity.	Five years	Differential diagnosis	NIV, Pune
6	Validation of early warning system for occurrence of Japanese encephalitis in Assam	Required as proactive research.	National relevance	Five years	Increase the confidence level of Early warning system of JE in Assam.	RMRC, Dibrugarh
7	Molecular identification of dengue and Japanese encephalitis virus vectors using genomic DNA.	Early diagnosis for better clinical management of cases	National relevance	Five years	Early detection system of JEV in Vectors	RMRC, Dibrugarh
8	The monitoring of JE virus activity in Tanjore & Tirunelveli zone, southern India, Development of a pictorial taxonomic key for dengue / Chikungunya vectors (Aedes mosquitoes) for field application.	Improvement of tools for correct identification of the vectors	National relevance	Five years	Tools for correct identification of JE vectors	RMRC, Dibrugarh
9	JE surveillance Network in Tamil Nadu	For early detection of outbreaks	National relevance	Five years	Data on JEV activity	CRME, Madurai
10	Vector and epidemiological survey for AES in Odisha	Contribution of JEV in AES in Odisha	National relevance	Five years	Would help in saving many human lives	RMRC, Bhubaneswar

11	Community based control of malaria and JE vectors in agricultural irrigating paddy fields using neem cake coated urea (NCU)	For the control of the Immature stages of JE vectors breeding in the paddy fields	Collaborative project with Agricultural college Madurai and CRME. CRME has earlier demonstrated on small scale the benefits of neem-coated urea.	Five years	Large scale field trial will bring out a better usage of the technique.	CRME, Madurai & Agriculture College, Madurai.
12	Task Force project on Insecticide resistance monitoring in culex vector under Vector Science Forum	Not off shoot	National relevance	Five years	Resistance to insecticides is to be monitored in vectors of dengue/ chikungunya, JE, filariasis and cutaneous leishmaniasis (wherever possible), in addition to those of malaria and visceral leishmaniasis.	CRME, Madurai NIV, Gorakhpur unit

4. Status of projects funded by other than ICMR							
Sr. No.	**Thematic Area and title of the Study**	**Objectives**	**Completed/ongoing with outcome of the study**	**Justification for continuation**	**Time frame**	**Institution**	**Source of Funding**
Basic Research							
1	Post marketing study to evaluate the safety and immunogenicity of single dose of JE vaccine Immunogenicity at Burdwan West Bengal.	This study was undertaken to study the persistence of anti-JEV neutralizing antibodies against the live attenuated vaccine SA14-14-2	Completed. The neutralizing antibody response was observed up to six months of vaccination which was gradually declined thereafter.			NIV, Pune	PATH
2	A prospective open-label, uncontrolled, single-centre, evaluation of viraemia in healthy flavivirus seronegative adults after primary vaccination with JE SA14-14-2 live attenuated vaccine	This study was undertaken to determine the protective efficacy of the live attenuated JE vaccine SA14-14-2 in healthy adults.	Completed. The vaccine was found safe and immunogenic in healthy adults.			A collaborative study between NIV Pune and KEM Hospital and Research Center, Pune	PATH
3	Susceptibility of *Culex tritaeniorhynchus* (Diptera: Culicidae) to Japanese encephalitis virus in Cuddalore district, Tamil Nadu.	The main objective was to find out the temporal changes, if any in vectorial capacity of *Cx. tritaeniorhynchus* to transmit Japanese encephalitis virus.	Completed. Understood the seasonality of virus infection in mosquitoes which is due to temporal changes in the susceptibility of vectors to virus.	completed	-	CRME, Madurai	DST
4	Population genetics of the predominant Japanese encephalitis vector *Culex tritaeniorhynchus*	To determine the genetic structure of *Culex tritaeniorhynchus* by specific enzyme markers and to determine the distinct genetic variation among geographic populations of this mosquitoes.	Completed. Genetic variability -Mean heterozygosity due to variations in the enzyme phenotypes measured genetic variation based on the detection of differences in migration of enzyme phenotype.	completed	-	CRME, Madurai	DST

5	Morphological and molecular characterization of the *Culex vishnui* subgroup mosquitoes with reference to biological and behavioural variations in JE vector *Culex tritaeniorhynchus*	To determine the inter specific and intra specific variations in *Culex vishnui* subgroup mosquitoes. To study the phylogenetic relationship of the members of the subgroup. To develop a molecular tool for the identification of different taxa	Highly variable region of rDNA among 60 specimens of *Cx. tritaeniorhynchus* collected from different study sites was analyzed to elucidate genetic variability of the species. The analysis showed the rDNA region of this species to be highly conserved with a genetic distance (K2P) of only 0.0014 among the specimens analyzed, indicating that the specimens from different sites belong to the same taxonomic clade	Project ended in 2011	2009-2011	VCRC, Puducherry	DST
6	Preparation of JE virus antigen in cell culture	To scale up antigen production of Indian isolates of JEV in established cell lines and to purify and evaluation of cell culture derived antigen.	Completed. Cell culture antigens were prepared and used in the screening of JEV antibodies in human samples. Validation by other institutes like NIV needs to be done before transfer of this technology to others.	completed		CRME, Madurai	DST
7	Genotypic characterization of Japanese encephalitis virus (JEV) in different- endemic areas of India and Vietnam: its implications on the vector-pathogen interactions.	1. To characterize Japanese encephalitis virus (JEV) from both humans and vector mosquitoes in India and Vietnam. 2. To determine phylogenetic relatedness amongst various genotypes variants.	Ongoing to find out the existing genotypes of JEV in India	New genotype discovery will help in developing new diagnostics and for vaccine production	2015	CRME, Madurai	DST