

Preface

The Desert Medicine Research Centre continued its endeavours in addressing the spectrum of Communicable Diseases, Non-communicable Diseases and Nutrition. The completion of 25 years of existence of DMRC coincided with the completion of the new laboratory cum administrative building of the centre and addition of the guest house at the centre. We were honoured to have to Dr. V. M. Katoch, the Secretary, Department of Health Research, Ministry of Health & FW and the Director-General, ICMR to join us at the function of celebration of silver jubilee of DMRC and to inaugurate the construction of new guest house.

Like preceding years, our major thrust this year also was focussed on communicable diseases such as Dengue, Malaria and Tuberculosis. In the area of Dengue, we focussed at basic and translation research aspects of dengue. While the basic research activities on development of molecular and genetic markers of vector competence of mosquitoes and molecular characterization of extrinsic peri-urban viruses in contributing to DHF were continued, a translational research module of prevention/control of dengue, based on our earlier research has also been developed. A malaria epidemic forecasting module has been tested and found useful under simulated and field conditions. In addition, evaluation of efficacy of different ovitraps for surveillance and control of dengue vector *Aedes aegypti*, was made and larvicidal potential of some of the desert plants especially *Solanum xanthocarpum* and *Calotropis procera* were undertaken. Sociological studies to understand the association between socio-economic factors and transmission of desert malaria were also undertaken. In the area of Tuberculosis research, study of developing a rapid culture for *Mycobacterium tuberculosis* from sputum samples were concluded with the observations that time required in testing drug susceptibility of *Mycobacterium tuberculosis* can be substantially reduced by supplementing LJ medium with sheep blood or by using blood agar in place of LJ medium.

In the area of non-communicable diseases (NCD), under a task force project on musculo skeletal diseases, a total population of 11365 in 1842 households in rural settings and 126083 in 4384 households of urban settings was screened. The data showed that 10.2 % of the subjects showed symptoms of the musculo skeletal diseases with almost equal magnitude in the populations of urban and rural settings. Epidemiological studies of Rheumatic Heart diseases (RHD) was another area of research on NCD during reported period. 167 patients of RHD were registered during the period for maintenance on secondary prophylaxis through the existing health system.

Under the continuing project of Nutrition monitoring survey on NNMB pattern, a total population of 3548 subjects were studied during the reported period.

Efforts have been made to improve upon the publications in the journals of better Impact Factor with almost half of publications during the year having published in the journals with IF 1.0 or more.

Having travelled through current year, DMRC is poised to step further towards the goal of focussed basic research, useful translational research targets and for adding advance state of art technology to our capacity strengthening efforts.

Dr. Bela Shah
Director-in-Charge

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1.1 Development and demonstration of a surveillance design for control of dengue vectors in Jodhpur, using GIS - *Vinod Joshi, Himmat Singh, Kirti Sharma, Bennet Angel, Anil Purohit and P. K. Dam*

Commencement: October, 2006

Duration: Two Years

Status: Ongoing

Objectives

1. Development of a surveillance design for dengue vectors as applicable for an arid town, Jodhpur, Rajasthan.
2. Demonstration of monitoring and control of dengue vectors using area specific design and involving local health agency and community participation.
3. Detection of virus maintaining foci in nature as crucial etiological niche contributing to maintenance and transmission of disease in a setting.

Progress of the Work

Dengue Action Plan developed for Jodhpur town (In consultation with CMHO)

A dengue prevention plan was developed and discussed with the Chief Medical & Health Officer, Jodhpur, for prevention/control of dengue and DHF in Jodhpur town. The stratification criteria developed by DMRC and the observations made on existing vectors and virus foci, as detected by the research team of DMRC, were used to demonstrate control of dengue in the town. The plan developed has been given in Annual Report 2007-08.

Demonstration of dengue control operations in Jodhpur town during February-March, 2008

The earlier research undertaken on the development of surveillance design for dengue under TDR, WHO project, was applied to demonstrate dengue control operations in Jodhpur town. The plan developed for Jodhpur town was implemented. The operations carried out, involved following steps:

- Based on our earlier observations of breeding and infectivity of *Aedes* mosquitoes, in all, 6 localities were selected for examining domestic and peri-domestic containers and for undertaking simultaneous larval control operations. Temephos, a larvicide used under the programme by state health department, was used to control the vector breeding.

- Control operations were executed in 1545 houses of 11 selected study localities through staff provided by the CMHO. The crucial sites of vector breeding which may expand and contribute enhanced vector burden during forthcoming rainy season, were identified and rigorously treated (Table 1).
- Post operations studies were made after 15-20 days which showed high mortality/poor viability of larvae in the treated habitats.

Findings

- Based on the observations that dengue virus maintains itself during inter epidemic periods through vertical transmission across mosquito generations, the larval control measures taken during February-March, 2008, showed the remarkable results of complete absence of dengue in July-October, 2008 till date, in Jodhpur town (As per official reports of Chief Medical & Health Officer, Jodhpur).
- DMRC has demonstrated first dengue control operations where elimination of larvae during February-March has resulted to absence of disease during transmission season.

Table 1. Details of sites and containers selected for larval control during February-March, 2008

Intervention date	Areas	Containers																		
		House Ex.	Cement Tanks	Ve+	UG	Ve+	OHT	Ve+	Plastic	Ve+	Clay	Ve+	Metallic	Ve+	coolers	Ve+	other	Ve+	Total	Ve+
21/02/08	Medical College	256	0	0	4	0	12	0	18	0	18	0	0	0	0	0	1	1	53	1
22/02/08	Mauriya Sansi colony	75	28	10	45	13	40	11	21	3	45	13	0	0	0	0	14	3	193	53
23/02/08	Masuriya Bhil colony	130	103	24	65	12	43	2	21	2	147	10	30	2	0	0	23	2	432	54
25/02/08	P. nagar jagdamba Cly	150	45	16	37	7	44	1	14	6	85	10	5	0	0	0	10	3	240	43
26/02/08	P. nagar Ambedkar Cly	140	59	23	60	19	107	3	38	3	257	16	22	1	0	0	35	5	578	70
27/02/08	Partap Nagar	100	36	13	39	8	17	1	19	2	105	9	17	1	0	0	18	3	251	37
28/02/08	Moti Chowk	200	134	21	75	3	98	2	23	5	145	0	13	0	0	0	24	9	512	40
29/02/08	Moti Chowk	120	67	13	23	0	43	1	12	3	135	0	12	1	0	0	12	7	304	25
03/03/08	Navchoukiya	150	72	35	23	2	74	5	23	6	112	1	3	0	0	0	23	2	330	51
04/03/08	Navchoukiya	163	161	32	50	4	76	6	27	3	200	3	7	0	0	0	12	1	533	49
05/03/08	Navchoukiya	61	56	9	5	2	4	1	18	2	85	0	4	0	0	0	19	0	191	14
	Total	1545	761	196	426	70	558	33	234	35	1334	62	113	5	0	0	191	36	3617	437
	Percent		25.7556		16.432		5.91		14.96		4.648		4.425		0		18.8			

1.2 Development of molecular and genetic markers of virus transmission competence of dengue vector species in Rajasthan - Vinod Joshi, Bennet Angel and Rashmi Chauhan

Commencement: October, 2005

Duration: Three Years

Status: Ongoing

Objectives

1. Determinations of mid gut proteins responsible for dengue virus transmission (horizontal and vertical) among mosquito vector species in different ecotypes of Rajasthan.
2. Location of gene controlling the implicated protein and study of frequency of its appearance through gene flow maps.
3. Extrapolation of observations in GIS across different ecotypes of Rajasthan to develop molecular markers of transmission risk of dengue in Rajasthan.

Progress of the Work

Experimental studies to confirm relationship of ovarian proteins of parent mosquitoes with transovarial transmission of dengue virus in F₁ progeny

Blood fed engorged female *Aedes aegypti* and *Aedes vittatus* were collected from dengue endemic localities. The females were allowed to lay the eggs. After egg laying head squash of each female was examined for IFA test to determine presence or absence of virus. Simultaneously, ovaries of the mosquitoes of individual mosquitoes were subjected to SDS-PAGE. The eggs obtained from fed females were immersed into water and adults of F₁ generation were obtained. The adults were then tested for IFA test to determine presence of vertically transmitted virus in them. Details are depicted in Table 1.

- The study of adults developed from eggs laid by the experimental pool showed that in two parent mosquitoes (one with four progeny and another with two progeny) where virus was present in head squash and 200 kDa protein was present in the ovary, in five out of six F₁ individuals of such combination, virus did not appear. In another combination, three parents (one with one progeny, another with four and yet another with three progeny) virus was present in head squash but 200 kDa protein was not present, in seven out of eight F₁ individuals of such combination in parents, virus was present.
- Our experimental studies on ovarian proteins and vertical transmission of virus had clearly shown that when there is a 200 kDa protein in ovary, parents do not pass virus to progeny except for one sample where even 200 kDa protein was present but one of the F₁ progeny had demonstrated vertically transmitted virus.

- The work reported in last report showed that the ovarian 200 kDa protein which did not pass the virus vertically was composed of Glycine as the N-terminal amino acid which is a special amino acid (**Glycine-Proline-Proline-Proline-Glutamine**). The 200 kDa protein sample of ovarian tissue which allowed vertical transmission of virus to progeny was composed of Serine (**Serine-Valine-Tryptophan-X-Alanine**) as its N-terminal amino acid which is a polar amino acid.
- The interpretation emerging from above observations is that 200 kDa protein composed of special terminal amino acid could be the blocking protein for internalization and further replication of dengue virus for mosquito cells. Whereas, same protein if composed of polar terminal amino acid may allow the virus transmission.
- This sequence when analysed for its protein information in the existing database using the BLAST program showed many results, one of which was identified as a transport protein SEC24 of *Culex pipiens quinquefasciatus*. Exploring this protein further predicted its putative orthologue sequence in *Aedes aegypti* as a putative protein SEC24.
- Since the protein with our above sequence worked out belongs to sample negative for virus transmission and as per the search we made, this is in the category of transporter proteins of *Culex pipiens quinquefasciatus*, this species being a non vector of dengue, we get further basis to believe that amino acid sequencing with Glycine as terminal amino acid may be a blocking protein for dengue transmission
- There is no study done so far on this aspect of vertical and horizontal transmission of dengue virus by *Aedes* mosquitoes. The field, laboratory and experimental observations generated in the present work highlight the membrane protein of 200 kDa molecular weight as composed of special amino acids as the marker of non transmission of virus by the vectors.

Identification of mosquito proteins from the amino acid sequence of 200 kDa protein

The present study is aimed to study molecular diversity of mid gut and ovarian proteins of *Aedes aegypti*, *Aedes albopictus* and *Aedes vittatus* with reference to their competence to transmit and maintain dengue viruses in a particular area. During the reported period, amino acid sequence of 200 kDa proteins associated as blocking protein for virus transmission by mosquitoes and in exceptional cases associated with transmission of virus through mosquito samples, *both* were further studied using the information about mosquito proteins available in genomic data. *The sequence of blocking protein was put to data base to identify complete protein.* The data base BLAST (Basic Local Alignment and Search Tool) program showed many results, one of which was identified as a transport protein SEC24 of *Culex pipiens quinquefasciatus*. Exploring the reports available on this protein further indicated a protein sequence to be its putative orthologue which is present in *Aedes aegypti* and is referred as putative protein SEC24. The gene and protein report of this protein is as follows (Figure 1):

VectorBase Gene ID	AAEL001273																					
Genomic Location	This gene can be found on SuperContig supercont1.28 at location 3,523,328-3,542,829. The start of this gene is located in contig_1923 .																					
Description	Sec24B protein, putative																					
Prediction Method	Genes were annotated by merging Ensembl and TIGR predictions sets. Ensembl automatic analysis pipeline using either a GeneWise/Exonerate model from a database protein or a set of aligned cDNAs/ESTs followed by an ORF prediction. GeneWise/Exonerate models are further combined with available aligned cDNAs/ESTs to annotate UTRs (For more information see V.Curwen et al., Genome Res. 2004 14:942-50). TIGR annotations were modeled based on a combination of Augustus (M. Stanke), GlimmerHMM (M. Pertea), Genie (D. Kulp), and Twinscan (M. Brent) gene predictions, coupled with protein and EST genome alignments.																					
Transcripts	AAEL001273-RA AAEL001273-PA novel transcript [Transcript info] [Exon info] [Peptide info] Features ▼ S'ctg supercont1.28 Length DNA(contigs) Vectorbase trans. Length 3.52 Mb 3.52 Mb 3.52 Mb 3.53 Mb 3.54 Mb 3.54 Mb 3.54 Mb 3.55 Mb Forward strand 39.50 Kb contig_1923 > < AAEL001273-RA Vectorbase (Novel Protein Coding) 39.50 Kb Reverse strand 3.52 Mb 3.52 Mb 3.52 Mb 3.53 Mb 3.54 Mb 3.54 Mb 3.54 Mb 3.55 Mb																					
Alignments	This gene can be viewed in genomic alignment with other species view genomic alignment with Pecan (3 mosquitoes)																					
Orthologue Prediction	The following gene(s) have been identified as putative orthologues: <table><thead><tr><th>Species</th><th>Type</th><th>Gene identifier</th></tr></thead><tbody><tr><td><i>Anopheles gambiae</i></td><td>1-to-1</td><td>AGAP005263 (Novel Ensembl prediction) [MultiContigView] [Align] No description [Target %id: 58; Query %id: 65]</td></tr><tr><td><i>Caenorhabditis elegans</i></td><td>1-to-1</td><td>ZC518.2 yeast SEC homolog family member (sec-24.2) [Source:RefSeq_peptide;Acc:NP_502354]</td></tr><tr><td><i>Culex pipiens</i></td><td>1-to-1</td><td>CPIJ002315 (Novel Ensembl prediction) [MultiContigView] [Align] transport protein SEC24 [Target %id: 79; Query %id: 59]</td></tr><tr><td><i>Drosophila melanogaster</i></td><td>1-to-1</td><td>CG1472 (CG1472) [MultiContigView] [Align] CG1472-PA [Source: RefSeq_peptide (NP_610531)] [Target %id: 52; Query %id: 52]</td></tr><tr><td><i>Homo sapiens</i></td><td>1-to-many</td><td>ENSG00000113615 Protein transport protein Sec24A (SEC24-related protein A) (Fragment). [Source:Uniprot/SWISSPROT;Acc:Q95486]</td></tr><tr><td></td><td>1-to-many</td><td>ENSG00000138802 Protein transport protein Sec24B (SEC24-related protein B). [Source:Uniprot/SWISSPROT;Acc:Q95487]</td></tr></tbody></table> MultiContigView showing all Homo sapiens orthologues View alignments of homologies.	Species	Type	Gene identifier	<i>Anopheles gambiae</i>	1-to-1	AGAP005263 (Novel Ensembl prediction) [MultiContigView] [Align] No description [Target %id: 58; Query %id: 65]	<i>Caenorhabditis elegans</i>	1-to-1	ZC518.2 yeast SEC homolog family member (sec-24.2) [Source:RefSeq_peptide;Acc:NP_502354]	<i>Culex pipiens</i>	1-to-1	CPIJ002315 (Novel Ensembl prediction) [MultiContigView] [Align] transport protein SEC24 [Target %id: 79; Query %id: 59]	<i>Drosophila melanogaster</i>	1-to-1	CG1472 (CG1472) [MultiContigView] [Align] CG1472-PA [Source: RefSeq_peptide (NP_610531)] [Target %id: 52; Query %id: 52]	<i>Homo sapiens</i>	1-to-many	ENSG00000113615 Protein transport protein Sec24A (SEC24-related protein A) (Fragment). [Source:Uniprot/SWISSPROT;Acc:Q95486]		1-to-many	ENSG00000138802 Protein transport protein Sec24B (SEC24-related protein B). [Source:Uniprot/SWISSPROT;Acc:Q95487]
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Figure 1. Picture of data base for identification of mosquito protein from observed amino acid sequence

Table 1. Details of experimental studies on relation of ovarian proteins & vertically transmitted virus

Species	Sample Number	Parent mosquito		Progeny mosquito			
		200kDa protein in ovary	Virus in head squash	F ₁ generation	Examined for IFA Test	IFA Test positive	IFA Test negative
<i>Aedes aegypti</i>	1	-	√	1	1	√	
	2	√	-	2	1		√
					1		√
	3	√	-	1	1		√
	4	√	-	4	1		√
					1		√
					1		√
					1		√
	5	-	-	2	1		√
					1		√
	6	-	-	2	1		√
					1		√
	7	√	-	4	1		√
					1	-	
					1	-	
					1	-	
	8	√	-	1	1		√
	9	√	-	2	1		√
					1		√
	10	√	-	2	1	√	
					1		√
	11	-	√	4	1	√	
					1	√	
					1	√	
					1	√	
	12	√	√	4	1		√
					1		√
					1	√	
					1		√
	13	-	√	3	1	√	
					1		√
					1	√	
	14	-	-	2	1		√
					1		√
	15	√	√	2	1		√
					1		√
	16	-	√	1	1	√	
	17	-	-	4	1		√
					1		√
					1		√
					1		√
<i>Aedes vittatus</i>	18	√	-	3	1		√
					1		√
					1		√
Total	18			44			

1.3 Molecular characterization of extrinsic peri-urban dengue viruses contributing to cause DHF: A new concept for mechanism of DHF - Vinod Joshi, Bennet Angel and Rashmi Chaubhan

Commencement: October, 2005

Duration: Three Years

Status: Ongoing

Objectives

1. To confirm occurrence of Zoonotic cycle (ZC) of Dengue virus among tree hole breeding mosquitoes and inhabiting monkey populations. Study of its occurrence and epidemiological significance in desert settings of Rajasthan.
2. Study of role of Zoonotic cycle in maintaining and/or amplifying endemic cycle of dengue fever.
3. Study of possible role of Zoonotic cycle as factor causing Dengue Haemorrhagic fever.
4. To undertake experimental studies to confirm monkey – mosquito cycle of dengue in laboratory models of monkeys.

Progress of the Work

Virus contained by the *Aedes* mosquitoes of different species from selected urban, peri-urban and rural areas of Jodhpur were typed (Table 1). The findings are as follows:

1. The mosquitoes belonging to species *Ae. vittatus* showed more in peri-urban localities. Den-2 and Den-3 strains were most common among infected samples of peri-urban area.
2. The PCR studies of infected mosquitoes and human patients of DHF, are likely to be made during forthcoming period. The PCR and genomic studies of mosquitoes and human patients of DHF will prove the contention.

Table 1. Distribution of virus types in mosquitoes of urban peri-urban and rural areas of Jodhpur

Setting	Species	Total tested	Total positive	% positive	Positive for DEN-1	% positive	Positive for DEN-2	% positive	Positive for DEN-3	% positive	Positive for DEN-4	% positive
Urban	<i>Aedes aegypti</i>	251	60	23.90	9	15.00	14	23.33	24	40.00	13	21.66
	<i>Aedes vittatus</i>	2	0	-	0	-	0	-	0	-	0	-
Peri-urban	<i>Aedes aegypti</i>	28	5	17.85	2	40.00	1	20.00	2	40.00	0	-
	<i>Aedes vittatus</i>	77	40	51.94	6	15.00	15	37.5	14	35.00	5	12.50
Rural	<i>Aedes aegypti</i>	18	2	11.11	1	50.00	0	-	1	50.00	0	-
	<i>Aedes vittatus</i>	67	5	7.46	3	60.00	2	40.00	0	-	0	-
Total	<i>Aedes aegypti</i>	297	67	22.55	12	17.91	15	22.38	27	40.29	13	19.40
	<i>Aedes vittatus</i>	146	45	30.82	9	20.00	17	37.77	14	31.11	5	11.11
Grand total		443	112	25.28	21	18.75	32	28.57	41	36.60	18	16.07

1.4 Confirmation of “Introduction, Transmission and Aggravation” conceptuality of Desert Malaria: Verification of research undertaken and softwares developed - Vinod Joshi, A. P. Dash, Bennet Angel, M. S. Chalgá and Himmat Singh

Commencement: December, 2005

Duration: Four Years

Status: Ongoing

Objectives

1. Study of feasibility and effectiveness of installing check post at sub centre/ village level to check in-migrated natives to prevent introduction of malaria.
2. Testing of malaria transmission prediction module software through prediction and its confirmation in the adopted study villages.
3. Computer programme development using introduction and transmission quantum of malaria in a group of villages to develop epidemic forecasting system.

Progress of the Work

The work undertaken so far by our group has shown that in desert settings of Rajasthan, Malaria appears every year afresh, it completes its one or two transmission cycles with the help of local vector fauna and then disappears. In certain pocket the outbreaks of disease appear more imported malaria meet with high vector density (DMRC unpublished data). The research undertaken in 26 villages of irrigated, semi-irrigated and non-irrigated villages of Jaisalmer and Jodhpur districts had shown that malaria is introduced into desert every year through in-migrated natives. Once introduced through in-migration, the transmission of malaria is controlled by a combined interactive resultant of mosquito density, human population density, cattle population density and carriers of active infection.

Based on the work undertaken during last 3 years following equation of inter-relationship between active cases, density of *An. stephensi*, density of *An. culicifacies*, density of humans, cattle density and resultant malaria was developed. The equation developed was as under:

$$TI = Ac (PMHst - Hd + PMHcu - Cd)$$

Where TI = Transmission Index; Ac = Active Cases; PMHst = density *stephensi*; PMHcu = density *culicifacies*; Hd = Human density; Cd = Cattle density

Based on the work done during last three years following conclusions were emerged:

- The simulation model developed is capable of processing varying values of all the parameters simultaneously as well as fixing all the values and varying value of one particular parameter.
- The software developed appears to process any existing value of vector, parasite and bait parameter very sensitively. Under real life conditions the values of any or all the parameters will be within range of simulated values, therefore, it can be validated under the field conditions for its possible application as the forecasting mechanism.

During reported period, a malaria out break appeared in the Pokaran town, Jaisalmer district and in the adjoining villages. The software developed was tested under field conditions. Following observations were made:

Testing in Urban area

Study Area	First study	Follow up
Pokaran city	9/8/2008	9/18/2008
No. of Ind.	126	109
Active cases	3	8
No. of .Cattle	53	50
No. Mosquitoes	117	15
Time Spent	57	55
<i>An. stephensi</i>	8	2
PMH	8.421053	2.181818
<i>An. culicifacies</i>	13	1
PMH	13.68421	1.090909
Prediction	Sporadic	
Observed Results	Sporadic	

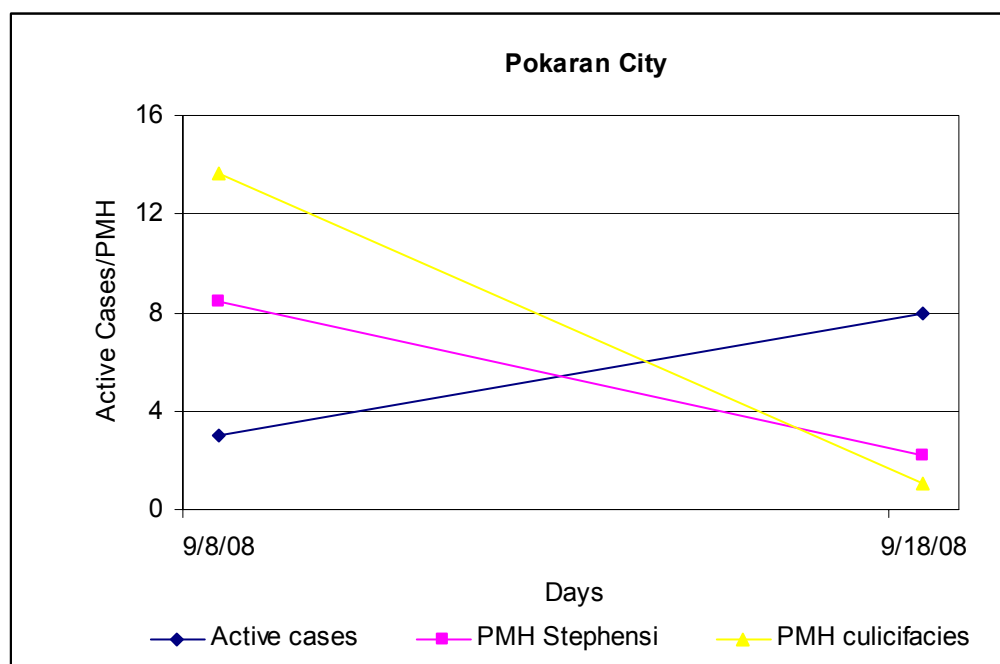


Figure 1. Transition of malaria magnitude from first to follow up study in rural area

Testing in Rural area

Study Area	First study	Follow up
Lawah	9/9/2008	9/19/2008
No. of Ind.	144	144
Active cases	5	19
No. of .Cattle	168	168
No. Mosquitoes	62	27
Time Spent	38	44
<i>An. stephensi</i>	0	2
PMH	0	2.727273
<i>An. culicifacies</i>	42	40
PMH	66.31579	54.54545

Prediction
Observed Results

Moderate
Moderate

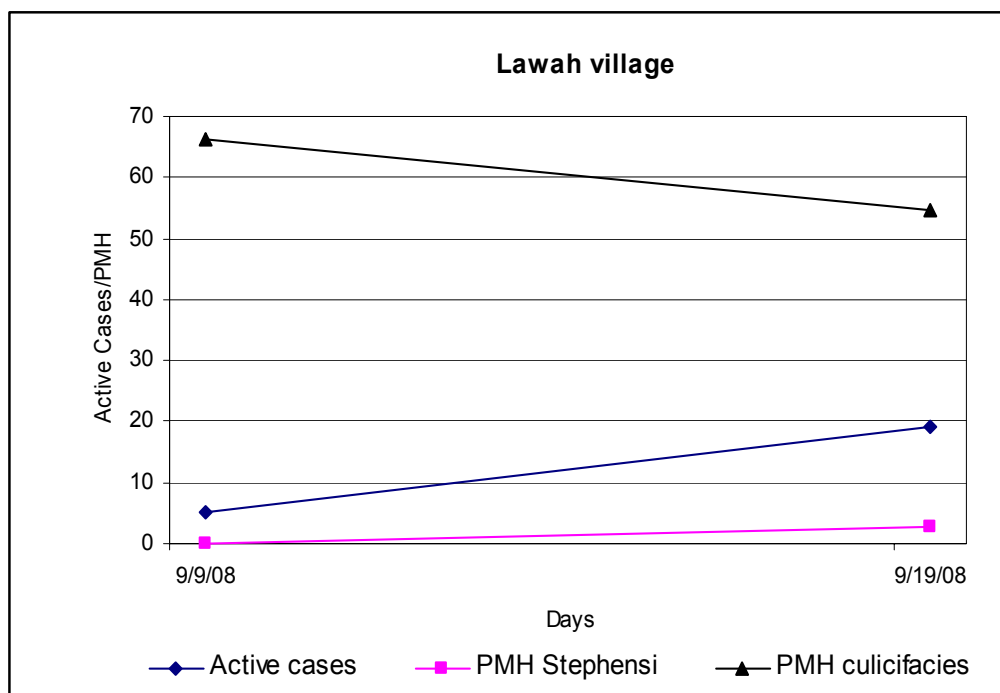


Figure 1. Transition of malaria magnitude from first to follow up study in rural area

Findings

- The malaria transmission prediction software developed has been found accurate and useful when tested under simulated and field conditions.
- The software developed has been submitted for patenting.

1.5 Study of rapid culture of *Mycobacterium tuberculosis* from sputum samples- *Murli L. Mathur and Aruna Solanki**

Commencement: May, 2007

Duration: Two years

Status: Completed

Objectives

1. To shorten the duration of time required for rapid culture of *Mycobacterium tuberculosis* from sputum samples, so that it can be used for rapid drug susceptibility testing.

Progress of work

LJ Medium:

Blood is a rich source of most nutrients which has been used to enrich to enhance growth of various bacteria. It was therefore thought to enrich LJ medium with blood to encourage the growth of MTB on it so as to reduce the time taken in appearance of macroscopic colonies of MTB. Simultaneously blood agar is also used comparing its performance with traditionally used LJ medium. Blood agar is commonly used in most clinical microbiology laboratories because it is inexpensive, simple to prepare and majority of bacteria can be grown on it. The use of blood agar media for the growth of Mtb was reported early in the last century but has not been mentioned in contemporary microbiology manuals.

Blood agar medium is used as a solid media for MTB culture. To make it a selective medium for growth of Mtb, we added malachite green to it and called it modified blood agar. Instead of unhemolysed blood, we used hemolysed sheep's blood which was centrifuged after hemolysis, for preparing blood agar. This was done to make the medium more homogeneous.

Primary Isolation of MTB from Sputum:- After treating with NALC-NaOH method, 35 sputum samples were inoculated on three slants of LJ medium supplemented with sheep blood as well as three slants of control LJ medium. Equal volume of same inoculum prepared from pellet of sputum was inoculated on each of these six slants. These were then incubated at 37°C and were observed daily for growth of MTB. Mean number of days taken on three experimental slants was compared with mean number of days on control slants using paired 't' test. Mean number of days taken in primary isolation of MTB from sputum sample on unsupplemented LJ medium was 39.1 ± 5.8 days which was not significantly different from that on LJ Medium supplemented with sheep blood (35.4 ± 6.4 days). The experiment was repeated twice on more sputum samples but statistically significant difference was not observed.

Growth of MTB from inoculum prepared from colonies grown from sputum:- Inoculum prepared from colonies of clinical isolates of MTB as well as *M. tuberculosis* H37Rv were separately inoculated on each of three slants of LJ medium supplemented with sheep blood and three slants of control unsupplemented LJ medium. These slants were kept in incubator and were observed everyday. Mean number of days taken on three experimental slants was compared with mean number of days on control slants using paired 't' test. The difference in mean number of days (6.7 ± 1.9) taken on supplemented medium (9.9 ± 2.7) was

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significantly less than that on unsupplemented medium ($p=0.001$). Thus approximately 32% time can be saved in subculture of MTB. For detection of drug sensitivity the inoculum prepared from MTB isolated from sputum samples are inoculated on drug containing and drug free LJ Medium. If this medium is supplemented with Sheep blood, approximately 32% time can be saved. The experiment was repeated thrice and similar results were obtained.



Figure 1. Growth of MTB on LJ Medium supplemented with sheep blood

Blood Agar

Equal volume of homogeneous inoculums prepared from smear positive sputum samples processed with N-acetyl l-cysteine sodium hydroxide method was inoculated on slopes of locally prepared 7% sheep blood agar and Löwenstein-Jensen medium. These were incubated at 37° C and were observed daily for growth visible with naked eyes. Time taken in growth of *Mycobacterium tuberculosis* was compared. LJ medium recovered 68 of 70 isolates (97.1%) as compared to 66 by blood agar (94.2%). The difference was not significant. Mean time to detect macroscopic colonies of *M. tuberculosis* on blood agar was 13.6 ± 5.2 days as compared to 20.4 ± 5.1 days on LJ medium ($p=0.0001$). Mean number of colonies was significantly more on blood agar than on LJ medium. This indicates that blood agar slants may be good substitute of LJ medium for rapid detection of *Mycobacterium tuberculosis* from sputum in resource limited settings. It may save about one third of time.

Modified Blood Agar

Inoculum was prepared from colonies of MTB grown from different samples of sputum as well as those of *M. tuberculosis* H37Rv. Each inoculum was kept separate in a tube in biosafety

cabinet. Fifty microliters of the same inoculum were inoculated on each of three slants of modified blood agar (using hemolysed blood and added malachite green) and three slants of Control (Unsupplemented LJ Medium). These were incubated at 37 °C and were observed every day for appearance of macroscopic colonies.

Mean number of days required for appearance of visible macroscopic colonies of MTB on modified blood agar was 4.90 days as compared to 8.71 days in control (unsupplemented LJ Medium). Thus the time required could be reduced to nearly half (56%). The experiment was repeated trice and similar results were obtained.



Figure 2. Colonies of MTB grown on Modified Blood Agar

Tetrazolium Microplate Assay (TEMA): Sputum samples of patients of tuberculosis were obtained from laboratory of District Tuberculosis Clinic and K. N. Chest Hospital. Sputum was processed with Modified Petroff's Method. It was centrifuged at 5000 rpm for 15 minutes and supernatant was discarded. The pellet was resuspended in distilled water and was divided into two tubes, which was again centrifuged and pellets were obtained in two tubes. After discarding supernatant, pellet of one tube was resuspended in supplemented 7H9 Middlebrook broth and pellet of other tube was suspended in unsupplemented 7H9 Middlebrook broth (Control). Polymyxin-B, trimethoprim, nalidixic acid and nystatin were also added to the broth. Wells of sterile 96 well microplate with lid were loaded with 200 microlitres of this inoculum in supplemented and control broth. The plate was sealed with parafilm and incubated at 37° C.

This day was taken as day 0. All work was carried out in biosafety cabinet class II. After three days the microplate was taken out from incubator and seal was opened in biosafety cabinet. Fifty microlitres of Tetrazolium Tween 80 mixture (1.5 ml of tetrazolium at a dilution of 1 mg/ml in absolute ethanol and 1.5 ml of 10% tween 80) was added to the first well of experimental row as well as control row and microplate was again sealed and kept in incubator at 37° C. If pink to red colour appeared in this well in 24 hours it was recorded, otherwise microplate was again opened in biosafety cabinet and tetrazolium was added to second wells of experimental as well as control row. The process was continued till colour appeared in control well. Number of days taken in appearance of visible pink to red colour in the well was recorded.

The contents of all well showing colour were inoculated on Muler Hinton Agar. Gram negative cocci grew on many plates of Muler Hinton Agar and therefore it could not be concluded that appearance of colour in TEMA plate was definite indicator of growth of MTB, it could also be due to growth of contamination cocci.

Microscopic Detection of Drug Sensitivity (MODS) MODS (Microscopic Observation Drug Susceptibility assay) is a liquid-culture based test that detects *Mycobacterium tuberculosis* and assesses isoniazid and rifampicin susceptibility directly from sputum samples. The method makes use of two important properties of *M. tuberculosis*: (1) markedly faster growth in liquid media than on solid media, and (2) an easily recognizable and characteristic microscopic cording appearance of that growth in liquid media. Using an inverted light microscope, 24 well plates inoculated with decontaminated sputum samples suspended in supplemented Middlebrook 7H9 medium are examined for micocolonies which can be detected in a median of 7 days, much earlier than macroscopic colony growth can be seen on solid medium. The incorporation of isoniazid and rifampicin in the testing process enables equally rapid MDR TB detection. The simplicity of the technique, the greater sensitivity of liquid over solid media culture for TB detection, the specificity of the characteristic growth of *M. tuberculosis*, the evaluation of drug susceptibility in a short timescale, and the low cost of reagents are the major advantages of the method.

Attempts were made to standardize this technique using sterile 24 well microplates and Middlebrook 7H9 broth as per standard protocol. Many repeated attempts were made but most of the times contaminants grew in the wells of the microplate and cords of MTB could not always be appreciated.

Probably tropical climate did not favour use of liquid culture medium for growth of MTB and therefore TEMA and MODS could not be successfully used.

Important Leads and Outcome

Results of preliminary work carried out indicate that the time required in drug susceptibility of *Mycobacterium tuberculosis* can be substantially reduced by supplementing LJ Medium with sheep blood or by using blood agar in place of LJ medium.

1.6 Evaluation of efficacy of different ovitraps for the surveillance and control of dengue vector, *Aedes aegypti* in Jodhpur city - Karam V. Singh and S. K. Bansal

Commencement: October, 2007

Duration: Three Years

Status: Ongoing

Objectives

1. To evaluate the efficacy of ovitraps in determining the population dynamics of *Aedes aegypti* in surveillance programme
2. To determine the efficacy of different ovitraps containing either *Bti* or temephos in reducing the immature stages of *Ae. aegypti* and consequently to bring down vector population.

Progress of the work

To evaluate the feasibility of ovitraps in arid situations and selection of study area, preliminary studies were conducted in 10 localities of Jodhpur city viz., Kudi Bhagtasni Housing Board, Railway Colony Bhagat ki Kothi, Indira Colony, Mahamandir, Soorsagar, Fidusar, Chopasni Housing Board, Mansuria colony, Pratap nagar and Bakra Mandi. The above colonies covered both old city areas as well as peripheral newly developed residential colonies.

Three types of ovitraps i.e. grass infusion (*Cynodon dactylon*), grass infusion with *Bacillus thuringiensis* var. *israelensis* (Bti) and grass infusion with cyfluthrin, were used during the studies. The studies revealed that 32.5 percent traps were positive showing immature stages of urban mosquito vector *Aedes aegypti* (Table 1). 57.5 percent traps were found positive for eggs, 3.8 percent for larval forms and 38.5 percent for both eggs and larval forms. The maximum no. of traps was found positive with grass infusion (G) only (46.7), followed by Control (36.1%), G+Bti (26.7%) and G+Cyfluthrin (14.3%). It indicates that grass infusion acts as oviposition attractant, whereas, Bti and Cyfluthrin both exhibited deterrent effect.

Table 1. Trap Positivity according to type of traps laid

Types	Total Traps Laid	Total +ive	% Positivity
Grass infusion	15	07	46.7
G + Bti	15	04	26.7
G + Cyfluthrin	14	02	14.3
Control	36	13	36.1
Total	80	26	32.5

Trap Positivity in relation to type of households was also analyzed and it was found that pucca houses exhibited high positivity, which may be due to availability of adult resting & breeding habitats. However, the difference was not found statistically significant.

The ovitraps positivity was also analyzed in relation to location of water storing devices and location of traps laid, It was found that maximum no. of traps were positive in the houses having overhead tanks (23.5%), followed by ground (18.6%) and underground tanks (17.6%).

The data on location of traps laid revealed that maximum no. of traps were found positive laid in side the bathrooms (34.6%) and minimum laid in the stair-cases. The order of preference was: bathroom > Courtyard>Verandah>Cattle sheds. It indicates that *Ae. aegypti* prefers dark and damp places for egg-laying.

Regarding the positivity of different types of ovitraps with respect to different immature stages, it was observed that in the ovitraps with only grass infusion and grass infusion and Bti, both eggs and larvae, either singly or combined were recorded, whereas, in case of traps with grass infusion and cyfluthrin only eggs were reported (Table 2), which indicate that this help either in killing larvae or preventing egg hatching. This needs verification.

Table 2. Traps positivity according to developmental stages of *Aedes aegypti*

Type of traps	Developmental stages		
	Eggs	Larvae	Both (Eggs+Larvae)
Grass infusion	05 (71.4%)	01(14.3%)	01(14.3%)
G + Bti	02(50.0%)	-	02(50.0%)
G + Cyfluthrin	02(100.0%)	-	-
Control	06(46.2%)	-	07(53.8%)
Total	15(57.7%)	01(3.8%)	10(38.5%)

Besides this, a field study to find out efficacy of different ovitraps on the basis of impact of colour and grass infusion (*Cyanodon dactylus*) on oviposition response of the *Aedes aegypti* was also conducted. The study revealed that grass infusion has more attractiveness than water. The positive prevalence of ovitraps was found 80% in case of ovitraps containing grass infusion and 57% those containing only water. This difference was found to be statistically significant ($p<0.001$). And the comparative data generated for ovitraps with and without oviposition attractant revealed that trap positivity, total no. of eggs collected, prevalence, egg density index and mean egg densities were recorded higher in case of grass infusion baited ovitraps.

Colour-wise analysis of ovitrap data revealed that relative occurrence of eggs, maximum no. of eggs was laid in Black coloured ovitrap in case of traps containing grass infusion. However in traps containing simple water the maximum no. of eggs were laid in the orange coloured ovitrap. For grass infusion ovitraps the prevalence was calculated 1.0 for Orange and Red colours, whereas the intensity and mean egg density results exhibited higher values in case of Black ovitraps, which indicates that the Orange and Red coloured ovitraps having grass infusion, as attractant, can be used for the detection of the presence of the vector species, but for quantification or correlation purposes with adult mosquitoes Black coloured ovitraps may be preferred. The results warrant detailed field investigations to validate the findings of this preliminary study.

For the field evaluation of different ovitraps Pratap nagar locality of Jodhpur city has been selected on the basis of preliminary survey of the *Ae. aegypti* population and positivity of ovitraps. In this locality, three groups of 30 houses each have been considered: one as control and two experimental for evaluation of lethal and non-lethal ovitraps.

1.7 Determination of larvicidal potential of active principle(s) of *Solanum xanthocarpum* against important mosquito vectors - S. K. Bansal and Karam V. Singh

Commencement: September, 2005

Duration: Three Years

Status: Completed

Objectives

1. Preparation of different solvent extracts from roots, leaves and fruits of *S. xanthocarpum* and isolation of their active constituents.
2. To identify different constituents of the extract and comparison of their larvicidal/repellent properties.

Progress of the Work

Susceptibility tests were carried out with larvae of different mosquito vectors viz. *Anopheles stephensi*, *Aedes aegypti* and *Culex quinquefasciatus*. For this purpose adults and larvae of all the mosquito species were collected from different areas of Jodhpur city and reared in the laboratory for further generations under controlled conditions of temperature ($28\pm 2^\circ\text{C}$) and humidity ($75\pm 5\%$). *S. xanthocarpum* has been selected for undertaking the proposed study. The different parts of this plant differ in their active constituents when extracted in different solvents. Seeds of this plant were shade dried between $30-40^\circ\text{C}$ for 10-15 days. Dried seeds were powdered and dissolved in different solvents and stock solutions and serial dilutions were made as per requirement. Third instar larvae of these mosquito species were tested as per standard WHO method for determining the baseline data on their susceptibility status. Experiments were carried out in 500 ml beakers containing 249 ml of water by using 20-25 larvae of each mosquito species. Mortality was noted after 24 and 48hr and corrected by using Abbott's formula. Average of four observations was taken and data subjected to log probit regression analysis.

Observations on the results of the larval susceptibility to methanol extracts of seeds for fresh, six and twelve months old samples were analyzed (Table 1). With all the mosquito species mortality was dose dependent i.e. mortality increased with increase in concentration. 24 & 48hr LC_{50} and LC_{90} values along with their fiducial limits, regression equation and chi-square were calculated. The 24hr LC_{50} values as observed for *An. stephensi*, *Ae. aegypti* and *Cx. quinquefasciatus* for the seed extracts were 73.7, 195.6, 251.2; 123.8, 377.6, 426.2 and 154.9, 520.0, 656.7 mg/l respectively. Comparative susceptibility observed on the basis of 24 & 48hr LC_{50} values showed that larvae of anophelines are much more susceptible than larvae of *Ae. aegypti* and *Cx. quinquefasciatus*. 48hr LC_{50} values were also found to be much less as compared to the 24hr LC_{50} values showing that efficacy was much more pronounced after 48hrs as compared to 24hrs. Experiments done after six and twelve months showed that efficacy was decreased 2-3 times as compared to the fresh samples.

For HPLC and Infra Red spectroscopic studies, cleaned and dried seeds were powdered and a known amount was extracted with petroleum ether ($40-60^\circ\text{C}$) as a solvent. The sample was subjected to HPLC and Infra Red spectroscopic studies at CDRI, Lucknow. The percent

composition of the different fatty acids so observed were Linoleic acid (32.96), Oleic acid (28.74), Stearic acid (18.44), Palmitic acid (08.58), Linolenic acid (04.32) Hydroxy acid (03.72) and Aracidic acid (03.14) (Table 2).

Susceptibility experiments were carried out with oil extracted from seeds of *S. xanthocarpum* with all the three species at a concentration range from 10 to 200 mg/l as per usual WHO procedure. The LC₅₀ and LC₉₀ values were obtained by probit regression analysis. It was observed that *An. stephensi* was more susceptible (24hr LC₅₀ 75.3 mg/l) as compared to *Ae. aegypti* (95.8 mg/l) and *Cx. quinquefasciatus* (138.9 mg/l) (Table 3 & 4).

Table 1. 24 and 48hr LC₅₀ values (mg/l) of extracts from seeds of *S. xanthocarpum* to larvae of different mosquito vectors

Mosquito species	Seeds 24 (48) hr LC ₅₀		
	FP	6m	12m
<i>Anopheles stephensi</i>	73.7	195.6 (95.6)	251.2 (172.4)
<i>Aedes aegypti</i>	123.8	377.6 (131.1)	426.2 (252.0)
<i>Culex quinquefasciatus</i>	154.9	520.0 (222.7)	656.7 (431.5)

FP- Fresh preparation; 6m- Six months storage; 12m-Twelve months storage

Table 2. Fatty acids composition of oil extracted from seeds of *S. xanthocarpum*

Fatty acids	Percent Composition
Linoleic acid	32.96
Oleic acid	28.74
Stearic acid	18.44
Palmitic acid	08.58
Linolenic acid	04.32
Hydroxy acid	03.72
Aracidic acid	03.14

Table 3. Larvicidal activity of *Solanum xanthocarpum* oil against different mosquito vectors

Concentrations (ppm)	Percent Larval Mortality		
	<i>Anopheles stephensi</i>	<i>Aedes aegypti</i>	<i>Culex quinquefasciatus</i>
Control	0.0	0.0	0.0
10	4.0	1.5	1.3
25	12.0	8.0	4.0
50	32.0	24.0	16.0
100	60.0	52.0	40.0
200	96.0	80.0	64.0

Table 4. Log probit regression analysis of the mortality data of larvae of different mosquito vectors to oil extracted from seeds of *S. xanthocarpum*

Mosquito Species	Regression Equation	Chi-Square (DF)	24hr LC₅₀ (Fiducial limits)	24hr LC₉₀ (Fiducial Limits)
<i>Anopheles stephensi</i>	Y=2.27x+0.74	1.11 (2)	75.3 (54.7-103.6)	275.7 (140.7-540.0)
<i>Aedes aegypti</i>	Y=2.35x-0.34	0.33 (2)	95.8 (69.6-131.9)	335.9 (166.3-678.5)
<i>Culex quinquefasciatus</i>	Y=2.24x-0.21	0.47 (2)	138.9 (96.9-199.0)	518.7 (235.7-1142.0)

1.8 Evaluation of some plant species found in the arid region for the larvicidal/repellant potential of their oils against the major mosquito vectors - S. K. Bansal, Karam V. Singh and M. R. K. Sherwani*

Commencement: April, 2008

Duration: Three Years

Status: Ongoing

Objectives

1. Determination of the larvicidal activity present in different parts of the plants after extraction in different organic solvents.
2. Extraction of essential oil present in different parts especially the fruits and seeds and evaluation of their larvicidal /repellant potential against mosquito vectors.
3. Identification of the active insecticidal constituents present in different parts by GC/MS and comparison of their larvicidal/repellant properties.

Progress of the Work

Preliminary Investigations have been carried out on *Tephrosea purpurea*, *prosopis juliflora* and *Citrus colocythis* while studies on some other plants are being going on. Larvicidal properties have been observed in some parts of these plants while repellant action of the oils present in the seeds and fruits are going on with adults of different mosquito species. The study will suggest the actual effective constituents of the plant extract which have larvicidal/repellent properties and later can be considered for the development of a commercial product.

Susceptibility tests were carried out with larvae of three mosquito species viz. *Anopheles stephensi*, *Aedes aegypti* and *Culex quinquefasciatus*. For this purpose larvae of all the three mosquito species were collected from different areas of Jodhpur city and reared in the laboratory for further generations under controlled conditions of temperature ($28\pm 2^\circ\text{C}$) and humidity ($75\pm 5\%$). The different parts of the plant differ in their active constituents when extracted in different solvents. Samples of roots, leaves and fruits were chopped and shade dried between $30-40^\circ\text{C}$ for 10-15 days. Dried plant material was powdered separately and dissolved in different solvents and stock solutions and duration and serial dilutions were made as per requirement. Third or early fourth instar larvae of these mosquito species were tested as per standard WHO method for determining the baseline data on their susceptibility status. Experiments were carried out in 500 ml beakers containing 249 ml of water by using 20-25 larvae of each mosquito species. Mortality was noted after 24 hr and corrected by using Abbott's formula. Average of four observations was taken and data subjected to log probit regression analysis.

Observations on the results of the larval susceptibility to methanol extracts of seeds of *Tephrosea* were analyzed (Table 1). With all the mosquito species mortality was dose and duration dependent i.e. mortality increased with increase in concentration. LC_{50} and LC_{90} values along

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with their fiducial limits, regression equation and chi-square were calculated. LC₅₀ and LC₉₀ values as observed for *An. stephensi*, *Ae. aegypti* and *Cx. quinquefasciatus* were 74.9, 63.2 & 47.0 and 382.3, 249.6 & 165.1 mg/l respectively. Comparative susceptibility calculated on the basis of LC₅₀ value showed that larvae of *Cx. quinquefasciatus* and *Ae. aegypti* were more susceptible than larvae of *An. stephensi*. Larval susceptibility tests were also carried out with methanol extracts of leaves of *Prosopis juliflora* with larvae of all the three mosquito species (Table 2). LC₅₀ and LC₉₀ values as determined for *An. stephensi*, *Ae. aegypti* and *Cx. quinquefasciatus* were 96.2, 128.1 & 118.8 and 344.2, 428.5 & 361.4 mg/l respectively which revealed that larvae of *An. stephensi* are more susceptible than the rest of the two species.

Experiments have also been carried out with the methanol extracts of leaves of *Tephrosia purpurea* and seeds of *Prosopis juliflora*. Experiments carried out up to 400 mg/l with leaves and seed extracts of these plant species show only up to 10-20% mortality indicating that active larvicidal principle may be present only in the seeds of *Tephrosia purpurea* and leaves of *Prosopis juliflora*. The 24 hr LC₅₀ values observed for methanol extracts of fruits of *Citrullus colocynthis* are 46.9, 74.9 and 97.6 mg/l for the three mosquito species respectively (Table 3). More preliminary studies are being carried out on some other desert plants found in this arid region.

Table 1. Log probit regression analysis of the mortality data of larvae of different mosquito vectors to seed extracts of *Tephrosia purpurea*

Mosquito species	Regression Equation	Chi-Square (DF)	LC ₅₀ (Fiducial Limits)	LC ₉₀ (Fiducial Limits)
<i>Anopheles stephensi</i>	Y=1.81x ± 1.61	1.68 (3)	74.9 (54.8-102.3)	382.3 (156.9-931.7)
<i>Aedes aegypti</i>	Y=2.15x ± 1.13	2.01 (3)	63.2 (48.8-81.9)	249.6 (133.6-466.0)
<i>Culex quinquefasciatus</i>	Y=2.35x ± 1.08	2.17 (3)	47.0 (35.7-61.9)	165.1 (90.0-303.0)

Table 2. Log probit regression analysis of the mortality data of larvae of different mosquito vectors to leaves extracts of *Prosopis juliflora*

Mosquito species	Regression Equation	Chi-Square (DF)	LC ₅₀ (Fiducial limits)	LC ₉₀ (Fiducial Limits)
<i>Anopheles stephensi</i>	Y=2.31x ± 0.41	1.54 (3)	96.2 (73.0-126.8)	344.2 (192.9-614.4)
<i>Aedes aegypti</i>	Y=2.45x ± -0.15	2.47 (3)	128.1 (101.7-161.4)	428.5 (254.8-721.1)
<i>Culex quinquefasciatus</i>	Y=2.65x ± -0.50	2.11 (3)	118.8 (93.7-150.7)	361.4 (212.4-614.8)

Table 3. Log probit regression analysis of the mortality data of larvae of different mosquito vectors to fruit extracts of *Citrullus colocynthis*

Mosquito species	Regression Equation	Chi-Square (DF)	LC₅₀ (Fiducial limits)	LC₉₀ (Fiducial Limits)
<i>Anopheles stephensi</i>	$Y=2.95x \pm 0.07$	0.86 (2)	46.9 (37.9-58.0)	127.6 (80.9-200.9)
<i>Aedes aegypti</i>	$Y=4.11x \pm 2.70$	3.43 (2)	74.9 (63.6-88.1)	153.4 (102.8-229.1)
<i>Culex quinquefasciatus</i>	$Y=5.12x \pm 5.18$	2.60 (2)	97.6 (87.2-109.3)	173.7 (132.9-227)

1.9 Malaria in children in desert - *S. P.Yadav and R. K. Kalundha*

Commencement: September, 2008

Duration: Three Years

Status: Ongoing

Objective

1. To know the current status of malaria in children in desert part of Rajasthan

Progress of the work

Jaisalmer district was having highest API among all the 12 desert districts. Among the PHCs of the district, Ramgargh PHC was on top as far as API is concerned. Therefore, villages of Ramgargh PHC in Jaisalmer district were selected for the study. During the reported period investigations were initiated in 12 villages namely Raghawa, Raimala, Seowa, Tejpala, Nagga, Bada, Sultana, Mokal, Sanu, Habur, Kakab and Lanala. Fever was used as proxy for malaria. Information was collected on pre-tested schedules. Questionnaires in schedules were in English but communicated to the respondent in Hindi or in local dialects *i.e.* Marwari for better understanding and communication between respondent and investigator. Based on this some observation are made.

In the studied villages, 1218 households have been covered. Total population of the surveyed households was 7232. The positive cases of malaria were 1441. About twenty two per cent 314 (21.8%) cases were children less than 15 years of age (Table 1).

Table 1. Distribution of malaria cases in the children according to age and sex

Age in years	Male		Female		Total	
	No.	%	No.	%	No.	%
0-1	22	16.1	27	15.3	49	15.6
1-5	74	54.0	97	54.8	171	54.5
5-10	23	16.8	31	17.5	54	17.2
10-15	18	13.1	22	12.4	40	12.7
Total	137	100.0	177	100.0	314	100.0

Table 1 depicts the current situation of malaria in infants in the rural population of desert.

Table 2. Distribution of malaria cases in the infants according to age and sex

Age in months	Male		Female		Total	
	No.	%	No.	%	No.	%
0- 3 months	1	4.5	3	11.1	4	8.2
3-6 months	4	18.2	4	14.9	8	16.3
6-9 months	7	31.8	8	29.6	15	30.6
9-12 months	10	45.5	12	44.4	22	44.9
Total	22	100.0	27	100.0	49	100.0

Findings

Seasonal studies were undertaken on the rainy and spring seasons during the months of April to July. A positive correlation was observed between vector population and transmission of disease.

1.10 A study of association between socio-economic factors and transmission of malaria in desert - S. P. Yada, A. K. Dixit and R. K. Kalundha

Commencement: October, 2007

Duration: Two Years

Status: Ongoing

Objectives

1. To study the socio-economic factors associated with malaria transmission in desert.
2. To find out the social solutions to control desert malaria.

Progress of the work

The API of Jaisalmer district was the highest among the desert districts of Rajasthan. Ramgarh PHC was on top among the district PHCs. Base line data of all the 60 villages of Ramgarh PHC were collected for understanding the factors associated with magnitude of disease. To accomplish the above objectives work was initiated and in 12 villages namely- Sanu, Seuwa, Tejpala, Bada, Naga, Sadhana, Raimala, Sultana, Mokal, Lanela, Habur and Kakab. All the households with malaria positive cases were included in the study. 876 households during the report period and 342 households in the last year could be covered during the survey. The data of previous year of the studied households updated during the year 2008 for the proper mixing of the data. Thus the data of total 1218 households were used for this report. Data pertaining to malaria cases during 6 years (2002-07) were collected with respect to each household from the MPWs/ANMs. Pre-tested schedules were used for data collection which were prepared in English but communicated in *Hindi* or in local dialect- *Marmari*. Head of the Household or a member more than 18 years of age who was present at the time of survey was interviewed. Fever was used for the proxy of malaria. Some observations were made based on the accomplished work.

Findings

Low Socio-Economic Group (LSEG) and High Socio-Economic Group (HSEG) both were comparable with socio-demographic characteristics such as age, sex, education, occupation, religion and castes. LSEG has the API all most 2 for the 6 consecutive years from 2002-2007 which is nearly 3 times more than the HSEG and presents alarming situation (Table 1) and the children are more affected. These preliminary observations indicate statistically significant effect of malaria on low socio-economic group of the community. LSEG was less aware about causation of malaria and its signs and symptoms as compare to HSEG. LSEG was less using

the preventive measures to prevent the mosquito bite as compared to HSEG (Table 2). This has direct relation with the level of knowledge about the disease and its preventive measures and affordability.

Table 1. Incidence of malaria in two different socio-economic group from 2002-07

Study Parameters	2002		2003		2004		2005		2006		2007	
	LSEG	HSEG	LSEG	HSEG	LSEG	HSEG	LSEG	HSEG	LSEG	HSEG	LSEG	HSEG
Population	3287	2817	3441	2956	3579	3073	3698	3180	3804	3263	3894	3338
ABER	7.0	7.6	7.1	8.3	7.2	8.1	7.4	8.8	6.9	8.0	7.1	8.1
(+) ve cases	154	46	255	38	158	59	244	49	218	26	158	36
Pf cases	92	7	143	10	80	9	111	15	76	5	51	8
API	4.7	1.6	7.4	1.3	4.4	1.9	6.6	1.5	5.7	0.8	4.1	1.1
Death	0	0	0	0	0	0	0	0	0	0	0	0

LSEG- Low Socio-Economic Group

HSEG- High Socio-Economic Group

ABER- Annual Blood slide Examination Rate

Pf- Falciparum

API- Annual Parasite Index

Table 2. Distribution of respondents using different preventive measures

preventive measures	LSEG		HSEG	
	No.	%	No.	%
Mosquito net	60	9.9	159	26.1
Odomos cream	7	1.1	40	6.6
Oils	36	5.9	64	10.5
Tortoise coil	9	1.5	52	8.5
Good night vapouriser	5	0.8	31	5.1
Smoke of cow-dung	61	10.0	133	21.8
Smoke of foliage	98	16.1	108	17.7
Nothing	333	54.7	22	3.6
Total	609	100.0	609	100.0

LSEG- Low Socio-Economic Group

HSEG- High Socio-Economic Group

1.11 A study of the suitable interventional methods for early detection of new PTB cases and bringing them for diagnosis and treatment under DOTS - *S. P. Yadav, A. K. Dixit and R. K. Kalundha*

Commencement: November, 2007

Duration: Three Years

Status: Ongoing

Objectives

1. To study of the suitable interventional methods for early detection of new PTB cases and bringing them for diagnosis and treatment under DOTS.
2. To study cost effectiveness and acceptability of such strategy in the community.

Progress of the work

All the PHCs of Jodhpur district were categorized according to the prevalence of PTB. Chamu PHC was on top. So it was decided to conduct study in Chamu PHC of Jodhpur district. Further more on the basis of population and as well as distance from the PHC, villages were selected for the study as the true representation of the universe. As per suggestions of the SAC 2008, collaboration was done with the In-charge PHC. The rapport was established with the In-Charge and staff of the Chamu PHC for their support and participation in the study whole heartedly. Village and community leaders were also contacted and explained the purpose of the study to establish the confidence among villagers for seeking their cooperation and participation in the study. The pre-tested schedules were used for collecting information on family composition, respiratory symptoms, KAP about disease, health seeking behaviour and so on for those who had chronic chest symptoms. The chronic symptomatic cases were interviewed separately. Three repeat visits were made to contact those who were not available on first or second visit. Based on data collected some observations and inferences were made. The population of surveyed households was 1514 in the age group 15-65 years. About fifteen per cent (14.9%) persons complained for chronic chest symptoms (Table 1). It seems high as compare to national percentage. It may be due to seasonal effect for long duration being winter season at the time of survey.

Table 1. Distribution of subjects according to chronic chest symptoms

Symptoms*	Number (N=1514)	Percentage
Cough	207	13.7
Sputum	105	6.9
Breathlessness	81	5.4
Wheeze	29	1.9
Blood in sputum	18	1.2
Chest pain	22	1.5
Any or a combination above symptoms	225	14.9

* Multiple symptoms: hence numbers do not add to 225

Prevalence of symptoms was given in table 2. Males (15.2%) had more chronic chest symptoms as compared to females (14.3%). The prevalence of symptoms was varied according to the level of the education and type of occupation of the study subjects (Table 2). Individual counseling was done for the motivation to report to nearest health facility under DOTS programme for diagnosis and treatment also.

Table 2. Prevalence of chronic chest symptoms among adults according to socio-demographic characteristics

Socio-demographic characteristics		Number	Chest symptomatic	Prevalence %
Age	15-44	1092	113	10.3
	45-65	422	112	26.5
Sex	Male	817	125	15.2
	Female	697	100	14.3
Education	Illiterate	663	129	19.5
	Up to primary	504	67	13.3
	Above primary	347	29	8.4
Occupation	Household work	498	74	14.9
	Student/Unemployed	198	24	12.1
	Service	175	18	10.3
	Business	146	23	15.8
	Labour	281	46	16.3
	Agriculture	216	40	18.5
Personal habit	Smoker	621	137	22.1
	Non-smoker	893	88	9.9

1.12 Studies on larvicidal ingredients of *Calotropis procera* and its possible role in blocking intracellular replication of dengue virus in mosquitoes - *Manju Singhi and Vinod Joshi*

Commencement: August, 2007 **Duration:** Two Years **Status:** Ongoing

Objectives

1. Study of possible impact of latex on intracellular structure and biochemical profile of *Ae. aegypti* treated with larvicidal compound and dengue virus.
2. Development of plant based antiviral and antilarval compound.

Progress of Work

During the report period the work was undertaken in the following three phases:

1. Acquisition of cell lines and their maintenance in the laboratory and treatment of virus infected cell lines with latex
2. Proteomic analysis of latex of *Calotropis procera* for its utility as virus blocking agent
3. Biochemical studies of trypsin inhibition in larvae treated with latex

Acquisition of cell lines and their maintenance in the laboratory and treatment of virus infected cell lines with latex

- The C6/36 cell lines were obtained from NCCS, Pune. The cell lines were maintained and two cycles of multiplications were obtained successfully under laboratory conditions of centre. Confluent monolayer of C6/36 were used for performing in-vitro anti-viral efficacy of plant extract.
- The cell lines were inoculated with viral suspension and number of fluorescence observed in the field were counted.
- The micro doses of extracted latex was suspended into cell lines and then virus inoculation was made and fluorescence fields were counted to study the impact of larvicidal component of latex on clearance of virus from cell lines.

Proteomic analysis of latex of *Calotropis procera* for its utility as virus blocking agent

- To pin point presence of a possible anti-viral component in latex of *Calotropis procera*, we assayed the latex for display of its proteins, employing SDS PAGE. Trypsin inhibitory enzyme of 20 kDa was observed in the latex, which could be useful in preventing digestion of viral envelop in mosquitoes.

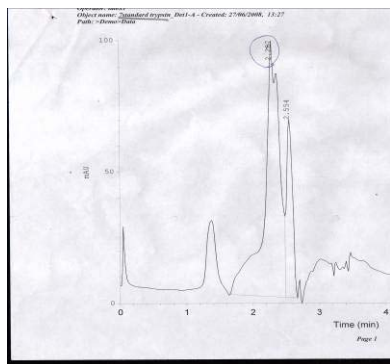
Biochemical studies of trypsin inhibition in larvae treated with latex

- The HPLC chromatographs were obtained for the assays of the mid gut of larvae treated with latex and not treated with latex.
- The curves showed that samples of mid gut of treated larvae showed inhibition of trypsin whereas in non treated samples relatively enhanced presence of trypsin was shown.

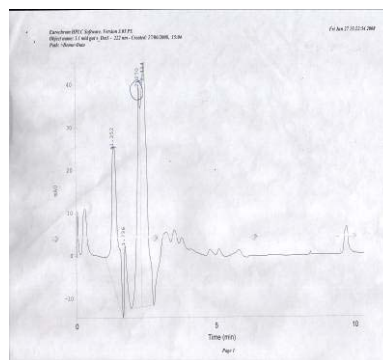
Findings

The present studies are in the initial phase of experiments and analysis. However, interesting initial findings have been observed as enumerated below:

1. Presence of trypsin inhibitory enzyme in the latex of *Calotropis procera* and biochemical analysis of larval assays treated with latex shows the evidence that treatment with latex could be trypsin inhibitory for larval mid guts.
2. The trypsin inhibition in mid guts could be useful action of latex causing biochemical incapacitation in larvae to break the virus envelop protein required for replication of virus.



Chromatogram standard trypsin



Chromatogram of methanol treated mid gut

1.13 Factors associated with malaria in block Banar of Jodhpur district, Rajasthan - P. K. Anand and R. Ramakrishnan*

Commencement: May, 2008

Duration: Seven Months

Status: Concluded

Objectives

1. To describe the cases of malaria in respect to their time, place and person characteristics
2. To identify the different factors associated significantly with the Malaria cases in block Banar

Progress of work

This study was carried out as dissertation project under post graduation programme, Master in Applied Epidemiology, running at National Institute of Epidemiology, Chennai. This is primary health centre based case control study. Sample of 48 cases and 144 controls estimated with 3 controls for each case. Cases and controls were matched with respect to age (+/- 5 years) and gender. We recruited 42 malaria cases and 84 matched controls, because of less recruitment of appropriate controls and limited time in dissertation. Analysis was done on 42 sets of matched case with controls. Cases were described in terms of their time, place and person characteristics. Matched Odds ratio estimated and conditional logistic regression analysis carried out in Epiinfo software to identify the factors associated with malaria. Complete dissertation report submitted to National Institute of Epidemiology, Chennai.

Out of all the 6 PHCs under study, only 3 PHCs Banar, Bisalpur and Fidusar recruited matched case and controls. Malaria cases recruited from the month of May till October. Out of these 6 months, 27 cases were recruited in the August and September months of rainy season. 95.2% of cases were above 5 years of age and 71.4% belonged to male gender (Table 1). 7.1 % cases occurred in persons of 45 years and above age group.

Matched analysis (Table 2): Adjusted OR (MH) value for history of recent travel was 3.6 with 95% confidence limits of 1.17-11. The corrected chi-square's p value for this association was 0.04. History of malaria in any family member within last one month had adjusted OR (MH) of value 9. Though the probability of this association was 0.06. The values of OR (MH) were 1.84, 2.1 and 4.33 for factors, not read news paper, presence of pond near home and presence of *Prosopis juliflora* plant near home respectively. Association of these factors was weak with $p < 0.2$ in all three.

Interaction: Values of crude OR was 1.55 for presence of juliflora alone near home. OR was 0 for the presence of pond alone near home. There was not even a single case with pond alone near home. Value was 2.87 if pond and juliflora both were available near home. Although in each of this case association was insignificant as, behaviour of crude OR was interesting to notice.

Conditional logistic regression (Table 3): OR of recent travel, presence of juliflora near home and history of malaria in family member were 4.89, 4.90 and 11.76 respectively, with $p < 0.05$. The values of coefficients were 1.5872, 1.5897 and 2.4652 for recent travel, juliflora near home and malaria in family respectively. The probability value of likelihood ratio of this model was 0.0012

*Guide and Scientist-E, NIE, Chennai

Table 1. General characteristic of malaria cases in block Banar, district Jodhpur, Rajasthan, India, 2008

Characteristics		Cases (n=42)	
		Number	Percentage
Age	<5 years	02	04.8
	5-14 years	07	16.7
	15-44 years	30	71.4
	45 & more	03	07.1
Gender	Male	30	71.4
	Female	12	28.6
Residence	Urban	19	45.2
	Rural	23	54.8
Occupation	Non earning	13	31.0
	Manual labour	21	50.0
	Sedentary occupation	08	19.0
Education	Illiterate	15	35.7
	Primary educated	18	42.9
	Secondary & above	09	21.4
Monthly financial income	Up to 1000 INR	18	42.9
	> 1000 INR	24	57.1
Religion	Hindu	36	85.7
	Muslim	06	14.3

Table 2. Association of selected exposures with malaria in Univariate analysis in block Banar, district Jodhpur, Rajasthan, India, 2008

Exposure factor	Discordant pairs where case is exposed	Discordant pairs where case is unexposed	Matched OR	95% CI	p value*
Illiteracy	15	12	1.25	0.49-3.1	0.81
Rural residence	08	13	0.61	0.20-1.8	0.26
Income < 1000 INR	06	14	0.42	0.13-1.3	0.08
Labor occupation	14	15	0.93	0.38-2.2	0.70
Don't read News paper	24	13	1.84	0.80-4.2	0.19
Don't view Tele vision	28	17	1.64	0.79-3.4	0.23
Unaware about malaria transmission	04	09	0.44	0.09-1.9	0.12
Unaware about malaria prevention	10	08	1.25	0.40-3.8	0.92
Don't use Antimosquito	17	12	1.41	0.57-3.4	0.60
History of recent travel	18	05	3.60	1.17-11	0.04
History of malaria in family	09	01	9.00	0.98-82	0.06
Mine work	06	06	1.00	0.25-3.9	0.74
Mud room	28	18	1.55	0.76-3.1	0.30
Pets at home	20	14	1.42	0.62-3.2	0.53
Pond near home	21	10	2.10	0.84-5.2	0.17
Prosopis Juliflora plant near home	13	03	4.33	0.83-22	0.12

* Corrected chi square p value

Table 3. Associated factors with malaria in Multivariate conditional logistic regression analysis in block Banar, district Jodhpur, Rajasthan, India, 2008

Exposure factors	Odd's ratio	95% Confidence Interval	P value
History of recent travel	4.89	1.33-17.93	0.0167
Presence of Prosopis juliflora plant near home	4.90	1.0025 - 23.97	0.0496
History of malaria in family member	11.76	1.11 - 124.41	0.0405

Findings

Malaria occurs in dry and wet season in Jodhpur. Malaria burden differs in different PHCs. Malaria is a disease of poor socioeconomic status in area. It affects almost each age group and both gender. History of malaria in any family member, recent travel within last one month and presence of juliflora plant near home are independently associated factors for malaria in this study area.

2.1 Epidemiology of Musculoskeletal Conditions in India - K. R. Haldiya, M. L. Mathur, N. C. Mathur* and Arvind Mathur*

Commencement: August, 2007

Duration: Two Years

Status: Ongoing

Objective

1. To study the magnitude and impact assessment of selected musculoskeletal disorders in adults (aged over 18 years) in the community with a focus on osteoarthritis, rheumatoid arthritis and spinal disorders.

Progress of the Work

This is the multi centric study of ICMR task force project. A total of 10 clusters were selected from Jodhpur district, of which five clusters selected from villages of Kaparda PHC of Billara tehsil and five from urban wards of Jodhpur city and Pipar city of Jodhpur. Out of five urban clusters, two clusters were selected from peri urban area of Billara Tehsil. Out of five villages and five urban wards, three villages and three urban wards have been surveyed. A total of 2384 households were covered with family size of 5.7. Out of 8038 individuals of 18 years and above, 6118 (76.1%) individuals were interviewed (Table-3). The coverage was similar in urban and rural areas. The coverage was lower in males (64.1%) as compared to females (88.3%). The list of selected villages and wards with their population is given in Table 1 & 2 as per census 2001. The training of selected staff and reliability exercises were carried out.

The preliminary analysis of data shows the prevalence of musculoskeletal symptoms was 10.2%. The prevalence of musculoskeletal disorders was 10.1% in urban area with 12.5% in females and 6.5% in males while in rural area, it was 10.5% with 8.8% in males and 11.9% in females. The major complaints were joint pain (71.9%) and backache among symptomatics (55.4%).

Table 1. List of selected villages and their population

Villages	Households	Population
Rawar	536	3536
Holpur kalan	171	981
Kaparda	454	2657
Binawas	351	2167
Ramasani	330	2024
Total	18 42	11365

Table 2. List of selected wards and their population from Urban area of Jodhpur District

Ward No.	Households	Population
Pipar city: 3	371	2211
Pipar city: 23	186	1203
Jodhpur: 4	7984	40590
Jodhpur: 26	2099	13117
Jodhpur: 49	4384	126083

*Collaboration: Departments of Orthopedics and Medicine, S.N. Medical College, Jodhpur

Table 3. Percentage distribution of study subjects according to their sex, coverage and residence

Study Subjects	Urban			Rural			Total		
	Male	Female	Total	Male	Female	Total	Male	Female	Total
Total Subjects	2659	2617	5276	1394	1368	2762	4053	3985	8038
Covered	1684 (63.3)	2334 (89.2)	4018 (76.2)	914 (65.6)	1186 (86.7)	2100 (76.0)	2598 (64.1)	3520 (88.3)	6118 (76.1)

Table 4. Age wise prevalence of musculoskeletal symptoms among study subjects according to residence

Age Groups (Yrs.)	Urban		Rural		Total	
	N	Symptoms	N	Symptoms	N	Symptoms
18-30	1754	34 (2.5)	808	26 (3.2)	2562	60 (2.3)
31-40	848	75 (8.8)	447	45 (10.1)	1295	120 (9.3)
41-50	616	100 (16.2)	340	63 (18.5)	956	163 (17.1)
51-60	408	91 (22.3)	274	46 (16.8)	682	137 (20.1)
61-70	265	62 (24.2)	163	32 (19.6)	428	94 (22.0)
71-100	125	42 (33.6)	68	9 (13.2)	193	51 (26.4)
Total	4018	406 (10.1)	2100	221 (10.5)	6118	627 (10.2)

Table 5. Age sex wise prevalence of musculoskeletal symptoms among study subjects of urban area of Jodhpur

Age Groups (Yrs.)	Male		Female		Total	
	N	Symptoms	N	Symptoms	N	Symptoms
18-30	727	12 (1.7)	1027	22 (2.1)	1754	34 (2.5)
31-40	325	13 (4.0)	523	62 (11.9)	848	75 (8.8)
41-50	260	22 (8.5)	356	78 (21.9)	616	100 (16.2)
51-60	182	23 (12.6)	228	68 (29.8)	408	91 (22.3)
61-70	127	20 (15.7)	138	44 (31.9)	265	64 (24.2)
71-100	63	20 (31.7)	62	22 (35.5)	125	42 (33.6)
Total	1684	110 (6.5)	2334	296 (12.5)	4018	406 (10.1)

Table 6. Age sex wise prevalence of musculoskeletal symptoms among study subjects of rural area of Jodhpur

Age Groups (Yrs.)	Male		Female		Total	
	N	Symptoms	N	Symptoms	N	Symptoms
18-30	330	7 (2.1)	478	19 (4.0)	808	26 (3.2)
31-40	191	12 (6.3)	256	33 (12.9)	447	45 (10.1)
41-50	160	18 (11.3)	180	45 (25.0)	340	63 (18.5)
51-60	134	24 (17.9)	140	22 (15.7)	274	46 (16.8)
61-70	64	13 (20.3)	99	19 (19.2)	163	32 (19.6)
71-100	35	6 (17.1)	33	3 (9.1)	68	9 (13.2)
Total	914	80 (8.8)	1186	141 (11.9)	2100	221 (10.5)

Table.7 Distribution of musculoskeletal symptoms according to the residence of subjects

Musculoskeletal Symptoms	Residence		Total N=627
	Urban N=496	Rural N=221	
Joint Pain	293 (72.1)	158 (71.5)	451 (71.9)
Joint Swelling	79 (19.5)	21 (9.5)	100 (15.9)
Joint Stiffness	96 (23.6)	46 (20.8)	142 (22.6)
Muscular Pain	45 (11.1)	22 (10.0)	67 (10.7)
Spine/Back pain	228 (56.2)	121 (54.8)	349 (55.7)

2.2 Jaivigyan Mission Mode Project on Control of Rheumatic Fever and Rheumatic Heart Diseases in Jodhpur district- *Murli L. Mathur, K. R. Haldiya, Sanjeev Sanghvi* and Aruna Solanki***

Commencement: March, 2007

Duration: Two and half years

Status: On going

Objectives

1. To determine the prevalence of RF/RHD in school children of age 5 to 14 years
2. To determine the profile of Group A Beta Hemolytic streptococcal strains prevalent in school children age 5 to 14 years
3. To study the Blood Pressure distribution in school children age 5 to 14 years
4. To establish a registry of RF/RHD cases in selected area
5. To monitor secondary prophylaxis to RF/RHD in selected area
6. To sensitize community for primary prophylaxis by providing relevant health education in selected area

Progress of work

Three adjacent tehsils namely Osian, Bhopalgarh and Bilara with lower socioeconomic development were selected to be included in the study area. The literacy rate in these tehsils is 43.8%, 45.0% and 54.3% respectively. Estimated population of these tehsils in year 2006 was 1019571 as shown in Table 1. The selected area has three community development blocks, two towns and 374 villages. It has 30 primary health centers (PHCs), four Block PHCs, two Community Health Centers and one referral hospital.

Table 1. Population of selected tehsils of Jodhpur district

Tehsil	Population in Year 2001			Expected population in 2006	No. of revenue villages
	Male	Female	Total	Total	
Osian	183431	169434	352865	411088	159
Bilara	129734	121109	250843	292232	92
Bhopalgarh	139909	131551	271460	316251	123
Total	453074	422094	875168	1019571	374

Involvement of State Health Services: Director, Medical and Health Services, Rajasthan, Joint Director, Medical and Health Services Jodhpur Zone and Chief Medical Officer, Jodhpur district were informed and briefed about the project. Co-operation of Medical Officers was sought for the project through official Channel.

*Professor of Cardiology, **Professor and Head, Department of Microbiology, Dr. S. N. Medical College, Jodhpur.

Training Programmes and Workshops Organised:

- Project Staff was trained as per their role in project. The trainings were imparted in biosafety, microbiological work, anthropological measurements, clinical examination of throat, throat swab collection, transportation, inoculation on blood agar, diagnosis of streptococcal sore throat and diagnosis of rheumatic heart disease.



Figure 1. Selected tehsils Osian, Bhopalgarh and Bilara in Jodhpur district

- Seven workshops were organised for Medical Officers working in study area and at Pediatrics Department of Teaching Hospital at Jodhpur. All the participants were briefed about streptococcal sore throat, rheumatic fever, rheumatic heart disease, objectives and activities of the project. They were requested to refer all suspects of rheumatic fever and rheumatic heart diseases to Dr. Sanjeev Sanghvi, Professor of Cardiologist, M. G. Hospital, Jodhpur.
- Two training programmes for health workers were organized. Fiftyfive and seventy workers were present at these programmes respectively. The health workers were explained about symptoms based on which rheumatic fever or rheumatic heart diseases can be suspected. They were briefed about the project and were requested to refer all suspects to the Medical Officer, who in turn would refer the suspects to Cardiologist at M. G. Hospital after examination.

School health survey: List of all schools including private schools, in the selected area was obtained from Offices of Education Department at Jodhpur. These included all primary schools, middle schools, secondary schools and senior secondary schools in the area. Every fifth school from list of all schools in rural area was selected for this purpose.

The team visiting school consisted of one Medical Officer and two field investigators. On reaching school, the team introduced itself to the Headmaster and sought cooperation of class teachers for school health examination. Teachers and students were explained the signs and symptoms of RF/RHD and facilities provided under Jai Vigyan Mission Mode Project. The role of teachers within school as well as in their residential areas for detecting suspected RF-RHD cases was also explained. IEC-1 was distributed to children and teachers. All girls of school were selected for examination and all boys of a class randomly selected. If number of students thus selected were less than 35, one more class was randomly selected and this continued till at least 35 students were selected. The team was seated on two benches viz. BENCH 1 (Field Assistant), BENCH 2 (Medical Officer). On bench one, Field investigator with the cooperation of class teacher, entered the name, age, sex, address etc. of student in school health form and allocated ID No. Another field investigator measured height, weight, waist circumference and hip circumference and recorded the same. Body weight was measured (to the nearest of 0.5 Kg) with the participant in standing position on weighing scale, feet about 15 cm apart and equally distributing weight on both lower limbs with minimum out wear and no footwear. Height was measured with the subject in standing position and with the head positioned so that the top of the external auditory meatus level with the inferior margin of the bony orbit. Waist circumference was measured in centimeters (cm) at the level of umbilicus to the nearest 0.1 cm. Hip circumference was measured at the trochanteric level in cm to the nearest 0.1 cm. MO took history if student was old enough to tell some thing. All students were given a printed paper which contained questions about their medical history. The child was asked to get it filled by his/her parent and return the same to the school teacher the next day. These papers were collected by the team after a few days. MO examined the student clinically and entered findings in school health form. For suspected RF-RHD cases, MO filled up the Referral Card and requested class teacher to call parents of the suspected case, who were then explained about disease of the child and were requested to consult senior cardiologist at M. G. Hospital with the referral card. MO took three readings of BP measurement of all students in sitting position with a minimum gap of 2 minutes per reading. Mercury Sphygmomanometer was used for this purpose. Every third child and the child with sore throat were asked to sit in a separate row for collection of throat swab. At the end MO collected throat swab of the selected children for transferring the same for to the laboratory.

Observations

Table 2. Age and sex distribution of students examined

Age in years	Males	Females	TOTAL	
			No.	Percent
5	199	169	368	5.8
6	270	218	488	7.7
7	290	297	587	9.3
8	379	403	782	12.4
9	468	428	896	14.2
10	474	401	875	13.8
11	455	378	833	13.2
12	403	328	731	11.6
13	282	248	530	8.4
14	131	98	229	3.6
TOTAL	3351	2968	6319	100.0

The team has visited 135 of selected schools in all, out of which fifty were covered before 1st April 2008 and remaining 85 were covered between 1st April 2008 and 31st March 2009. Of these 135 schools, 51 were from Osian tehsil, 34 were from Bhopalgarh Tehsil and 50 were from Bilara tehsil. In all 6319 students of 5-14 years from these schools were examined. Throat swabs were collected from 354 students.

Table 3. Median Weight of students according to age and sex

Age in years	Males			Females		
	N	Median Weight in Kg.	WHO Ref Median	N	Median Weight in Kg.	WHO Ref Median
5	199	17.0	18.5	169	16.0	18.3
6		18.0	20.5	218	17.5	20.2
7		20.4	22.9	297	20.0	22.4
8	378	22.8	25.4	403	21.8	26.0
9		24.9	28.1	428	24.0	28.2
10	474	27.0	31.2	401	27.0	31.9
11	455	28.8	-	378	29.4	-
12	403	31.6	-	328	32.5	-
13	282	34.0	-	248	34.6	-
14	131	37.7	-	98	37.7	-

Table 4. Median height of students according to age and sex

Age in years	Males			Females		
	N	Median Height in Cms.	WHO Ref Median	N	Median Height in Cms.	WHO Ref Median
5	199	109.8	110.3	169	108.3	109.6
6		115.1	116.0	218	113.4	115.1
7		122.3	121.7	297	120.0	120.8
8	378	126.5	127.3	403	125.4	126.6
9		131.5	132.6	428	130.2	132.5
10	474	136.7	137.8	401	135.4	138.6
11	455	140.2	143.1	378	140.7	145.0
12	403	144.7	149.1	328	145.3	151.2
13	282	148.6	156.0	248	147.5	156.4
14	131	152.3	163.2	98	150.5	159.8

Table 5. Median BMI of students according to age and sex

Age in years	Males			Females		
	N	Median BMI	WHO Ref Median	N	Median BMI	WHO Ref Median
5		13.9	15.3	169	13.8	15.2
6		13.7	15.3	218	13.1	15.3
7		13.9	15.5	297	14.0	15.4
8		14.1	15.7	403	13.9	15.7
9		14.3	16.0	428	14.2	16.1
10		14.6	16.4	401	14.7	16.6
11	455	14.7	16.9	378	15.1	17.2
12	403	15.3	17.5	328	15.5	18.0
13	282	15.5	18.2	248	16.0	18.8
14	131	16.2	19.0	98	16.8	19.6

Table 6. Mean hip circumference of students according to age and sex

Age in years	Males		Females	
	N	Mean Hip Circ. in Cms.	N	Mean Hip Circ. In Cms.
5	199	56.9 ± 3.8	169	56.4 ± 3.8
6	270	58.5 ± 3.9	218	58.2 ± 3.9
7	290	60.0 ± 3.7	297	60.2 ± 4.2
8	378	62.2 ± 4.5	403	62.0 ± 4.6
9	468	64.0 ± 4.6	428	64.4 ± 5.3
10	474	66.3 ± 5.3	401	67.0 ± 5.9
11	455	68.0 ± 5.6	378	69.9 ± 6.3
12	403	70.5 ± 6.1	328	72.2 ± 6.3
13	282	72.1 ± 5.6	248	74.2 ± 6.3
14	131	74.5 ± 6.3	98	76.4 ± 5.8

Table 7. Mean waist circumference of students according to age and sex

Age in years	Males		Females	
	N	Mean Waist Circ. In Cms.	N	Mean Waist Circ. in Cms.
5	199	50.0 ± 3.4	169	48.8±3.4
6	270	50.8 ± 3.9	218	50.2±4.1
7	290	51.8±3.4	297	51.1±3.9
8	378	53.3±3.9	403	52.1±4.3
9	468	54.2±4.1	428	53.2±4.2
10	474	56.1±4.5	401	55.7±5.1
11	455	57.1±5.1	378	57.5±5.7
12	403	59.5±5.7	328	59.2±5.5
13	282	60.7±4.9	248	60.7±5.7
14	131	62.7±5.4	98	61.2±5.7

Table 8. Mean waist hip ratio (WHR) of students according to age and sex.

Age in years	Males		Females	
	N	Mean WHR	N	Mean WHR
5	199	0.88±0.03	169	0.87±0.04
6	270	0.87±0.04	218	0.86±0.05
7	290	0.86±0.04	297	0.85±0.04
8	378	0.86±0.04	403	0.84±0.05
9	468	0.85±0.04	428	0.83±0.04
10	474	0.85±0.05	401	0.83±0.04
11	455	0.84±0.04	378	0.82±0.05
12	403	0.84±0.06	328	0.82±0.05
13	282	0.84±0.05	248	0.82±0.05
14	131	0.84±0.05	98	0.80±0.05

Table 9. Distribution of school children (5-14) according to Waist-Hip Ratio (WHR)

Percentile	WHR	
	Males	Females
5th percentile	0.78	0.75
10 th percentile	0.80	0.77
20 th percentile	0.82	0.79
30 th percentile	0.83	0.81
40 th percentile	0.84	0.82
50 th percentile	0.85	0.83
60 th percentile	0.86	0.85
70 th percentile	0.87	0.86
80 th percentile	0.89	0.87
90 th percentile	0.91	0.89
95 th percentile	0.92	0.91

Table 10. Mean Systolic Blood Pressure of students according to age and sex

Age in years	Males		Females	
	N	Mean SBP in mm Hg	N	Mean SBP in mm Hg
5	199	109.4±11.3	169	109.3±11.8
6	270	109.9±12.0	218	112.4±11.8
7	290	112.1±11.7	297	111.4±11.4
8	378	113.5±10.7	403	112.4±12.0
9	468	113.8±10.5	428	114.7±11.4
10	474	114.4±12.7	401	115.4±11.0
11	455	115.3±10.8	378	117.6±10.6
12	403	116.2±10.5	328	118.7±11.3
13	282	117.5±11.3	248	118.7±10.5
14	131	116.4±10.4	98	118.7±11.2

Table 11. Mean Diastolic Blood Pressure of students according to age and sex

Age in years	Males		Females	
	N	Mean DBP in mm Hg	N	Mean DBP in mm Hg
5	199	69.0 ± 6.9	169	68.7 ± 6.6
6	270	69.4 ± 6.9	218	73.6 ± 5.0
7	290	71.0 ± 7.1	297	70.9 ± 6.4
8	378	71.6 ± 7.2	403	71.5 ± 7.3
9	468	72.0 ± 6.4	428	72.8 ± 7.0
10	474	73.0 ± 7.0	401	72.8 ± 7.3
11	455	73.2 ± 6.8	378	74.5 ± 7.0
12	403	74.1 ± 7.0	328	74.6 ± 6.7
13	282	74.8 ± 7.2	248	74.9 ± 6.4
14	131	74.0 ± 6.8	98	75.2 ± 6.9

Throat swabs were collected and transported to the laboratory where these were inoculated on blood agar. Throat swabs collected during July to September were processed same day as this was suited to school timings. From October, 2007 to December 2007 as the school time was changed the samples were collected and transferred to filter paper strips which could be processed within one week of sample collection. Any colony of Beta Hemolytic Streptococcus did not grow from throat swabs up to December 2007.

From January 2008 blood agar plates were carried to schools, where they were directly inoculated from the throat swabs immediately after collection of throat swab. Then 3 or 4 sets of streaks were spread from the primary inoculum with a sterile wire loop to obtain well-spaced single colonies. The plates were then incubated overnight at 37°C in Candle jar.

After required time of incubation the plates were observed for morphological identification of the colonies and the appearance of hemolytic zone around the colonies of beta hemolytic streptococci (BHS). Gram's staining was done for microscopic examination of colonies. The observations were recorded in laboratory reporting forms. Morphological and microscopic

observation of colonies appeared on blood agar plate showed variety of microorganism including yeast, alpha hemolytic bacteria, gram positive bacilli, gram negative cocci.

The growth of Beta-hemolytic streptococci was observed in plates prepared from nine throat swabs after From January 2008 to March 2008. All these nine colonies were sent to PGI Chandigarh. These were of Gr. A/C/G beta-hemolytic streptococci. However, many other plates showed growth of alpha-hemolytic streptococci, yeast and fungi. Out of these nine strains eight were of GAS. From April 2008 to March 2009, forty more throat swabs yielded growth of GAS. All these strains of sent to PGI.

Registered RF/RHD cases

Among 167 registered cases proportion of females was 55.7%. According to severity of RHD disease at time of registration, 61.5% were severe cases, 27.0% moderately severe and 11.5% mild.

Table 12. Patients registered in different categories

Categories		No. of patients
1A.	ARF with carditis	02
1B.	ARF without carditis	01
2A.	RHD with rheumatic activity	05
2B.	RHD without rheumatic activity	156
3A	Documented past RF without RHD	02
3B.	Not Documented past RF without RHD	01
Total no. of RF/RHD cases registered		167

Table 13. Age and sex wise trend of registered RF/RHD Cases

Age Group	No. of RF/RHD Cases			
	Males	Females	Total	
			No.	%
0-4	0	0	0	0.0
5-9	0	0	0	0.0
10-14	10	11	21	12.6
15-19	6	4	10	6.0
20-24	7	10	17	10.2
25-29	7	10	17	10.2
30-34	8	15	23	13.8
35-39	8	9	17	10.2
40-44	8	11	19	11.4
45-49	8	5	13	7.8
50-54	4	2	6	3.6
55-59	2	3	5	3.0
60-64	3	3	6	3.6
65-69	1	0	1	0.6
70-74	0	0	0	0.0
75-79	0	0	0	0.0
80 & above	0	1	1	0.6
TOTAL	74	93	167	100.0

Table 14. Month wise registration of RHD cases

S. No.	Month	No. of RF/RHD Cases registered
1.	May 2007	06
2.	Jun 2007	06
3.	Jul 2007	05
4.	Aug 2007	03
5.	Sep 2007	04
6.	Oct 2007	04
7.	Nov 2007	16
8.	Dec 2007	13
9.	Jan 2008	12
10.	Feb 2008	10
11.	Mar 2008	10
12	Apr 2008	12
13	May 2008	08
14	Jun 2008	02
15	Jul 2008	11
16	Aug 2008	06
17	Sep 2008	08
18	Oct 2008	02
19	Nov 2008	03
20	Dec 2008	04
21	Jan 2009	08
22	Feb 2009	07
23	Mar 2009	07
	Total	167

Table 15. Final diagnosis (ECHO diagnosis) of RF/RHD patients seen by Cardiologist according to Specific Valve involvements

RHD- Valve involved	Males	Females	Total
Mitral Alone	40	55	95
Mitral+Aortic	14	13	27
Mitral+Aortic+Tricuspid	6	10	16
Mitral+ Tricuspid	9	11	20
Aortic alone	3	0	3
RF (Documented/Undocumented)	2	4	6
Total	74	93	167

Monitoring of Secondary Prophylaxis (SP):

Out of 167 registered cases of RF/RHD, thirteen have died, 26 are not advised by cardiologist to take SP as they are above age of 45 years or have other problem. Out of remaining 128 patients put on SP, information from 86 (67.2 %) subjects, is available that all of them have taken last dose of due SP in April 2009, none of them have stopped SP. Two patients have migrated and could not be followed. Remaining 32.8% could not be contacted and information about them is not available, whether they have taken last due dose of SP or not.

Important Leads and Outcome

Preliminary data indicate that prevalence of RF/RHD in school age group is around one per thousand in the selected area. The data about blood pressure, height, weight, BMI, waist circumference, hip circumference and WHR are expected to reveal more.

2.3 Dermatoglyphic Patterns in Diabetes Mellitus Patients and Non-Diabetics: A preliminary study- *P. K. Dam, Vinod Joshi, Anil Purohit and Himmat Singh*

Commencement : June, 2006

Duration: Three years

Status: Ongoing

Objectives

1. To identify patterns of dermal ridges on the fingertips and palms of diabetics and non-diabetics
2. To focus the predictive strength of dermatoglyphics of diabetics.

Progress of the Work.

This was suggested during 17th SAC, that the present study should explore the diabetic situation in other communities, in addition to the Mathur families. Therefore, attempt was made to collect data pertaining to (a) Anthropometric Measurements (height, weight, waist & hip circumference) to calculate BMI & wist-hip ratio and (b) Family history of Diabetes (to assess the occurrence of Diabetes in first / second degree relatives) were undertaken among 70 respondents belonging to Brahmin community and 70 respondents belonging to Mathur Community. Out of these, in present Annual Report, findings of 50 respondents each from Brahmin (case: 26, control: 24) and Mathur (case:17, control:33) communities have been analyzed and presented (Table1-4).

Among the cases in 34-41 yrs. age group,5.9% and 11.5% of the male respondents were observed as diabetic in Brahmin and Mathur families respectively, whereas no female diabetic case were observed among the Brahmins in 34-41 yrs. age group. Among the Mathurs,5'9% female diabetic cases were observed in 34-41 age group (Table 4).BMI profile among the diabetics of both the communities reveal that among the Mathurs 'Obese ii' case have not been observed in 34- 41 years and > 42 years age group respectively. Among the Brahmins, 7.7% and 23.1 % are observed as 'Obese ii' in 34- 41 years age group & >42 years age group.

Contrastingly, 17.6% and 5.9% of the diabetics in >42 yrs. age group among the Mathurs are observed as having 'low weight normal' and 'CED 1' BMI respectively, which are not found among the Brahmins. >50% of the cases in both the study groups are found as having 'Obese 1' and 'Obese ii' BMI in >42 yrs. age group (Table.3).

Out of 105 families of Mathurs (total family members:500), 78 were found as afflicted with T2DM,and 64.8% was having family history of T2DM (Fig.1).Out of 78 families of Jain groups (total family members:343),60 were total T2DM cases and 70.5% revealed family history positive for T2DM.Out of 80 families of the Brahmin group,64 were total T2DM cases,and 68.8% was having positive family history for T2DM. This indicates that current occurrence of T2DM is no less than others, but family history suggests that T2DM is more contemporary amongst the Mathurs.

Table 1. Frequencies of finger pattern types in 19 female T2DM cases and 12 Female controls

Pattern	T2dm		Controls	
	Total	%	Total	%
Whorls	72	38.3	60	50.0
Ulnar loop	112	59.6	51	42.5
Radial loop	3	1.6	5	4.2
Arches	1	0.5	4	3.3
Total readable prints	188		120	

Table 2. Frequencies of finger pattern types in 15 male T2DM cases and 8 male controls

Pattern	T2dm		Controls	
	Total	%	Total	%
Whorls	85	56.7	46	57.5
Ulnar loop	62	41.3	32	40.0
Radial loop	0	0	1	1.25
Arches	3	2.0	1	1.25
Total readable prints	150		80	

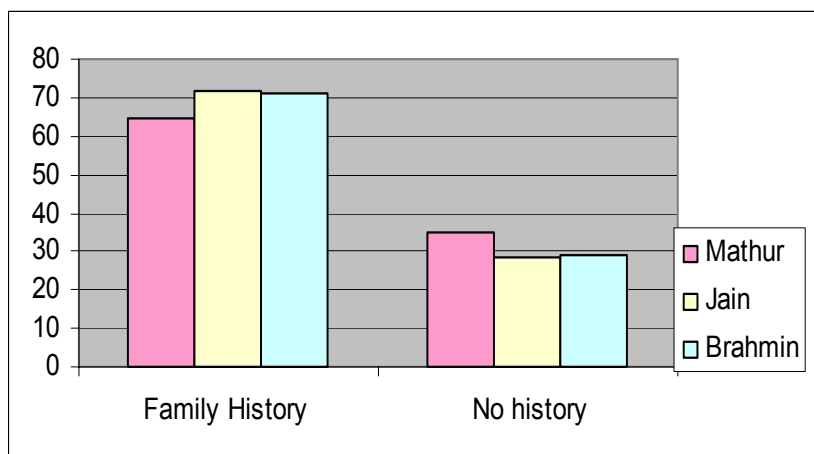
Table 3. Percentage of age wise distribution of BMI among Diabetics of Brahmin and Mathur

BMI	Brahmin (n = 26)				Mathur (n = 17)			
	18 - 25	26 - 33	34 - 41	> 42	18 - 25	26 - 33	34 - 41	> 42
17- 18.5 (CED 1)	0	0	0	0	0	0	0	5.9
18.5 – 20 (Low weight normal)	0	0	0	0	0	0	5.9	17.6
20 – 25 (Normal)	0	0	0	7.7	0	0	0	11.8
25 – 30 (Obese 1)	0	0	3.8	57.7	0	0	5.9	52.9
> 30 (Obese ii)	0	0	7.7	23.1	0	0	0	0

Table 4. Percentage of age and sex distribution of T2DM cases among the respondents of Brahmin and Mathur Families.

Age group	Brahmins (n = 26)			Mathur (n=17)		
	Male	Female	Total	Male	Female	Total
18-25	0	0	0	0	0	0
26-33	0	0	0	0	0	0
34- 41	11.5	0	11.5	5.9	5.9	11.8
>42	53.8	34.6	88.5	47.0	41.2	88.2

Figure 1. Frequency distribution of Family History of T2DM in different communities



Community	Family History	No history
Mathur	64.8	35.2
Jain group	71.6	28.4
Brahmin group	71.2	28.8

2.4 The Anthropological Survey of India Study of Bio-Cultural Risk Factor Assessment for Type 2 Diabetes: Consortium for the Family Studies of Genetics of Type 2 Diabetes in Indian Populations- *A. P. Dash, P. K. Dam and K. R. Haldiya*

Commencement : June, 2008

Duration: Three years

Status : On going

Objectives

1. Initiate a consortium for multidisciplinary genetic epidemiologic studies of type 2 diabetes (T2DM) and its correlated disease conditions such as obesity and the metabolic syndrome (MS) in three genetically and culturally diverse endogamous caste communities at high risk for T2DM and related phenotypes from the states of Karnataka, Andhra Pradesh, and Rajasthan by establishing a mutually beneficial collaboration between Indian and US scientists that have extensive experience in the fields of anthropology, biochemistry, endocrinology, epidemiology, genetics, medicine, nutrition, physical education, public health, and psychology. These activities will be carried out as part of the Anthropological Survey of India Study of Bio-Cultural Risk Factor Assessment for Type 2 Diabetes.
2. Conduct a complex pedigree-based study from each of the three communities to examine the genetic determination of susceptibility to T2DM and its related disease conditions by recruiting 1,000 individuals from about 50 large families from each community that will be ascertained on type 2 diabetic probands. Approximately 50 families with a minimum of 20 individuals per pedigree will be included. Thus, for a given community, the total target is 1,000 individuals. All first, second, and third degree relatives of probands aged 18 years or above will be asked to participate in a given study.
3. Collect phenotypic information relating to T2DM and its related phenotypes such as obesity, adipocytokines, impaired fasting glucose, impaired glucose tolerance, insulin resistance, dyslipidemia, albuminuria (micro and macro), markers of non-alcoholic fatty liver disease, markers of inflammation, hypertension, and MS.
4. Collect information on shared and non-shared environmental factors, including household information, diet (24-hr recall method), physical activity, cigarette smoking, alcohol consumption, socioeconomic status, and psychological/behavioral attributes (e.g., depression, anxiety, and positive well-being).
5. Perform the whole-genome genotyping using the new Illumina Human1M BeadChip which contains information from approximately 1 million single nucleotide polymorphisms (SNPs) and generate a high-density SNP-based linkage panel (>6,000 SNPs).
6. Localize genes influencing variation in T2DM, obesity, MS, and their related phenotypes by uniquely conducting both association- and linkage-based genome screenings for a given phenotype using extended pedigree data and variance components analytical technique and its extensions.

7. Perform multipoint multivariate linkage analysis to identify genetic locations with common genetic influences (i.e., pleiotropy) on a given trait-pair (e.g., T2DM and obesity) and conduct a modified multipoint linkage analysis that examines genotype-by-environment (e.g., physical activity or diet) influences on T2DM and its related phenotypes.
8. As a part of T2DM gene discovery efforts, perform a preliminary analysis to identify the most likely functional variations within the two best positional candidate genes for T2DM identified in Aim 6. Selected SNPs that are most strongly associated with T2DM will be validated in a replication sample.
9. Establish a resource for long term maintenance and storage of DNA and cell lines.

Progress of the Work

Study design, materials and methods were determined during 1st Meeting held on June, 2008.

Meeting of the Ethics Committee held at Desert Medicine Research Centre, Jodhpur, on 22 August, 2008 and ethical clearance was granted to the study by Ethics Committee.

Finalization and field testing the schedules of Demographic / Pedigree chart / Anthropometric data were accomplished during first workshop organized at DMRC Jodhpur in November, 2008 along with practical demonstration of Auto Analyzer.

Thereafter, demographic/pedigree data collection by finalized schedules were initiated among the recruited probands of Mathur families that were previously identified as having T2DM & inhabiting Jodhpur city / adjoining areas. Blood sugar status determination of the family members of the probands were also undertaken.

The protocols/schedules on Dietary intake/ Psychological status/ Physical activity, were finalized in the Second workshop during February, 2009, conducted at Andhra University, Visakhapatnam. In DMRC Jodhpur, the finalized version of Psychological Schedules were translated into Hindi following the standardized protocol recommended by Anthropological Survey of India.

Following this, it was decided in the 2nd workshop that the Third workshop will be conducted at Mysore to discuss and finalize the protocols/schedules of biochemical and microbiological investigations, during June 2009. Memorandum Of Agreement (MOA) will be signed between Anthropological Survey of India and the six participating Institutes including DMRC Jodhpur during Third Workshop.

To cover 1000 individuals from 50 proband's family (sample size that was decided for DMRC Jodhpur to cover), data collection for pedigree and demographic aspect is ongoing, out of which family data from four (4) probands have been collected, analyzed and presented (Table.1).

Table. 1. Frequency distribution of T2DM cases and non-diabetics in bilateral pedigrees of 4 probands along with deduction factor.

Proband	T2DM Male	T2DM Female	Control male	Control female	Total Respondents in the family (Family=House hold+kin)	Deduction factor				
						Deceased	below 18 yrs	Respondents migrated	Non cooperative families	Total Respondents interviewed
1	22	24	123	110	279	70	26	52	35	96
2	10	15	95	84	204	22	17	37	20	108
3	15	11	111	92	229	30	14	72	5	108
4	1	7	62	45	115	5	0	21	0	89
Total	48	57	391	331	827	127	57	182	60	401

3.1 Nutrition Monitoring Survey on NNMB pattern in Jodhpur district of Rajasthan - *Madhu B. Singh, J. Lakshminarayana and Ranjana Fotedar*

Commencement: January, 2005

Duration: Long Term

Status: Ongoing

Objectives

1. To develop continuous monitoring service to study the nutritional status, dietary habits, food availability and the effect of changing social and environmental factors on the health status of the population.
2. Aim at doing comparisons with other states data so as to assess the percentage of variation among the states.

Progress of the work

Project activities included working on sampling plan in detail. Similar sampling design and protocol was adopted for the Nutrition Monitoring type of survey for carrying out in Rajasthan, as it is being done in other states where NNMB is in operation. The sampling adopted here was two stage stratified random sampling method in which the villages in selected district formed the first stage units (FSU's) while in the village households (HH's) formed the second stage units. For the study purpose the district has been divided in to different strata in rural areas as per the tehsils with agro-economic regions and based on the population size of the village i.e. <2000 and ≥ 2000 populated villages. In the urban area three wards were selected.

From each stratum i.e. Tehsil, five villages were chosen randomly for the purpose of the survey in different direction one each from North, South, East, West and central part, to have proper representation of the tehsils in the district. Households in each village have been selected by adopting cluster sampling procedure for the purpose of the survey. A total of five clusters of four households each were selected from each village. Generally, the households in a village can be divided into natural "groups/areas" by geographical location such as streets/mohallas/areas. The SC/ST population often lives in a separate group/area in the villages. One cluster was selected from SC/ST group/area while the remaining 4 clusters were selected by systematic random sampling procedure, probability proportion to size of the group. In each cluster, by selecting a random start, 4 contiguous households were covered. For logistic reasons Jodhpur district was decided to be covered first and later on to expand horizontally in other districts of the state in the similar pattern.

Second phase has been completed in which 30 villages were covered from six tehsils of Jodhpur district i.e. Jodhpur, Osian, Phalodi, Shergarh, Bilara and Bhopalgarh (five villages from each tehsil), covering 600 households. Third phase has been started in which 50 percent new villages and 50 percent old villages have been selected randomly in view to cover variation. A total of 25 villages were covered from five tehsils of Jodhpur district i.e. Jodhpur, Osian, Shergarh, Bilara and Bhopalgarh, covering 500 households. The entire selected household was examined for Socio-demographic and Socio- economic aspects. All the members in the household have been examined for nutritional deficiency signs, anthropometric measurements (Height, weight, arm circumference and FFT), Dietary intake (24 hours recall method) and examination of nutritional

morbidities in preceding 15 days. Dietary intakes of the individuals information were recorded in alternate houses i.e.10 households from each village are covered.

Analysis of 600 households covering 3548 individuals of the second phase has been done. Table 1 showed age and sex wise distribution of population (1802 males and 1746 females). Analysis revealed that 93.8 percent of population was Hindus and 5.0 percent Muslims. Nuclear families were significantly more (68.7 %) as compare to 20.0 % joint families (Tables 2). Table 3 revealed that illiteracy is significantly high in females (59.2 %) than males (33.2 %). Higher education (Above 12th class) is very low in this area.

Table 1. Age and sex wise distribution of population covered

Age group	Males	%	Females	%	Total	%
0-5	292	16.2	289	16.6	581	16.4
6-9	190	10.5	188	10.8	378	10.7
10-14	251	13.9	256	14.7	507	14.3
15-17	128	7.1	114	6.5	242	6.8
18– 29	355	19.7	336	19.3	691	19.5
30-39	214	11.9	202	11.6	416	11.7
40-49	157	8.7	153	8.8	310	8.7
50-59	100	5.6	100	5.7	199	5.6
>=60	115	6.3	108	6.2	222	6.3
Total	1802	100.0	1746	100.0	3548	100.0

Table 2. Distribution of households according to type of family

Type of family	N	%
Nuclear	412	68.7
Extended Nuclear	68	11.3**
Joint	120	20.0**
Pooled	600	100.0

** P<0.01

Table 3. Distribution of population according to educational status

Educational status	Males	%	Female	%	Total	%
Illiterate	599	33.2	1034**	59.2	1633	46.0
Read & Write	19	1.1	18	1.0	37	1.0
1-4 Standard	499	27.7	387**	22.2	886	25.0
5-8 Standard	302	16.7	151**	8.7	453	12.8
9-12 Standard	247	13.7	46**	2.6	293	8.3
College	59	3.3	13**	0.7	72	2.0
N. A.	77	4.3	97	5.6	174	4.9
Pooled	1802	100.0	1746	100.0	3548	100.0

** P<0.01

Main morbidities observed in population were acute respiratory infection (11.3 %) and fever (8.5 %) followed by diarrhea (1.4 %). Both the morbidities i.e. acute respiratory infection and fever were significantly higher in females than males (Table 4). Regarding nutritional deficiency signs, it is observed that discoloration and sparseness of hair, a sign of protein calorie malnutrition was observed to be high i.e. 7.0 percent which was significantly higher in females than males. Marasmus was observed only in females (0.05 %). Angular stomatitis, cheliosis and glossitis were ranging from 0.6 to 2.9 percent. Vitamin A deficiency (Night blindness) was 0.2 percent, higher in females than males. Dental caries (33.8 %) and dental fluorosis (24.3 %) observed high in this area. Females suffered significantly more from dental caries and dental fluorosis than males (Table 5). Koilonychia, a sign of anemia, was observed only in females (0.1 %).

Table 4. Distribution of population according to Morbidity

Morbidity	Males N=1802	%	Females N= 1746	%	Total N=3548	%
N.A.D.	1505	83.5	1310	75.0	2815	79.3
Fever	110	6.1	191**	10.9	301	8.5
Diarrhoea	20	1.1	30	1.7	50	1.4
Dysentery	4	0.2	8	0.5	12	0.3
A. Res. Infection	181	10.0	219*	12.5	400	11.3
Measles	0	0.0	1	0.1	1	0.0
GIT	50	2.8	62	3.6	112	3.2

** P<0.01 * P<0.05

Table 5. Distribution of population according to Nutritional deficiency signs

Deficiency Signs	Males N=1802	%	Females N= 1746	%	Total N=3548	%
Hair Discolour	70	3.9	180*	10.3	250	7.0
Hair Sparseness	3	0.2	7	0.4	10	0.3
Emaciation	0	0.0	2	0.1	2	0.05
Marasmus	0	0.0	1	0.05	1	0.02
Night blindness	3	0.2	5	0.3	8	0.2
Angular Stomatitis	6	0.3	14*	0.8	20	0.6
Chelosis	44	2.4	60*	3.4	104	2.9
Glossitis	19	1.1	31	1.8	50	1.4
Koilonychia	0	0.0	2	0.1	2	0.05
Gums-spongy Bleeding	16	0.9	41*	2.3	57	1.6
Dental Caries	441	24.5	759*	43.5	1200	33.8
Dental Fluorosis	330	18.3	531*	30.4	861	24.3

* P<0.01

The weights of pre-school children were expressed as percent of NCHS standards and categorized into different nutritional grades, based on Gomez classification (Tables 6-7). The overall prevalence of under nutrition was very high i.e. 79.1 percent which was higher in ST community (89.2 %) in comparison to OBC communities (75.8 %). The overall prevalence of severe under nutrition was high i.e. 6.9 percent. Under nutrition was higher in joint

families (82.1 %) and observed maximum in kutcha houses (80.4 %) followed by pucca houses though statistically insignificant. The percent prevalence of severe under nutrition was higher in low income group (9.8 %) than high income group where as reverse trend was observed in case of mild under nutrition i.e. more in high income group (53.3 %) (Fig. 1).

Table 6. Distribution of 1-5 years children according to Gomez distribution and Type of family

Type of Family	N	Nutritional Grades*			
		Normal	Mild	Moderate	Severe
1. Nuclear	250	20.8	42.8	30.8	5.6
2. Extended	43	27.9	34.9	27.9	9.3
3. Joint	95	17.9	41.1	31.6	9.5
Pooled	388	20.9	41.5	30.7	6.9

$\chi^2_{1,2} = 23.2$ $P < 0.01$

$\chi^2_{1,3} = 36.2$ $P < 0.01$

$\chi^2_{2,3} = 10.4$ $P < 0.01$

* NCHS Standards

Fig. 1. Gomez Classification according to Per Capita Income (Rs.) in preschoolers

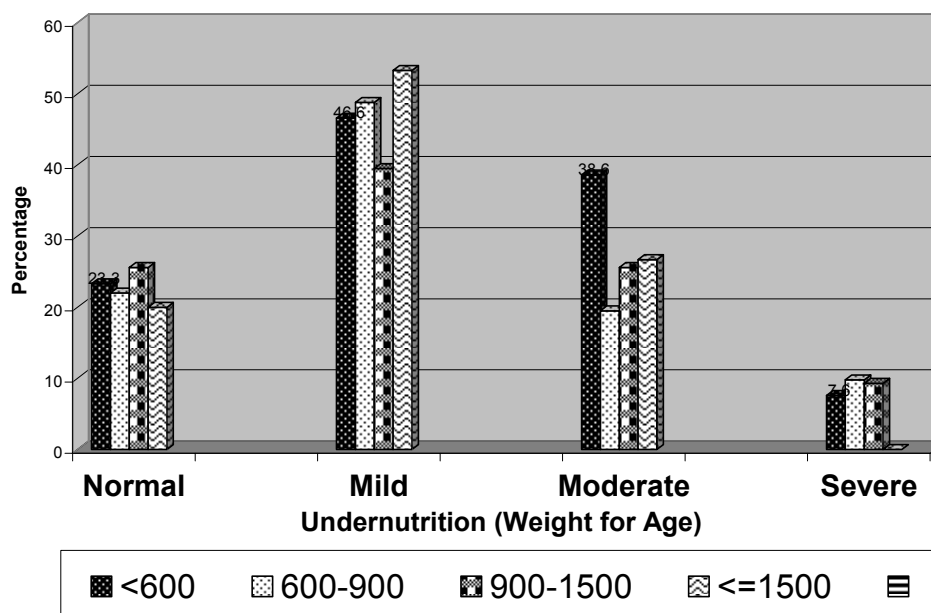


Table 7. Distribution of 1-5 years children according to Gomez distribution and type of house

Type of House	N	Nutritional Grades*			
		Normal	Mild	Moderate	Severe
1. Kutcha	46	19.6	34.8	43.5	2.1
2. Semi Pucca	159	22.0	40.9	28.9	8.2
3. Pucca	183	20.2	43.7	29.0	7.1
Pooled	388	20.9	41.5	30.7	6.9

$P > 0.05$, * NCHS Standards

The extent of different types of malnutrition viz. stunting (Height for age) and under nutrition (Weight for age) were computed by adopting standard deviation classification using NCHS standards. All the children with any of the above anthropometric measurement less than Median-2SD of NCHS values were considered as undernourished. Prevalence of under nutrition computed using Gomez classification and SD classification differ as the cut off values are different.

Stunting (Height for age) was 62.1 percent in preschoolers with the prevalence of severe stunting 39.4 %, which needs attention. It's higher than NNMB (49.3 %) and NFHS III (33.7%- up to 3 years) where as lower than DMRC Phase I (71.6 %) as shown in Fig. 2.

Underweight (Weight for age) in preschoolers observed 59.5 %, higher than NFHS III (44.0 %). The proportion of severe underweight was high (28.8 %) (Fig. 3) Both stunting and underweight were observed higher in males than females though statistically insignificant. Declining trend has been observed in stunting and underweight in comparison to Phase one study.

Fig. 2. Standard Deviation Classification for Height for Age in preschoolers

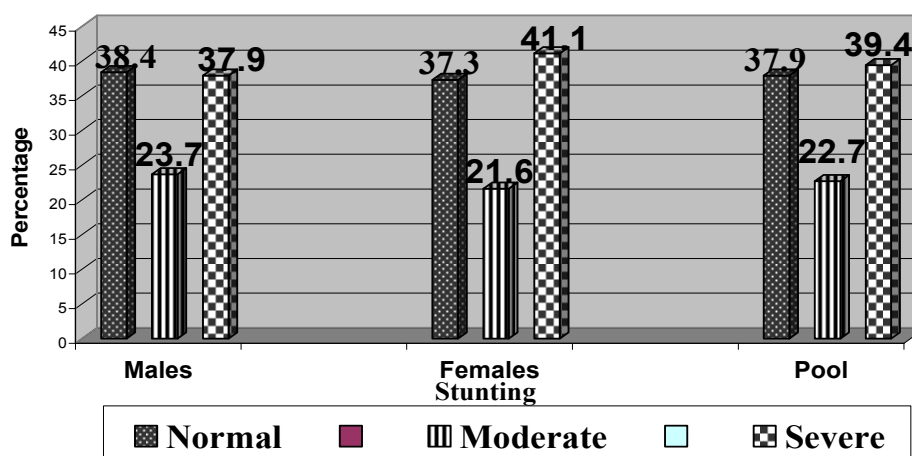
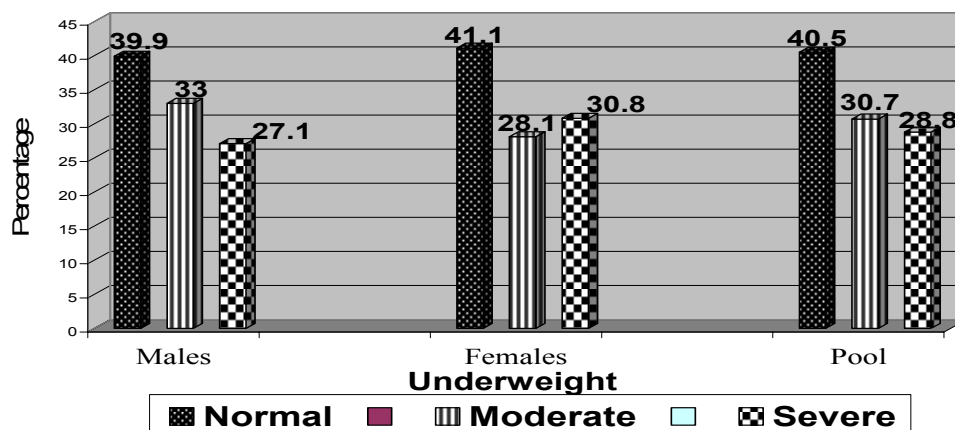


Fig. 3. Standard Deviation Classification for Weight for Age in Preschoolers



The distribution of adults according to BMI grades have been shown in Tables 8 to 10. At the aggregate level, 54.8 percent had normal BMI (18.5-25.0), while 34.4 percent had chronic energy deficiency (Fig. 4). Severe chronic energy deficiency was significantly higher in joint families (9.5 %) than nuclear families (6.1 %) and maximum in pucca houses (13.3 %). Chronic Energy deficiency declined with increase of per capita income i.e. 41.2 percent in low income (<300 Rs.) and 31.8 percent in high income group (≥ 1500 Rs.).

Table 8. Distribution of adults (≥ 18 years) according to BMI classification

Type of Family	Obese II ≥ 30	Obese I 25-30	Normal 20-25	Low wt Normal 18.5-20	CED I 17-18.5	CED II 16-17	CED III <16
Male N=316	2.2	6.3	36.4	19.0	19.0	9.2	7.9
Female N=657	3.2	8.7	32.9	21.6	17.4	9.6	6.6
Pooled N=973	2.8	7.9	34.0	20.8	17.9	9.5	7.0

$$\chi^2_{1,2} = 84.2 \quad P < 0.01$$

Fig. 4. Body Mass Index in adults

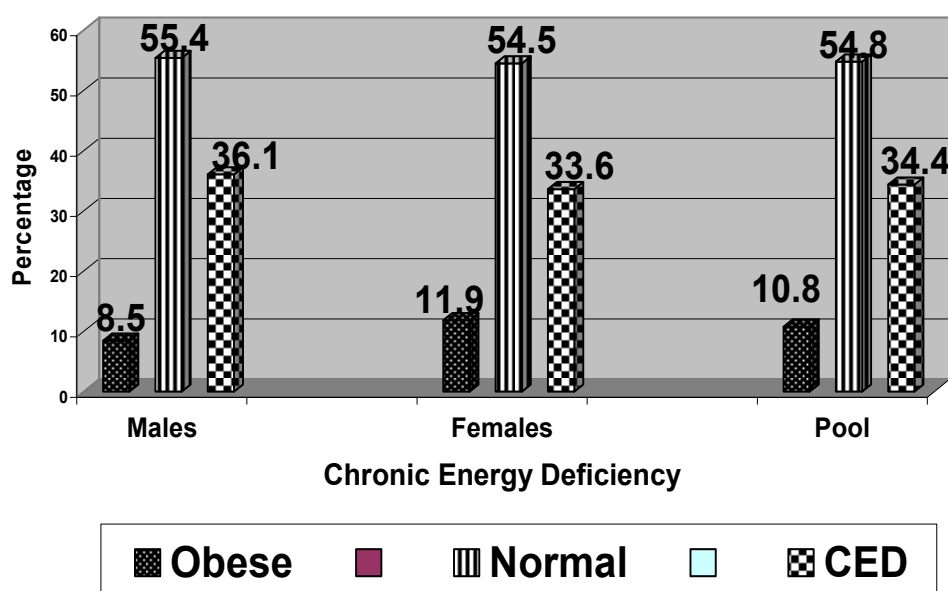


Table 9. Distribution of adults (≥ 18 years) according to BMI classification and Type of family

Type of Family	Obese II ≥ 30	Obese I 25-30	Normal 20-25	Low wt Normal 18.5-20	CED I 17-18.5	CED II 16-17	CED III <16
1.Nuclear N=575	2.5	7.5	33.6	21.7	18.6	9.9	6.1
2.Extended N=136	2.2	8.8	32.4	20.6	18.4	11.0	6.6
3. Joint N=262	3.8	8.4	35.9	18.8	16.0	7.6	9.5
Pooled N=973	2.8	7.9	34.0	20.8	17.9	9.5	7.0

$$\chi^2_{1,2} = 114.0 \quad P < 0.01 \quad \chi^2_{1,3} = 101.3 \quad P < 0.01 \quad \chi^2_{2,3} = 44.0 \quad P < 0.01$$

Table 10. Distribution of adults (>=18 years) according to BMI classification and type of house

Type of House	Obese II >=30	Obese I 25-30	Normal 20-25	Low wt Normal 18.5-20	CED I 17-18.5	CED II 16-17	CED III <16
1.Pucca N=75	1.3	4.0	24.1	22.7	21.3	13.3	13.3
2.Kutchha N=351	0.6	6.3	30.2	26.4	20.5	9.1	10.0
3. Mixed N=547	4.6	9.5	37.8	18.9	15.7	9.1	4.4
Pooled N=973	2.8	7.9	34.0	20.8	17.9	9.5	7.0

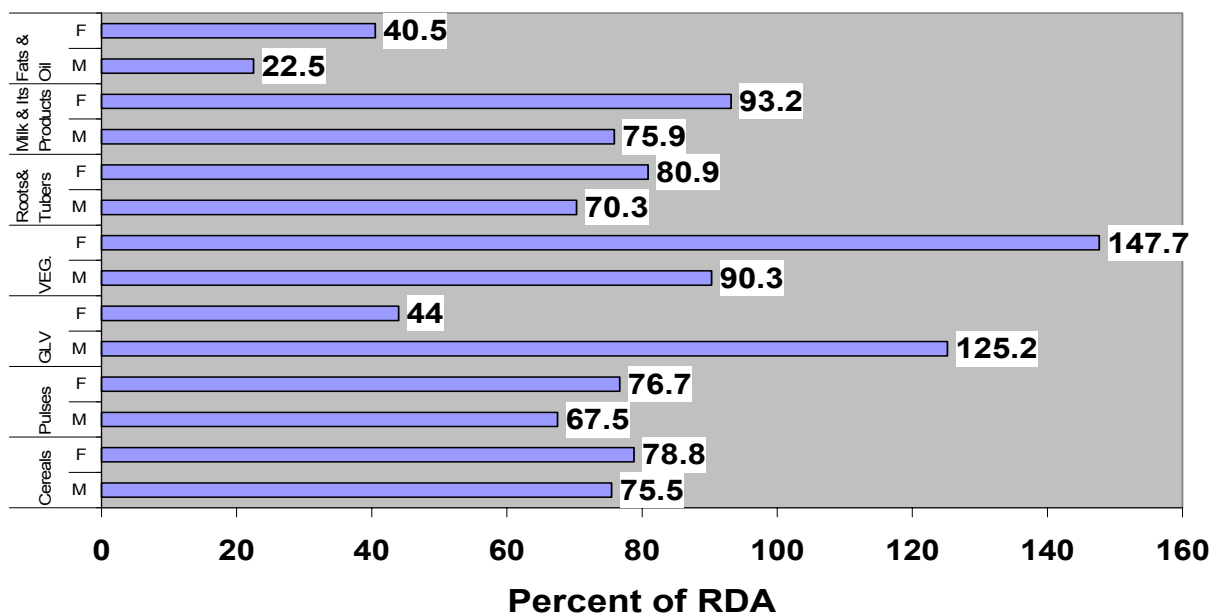
$$\chi^2_{1,2} = 9.79 \quad P < 0.01 \quad \chi^2_{1,3} = \text{NS} \quad \chi^2_{2,3} = 317.5 \quad P < 0.01$$

Dietary intake was collected on 873 adults (439 males and 434 females) and 759 children (375 males and 384 females). Dietary analysis revealed that diet of adults was marginally low in consumption of Cereals & Millets i.e. 75.5 and 78.8 percent of RDA in males and females respectively (Fig. 5) where as 80 percent in DMRC phase I. Deficit was more in males (24.5 %) than females (21.2 %). In children deficit ranging from 6.6 to 34.0 and 4.0 to 31.0 percent in males and females respectively. Maximum deficit was in 4-6 years age group. Consumption of Pulses & Legumes was very low in children i.e. deficit ranges from 35.6 to 64.4 % of RDA where as in adults it was 23.3 to 32.5 percent. In consumption of Fats & Edible Oil, deficit ranges from 49.6 to 89.2 % of RDA in children where as in adults it was 68.5 percent. Green Leafy Vegetables deficit was more in children and adult females i.e. ranges from 19.1 to 60.4 percent of RDA in children and 56.0 percent in adult females. Consumption of Roots & Tubers meets the requirement in most of age groups. Deficit in consumption of milk and its products was more in children than adults i.e. 31.3 to 60.0 percent in children where as 15.4 percent in adults.

Average energy intakes (calories) was less than RDA in all age groups i.e. 1-3, 4-5, 7-9, 10-12, 13-15, 16-17 years and adults ranging from 53.5 to 85.9 percent, lower than DMRC phase I (59.0 to 99.2 %). Calorie deficit was observed more in children (14.1 to 46.5 %) than adults (19.7 to 27.9 %) and observed highest in 1-3 years group (42.1 to

46.5 %) followed by 4-6 and 7-9 years groups. Deficiency in protein intakes was observed only in 13-15 (13 %) followed by 16-17 years and 10-12 (only girls) years age group. Diet of preschoolers and juvenile adolescents was deficient in fats (6.6 to 42.3 %), where as range in DMRC phase I was 19.6 to 34 percent and observed highest in 1-3 years group (41.0 to 42.3 %) followed by 4-6 and 7-9 years groups. Vitamin A intakes (β carotene) in their diet was below the RDA only in preschoolers and juvenile adolescents, ranging from 2.9 to 28.5 percent. Preschoolers suffered from Thymine and Riboflavin deficiencies in their diet where as Niacin consumption was deficient in diet of all age groups of children, more in preschoolers (30 to 47.4 %). Diet of all individuals in all age groups was deficient in intakes of Vitamin C (17.9 to 76.7 %), maximum in 1-3 years age group (66.4 to 76.7 %) and minimum in adults (17.9 to 25.0 %).

Food Intake (% RDA) among Adults



Trends revealed that high illiteracy, poor economic conditions along with high deficiencies in their diet played important role for higher nutritional deficiencies, under nutrition and stunting especially more in children and chronic energy deficiency in adults of this region. After completion of third phase, fourth phase will be started in which 50 percent new villages and 50 percent old villages will be selected randomly in view to cover variation. Comparison of results of different Phases will be done in order to develop continuous monitoring service to study the nutritional status, dietary habits, food availability and the effect of changing social and environmental factors on the health status of the population and will continue year wise

3.2 Impact assessment of consumption of the two electrolyte products on the mineral profile and general health & nutrition profile of Adult population in desert areas of Rajasthan- *Madhu B. Singh and K. S. Premavalli**

Commencement: August, 2008

Duration: Nine months

Status: Ongoing

Project Coordinators: Dr. Bela Shah, Director-in-Charge, DMRC, Jodhpur & Dr. A. S. Bawa, Director, Defence Food Research Laboratory, Mysore

Objectives

1. To observe the impact of supplementation of the two electrolyte products on mineral profile (blood) of Adults of the desert areas
2. To study the effect of supplementation of the two electrolyte products on the general health & nutrition profile of Adults of the desert areas

Progress of the work

In this project, two villages i.e Salwa and Kharwali forming a contiguous field in rural part of Jodhpur, a desert district have been selected. In sampling, based on the reported 40 percent prevalence of MDDs based on Urinary Iodine Excretion level (UIE levels) in desert area, we need to administer 112 adults for administration of one product. Allowing for non-response error, we need to cover 130 adults for administration of one preparation. The HHs of these villages first listed fully, then allocated randomly in to three groups. Each group was then administered one product. Norms of double blind trial were adhered to. Two products i.e. two natural electrolytes, tender coconut water and Ashgourd juice have been developed by the Defence Food Research Laboratory, Mysore and the two products have been taken up for the study. These products have been supplied by DFRL, Mysore. Double blind randomized controlled trial have been conducted to test the efficacy of the two products separately i.e. Group one- tender coconut water and group two- Ashgourd juice. Third group was controlled group to which mineral water was supplied.

In this project, 390 adults (15-45 years) have been registered and categorized into three groups for supplementation of two products and mineral water. The data was collected on 390 adults (15-45 years) from selected HHs located in two villages. The data have been collected on all the parameters mentioned below in all the enrolled adults before initiation and also after the completion of the intervention program in this area. These villages were visited to supply the products to all enrolled adults for two months.

Information for demographic and socio-economic aspects have been collected. Before and after application, all the enrolled adults have been examined for the general health and nutrition profile i.e. morbidity profile for last 15 days and anthropometry i.e. height and weight.

* Scientist "F", Defence Food Research Laboratory, Mysore

After the completion of the intervention program in this area, overall non response was 17.7 percent. Non response for Product No. 1, Product No. 2 and Product No. 3 were 17.4, 21.1 and 14.4 percent respectively. A total of 321 adults were examined for all parameters after intervention. Reasons for non response were 'non co-operation', 'not good taste' and 'gone outside due to urgent work'.

Analysis of blood and urine samples is under progress for estimation of mineral profile of Na and K. Data collected in the field will be computerized under the supervision of biostatistician. The computer program will be developed in FOX- PRO and EPI-INFO-2002 software will be used for analyzing the data. As regards to the specific analysis, the extent of increase in mineral profile of Na and K will be carried out and the same will be compared before and after the initiation of the supplementation of two products using students t-test in order to observe its impact.

Expected Outcome

Consumption of two electrolyte products will enhance mineral profile (K & Na) in human blood samples of adults in desert areas of Rajasthan which will help in promotion of the health and nutrition status of adults residing in desert areas who are in a constant state of stress due to extreme environmental conditions of desert area which in turn affects the mineral (K, Na) and general health profile of the residents of this area. This will be helpful to health functionaries in designing operational pack rations for residents of this area.

3.3 Assessment of Iodine deficiency disorder, Anemia and Nutrition Intervention in school age children of Jodhpur district of Rajasthan

*Madhu B. Singh and K. S. Premavalli**

Commencement: July, 2008

Duration: Two years

Status: Ongoing

Project Coordinators: Dr. Bela Shah, Director-in-Charge, DMRC, Jodhpur & Dr. A. S. Bawa, Director, Defence Food Research Laboratory, Mysore

Objective

1. To study the distribution and magnitude of the Iodine deficiency disorders and anemia in school age children of Jodhpur district.
2. Assessment of the extant of use of iodized salt by the community of Jodhpur district.
3. To study the effect of supplementation on micronutrient deficiency disorders

Progress of work

Project activities included working on sampling plan in detail. According to WHO/ UNICEF/ ICCIDD, for school based survey, 30 cluster sampling approach need to be adopted keeping in view the operational feasibility. Recently DGHS (2005) has given new guidelines for sampling according to which sample size is calculated on the basis of prevalence of IDD as 10 %, level of confidence - 95 %, relative precision - 20 % and design effect - 2. Using formula $(Z\alpha)^2 Q / (L^2) P$, sample size worked out to be 1800 children from the district or $1800 / 30 = 60$ children per cluster for goiter and cretinism. Salt and urine samples along with blood on filter paper for haemoglobin estimation have been collected from 30 % of the children examined. At first step, listing of all the government and private schools with children 6-11 years of age from both rural and urban areas have been listed from district education office of Jodhpur. Secondly cumulative enrollment has been determined. Finally schools have been selected using PPS sampling technique as recommended by WHO. In the selected schools, children have been selected randomly using Tippets random number table. Equal proportion of boys and girls and proportionate distribution of children from 6-11 years have been covered in the selected school. Till now data has been collected from 23 schools covering 1380 school age children.

From each selected child, information for socio-demographic aspects has been collected. Each child interviewed / examined for the nutritional deficiency signs. Iodine deficiency disorders have been assessed by clinical examination of thyroid gland using the standard method as recommended by the joint WHO / UNICEF / ICCIDD consultation. A casual urine sample has been collected for estimation of Urinary Iodine Excretion (UIE) levels to assess the Iodine nutriture status. Iodine have been determined by wet digestion method using standard laboratory technique. UIE level less than 10 mcg/dl have been considered as indicator of iodine deficient nutriture. 30 percent of children selected randomly were requested to bring their urine

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sample and sample of 20 gm. salt consumed in their families in auto seal LDPE pouches. Iodine content of salt sample was estimated using standard iodometric titration method. Salt samples having iodine content less than 15 ppm was classified as with inadequate iodine. Anemia was assessed by clinical signs (platinichia and koilonichia). Hemoglobin levels were estimated using Cyanmethaemoglobin technique and have been classified as per WHO classification.

Nutrition intervention will be conducted on 15 percent of school children i.e. 270. These children will be supplied with supplements daily for the period of 90 days. These supplements have been developed in Defence Food Research Laboratory, Mysore. These products will be supplied by DFRL, Mysore. These children will be re-examined after period of 90 days supplementation for all above mentioned parameters observing the impact of the supplements on reduction of micronutrient deficiency disorders on school children of Rajasthan.

Analysis of 1380 school age children (659 boys and 721 girls) has been done so far (Table 1). Children belonging to Hindu religion were 99.5 percent. Most of children's parents (47.1 %) belong to agriculture occupation followed by labour (29.8 %) and service (12.5 %). Sickness at the of survey was 10 percent which was higher in males (9.9 %) than females (8.7 %). Regarding nutritional deficiency signs, it is observed that discoloration of hair, a sign of protein calorie malnutrition was observed to be high i.e. 41.9 percent. Total goiter rate was 5.3 percent. Night blindness was 0.4 percent

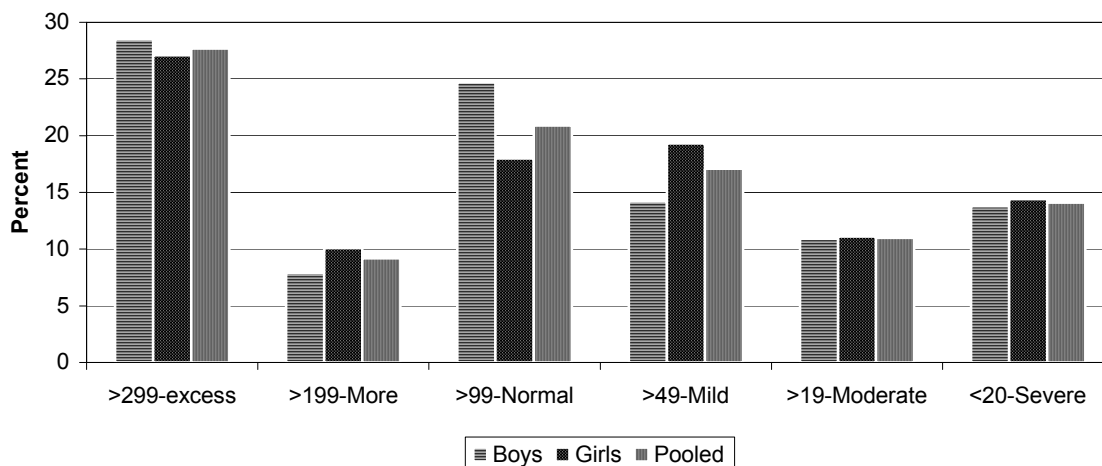
Table 1. Distribution of children according to age and gender

Age Group	Males	%	Females	%	Total	%
6+	86	13.1	86	11.9	172	12.2
7+	86	13.1	139	19.3	225	16.3
8+	108	16.34	141	19.6	249	18.0
9+	99	15.0	107	14.8	206	14.9
10+	146	22.2	128	17.8	274	20.0
11+	134	20.3	120	16.6	254	18.4
Total	659	100.0	721	100.0	1380	100.0

Stunting (Height for age) was 29.1 percent in school age children with the prevalence of severe stunting 7.7 %, which needs attention. Underweight (Weight for age) in school age children observed 43.1 %. Both stunting and underweight were observed higher in males than females though statistically insignificant.

Epidemiological criteria, as prescribed by WHO, for assessing iodine nutrition is based on median urinary iodine concentrations / levels as shown in figure 1. Analysis of 545 urine samples has been done so far. Median urinary iodine value was 140.0 mcg/l. Nearly 28.1 percent school age children suffered from mild to moderate iodine deficiency disorder. Severe iodine deficiency disorder was observed in 14.1 percent school age children.

Distribution of school age children according to UIE level



Iodine content of 470 salt samples has been estimated using standard iodometric titration method. Overall high proportion of children (72.6 %) consumed salt having inadequate iodine content i.e. less than 15 ppm (Table 2). High proportion of children i.e. 32.3 percent consumed salt having negligible iodine content (Less than 5 ppm).

Table 2. Distribution of children according to Iodine content in edible salt

Age Group	Normal >15ppm		Mild 7 - <15 ppm		Moderate 5 - <7 ppm		Severe <5 ppm	
	N	%	N	%	N	%	N	%
Boys N=192	44	22.9	55	28.6	25	13.0	68	34.9
Girls N=278	85	30.6	76	27.3	33	11.9	84	30.2
Total N=470	129	27.4	131	27.9	58	12.4	152	32.3

P<0.05

Analysis is under process. Data will be collected from 7 schools covering 420 school age children. Nutrition intervention will be conducted on 10 percent of school age children i.e. 260. These children will be re-examined after period of 90 days supplementation for all above mentioned parameters observing the impact of the supplements on reduction of micronutrient deficiency disorders on school children of Rajasthan.

3.4 Estimating and Adjusting for Publication Bias of Vitamin A Deficiency (VAD) in Meta Analysis- *J. Lakshminarayana and Madhu B .Singh*

Commencement: August, 2008

Duration: One year

Status: On going

Objective

1. To estimate & detect the effect of publication bias in meta-analysis of vitamin A deficiency and adjusting for the bias.

Progress of the work

Meta Analysis is collection of data systematically and synthesizing the results from individual studies to estimate overall size. Studies with positive results are more likely to get published than those studies with negative results. By using only the results of more positive studies leads to publication bias. We have used different methods to detect this publication bias.

Materials and method

The data of meta analysis was collected through literature search for the Vitamin A Deficiency (VAD). The systematic review of these meta analysis were carried out. The data consists of 15 publications to study the effect of Vitamin-A among school children. The duration was considered to be for a period of 10 years. We have used a binomial model to estimate the results and rank correlation method proposed by Begg and linear regression method proposed by Egger & Trim and Fill method to evaluate bias. The publication bias was adjusted by using "Trim & Fill method. The analysis was done using EPI META package.

Methods for detection of Publication bias

Funnel plot

Begg's Rank Correlation method

Egger's Regression method

Trim and Fill method (which also adjust for it)

A funnel plot is a plot of each trial's effect size against measure of its size, such as the precision, the overall sample size, or the standard error.

Trim and Fill method

It is a method to evaluate bias in funnel plots.

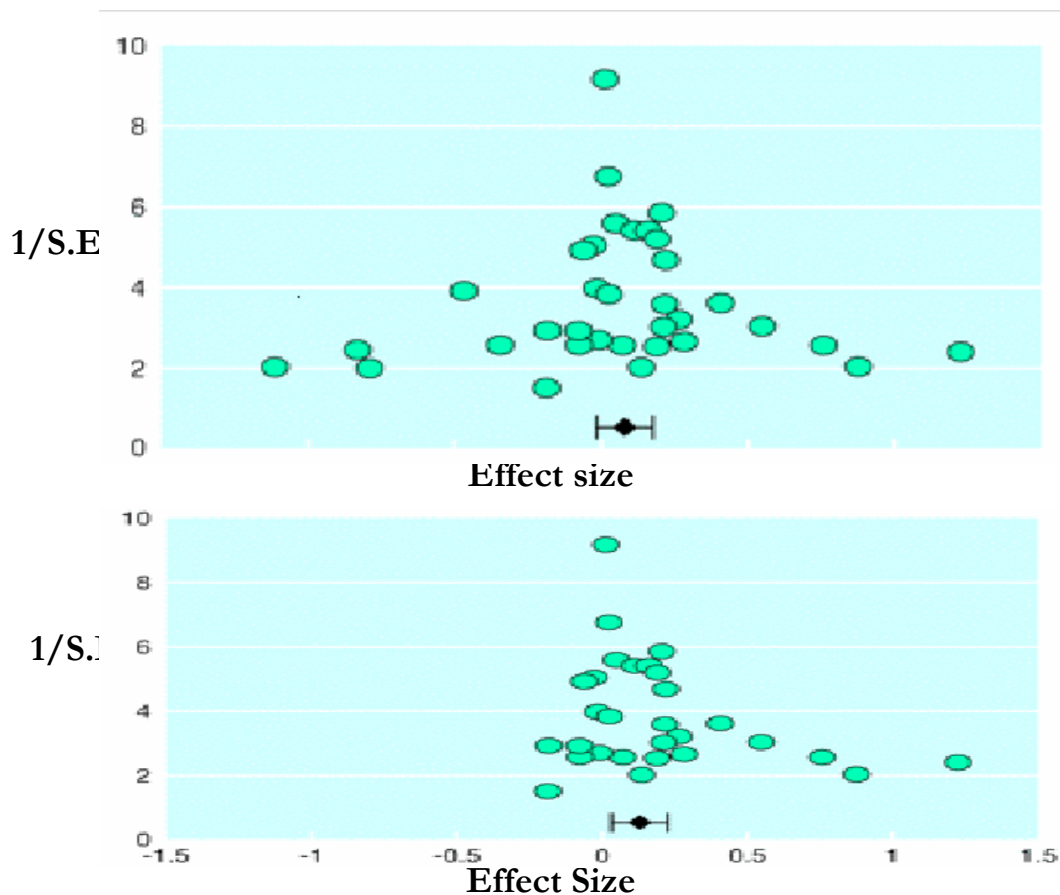
Firstly, the number of "asymmetric" trials on the right side of the funnel is estimated.

It trims off or removes these trials from the funnel leaving a symmetric remainder to estimate the true center of the funnel.

The trimmed trials are then replaced and their missing counterparts imputed or "filled".

Results

The two sided P values from the methods of Begg and Eger are 0.04 and 0.05 indicating the possible existence of publication bias. By Trim & Fill method ignoring publication bias the fixed effect model was fitted to see the overall estimates of pooled Odds Ratio is 1.16 with 95% CI (0.69, 1.82). A Funnel plot was plotted which Asymptotical, indicating the presence of publication bias was. When Trim & fill method was applied, one study which had missing variables was filled and we obtain an overall estimate of pooled odd ratio 0.98 with 95% CI (0.42, 1.56).



Conclusions

The overall estimate of pooled odds ratio is reduced in the adjusted model using Trim & Fill method. The meta analysis of conclusions drawn improved the bias among the studies after fitting the funnel plot technique.

3.5 Nutritional Status along with morbidity and mortality of Neonates & Infants in Jodhpur District- *Ranjana Fotedar, Madhu B. Singh and J. Lakshminarayana*

Commencement: September, 2007

Duration: Two years

Status: On going

Objectives

1. To study profile of Health and Nutritional Status of neonates by means of anthropometry, and clinical examination for nutritional deficiency signs along with feeding practices and their follow up for 1 year age group.
2. To study of types of morbidity, mortality and their cause among neonates and infants.

Progress of the Work

Registered newborns till December, 2008 are 250 newborns (0-7 days) from 24 villages. So far 58 newborns have been completed up to the age of 12 months (1 year) out of 250 newborns. 150 newborns have been followed up at the interval of one month and will continue up to one year of age. All the newborns are clinically examined for morbidity, mortality and nutritional deficiency signs, anthropometry (height i.e. Recumbent length, weight, arm circumference, chest circumference, head circumference and fat fold triceps following standard techniques of WHO) and feeding practices. Infant feeding practice comprising of both the breast feeding as well as complementary feeding have been recorded. The socio demographic / economic information have also been recorded from mother of each child registered in the study. The feeding practices of new born and socio-cultural / economic causes responsible for mortality are also recorded from mother of each child registered in the study. Each registered child has been followed up every month for above mentioned clinical examination. This process of registration will continue till the required number of 300 newborns (0 – 7 days) will be attained.

Preliminary analysis of 250 new born (123 males & 127 females) has been done. The follow up of 58 neonates have been completed upto the age of 12 months (1 year) out of 250 newborns.

Sickness at the time of survey in new born was 6.4 percent. Main morbidities observed were Fever-viral, Common Cold & Cough, Stomach ache, Diarrhea, Vomiting, Constipation, Respiratory Problem, Eye Infection, Skin Infection (Boils). One case of congenital abnormality of Bilateral cleft lip & hard palate has been observed.

It has been observed that 82.8 percent new born had normal weight at the time of birth whereas 17.2 percent were under weight (Table 1). Mortality observed in 10 cases out of 250 new born. Distribution of New Born according to mortality has been shown in Table 2.

Table 1. Distribution of New Born According to Weight for Age

New Born (0-7 days) N=250	Nutritional Grades		
	Normal ≥2.5 Kg	Mild < 2.5 – 2.0 Kg	Severe < 2 Kg
Number	207	36	7
Percentage	82.8	14.4	2.8

Table 2. Distribution of New Born according to mortality

Duration	Symptoms
0-3 days	Fever
0-7 days	Premature delivery (7 months) under weight = 1.3 Kg. (very small size baby)
14 days – 1 month	Under weight 2.2 Kg. (Unknown)
0-9 days	Baby had stiffed body convulsion fever (Bi-lateral cleft lip & hard palate; baby stopped suckling milk six days before death.
0-23 days	Baby suckling normal cause not known. Fever
0-4 days	B/F left 2 days before death, premature delivery (7 months) under weight 1.7 Kg.
0-19 days	Under weight 2.0 Kg Fever Jaundice, Diarrhea, Stopped suckling 2 days before death
0-13 days	Stopped suckling 1 day before death. Pneumonia & Fever
9 months & 7 days	Pneumonia, High Fever & Asthma
10 months & 9 days	High Fever, Asthma & Pneumonia

The project is ongoing and the survey would be carried out to cover remaining registration of 50 newborns (0 – 7 days) belonging to villages of Salawas CHC of Jodhpur district. Each registered child has been followed up every month for above mentioned clinical examination. This process of registration will continue till the required number of 300 newborns (0 – 7 days) will be attained.

4. Papers Published/Accepted

1. Angel B, Sharma K and Joshi V. Association of ovarian proteins with transovarial transmission of dengue viruses by *Aedes* mosquitoes in Rajasthan, India. *Indian J. Med. Res.*, 2008; 128: 181-184. IF: 1.670
2. Angel B and Joshi V. Distribution of dengue virus types in *Aedes aegypti* and possible risk of DHF in dengue endemic districts of Rajasthan, India. *Indian J. Med. Res.*, 2008 (In Press). IF:1.670
3. Bansal SK, Singh Karam V and Suresh Kumar. Larvicidal activity of the extracts from different parts of the plant *Solanum xanthocarpum* against important mosquito vectors in the arid region. *J Environ. Biol.*, 2008 (In Press) IF: 1.357
4. Bansal SK and Singh Karam V. Evaluation of the larvicidal efficacy of *Solanum xanthocarpum* storage against vector mosquitoes in north western Rajasthan. *J Environ. Biol.*, 2008 (In Press) IF: 1.357
5. Lakshminarayana J, and Singh Madhu B, Opium Consumption among rural population in western Rajasthan: Some observations from the study. *Journal of Human Ecology*, 2009 (In Press).
6. Lakshminarayana J, Haldiya KR, Singh Madhu B, Opium Consumption and its indirect effect on the fertility of women in western Rajasthan. *Annals of Arid Zone*, 2008 (In Press). IF: 0.26
7. Sharma K, Angel B, Singh H, Purohit A and Joshi V. Entomological studies for surveillance and prevention of dengue in arid and semi-arid districts of Rajasthan, India. *J. Vector Borne Dis.*, 2008; 45: 140–149.
8. Singh Madhu B, Fotedar R and Lakshminarayana J. Micronutrient deficiency Status among women of desert areas of western Rajasthan, India. *Pub. Health Nutrition*, doi: 10.1017/S1368980008002395. Published online by Cambridge University Press, UK, 2008. IF: 2.123
9. Singh Madhu B, Lakshminarayana J and Fotedar R. Chronic energy deficiency and its association with dietary factors in Adults of drought affected desert areas of Western Rajasthan, India. *Asia Pac J Clin Nutr.*, 2008; 17(4): 580-585. IF: 1.015
10. Singh Madhu B, Fotedar R and Lakshminarayana J. Camel Milk Consumption Pattern and Its Association with Diabetes among Raika Community of Jodhpur District of Rajasthan. *Ethno-Med.*, 2008; 2 (2): 03-105.
11. Singh Madhu B, Lakshminarayana J and Fotedar R. Nutritional status of adult population of Raika community in Jodhpur, desert district, of Rajasthan. *Journal Human Ecology*, 2008 (In Press).
12. Singhi, Manju and Joshi, Ramesh. Famine Foods of Arid Rajasthan : Utilization, perceptions and need to integrate social practices by bio-resolutions. *Studies on Ethno-Medicine*, 2008 (In Press).

5. Workshops / Conferences / Symposia / Scientific Meetings attended/ participated / organized by the scientists

Dr. Murli L. Mathur, Scientist F

- Delivered invited talk on 'Prevention of Silicosis in Sandstone Quarry-workers (Management of Occupational Health and Safety Delivery in Sandstone Quarry Workers: Scopes and Challenges)' as a resource person, at WHO Sponsored National Workshop on 'Management of Occupational Health and Safety Delivery: Scopes and Challenges', held on 30th September and 1st October 2008 at NIOH, Ahmedabad.
- Delivered invited Lecture on 'Environmental interventions in silicosis: DMRC experience' as Guest Faculty in three day workshop on "Silica Exposure: Disease and Management" on 15-17 January 2009 at NIOH, Ahmedabad.
- 63rd National Conference on Tuberculosis and Chest Diseases and First International Conference of South East Asia Region (The Union) SEAR-NATCON 2008) organized by The Tuberculosis Association of India New Delhi held from September 8-10, 2008 and presented paper entitled 'Glutaraldehyde Test for diagnosis of Pulmonary tuberculosis'.

Dr. Karam V. Singh, Scientist E

- 'Second International Forum for Sustainable Management of Disease Vectors' held at Beijing, China from November 2-4, 2008 and presented a paper entitled 'The Impact of Indoor Residual Spray on the Insecticide Susceptibility Status of Malaria Vectors (Rajasthan: India)'.
- '4th World Congress on Leishmaniasis', being organized by CDRI, Lucknow from 3rd to 7th February, 2009, and presented a paper entitled 'Faunal diversity of sand fly species prevalent in cutaneous leishmaniasis endemic areas of Rajasthan, India'.
- Brainstorming meeting for finalization of components and action programme under All India Coordinated Project on *Jatropha* funded by Indian Council of Forest Research and Education (ICFRE), at Arid Forest Research Institute, Jodhpur on 5th September, 2008.
- 'Akhil Bhartiya Rajbhasha Sammelan and Workshop' at Hyderabad from 27th to 29th December, 2008, organized for promotion & use of Rajbhasha and development of soft wares and tools for the purpose.
- Organized 'Refresher Training Course for Laboratory Technicians' sponsored by State Health Society (NVBDGP), Government of Rajasthan, at DMRC, Jodhpur in two batches during April, 2008.
- International Symposium on Tribal Health, organized by RMRC for Tribals, Jabalpur from 27th Feb. to 1st March, 2009 and presented a paper entitled 'Mapping of Insecticide Resistance in Malaria Vectors of Western Rajasthan'.

Dr. S. K. Bansal, Scientist E

- Organized 'Refresher Training Course for Laboratory Technicians' sponsored by State Health Society (NVBDCP), Government of Rajasthan, at DMRC, Jodhpur in two batches during April, 2008.
- 'Akhil Bhartiya Rajbhasha Sammelan and Workshop' at Hyderabad from 27th to 29th December, 2008, organized for promotion & use of Rajbhasha and development of soft wares and tools for the purpose

Dr. Madhu B. Singh, Scientist E

- 'International Conference on Iron Deficiency' held at Gyan Sarover, Mount Abu, Rajasthan from 4th to 8th December, 2008 and presented a paper entitled 'Anemia and their association with diet and other related factors among women of desert areas of western Rajasthan'.
- Training Programme on 'Intellectual Property Rights (IPRs) and Related WTO Issues' organized by CUTS International with the support from the Department of Science and Technology (DST), Government of India from November 03-07, 2008 at Jaipur.
- International Symposium on Tribal Health, to be organized by RMRC for Tribals, Jabalpur from 27th Feb. to 1st March, 2009 and present a paper entitled 'Studies on Micronutrient deficiency disorders in different communities among pregnant and lactating women of desert areas of western Rajasthan'.

Dr. A. K. Dixit, Scientist E

- ISMS conference held at Nainital from November 7-9, 2008 and presented the paper entitled 'Household distribution of disease burden and its household level risk factors: A study in desert'.

Dr. J. Lakshminarayana, Scientist D

- ISMS conference held at Nainital from November 7-9, 2008 and presented a paper entitled on 'Opium consumption among rural population in western Rajasthan: some observations from the study'.

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17. Sh. Khushal Singh, Attendant
18. Sh. Jodha Ram, Attendant
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20. Sh. Ladu Ram, Peon
21. Smt. Sohni Devi, Peon
22. Smt. Sua Devi, Sweeper

E. PROJECT STAFF

❖ Development of molecular and genetic markers of dengue vectors species in Rajasthan, India

1. Ms. Bennet Angel, Senior Research Fellow
2. Ms. Rashmi Chauhan, Junior Research Fellow

❖ Evaluation of Immunization Coverage against Vaccine Preventable Diseases in Western Rajasthan

1. Sh. Oma Ram Choudhary, Field Investigator
2. Sh. Piyush Raj Joshi, Field Investigator
3. Sh. Mohd. Kasim, Field Investigator

❖ Jai Vigyan Mission Mode Project on Control of Rheumatic Fever and Rheumatic Heart Diseases in Jodhpur District

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4. Ms. Jyoti Gaur, Field Investigator
5. Ms. Ruchika Sharma, Field Investigator

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❖ **Nutrition Profile Project**

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