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(2011-2012)

DESERT MEDICINE RESEARCH CENTRE
(Indian Council of Medical Research)
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डॉ विश्व मोहन कटोच
एम.डी. एफ एन ए एससी. एफ ए एम एस. एफ ए एससी. एफ एन ए
सचिव, भारत सरकार
(स्वास्थ्य अनुसंधान विभाग)
स्वास्थ्य एवं परिवार कल्याण मंत्रालय एवं
महानिदेशक, आई सी एम आर
Dr. Vishwa Mohan Katoch
MD, FNASc, FAMS, FASc, FNA
Secretary to the Government of India
(Department of Health Research)
Ministry of Health & Family Welfare &
Director-General, ICMR

सत्यमेव जयते

FOREWORD

It gives me immense pleasure in writing the foreword for the Annual Report-2011-12 of Desert Medicine Research Centre (DMRC) containing description of various scientific activities of the Centre. The report presents information on the research programmes of DMRC on dengue transmission in mosquitoes; investigations pertaining to malaria and dengue outbreaks; pyrazinamide sensitivity of *M. tuberculosis*; identification of biological forms of *An. stephensi*; Susceptibility of urban vectors of malaria and dengue to insecticides being used in national programme; and exploring larvicidal potential of certain indigenous desert plants against vector mosquitoes. Association between socio-economic factors, treatment seeking behavior and early detection of new pulmonary tuberculosis cases have been investigated. The report also covers studies on polymorphisms in duffy blood group genes of *P. vivax* malaria patients, mosquito age determination and risk assessment of Japanese Encephalitis virus emergence. Prevalence of diabetes mellitus and impaired glucose tolerance in the Raika community has been assessed.

Studies on consumption pattern of different recipes of pearl millet, assessment of iodine deficiency disorders in school children, nutritional status along with morbidity and mortality of neonates and infants, are some other key areas dealt in the report.

I hope that the report will provide the readers specially the policy makers useful information on the programmes and contributions of DMRC in various areas of desert health. The ongoing research activities at the Centre would not only be beneficial to current national health programmes but also widen the horizon of DMRC research.

(V. M. Katoch)

PREFACE

The Centre during the report period has been engaged in research pertaining to Dengue/ DHF, Malaria and Tuberculosis among the communicable and Musculoskeletal disorders, RF/RHD, Diabetes Mellitus and Coronary Heart Diseases among non-communicable diseases in general. Under communicable diseases the projects were confined to: development of probe to detect dengue transmission as early warning tool; testing a module of dengue control; investigations of dengue and malaria outbreaks, and factors affecting incidence and management of malaria. Polymorphisms in Duffy blood group genes of *Plasmodium vivax*; age determination of mosquitoes; insecticide susceptibility of urban mosquito vectors; risk assessment of JE virus emergence have been other important priority areas of research. The aspects of early diagnosis and detection of new PTB cases have been dealt to reduce the burden of the disease in the region.

The projects on prevalence of diabetes mellitus and impaired glucose tolerance in the Raika community and identification of coronary artery diseases have been carried-out under non-communicable diseases. Studies on pearl millet consumption; assessment of iodine deficiency disorders, anemia and nutrition intervention in school age children, and nutritional status, morbidity and mortality of neonates and infants have been carried-out for the benefit of the most vulnerable groups of the society.

As a part of 'Centenary Celebrations of ICMR' the Centre organized 'ICMR-INSERM Workshop on Gene Environment Interactions, Epi-genetics Nutrition and Drugs in Diabetes', a Conference for dissemination of findings of pearl millet project' and an invited talk on 'Malnutrition'. The work of the Centre was projected in '3rd Vision Rajasthan, 2012' and adjudged first among scientific institutions.

Ph. D. and M. Sc. dissertation programmes in the Centre is helping the scientists in brushing up their knowledge besides developing trained man power in the region. Publication of quarterly magazine 'Chetna', both in Hindi and English to disseminate the scientific knowledge and activities of the Centre, has become a regular feature of the Centre.

The Centre is engaged to explore research needs of the region and transform the ideas to ground realities for the benefit of the society, and also strengthen the resources in the form of trained man power and laboratory support.

Dr. Bela Shah
Director-In-Charge
Scientist , 'G' & Head
Division of Non-Communicable Diseases

CONTENTS

1.1 Development of probe to detect dengue transmission or blocking proteins in mosquitoes as early warning tool of transmission risk of dengue in endemic areas- a fellowship project

Principal Investigator: *Dr. Vinod Joshi, Scientist 'F'*
Research Staff: *Dr. Bennet Angel, Research Associate*
Commencement: February, 2010 **Duration:** Three Years **Status:** Ongoing
Funding Agency: Indian Council of Medical Research (Extramural)

OBJECTIVES

1. Establishment of a cause & effect relationship between mid gut proteins and horizontal transmission and ovarian proteins, and vertical transmission of dengue virus by mosquito vectors in *in vitro* and *in vivo* experiments. Implication of proteins for facilitating and blocking of virus replication by mosquito cells
2. Sequencing of observed proteins, determining complete peptide, synthesis of peptide and development of specific antibodies against peptide
3. Application of transmission specific protein antibodies as probe to detect the field collected mosquitoes for developing early warning risk of dengue transmission in an area

PROGRESS

Entomological investigations: *Aedes* larvae were collected from different localities of Jodhpur. They were reared into adults under laboratory conditions. The mid gut and ovary of these mosquitoes were dissected out and subjected to SDS-PAGE for studying the protein profile.

Virological investigations: Simultaneous with dissecting mid gut and ovarial tissues from field collected and laboratory reared mosquitoes, head squash of individual mosquito was subjected to Indirect Fluoresce Antibody Test (IFAT) for detecting the presence or absence of dengue antigen.

Proteomic studies of mid gut and ovarian tissues showing virus positive and negative results: The mid gut and ovarian tissues of mosquitoes showing negative results of virus presence in IFA Test as well as RT-PCR tests were subjected to SDS-PAGE for all the three species of *Aedes* mosquitoes *viz*., *Aedes aegypti, Aedes albopictus and Aedes vittatus.*

Among 365 mosquitoes belonging to *Ae. vittatus,* the midgut tissues were subjected to SDS-PAGE, 35 showed presences of proteins in the range of 200 kDa. Similarly 19 out of 110 specimens of *Ae. albopictus* and 12 out 114 specimens of *Ae. aegypti* showed the 200 kDa protein bands.

Same pools of mosquitoes (589 specimens) were also dissected to collect the ovarian tissues from them. Ovaries of 155 mosquitoes from 365 *Ae. vittatus*, 24 ovaries from 110 *Ae. albopictus* and 58 from 114 specimens of *Ae. aegypti* showed the protein bands in 200 kDa range.

The protein bands displayed in the mid gut as well as ovarian samples are shown in the Fig.1-3. The proteins bands of 200 kDa were sliced from the gels and were subjected to sequencing and subsequent protein identification employing In-Gel tryptic digestion method followed by MS/MS analysis.

Protein Identification from 200 kDa protein bands of mosquito mid gut and ovaries and derivation of amino acid sequencing to determine complete peptide: The protein bands in the range of 200 kDa molecular weight as appeared in SDS-PAGE assays of mid gut and ovarian samples were sliced from the gels and were identified commercially at Centre for Cellular and Molecular Biology, a part of National Centre for Biological Sciences, Bangalore, using Mass Spectroscopy (RPLC-MS/MS).

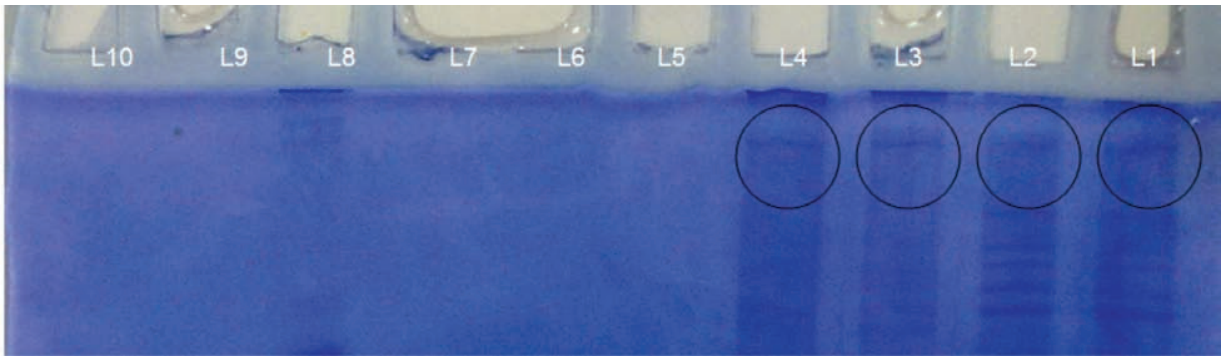


Fig. 1. SDS-PAGE assay of mid gut and ovarian tissues of *Ae. aegypti

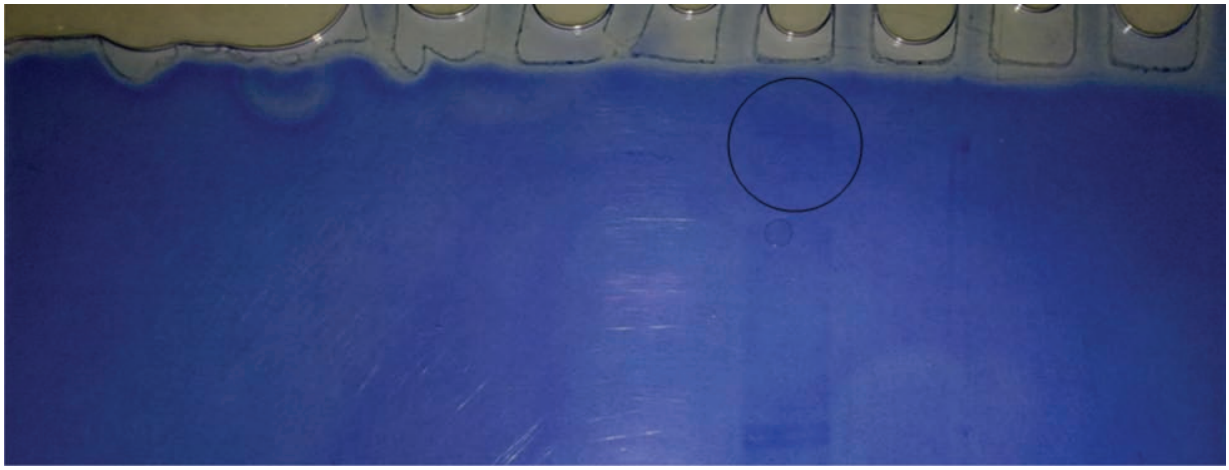


Fig. 2. SDS-PAGE assay of mid gut tissues of *Ae. albopictus

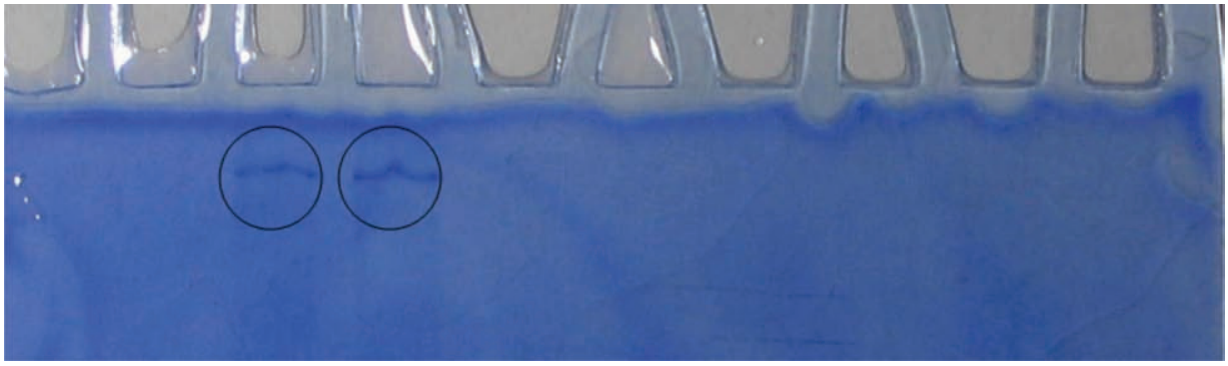


Fig. 3. SDS-PAGE assay of mid gut tissues of *Ae. vittatus

* The bands in the circle were used for protein identification using MS/MS technique

Table 1 depicts the details of proteins identified from ovarian and mid gut tissues of *Ae. aegypti, Ae. albopictus* and *Ae. vittatus*. 30-50 proteins were identified in the samples sent. But as shown in Table 3 there appeared one common protein of 224.1 kDa which was found to be present in ovarian tissues of all the three species. The same protein also appeared in the mid gut tissue of *Ae. aegypti*. This protein identified as 'Myosin heavy chain, non muscle or smooth muscle' protein of *Ae. aegypti* origin (identified on MASCOT using Swis-Prot database), was consisting of 1963 amino acids. Keeping in view the physical mode of internalization of dengue virion into mosquito cell, this protein (Myosin) appears to be protein of our choice playing possible role in blocking internalization of dengue virion into ovarian tissues of all three species and mid gut cells of *Ae. aegypti*. Since this was the common protein present in the ovaries of all the three species and mid gut cells of *Ae. aegypti*, the complete sequence of this protein was searched using accession number provided by the sequencing agency. The full sequence of Myosin was therefore, derived and the whole peptide of 1963 amino acids was determined as shown in Table 2.

Table 1. 200 kDa bands from mid gut & ovarian tissues of *Aedes* Species

***Aedes* species**	**Protein name**	**Molecular Weight (kDa)**	**No. of amino acids**	**Tissue of protein**	**Accession number**
Ae. aegypti	Myosin heavy chain, non muscle or smooth muscle *(Ae. aegypti)*	224.1	1963	Mid gut	157111095
Ae. aegypti ***Ae. vittatus*** ***Ae. albopictus***	Myosin heavy chain, non muscle or smooth muscle *(Ae. aegypti)*	224.1	1963	Ovary	157111095

As discussed above (Table 3) we observed a common protein in ovarian tissues of all the three species and mid guts of *Ae. aegypti*. However, in samples detailed above, a protein

could not be identified in mid gut tissues of *Ae. vittatus* and *Ae. albopictus*. Table 4 shows similar protein identified in the mid gut samples from *Ae. vittatus* and *Ae. albopictus* but of *Drosophila melanogaster* origin. The protein identified was similar to the above with a difference of 0.2 kDa i.e. of 224.3 kDa and an amino acid sequence less than one amino acid from the earlier one i.e. of 1962 amino acids. Using the accession number provided by sequencing agency we derived the full sequence of this protein as shown in the data (Table 4).

During reported period we have identified two proteins; one being present in ovarian samples of all the three species and mid gut samples of *Ae. aegypti*. Another protein we have identified was present in mid gut samples of two species *viz*., *Ae. vittatus* and *Ae. albopictus*. Both the proteins are Myosin heavy chain proteins consisting of 1963 and 1962 amino acids respectively and their complete sequence has been obtained.

Table 2. Amino Acid sequence of complete peptide of 224.1 kDa

1	mpkpvvqvgd dpdpsewlfv sleqkridqs kpydakkacw vpdekegyvl geikatkgel
61	vtvglpggee knfkkelisq vnppkfekve dmadltylne aavlhnlrqr yyskliytys
121	glfcvvinpy krwplytlrv akmyrgkrrn evpphlfavs dgayvnmltn henqsmlitg
181	esgagktent kkviayfati gastkkeess ekkasledqv vqtnpvleay gnaktvrndn
241	ssrfgkfiri hftgsgklag adietyllek arvisqqsle rsyhifyqmm sgsvkglkdm
301	cflsndiydy ynvaqgkiti pnvddgeecl ltdeafnvlg ftqeekdniy kitaavmhmg
361	gmkfkqkgre eqaeadgmee gdrvakllgc vtedlyknll kprikvgaef vtkgqnkdqv
421	tnavgalckg ifdrlfkwlv kkcnetldtq mkrvqfigvl diagfeifdy ngfeqlcinf
481	tneklqqffn hhmfvleqee ykkeginwaf idfgmdllac idliekpmgi lsileeesmf
541	pkatdqtfae klmnnhlgks apfqkpkppk pgcqaahfai ghyagvvsyn itgwleknkd
601	plndtvvdqf kkgqnklvve ifadhpgqsg gadagggkgg rgkkgagfat vsssykeqln
661	nlmttlkstq phfvrciipn elkqtglida hlvmhqltcn gvlegiricr kgfpnrmmyp
721	dfklrykiln pkaaeeqkep knvadvilts igldtesyrl ghtkvffrag vlgqmeefrd
781	erlskimswm qswcrgylar kefkkmqeqr valetvqrnl rkymklrtwa wwklwqkvkp
841	llnvsrvedq iaeleskaqk aqeafekeek arkelealns kllaektall dslsgekgal
901	qdfqektakl taqkndlenq lrdtqerlsq eedarnqlmq tkkkleqeig gqkkdaedle
961	lqiqkieqdk askdhqirnl ndeiahqdel inklnkekkm qgevnqktae elqaaedkvn
1021	hlnkvkakle qtldeledsl erekklrgdv ekakrkvegd lkltqeavad lernkkeleq
1081	timrkdkeis alsakledeq slvgktqkqi kelqgrieel eeeveaerqa rakaekqrad
1141	lareleelge rleeaggats aqielnkkre aelaklrrdl eesniqhegt lanlrkkhnd
1201	avaemaeqvd qlnklktkae kersqyyaem ndarlsldhm anekaaqekv akqlqhtlne
1261	vqgkldetnr tlndfdsakk klsiensdll rqledaesqv sqlskikisl tqqledtkrl
1321	adeesrerat llgkfrnleh dldslreqve eeaegkadiq rqlskanaea qlwrtkyese
1381	gvaraeelee akrklqarla eaeetiesln qkcvalektk qrlstevedl qlevdratsi
1441	anaaekkqka fdkiigewkl kvddlaaeld asqkecrnys telfrlkgay eegqeqleav
1501	rrenknlade vkdlldqige ggrniheiek srkrleaekd elqaaleeae aaleqeenkv
1561	lraqlelsqv rqeidrriqe keeefentrk nhqraldsmq asleaeakgk aealrmkkkl
1621	eadineleia ldhankanae aqknikryqq qlkdvqsale eeqrarddar eqlgiserra
1681	nalqnelees rtlleqadrg rrqaeqelsd aheqlnevsa qnasiaaakr kleselqtlh
1741	sdldellnea knseekakka mvdaarlade lraeqdhaqt qeklrkaleq qikelqvrld
1801	daetnalkgg kkaiqkleqr vreleselds eqrrhtdaqk nlrkserrik eltfqseedr
1861	knhermqdlv dklqqkikty krqieeaeei aalnlakfrk aqqeleeaee radiaeqaat
1921	kfrtkggrag svqrgaspap qrqpsvmpgl aglnfptfdd hgf

Table 3. Amino Acid sequence of complete peptide of 224.3 kDa

1	mpkpvanqed edptpylfvs leqrridqsk pydskkscwi pdekegyllg eikatkgdiv
61	svglqggevr diksekvekv nppkfekied madmtvlntp cvlhnlrqry yakliytysg
121	fcvainpyk rypvytnrca kmyrgkrrne vpphifaisd gayvdmltnh vnqsmlitge
181	sgagktentk kviayfatvg askktdeaak skgsledqvv qtnpvleafg naktvrndns
241	srfgkfirih fgptgklaga dietylleka rvisqqsler syhifyqims gsvpgvkdic
301	lltdniydyh ivsqgkvtva siddaeefsl tdqafdilgf tkqekedvyr itaavmhmgg
361	mkfkqrgree qaeqdgeeeg grvsklfgcd taelyknllk prikvgnefv tqgrnvqqvt
421	nsigalckgv fdrlfkwlvk kcnetldtqq krqhfigvld iagfeifeyn gfeqlcinft
481	neklqqffnh imfvmeqeey kkeginwdfi dfgmdllaci dliekpmgil sileeesmfp
541	katdqtfsek ltnthlgksa pfqkpkppkp gqqaahfaia hyagcvsyni tgwleknkdp
601	lndtvvdqfk ksqnklliei fadhagqsgg geqakggrgk kgggfatvss aykeqlnslm
661	ttlrstqphf vrciipnemk qpgvvdahlv mhqltcngvl egiricrkgf pnrmmypdfk
721	mryqilnprg ikdldcpkka skvliestel nedlyrlght kvffragvlg qmeefrderl
781	gkimswmqaw argylsrkgf kklqeqrval kvvqrnlrky lqlrtwpwyk lwqkvkplln
841	vsriedeiar leekakkaee lhaaevkvrk elealnakll aektalldsl sgekgalqdy
901	qernakltaq kndlenqlrd iqerltqeed arnqlfqqkk kadqeisglk kdiedlelnv
961	qkaeqdkatk dhqirnlnde iahqdelink lnkekkmqge tnqktgeelq aaedkinhln
1021	kvkakleqtl deledslere kkvrgdveks krkvegdlkl tqeavadler nkkeleqtiq
1081	rkdkelssit akledeqvvv lkhqrqikel qarieeleee veaerqarak aekqradlar
1141	eleelgerle eaggatsaqi elnkkreael sklrrdleea niqhestlan lrkkhndava
1201	emaeqvdqln klkakaehdr qtchnelnqt rtacdqlgrd kaaqekiakq lqhtlnevqs
1261	kldetnrtln dfdaskkkls iensdllrql eeaesqvsql skikislttq ledtkrlade
1321	esreratllg kfrnlehdld nlreqveeea egkadlqrql skanaeaqvw rskyesdgva
1381	rseeleeakr klqarlaeae etieslnqkc iglektkqrl stevedlqle vdranaiana
1441	aekkqkafdk iigewklkvd dlaaeldasq kecrnystel frlkgayeeg qeqleavrre
1501	nknladevkd lldqigeggr niheiekark rleaekdelq aaleeaeaal eqeenkvlra
1561	qlelsqvrqe idrriqekee efentrknhq raldsmqasl eaeakgkaea lrmkkklead
1621	ineleialdh ankanaeaqk nikryqqqlk diqtaleeeq rarddareql giserranal
1681	qneleesrtl leqadrgrrq aeqeladahe qlnevsaqna sisaakrkle selqtlhsdl
1741	dellneakns eekakkamvd aarladelra eqdhaqtqek lrkaleqqik elqvrldeae
1801	analkggkka iqkleqrvre leneldgeqr rhadaqknlr kserrvkels fqseedrknh
1861	ermqdlvdkl qqkiktykrq ieeaeeiaal nlakfrkaqq eleeaeerad laeqaiskfr
1921	akgragsvgr gaspaprats vrpqfdglaf pprfdlapen ef

Table 4. Protein details of 200 kDa bands from mid gut tissues of *Aedes* species

***Aedes* species**	**Protein name**	**MW**	**No. of amino acids**	**Tissue of protein**	**Accession number**
Ae. vittatus ***Ae. albopictus***	Myosin Heavy Chain, muscle (*D. melanogaster*)	224.3	1962	Mid gut	P05661

FINDINGS

1. We have established the relationship between presence of 200 kDa protein bands and absence of virus transmission by mosquitoes.
2. From virus negative mosquito samples we have obtained protein bands and have slice them from gel and have identified as Myosin heavy chain proteins.
3. The myosin heavy chain proteins obtained from mid gut and ovarian tissues of virus negative mosquito samples of all three species were sequenced and a complete peptide molecule has been constructed.
4. As a part of next year's work, we propose to develop antibodies against these two peptides to develop a testing probe for vector competence of mosquito vectors of dengue.

1.2 Report on Investigations of Outbreak of Dengue in Jodhpur town

Principal Investigator: *Dr. Vinod Joshi, Scientist 'F'*
Research Staff: *Dr. Bennet Angel, Research Associate, Ms. Annette Angel, Investigator and Ms. Nidhi Vyas, Investigator*
Commencement: September**,** 2011 **Duration:** Six Months **Status:** Completed
Funding: Desert Medicine Research Centre (Intramural)

OBJECTIVES

1. Characterization of outbreak of dengue fever in Jodhpur town
2. Serological and molecular biological studies on the patients showing peculiar clinical conditions
3. Correlation of extrinsic and intrinsic viral load and types in human patients and mosquitoes of patient's houses
4. Explanation of outbreak and clinical conditions and suggestions of better clinical and public health management through research

PROGRESS

Characterization of Outbreak through molecular diagnosis: The research team of DMRC collected 324 serum samples from medical college hospitals of Jodhpur, belonging to patients clinically suspected of infected of dengue, by treating physicians. Serum samples were brought to the laboratory of Virology & Molecular Biology, DMRC. The details of the diagnosis as made by hospital laboratory were recorded from hospital records. To characterize the outbreak as being caused by dengue virus, Real Time PCR (RT-PCR) was performed on selected samples, using primers and probes obtained from National Institute of Virology, Pune. Following are the details of tests conducted:-

Observations:

1. The data depicted in Table 1 show results of 42 selected serum samples, 26 of whose tests results as tested by hospital laboratory were +ve for either IgM or IgG antibodies as shown by the strip test. The analysis of hospital results showed 5 out of 26 samples +ve for IgM but negative for IgG, showing that current infection is first infection of dengue. Similarly 5 out of 26 samples analyzed showed IgM –ve but IgG positive, indicating past infection of dengue but no current infection. Out of 26 samples, 11 samples showed IgM and IgG antibodies both, which indicates presence of past as well as present infection of dengue. 2 of the 16 samples reported as negative by the hospital showed +ve for IgM while 3 showed +ve for IgG and 1 for NS1 (Table 1).
2. The tests undertaken by DMRC laboratory focused on testing NS1 (Non-Structural) protein of dengue virus by strip test, IgM antibodies by Mac-ELISA and Real Time PCR test (Figure 1-3).

Table 1. Molecular diagnosis and serological investigations of serum samples of patients suspected of infected from dengue fever

Patient Code	Age	Sex	Hospital tests		DMRC Laboratory investigations		
			IgM	IgG	IgM (Mac-ELISA)	NS_1 antigen	RT- PCR
1/11/MDM	NK	M	+	+	+	-	+
2/11/ MEMO	25	M	+	+	-	-	+
3/11/ MDM	25	M	+	+	-	-	-
4/11/ MDM	14	F	+	+	-	-	-
5/11/ MDM	21	M	-	+	-	-	-
6/11/ MDM	15	F	-	+	+	-	+
7/11/ MDM	NK	M	+	+	+	-	+
8/11/ MDM	23	M	+	+	-	-	+
9/11/ MDM	14	F	+	+	+	-	+
10/11/ MDM	19	M	+	+	-	-	+
11/11/ UMD	8	F	-	-	+	-	+
12/11/UMD	NK	M	+	+	+	+	+
13/11/UMD	8	F	+	-	-	-	+
14/11/ UMD	7	M	+	-	-	-	+
15/11/ MDM	19	M	-	+	-	-	+
16/11/ UMD	10	F	+	-	+	-	+
17/11/UMD	11	F	-	+	-	-	+
18/11/UMD	5	M	+	-	-	-	+
19/11/UMD	10	M	+	+	+	-	+
20/11/UMD	9	M	-	-	-	+	+
21/11/MGH	13	M	-	-	-	+	+
22/11/UMD	16	M	-	-	+	+	+
23/11/MGH	39	M	+	+	-	-	-
24/11/UMD	4	F	-	+	+	-	+
25/11/UMD	7	M	-	-	+	+	+
26/11/UMD	18 Mths.	F	+	-	+	-	ND
27/11/MGH	80	M	-	-	-	-	+
28/11/MGH	NK	F	-	-	-	-	-
29/11/MGH	20	M	-	-	-	-	-
30/11/MGH	50	F	-	-	-	-	+
31/11/MGH	NK	M	-	-	-	-	+
32/11/MGH	30	M	-	-	-	+	+
33/11/MDM	19	M	-	-	-	-	-
34/11/MDM	20	F	-	-	-	-	-
35/11/MGH	19	F	-	-	-	-	-
36/11/MGH	16	F	-	-	-	-	-
37/11/MDM	17	M	-	-	-	-	+
38/11/MDM	45	M	-	-	-	-	-
39/11/MDM	18	M	-	-	-	-	-
40/11/MGH	NK	F	-	-	-	-	+
41/11/MGH	13	M	-	-	-	-	-
42/11/MGH	NK	M	-	-	-	ND	ND

3. Only 7 out of 42 serum samples selected showed presence of NS1 viral protein. Out of 7 cases positive for NS1 only two samples showed +ve tests for IgM and IgG antibodies where as all remaining 5 samples positive for NS1 showed negative results for IgM and IgG antibodies there by indicating that most of the samples positive for NS1 are devoid of both IgM and IgG antibodies and hence represent the first infection of dengue. This observation explains that there may not be cross infection of two DEN strains and possibly also explains why during present outbreak, cases of DEFT were not reported. Wherever, NS1 protein has been detected, in all such cases RT-PCR test showed +ve results.
4. Out of 26 samples tested positive for IgM and/or IgG by hospital laboratory, 21 samples showed positive results for Real Time PCR. In 9 out of 26 samples which were tested negative for IgM antibodies were observed positive for RT-PCR. The observations show the need for molecular diagnosis of cases for accurate diagnosis and subsequent case management.

Inferences:

1. The tests undertaken for molecular diagnosis of dengue in suspected cases establish that the present outbreak was caused by dengue virus.
2. Our studies show that number of cases which showed negative test results for IgM and IgG antibodies showed positive results for RT-PCR. The present practice of detection of dengue through IgM and IgG antibodies need to be advanced to molecular diagnosis of virus for exact disease magnitude and subsequent better case management in the hospitals.

Expected Impact of new knowledge generated:

1. The advance knowledge generated by DMRC on molecular diagnosis of samples, the viral outbreak has been characterized as being caused by dengue virus. Our studies will sensitize disease specific public health measures to handle the future outbreaks of dengue.
2. Our molecular biological studies have shown that present practice of diagnosing dengue infection through IgM and IgG antibodies may not highlight true quantum of disease burden and that present practice of serological diagnosis need be coupled with molecular diagnosis.

Detailed study of serological and molecular parameters: Serological and molecular parameters of 42 serum samples of suspected cases of dengue were clustered to understand the disease pathogenicity. Following are the observations:

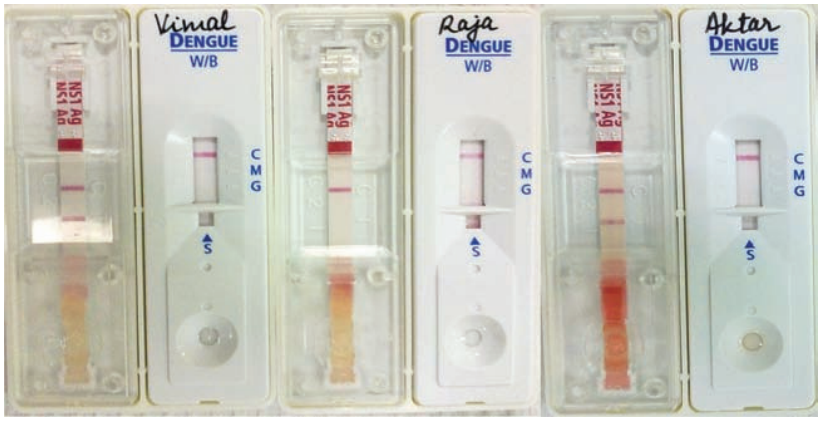


Fig.1. Dengue rapid test strip results for NS1, IgM and IgG.

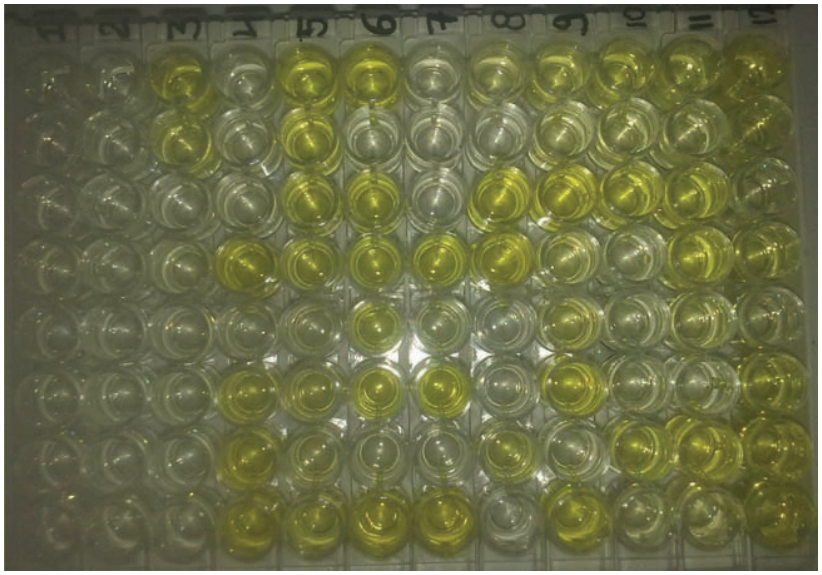


Fig. 2. Dengue Mac-ELISA test results for IgM antibodies

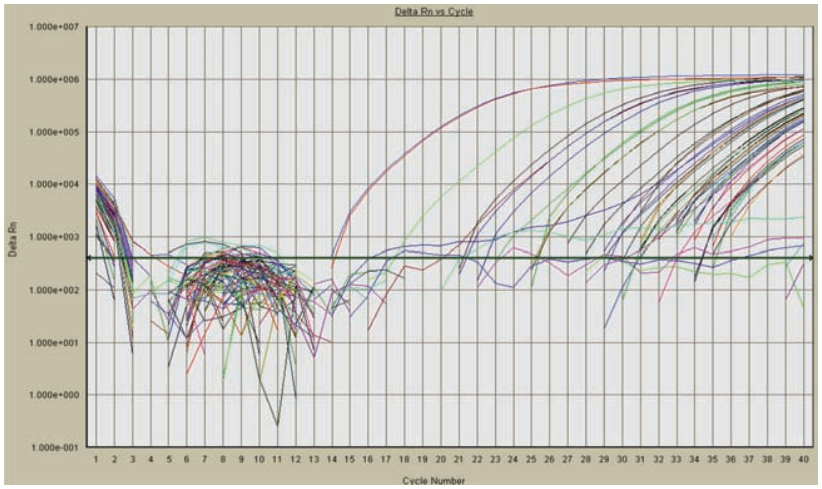


Fig. 3. RT-PCR graphs of serum and respective mosquitoes of the Dengue infected individuals.

Observations:

a. IgM +ve; IgG –ve, RT-PCR +ve

This condition denotes presence of current infection and evidence of antibody response against current infection (presence of IgM antibodies). The condition also indicates absence of antibodies against previous infection. In 4 of such samples RT-PCR results were positive.

b. IgM +ve; IgG –ve, RT-PCR -ve

This condition denotes presence of current infection and evidence of antibody response against current infection (presence of IgM antibodies). The condition also indicates absence of antibodies against previous infection. In none of such conditions RT-PCR was -ve. It means that if we observe IgM antibodies and no IgG in none of such conditions RT-PCR result will be –ve.

c. IgM +ve; IgG +ve, RT-PCR +ve

This combination indicates antibody response against current infection of dengue and antibody presence against past infection. The condition denotes +ve result of RT-PCR. During present studies we observed 11 such conditions.

d. IgM –ve; IgG +ve; RT -PCR +ve

This condition shows absence of anti body response against current infection, presence of antibodies against past infection and RT PCR +ve. We observed such condition for 5 of samples studies.

e. IgM –ve; IgG –ve; RT PCR +ve and NS1 +ve

This condition indicates absence of antibodies against current as well as past infection but a +ve result for PCR. We observed 5 such conditions where even none of antibodies was present but viral genome was observed. In all such cases NS1 antigen was present.

Inference:

1. For explaining the severe clinical conditions of patients of dengue *viz;* fall in platelet count, persistence of fever and bleeding etc., or anticipating such conditions in a patient, a condition of lack of antibodies presence and presence of viral genome could be crucial along with presence of NS1 viral protein.

Expected Impact of new knowledge generated: The new knowledge generated by DMRC on simultaneous investigations of parameters such as IgM, IgG, NS1 and RT-PCR and understanding of patient's clinical condition based on cumulative impact of these parameters, will help treating physicians in understanding the causes of severity of dengue cases.

Virus isolations from mosquitoes and correlation of extrinsic and intrinsic viraemia:

Aedes mosquitoes were collected from domestic premises of 42 patients suspected to be infected of dengue fever. The residential addresses of the patients were obtained from hospital records and collection of larval and adult *Aedes* mosquitoes was made during the period of illness of the patients. Details of virus isolations from larval and adult mosquitoes employing Indirect Fluorescence Antibody Test (IFAT) and RT-PCR are shown in Table 2.

Observations:

1. The observations depicted in Table 2 shows correlation of molecular presence of dengue virus from human patients and from mosquito collected from the houses of the patients. Out of 41 serum samples tested for presence of viral genome 27 was observed +ve for RT-PCR. Screening of houses of the 27 patients showing +ve results for dengue virus genome, 19 houses showed mosquitoes carrying the dengue virus while 8 mosquito samples shoed –ve results for presence of virus.
2. Out of 27 houses +ve for human dengue, 19 showing presence of virus in mosquito fauna is a high vector infectivity rate which is likely to continue across generations of mosquitoes through transovarial transmission of virus.

INFERENCES & EXPECTED IMPACT OF NEW KNOWLEDGE GENERATED:

1. The high vector infectivity rate by dengue virus could establish the persistent foci of virus presence in nature in the endemic localities. These foci may serve as a cause for prospective emergence of disease during time to come.
2. The extrinsic viral foci interphasing between current and prospective dengue outbreaks carries crucial etiological significance and should be targeted for the elimination.
3. The infected mosquito foci of dengue virus could be treated for elimination of interphasing virus to prevent prospective outbreaks.

Table 2. Correlation of Molecular isolation of intrinsic (from human serum samples) and extrinsic (from mosquito samples) viruses in study settings

Patient Code	Age	Sex	RT-PCR results of human sera virus	Adult mosq-uitoes	Larval mosquito	IFAT results	RT- PCR results of mosquito virus
1/11/MDM	NK	M	+	-	+	+	+
2/11/ MDM	25	M	+	+	+	+	+
3/11/ MDM	25	M	-	+	+	+	+
4/11/ MDM	14	F	-	+	+	+	-
5/11/ MDM	21	M	-	-	+	-	+
6/11/ MDM	15	F	+	+	-	-	+
7/11/ MDM	NK	M	+	+	+	+	-
8/11/ MDM	23	M	+	+	-	-	+
9/11/ MDM	14	F	+	+	+	+	+
10/11/ MDM	19	M	+	-	+	-	-
11/11/ UMD	8	F	+	-	+	-	+
12/11/UMD	NK	M	+	+	-	+	+
13/11/UMD	8	F	+	+	-	+	-
14/11/ UMD	7	M	+	-	+	+	-
15/11/ MDM	19	M	+	+	+	+	+
16/11/ UMD	10	F	+	+	-	-	+
17/11/UMD	11	F	+	-	+	-	+
18/11/UMD	5	M	+	+	+	-	+
19/11/UMD	10	M	+	-	+	-	+
20/11/UMD	9	M	+	+	+	+	+
21/11/MGH	13	M	+	+	+	+	+
22/11/UMD	16	M	+	+	+	+	-
23/11/MGH	39	M	-	+	+	+	-
24/11/UMD	4	F	+	-	+	+	-
25/11/UMD	7	M	+	+	+	+	+
26/11/UMD	18M	F	ND	+	-	ND	ND
27/11/MGH	80	M	+	+	-	-	+
28/11/MGH	NK	F	-	+	+	+	+
29/11/MGH	20	M	-	-	+	+	+
30/11/MGH	50	F	+	+	+	+	-
31/11/MGH	NK	M	+	+	+	+	+
32/11/MGH	30	M	+	-	+	+	+
33/11/MDM	19	M	-	+	-	-	-
34/11/MDM	20	F	-	+	+	+	-
35/11/MGH	19	F	-	-	+	-	-
36/11/MGH	16	F	-	-	+	+	+
37/11/MDM	17	M	+	+	-	-	-
38/11/MDM	45	M	-	-	+	-	+
39/11/MDM	18	M	-	-	+	+	+
40/11/MGH	NK	F	+	-	+	-	+
41/11/MGH	13	M	-	+	-	+	-
42/11/MGH	NK	M	ND				

1.3 Report on outbreak of fever/malaria cases in Bap village, Jodhpur District, Rajasthan

Principal Investigator: *Dr. Vinod Joshi, Scientist 'F'*
Co-Investigators: *Dr. K. R. Haldiya, Scientist 'F', Dr. M. L. Mathur, Scientist 'F' and Dr. S. P. Yadav, Scientist'E'*
Research Staff: *Dr. Bennet Angel, Research Associate, Ms. Annet Angel, Investigator and Ms. Nidhi Vyas, Investigator*
Commencement: September, 2011 **Duration:** Three Months **Status:** Completed
Funding: Desert Medicine Research Centre (Intramural)

OBJECTIVES

1. To undertake investigation of reported malaria outbreak in study area and establish causes of enhanced transmission leading to outbreak.
2. Demonstrate intervention at public health level to achieve control over ongoing outbreak.
3. Recommendations to state health department as guideline for future outbreaks.

PROGRESS

An outbreak of fever cases with suspected illness caused by malaria/viral pathogens was reported in the News Papers in jodhpur from 20.9.11 till 22.9.2011. The Joint Director, Jodhpur Zone, Department of Health & Family Welfare, Govt. of Rajasthan was contacted and offered expertise to investigate the situation to establish the cause of outbreak occurrence.

Situation analysis: Affected area Bap is situated in the north-west of Jodhpur district, Rajasthan (Fig. 1). A retrospective analysis of the malaria cases as reported in last three years in the records of the Primary Health Centre, Bap, Jodhpur District, Rajasthan was analyzed. It was observed that almost same numbers of malaria, as reported during current period, have been reported during last three years also. Epidemiologically, the same situation of malaria magnitude is persisting in the Bap area.

Investigations into affected villages - House Surveys: Team of scientists of DMRC, Jodhpur visited two villages Kanasar and Rawra from where malaria cases were reported to Bap PHC in abundance. The joint investigation team of DMRC scientists and staff of the state health department posted in Bap PHC visited the houses of patients. In the affected houses, the investigations were made with respect to blood report and treatment given to the patients of malaria by the treating physicians. Entomological investigations were made on the type of adult and larval mosquito species present in the houses, their densities, status of insecticide spray and breeding sites of mosquitoes.

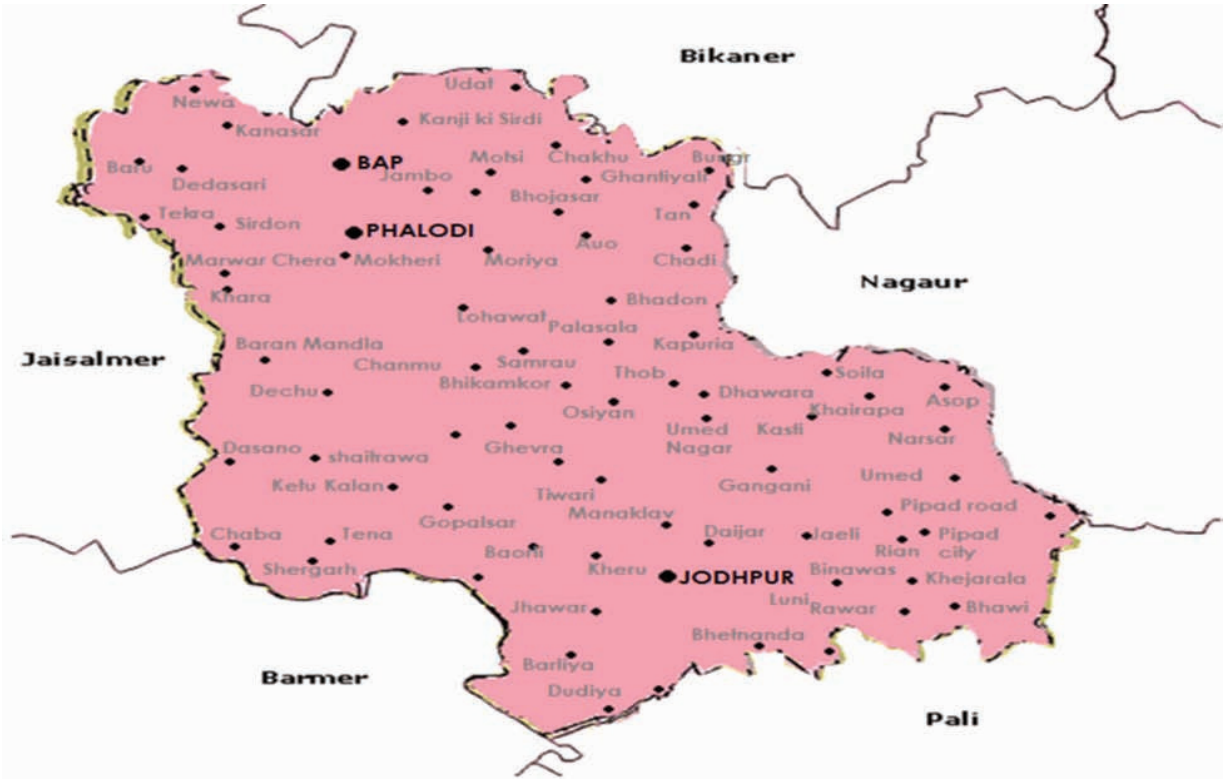


Fig. 1. Map of Jodhpur district, Rajasthan showing study areas

Investigations into affected villages -Village Pond Surveys: To ascertain the breeding habitats of anopheline mosquitoes in the malaria affected villages, the inter-stone spaces on the banks of village ponds were investigated for the presence of larvae. It was also studied how the water from the village pond were drawn and transferred to household water storage tanks.

OBSERVATIONS

1. Based on the reports of the blood slides taken by the hospital, the present outbreak was observed to be caused by malaria. Since almost similar numbers of cases were reported to BAP PHC every year during last three years, the present outbreak was not involving sudden and unexpected number of malaria cases.
2. Two anopheline species *viz*., *An. stephensi* and *An. subpictus* were observed during the course of investigation. The density of *An. Stephensi* was 24 PMH while that of *An. subpictus* was 40 PMH.
3. DDT spray was observed in the malaria affected houses but in spite of DDT spray, live adult anopheline mosquitoes were present.
4. No indoor breeding of mosquitoes was observed in the affected villages.
5. The mosquito breeding was observed in the gaps of inter-stone spaces forming banks of bond where water current inflows and outflows along with waves.

6. The village inhabitants appeared to import mosquito larvae from the village ponds to their house hold storage tanks of water.
7. In all 13 villages in and around Bap village, contributed to the current magnitude of malaria. It was observed that the cases reported in Bap PHC were treated by the available physicians.

Interventions by the DMRC scientists for controlling the malaria situation: Public health interventions to control malaria cases in Bap and adjoining villages were discussed by the DMRC scientists with the state health authorities. Following control strategy of malaria was suggested:

1. It was reported by the state health officers deputed in Bap village that all 13 affected villages were contributing malaria cases. DMRC suggested that we should simultaneously impose the parasite containment in cases through their door to door supply of anti malarial drugs in all 13 villages and that case treatment should be coupled with vector control (adult & larval) to check further transmission.
2. For pin pointing the active breeding sites in the village/houses, the state health staffs were trained by DMRC scientists.
3. The field staff of state health department were accompanied by DMRC scientists to the affected villages and an on the spot demonstration of anti-larval applications was given.

1.4 Translational Research for development and testing of ICMR-DMRC module of Dengue control for Rajasthan

Principal Investigator: *Dr. Vinod Joshi, Scientist 'F'*
Co-Investigator: *Dr. Manju Singhi, Scientist 'C'*
Research Staff: *Ms. Annette Angel, Invesgigator, Ms. Nidhi Vyas, Investigator, Mr. Rameshwar Lal, Investigator, Mr. Narendra Vyas, Investigator, Mr. Ajay Vyas, Investigator and Mr. Gajendra Singh, Investigator*
Commencement: March, 2011 **Duration:** Two Years **tatus:** Ongoing
Funding Agency: Indian Council of Medical Research–Translational Research **(**Extramural)

OBJECTIVES

1. Development of an ICMR-DMRC Dengue Control Plan for 7 zones of department of health & family welfare of Rajasthan
2. Demonstration of Dengue control/prevention in collaboration with the state health authorities using the larvicide used in programme
3. Demonstration and monitoring of disease control/ elimination for two consecutive years and handing over the documental manual of dengue control plan for Rajasthan

PROGRESS

Based on our earlier research that mosquitoes maintain dengue viruses in dengue endemic settings through the mechanism of Transovarial transmission (TOT), a subsequent translational research was conceived which was aimed to trace the virus activity in the larval mosquitoes in pre-transmission period (March-July) and get virus containing breeding habitats treated by the larvicide through state health department, so that a control over the disease transmission could be achieved in the prospective transmission season of dengue.

Entomological surveillance: During reported period the surveillance of breeding of larval *Aedes* has been undertaken in 9 district headquarters (Town/City). The areas namely Jalore, Pali, Sirohi, Barter, Jaisalmer, Jodhpur, Jaipur, Alwar and Dausa have been surveyed taking representative sampling from all the municipal wards of these towns. In all 193 households in Jalore, 302 in Pali, 492 in Sirohi, 335 in Barmer, 307 houses in Jaisalmer, 1852 in Jodhpur, 1568 in Jaipur, 403 in Alary and 309 in Dausa were screened. Maximum breeding was observed in Jaipur town (23.7 %). The details are provided in Table 1.

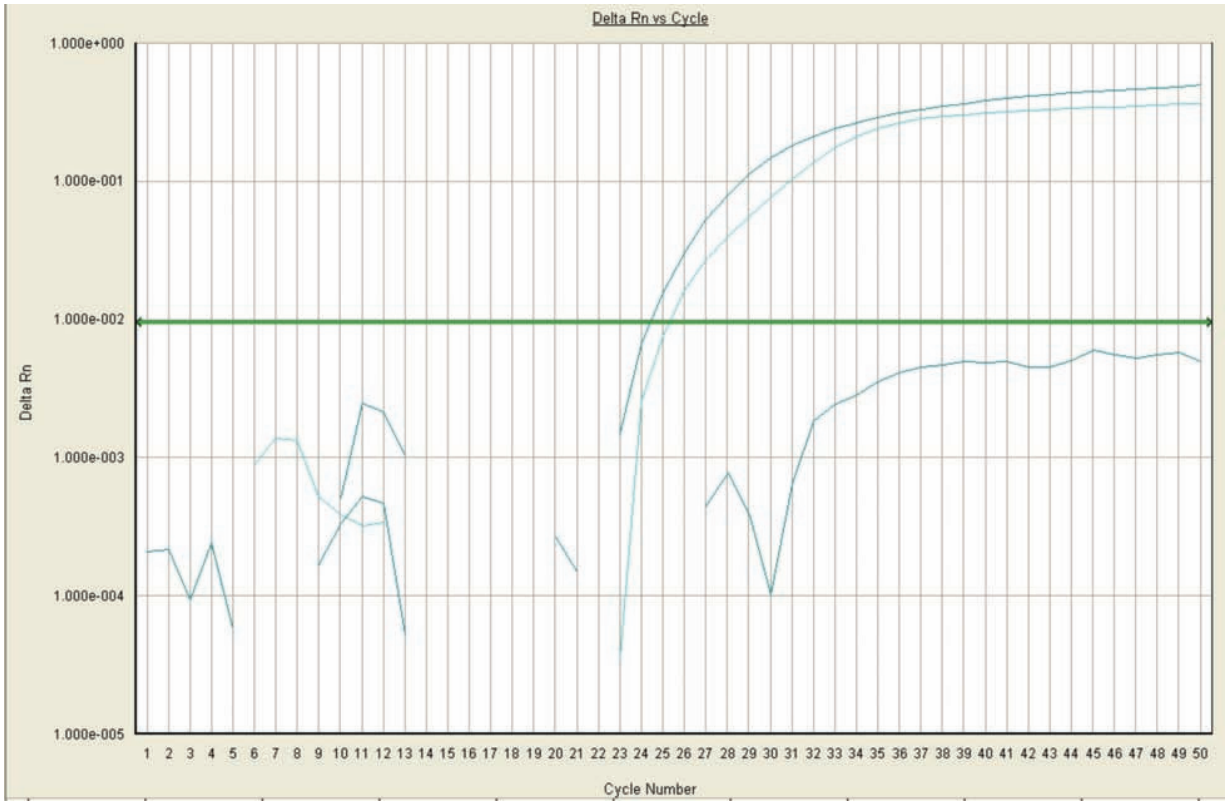
Virological investigations using IFAT: The field collected larvae were reared into adults under laboratory conditions. The laboratory reared adults were subjected to Indirect Fluorescence Antibody Test (IFAT) to screen mosquitoes carrying dengue antigen. Maximum mosquito infections were observed in Barmer town (53.6%). The details are provided in Table 1.

Table 1. Positive breeding containers and virus activities detected at household level in 9 district towns

Study Area	No. of Houses surveyed	No. Breeding +ve	% Breeding +ve	Conta-iners for Virus +ve	% Virus +ve	Total mosquitoes assayed	Positive for Dengue Virus	% +ve
Jalore	193	15	7.7	11	0.84	168	27	16.07
Pali	302	40	13.2	25	1.06	209	40	19.13
Sirohi	492	53	10.7	45	1.46	335	88	26.26
Barmer	335	28	8.3	21	1.38	82	44	53.65
Jaisalmer	307	18	5.8	9	0.58	44	16	36.36
Jodhpur	1852	226	12.2	258	2.56	866	212	24.48
Jaipur	1568	373	23.7	256	3.29	916	195	21.28
Alwar	403	40	9.92	42	2.05	98	27	27.55
Dausa	309	23	7.44	23	1.19	80	24	30.00
Total	**5761**	**816**	**14.1**	**690**	**2.18**	**2798**	**673**	**24.05**

Virological investigations using RT-PCR: Pools of mosquitoes as well individual mosquito body remnants showed positive results for IFAT were subjected to RT-PCR. All the individual mosquitoes showing IFAT positive showed positive result of presence of dengue viral genes (Fig.1).

Development of Early warning system and translation of observations into preventive actions: The virus activities as observed in the larvae of a particular house were used as indicators of prospective risk of dengue in a particular household. The details of address of house and key container where virus activity was recorded, communicated to state health department for taking anti-larval measures.



Well	Sample name	Detector	Task	Ct
E4	82 2F	Dengue	Unknown	24.32
F4	82 2 F	ACTB gene	Unknown	25.23

Fig. 1. RT-PCR curves showing +ve results of IFAT +ve mosquitoes

FINDINGS

1. Of the total houses screened for *Aedes* breeding, the percentage of breeding positive houses ranged from 5.8% minimum to 23.7% maximum. Data show that during pre-transmission season, mosquito breeding is confined to very limited breeding foci.
2. At household level, during pre-rain period the mosquitoes infected of dengue antigen ranged from 2.9 to 8.3% only. The observations clearly indicate that pre-rain period is the interphase of extrinsic virus activity and a translational approach of intervention during this time will eliminate the infected clusters of mosquitoes.

1.5 Study of pyrazinamide sensitivity of *Mycobacterium tuberculosis* as compared to nicotinamide sensitivity

Principal Investigator: *Dr. M. L. Mathur, Scientist 'F'*
Co-Investigator*: Dr. Aruna Solanki, Prof. & Head, Dept. of Microbiology, Dr. S. N. Medical College, Jodhpur.*
Commencement: March, 2011 **Duration:** One Years **Status:** Completed
Funding Agency: Indian Council of Medical Research –Translational research (Extramural)

OBJECTIVE

1. To find out utility of results of proportion method using nicotinamide against Wayne Method for pyrazinamide resistance in tuberculosis

PROGRESS

In tuberculosis, detection of resistance to pyrazinamide is difficult because an acidic pH of the medium is required to demonstrate activity of the drug and *Mycobacterium tuberculosis* strains do not grow well at acidic pH of 5.5. Secondly the use of too large an inoculum (over 10^7 bacilli/ml) leads to release of ammonia that increases pH and interferes in activity of pyrazinamide (PZA). As a result of these difficulties, there is lack of laboratories performing PZA susceptibility testing. This might be leading to gross under reporting and under estimation of resistance to pyrazinamide in tuberculosis. It was shown in initial observations at DMRC that if nicotinamide in LJ medium is used in place of pyrazinamide, it gives results similar to gold standard Wayne Method. Present study, therefore, used nicotinamide instead of pyrazinamide in proportion method using LJ medium for studying sensitivity to pyrazinamide on a larger sample. This proposed method would not require acidic pH in LJ medium and therefore, all strains of MTB will easily be tested for sensitivity using this method. The method is likely to prove useful for diagnosis of X-DR TB in RNTCP.

One Senior Research Fellow and one Field Investigator had joined in May 2012. They went to National Institute for Research in Tuberculosis (earlier known as TRC), Chennai along with PI, for two weeks training in culture and sensitivity of *M. tuberculosis* from 4th July 2011. In addition to practice of biosafety, they learned washing, staining, preparation of culture media, collection and processing of sputum samples and processing of extra pulmonary samples and proportion method for drug sensitivity of MTB.

Daily smear positive sputum samples of patients of K. N. Chest Hospital were collected with all biosafety precautions. Project staff prepares LJ medium slants without drug and with varying concentrations of Nicotinamide as per WHO guidelines. Sputum samples

are processed with Modified Petroff's Method as per TRC protocol and then the same are inoculated on drug free LJ medium. Then all inoculated LJ slants are incubated in incubator at 37° C. So far 290 (two hundred ninety) smear positive sputum samples have been inoculated.

When sufficient growth of *M. tuberculosis* was observed on LJ slants, sensitivity tests with proportion method using varying concentrations of nicotinamide (4, 5, 6, 7, 8 and 9 mg/ml) were carried out. For proportion method, TRC Protocol was followed. Results of this proportion method are being observed after 42 days of inoculation. So far results of 103 samples have been read after 42 days. Sensitivity to pyrazinamide by Wayne method (Gold Standard) was also studied in all these 103 samples. Wayne method was positive (means sensitive to pyrazinamide) for 87 and negative (means resistance to pyrazinamide) for 16 of these. Results of proportion method using different concentrations of nicotinamide in LJ medium are summarized in Table 1.

Table 1. Results of proportion method using different concentrations of Nicotinamide and results of Wayne Method

Conc. of Nicotinamide in LJ medium for Proportion Method		Results of Wayne Method		Performance of Nicotinamide Method	
		Positive (Sensitive to PZA)	Negative (Resistant to PZA)	Sensitivity (%)	Specificity (%)
4mg/ml	R	17	7	43.7	
	S	68	9		80.0
	C	2	0		
5mg/ml	R	11	5	33.3	
	S	75	10		87.2
	C	1	1		
6mg/ml	R	6	4	25.0	
	S	79	12		93.0
	C	2	0		
7mg/ml	R	5	3	18.7	
	S	80	13		94.1
	C	2	0		
8mg/ml	R	5	2	12.5	
	S	80	14		94.1
	C	2	0		
9mg/ml	R	2	1	6.2	
	S	83	15		97.6
	C	2	0		
TOTAL		**87**	**16**		

R=Resistant; S=Sensitive; C=Contamination

As shown in Table 1, Specificity of nicotinamide method was 80.2% to 97.5%, however sensitivity of this method using nicotinamide 4-9 mg/ml LJ medium was low, and better towards lower concentrations of nicotinamide. It was, therefore, thought that lower concentrations of nicotinamide would be more useful. The sensitivity testing of all strains has been repeated using nicotinamide 0.5, 1.0, 2.0 and 3.0 mg/ml LJ medium. Results are awaited. Results of DST of 23 strains with lower concentrations of nicotinamide (0.5 to 3 mg/ml) are available, which are shown in Table 2.

Table 2. Results of proportion method using different concentrations of Nicotinamide and results of Wayne Method

Conc. of Nicotinamide in LJ medium for Proportion Method		Results of Wayne Method		Performance of Nicotinamide Method	
		Positive (Sensitive to PZA)	Negative (Resistant to PZA)	Sensitivity (%)	Specificity (%)
0.5 mg/ml	R	6	11	100.0	
	S	5	0		45.5
	C	1	0		
1.0 mg/ml	R	1	9	81.8	
	S	11	2		91.7
	C	0	0		
2.0 mg/ml	R	1	8	72.7	
	S	11	3		91.7
	C	0	0		
3.0 mg/ml	R	1	7	63.6	
	S	11	4		91.7
	C	0	0		
Total		**12**	**11**		

R=Resistant; S=Sensitive; C=Contamination

As shown in Table 2, Specificity of nicotinamide method was 91.7% and sensitivity was 81.8% at 1.0 mg/ml LJ medium, which might be in acceptable limits. The data on lower concentrations of nicotinamide are encouraging and more results are awaited. The work is continuing and conclusions could only be drawn after getting observations of sufficient number of samples.

1.6 Development of molecular markers for the identification of Biological forms of *Anopheles stephensi* prevalent in arid areas of Rajasthan

Principal Investigator: *Dr. Karam V. Singh, Scientist 'F'*
Co-Investigators: *Dr. S. K. Bansal, Scientist 'F' and Dr. Himmat Singh, Research Assistant*
Research Staff: *Mr. Robin Marwal, Junior Research Fellow and Ms. Anusha Mishra, Research Assistant*
Commencement: February, 2010 **Duration:** Two Years **Status:** Ongoing
Funding Agency: Ministry of Environment and Forest (Extramural)

OBJECTIVES

1. Identification of biological forms *of Anopheles stephensi* using molecular tools and studies on their bionomics and distribution in different arid environs

PROGRESS

The studies during the report period have been carried out in Jodhpur, Jaisalmer and Barmer districts, as per project proposal. During the studies the main emphasis has been given on the investigations related to the distribution and bionomics of *An. stephensi* bioforms, present in the study areas. In each district two types of locations i.e. urban and rural were selected for the investigations and entomological surveys were carried out for the collection of adult as well as immature stages of *An. stephensi* bioforms, besides recording the data on associated factors. Two bioforms of *An. stephensi* species i.e. 'type' and variety *mysorensis* have so far been recorded from the study areas. The type and variety *mysorensis* have been differentiated on the basis of egg ridges. The type form has been identified as the eggs having ride count ≥ 16 and the var. *mysorensis* with ≤14 ridge count. In Jodhpur district, Public park, Rameshwar nagar and Bamba mohalla have been selected as urban localities, whereas, Jhalamund and Mahadev Nagar as rural. In Barmer district, the urban locations included Station road areas, whereas, Baitu, Jasol and Marudi villages as rural. In Jaisalmer district, Hanuman chowk represented urban area and Raghwa and Tejpala villages the rural ones. The breeding of both type and variety *mysorensis* was observed mainly in cemented tanks and type was found breeding in clean waters and variety *mysorensis* in turbid water.

Regarding morphological variations, the egg of var. *mysorensis* was found having both average length and width smaller than the length and width of the eggs of the type form Similarly, the Spiracular Index was also found smaller in case of var. *mysorensis* in comparison to type form.

During the report period, the insecticide susceptibility status of *An. stephensi* type and

var. *mysorensis* was determined using WHO Test Kit against DDT and Malathion. The experiments revealed that both type and var. *mysorensis* have developed resistance against DDT and Malathion (Table 1). Var. *mysorensis* exhibited high degree of resistance (Mortality 19.23%) against DDT, whereas, the high degree of resistance in case of Malathion was recorded against type form (mortality 8.33%). Against synthetic pyrethroid Cyfluthrin, both type and var. *mysorensis* were found susceptible.

Table 1. Insecticide susceptibility status of *An. stephensi* type and *mysorensis* forms against DDT and Malathion in study area

Insecticide used	Discriminating dose (%)	Exposure time (hrs)	Bioforms tested	Percent mortality (%)	Insecticide susceptibility Status*
DDT	4.0%	1.0	Type	55.0	Resistant
			mysorensis	19.2	Resistant
Malathion	5.0%	1.0	Type	8.3	Resistant
			Mysorensis	63.6	Resistant

*Insecticide Susceptibility WHO criteria: Percent mortality ≥98- Susceptible, 80-97.9- Intermediate Resistant, <80- Resistant

Table 2. Insecticide susceptibility status of *An. stephensi* var. *mysorensis* against two larvicides

Name of larvicide (Formulation)	Regression Equation	χ^2 Value (df)	LC_{50} with fiducial limits (ppm)	LC_{90} with fiducial limits (ppm)
Abate (EC)	Y= -14.82+16.21x	46.84* (6)	0.1006 (0.0696-0.1510)	0.2002 (0.1520-0.3375)
Bti (WP)	Y= -08.79+08.60x	48.60* (4)	0.0600 (0.0198-0.0225)	0.149 (0.0887-0.750)

*Heterogeneity of the response was found significant

The insecticide susceptibility status of larvae of An. stephensi var. mysorensis was determined against two common larvicides, Abate (OP compound) and Bacillus thuringiensis var. israelensis (Bti - a biolarvicide) following WHO method, in the laboratory (Temp. 28±2°C, RH 70±5%). Abate was used as emulsifiable concentrate (EC), whereas, Bti as wettable powder (WP). During the experiments mortality concentration data was obtained which was subjected to log-probit analysis for the calculation of LC50, LC90, and $\chi 2$ values (Table 2). The lethal concentrations determined during the experiments revealed that the species is still susceptible to both Abate and Bti and both can be used in the control programme as larvicide.

1.7 Current status of susceptibility of *Aedes aegypti* and *Anopheles stephensi* against larvicides/insecticides being used in National Programme in Rajasthan

Principal Investigator: *Dr. Karam V. Singh, Scientist 'F'*
Co-Investigators: *Dr. S. K. Bansal, Scientist 'F' and Dr. Himmat Singh, Research Assistant*
Commencement: October, 2010 **Duration:** Two Year **Status:** Ongoing
Funding: Desert Medicine Research Centre (Intramural)

OBJECTIVES

1. To determine the current status of susceptibility of *Ae. aegypti* and *An. stephensi* in rural, with particular reference to arid situations, and urban areas of Rajasthan against conventional insecticides being used under national control programme
2. Determination of biochemical mechanisms involved in the development of insecticide resistance, if any

PROGRESS

The studies were carried-out in 9 districts of Rajasthan state *viz.,* Ajmer, Alwar, Barmer, Bikaner, Jaipur, Jaisalmer, Jodhpur, Pali and Udaipur and in all the districts, except Jodhpur district, only urban localities were considered during the report period. During the field investigations, the main emphasis was given on the collection of adult and larval forms of both the vector species in different habitats potential for their resting and breeding.

Susceptibility tests were carried out with the adults and larvae of two mosquito species *viz*., *An. stephensi* and *Ae. aegypti,* collected from the field or reared in the Insectary under controlled conditions of temperature (28–2°C) and humidity (75–5%). Unfed females and third or early fourth instar larvae of these mosquito species were tested as per standard WHO methods for the determination of the baseline data on their susceptibility status using diagnostic doses of the test insecticides/larvicides. The tests were conducted against adulticides *viz*. DDT, Malathion, Fenitrothion, Propoxur and Cyfluthrin and Abate larvicide. Four to five replicates of each observation were made and the data was subjected to log probit regression analysis for the determination of lethal concentrations at 50 and 90 percent levels. WHO criteria was followed for the determination of the susceptibility status of individual species.

An. stephensi adults of districts Jodhpur and Bikaner were tested against the diagnostic doses of Fenitrothion, Propoxur and Cyfluthrin and the results of the tests revealed that this species, in Jodhpur district, was found susceptible to Fenitrothion and developed intermediate resistance to Propoxur, however, in Bikaner the species was found susceptible to Cyfluthrin (Table 1).

Table 1. Susceptibility status of *An. stephensi* against Fenitrothion, Propoxur and Cyfluthrin

Districts	Insecticide & Diagnostic Dose	Exposure time in Hours	Mortality %	Susceptibility Status*
Jodhpur	Fenitrothion,1.0%	2	100.0	S
	Propoxur, 0.1%	1	92.0	IR
Bikaner	Cyfluthrin, 0.15%	1	100.0	S

*S- Susceptible, IR- Intermediate Resistant

The susceptibility status of *An. stephensi* was also determined against Abate larvicide in the urban areas of Ajmer, Bikaner, Jaisalmer and Jodhpur (rural also) and the species was found totally susceptible to Abate. The lethal concentrations were determined at 50 and 90 percent levels against *An. stephensi* in all the four districts and it was found that at 90 percent level, Abate was maximum effective in Jodhpur urban, followed by Jodhpur rural, Jaisalmer, Bikaner and Ajmer (Table 2). The heterogeneity of the response of *An. stephensi* populations tested in all the cities and was found statistically significant in Ajmer and Jodhpur urban areas.

Table 2. Determination of lethal concentrations of Abate larvicide against *An. stephensi*

Districts	Regression equation	LC_{50} (ppm)	LC_{90} (ppm)	Chi Square (df)
Ajmer - Urban	Y= -0.67+ 3.07X	0.188	0.540	8.23*(3)
Bikaner - Urban	Y= -1.49+5.06X	0.029	0.389	2.41(4)
Jaisalmer - Urban	Y= -1.77+4.86X	0.170	0.293	3.90(4)
Jodhpur -Urban	Y= -0.80+13.86X	0.061	0.158	136.5*(7)
Jodhpur -Rural	Y= -1.78+ 3.15X	0.113	0.195	2.02(2)

*Heterogeneity of the response was found statistically significant

Ae. aegypti adults, reared from the larvae collected from urban localities of Barmer, Jaisalmer and Udaipur, were tested for their insecticide susceptibility status against DDT, Malathion and Cyfluthrin (Table 3). The testes conducted in Jaisalmer and Udaipur districts revealed that *Ae. aegypti* adults are still susceptible to all the three adulticides, whereas, the adults from Barmer were found susceptible to Malathion and Cyfluthrin, but resistant to DDT (Table 3).

Table 3. Susceptibility status of *Ae. aegypti against* DDT, Malathion and Cyfluthrin

Districts	Insecticide & Diagnostic Dose	Exposure time (Hrs)	Mortality%	Susceptibility Status*
Barmer	DDT, 4.0%	1	64.1	R
	Malathion, 5.0%	1	100.0	S
	Cyfluthrin, 0.15%	1	100.0	S
Jaisalmer	DDT, 4.0%	1	98.5	S
	Malathion, 5.0%	1	100.0	S
	Cyfluthrin, 0.15%	1	100.0	S
Udaipur	DDT, 4.0%	1	98.6	S
	Malathion, 5.0%	1	100.0	S
	Cyfluthrin, 0.15%	1	100.0	S

*S- Susceptible, R- Resistant

Table 4. Susceptibility status of *Ae. aegypti* against Abate

Districts	Diagnostic Dose	No. exposed	No. Dead	Mortality%	Susceptibility status
Alwar	0.02 ppm	100	53	53.0	R
Bikaner	0.02 ppm	100	23	23.0	R
Jodhpur	0.02 ppm	50	41	82.0	IR
Pali	0.02 ppm	100	96	96.0	IR
Udaipur	0.02 ppm	50	26	52.0	R

*R- Resistant, IR- Intermediate Resistant

The susceptibility status of *Ae. aegypti* was also determined against Abate larvicide in the urban areas of Alwar, Bikaner, Jodhpur, Pali and Udaipur cities and the species was found totally resistant to Abate in Alwar, Barmer, Bikaner, and Udaipur, however, intermediate resistant in Jodhpur and Pali districts (Table 4).

The lethal concentrations of Abate were also determined at 50 and 90 percent levels against *Ae. aegypti* in seven districts and it was found that at 90 percent level, Abate was maximum effective in Jaisalmer, followed by Udaipur, Bikaner, Alwar, Ajmer, Jaipur and Jodhpur (Table 5). The Chi-square values revealed that heterogeneity of the response of *Ae. aegypti* population in Ajmer, Alwar, Jaipur and Udaipur cities was found statistically significant.

Table 5. Determination of lethal concentrations of Abate larvicide against *Ae. aegypti*

Districts	Regression equation	LC_{50} (ppm)	LC_{90} (ppm)	Chi Square(df)
Ajmer	Y= -0.58+3.07X	0.032	0.104	9.0*(3)
Alwar	Y= -0.77+8.60X	0.032	0.085	29.0*(5)
Bikaner	Y= -1.96+7.88X	0.027	0.045	4.1(3)
Jaisalmer	Y= -1.23+4.04X	0.009	0.019	4.4(5)
Jaipur	Y= -0.64+6.82X	0.036	0.106	21.1*(4)
Jodhpur	Y=-0.53+16.44X	0.024	0.820	5.1(8)
Udaipur	Y= -1.08+6.98X	0.013	0.029	40.2*(5)

*Heterogeneity of the response was found statistically significant

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

Principal Investigator: *Dr. S. K. Bansal, Scientist 'F'*
Co-Investigator: *Dr. Karam V. Singh, Scientist 'F'*
Commencement: June, 2009 **Duration:** Three Years **Status:** Completed
Funding: Desert Medicine Research Centre (Intramural)

OBJECTIVES

1. Determination of the larvicidal activity present in different parts of the plants after extraction in different organic solvents
2. Extraction of oils 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 and comparison of their larvicidal/repellant properties

PROGRESS

Studies are being carried out on *Withania somnifera*, also known as Ashwagandha, a plant in Solanaceae or nightshade family. Fruits, leaves and seeds of this plant have been traditionally used for the Ayurvedic system as aphrodisiacs, diuretics and for treating memory loss. Larvicidal properties have been observed in some parts of this plant 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 stephens*i, *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. 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°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, acetone and petroleum ether extracts of fruits without seeds of *W. somnifera* are given in Tables 1-3. With all the mosquito species mortality was dose and duration dependent i.e. mortality increased with increase in concentration. 24 and 48 hr LC_{50} values along with their 95% confidence limits, LC_{90} values, regression equation and chi-square were calculated. 24 and 48 hr LC_{50} values as observed for *An. stephens*i, *Ae. aegypti* and *Cx. quinquefasciatus* for methanol extracts were 88.4, 404.4 & 1030.0 and 54.9, 238.6 & 474.7 mg/l (Table 1); for acetone extracts were 80.2,97.6 & 146.6 and 60.8, 53.5 & 87.7 mg/l (Table 2) while for petroleum ether extracts were 30.0, 44.5 & 54.2 and 22.4, 27.8 & 33.0 mg/l (Table 3) respectively. The results suggest that the petroleum ether extracts were more effective followed by acetone and methanol extracts against all the mosquito species tested. Anophelines were found more susceptible than the culicine species when tested with fruit without seeds against all the three solvents tested in the present study.

Larval susceptibility tests were also carried out with methanol, acetone and petroleum ether extracts of seeds of *W. somnifera* and the results are given in Tables 4-6. 24 and 48 hr LC_{50} values as determined for *An. stephens*i, *Ae. aegypti* and *Cx. quinquefasciatus* for methanol extracts were 245.5,769.0 & 1169.0 and 141.6, 402.6 & 609.5 mg/l (Table4); for acetone extracts were 188.1, 777.5 & 822.5 and 101.6, 650.2 & 718.9 mg/l (Table5) while for petroleum ether extracts were 140.3, 822.9 & 778.4 and 81.6, 765.8 & 668.9 mg/l (Table6) respectively. The results here also suggest that the petroleum ether extracts were more effective followed by acetone and methanol extracts against all the mosquito species tested. Anophelines were found more susceptible than the culicine species when tested with seeds of *W. somnifera* against all the three solvents tested in the present study. 48hr LC_{50} values were much less as compared to 24hr LC_{50} indicating that plant extracts are much effective after 48hr instead of after 24hr (Table 7).

More experiments are being carried out with different aqueous and organic solvent extracts with different parts of this plant species in order to see the comparative efficacy of these solvents on these mosquito species in this arid region.

Table 1. Log probit regression analysis of the mortality data of larvae of different mosquito vectors to methanol extracts of fruit without seeds of *W. somnifera*

Mosquito Species	Regression Equation	χ^2, df =3 ***(p value)***	24hr LC_{50} **(LC_{90})** (95% C. limits)	48hr LC_{50} **(LC_{90})** (95% C. limits)
An. stephensi	Y=2.14x +0.83	0.19 ***(.979)***	88.4 **(349.3)** (66.1-118.2)	54.9 **(215.6)** (41.1-73.5)
Ae. aegypti	Y=1.17x +1.95	0.23 ***(.973)***	404.4 **(5029.0)** (226.3-722.8)	238.6 **(1999.0)** (157.5-361.6)
Cx. quinquefasciatus	Y=1.20x +1.38	0.04 ***(.998)***	1030.0 **(12010)** (437.7-2426)	474.7 **(4024.0)** (292.3-771.1)

24 and 48hr LC_{50} and LC_{90} values along with their 95% Confidence limits are in mgl^{-1}
χ^2 - Chi Square; df - Degree of Freedom; Values in bold in parentheses are the LC_{90} values
Y and x are the Expected Probits and Log Concentrations respectively

Table 2. Log probit regression analysis of the mortality data of larvae of different mosquito vectors to acetone extract of fruits without seeds of *W. somnifera*

Mosquito Species	Regression Equation	χ^2 (df =3) ***(p value)***	24hr LC_{50} **(LC_{90})** (95% C. limits)	48hr LC_{50} **(LC_{90})** (95% C. limits)
An. stephensi	Y=1.88x +1.42	0.41 ***(.938)***	80.2 **(384.0)** (59.9-107.3)	60.8 **(270.5)** (47.1-78.5)
Ae. aegypti	Y=1.86x +1.30	0.14 ***(.986)***	97.6 **(475.8)** (70.8-134.6)	53.5 **(228.5)** (39.4-72.6)
Cx. quinquefasciatus	Y=1.99x +0.68	1.40 ***(.706)***	146.6 **(643.7)** (108.1-198.9)	87.7 **(425.6)** (64.9-118.7)

24 and 48hr LC_{50} and LC_{90} values along with their 95% Confidence limits are in mgl^{-1}
χ^2 - Chi Square; df - Degree of Freedom; Values in bold in parentheses are the LC_{90} values
Y and x are the Expected Probits and Log Concentrations respectively

Table 3. Log probit regression analysis of the mortality data of larvae of different mosquito vectors to petroleum ether extracts of fruit without seeds of *W. somnifera*

Mosquito Species	Regression Equation	χ^2 (df =3) ***(p value)***	24hr LC_{50} **(LC_{90})** (95% C. limits)	48hr LC_{50} **(LC_{90})** (95% C. limits)
An. stephensi	Y=2.03x +2.00	0.88 ***(.830)***	30.0 **(127.7)** (22.4-40.2)	22.4 **(71.6)** (17.4-28.7)
Ae. aegypti	Y=1.82x +2.00	1.57 ***(.666)***	44.5 **(225.3)**(32.7-60.7)	27.8 **(176.7)**(19.5-39.8)
Cx. quinquefasciatus	Y=2.28x +1.05	0.81***(.847)***	54.2 **(197.7)** (41.8-70.4)	33.0 **(107.4)** (24.7-44.1)

24 and 48hr LC_{50} and LC_{90} values along with their 95% Confidence limits are in mgl^{-1}
χ^2 - Chi Square; df - Degree of Freedom; Values in bold in parentheses are the LC_{90} values
Y and x are the Expected Probits and Log Concentrations respectively

Table 4. Log probit regression analysis of the mortality data of larvae of different mosquito vectors to methanol extracts of seeds of *W. somnifera*

Mosquito Species	Regression Equation	χ^2 (df =3) ***(p value)***	24hr LC_{50} **(LC_{90})** (95% C. limits)	48hr LC_{50} **(LC_{90})** (95% C. limits)
An. stephensi	Y=1.22x +2.08	0.16 ***(.983)***	245.5 **(2747.0)** (140.2-429.9)	141.6 **(1265.0)** (92.4-226.6)
Ae. aegypti	Y=1.34x +1.12	0.41 ***(.938)***	769.0 **(6902.0)** (369.4-1601.0)	402.6 **(2839.0)** (256.3-632.5)
Cx. quinquefasciatus	Y=1.54x +0.28	3.13 ***(.372)***	1169.0 **(7949.0)** (566.9-2409.0)	609.5 **(5525.0)** (347.0-1070.0)

24 and 48hr LC_{50} and LC_{90} values along with their 95% Confidence limits are in mgl^{-1}
χ^2 - Chi Square; df - Degree of Freedom; Values in bold in parentheses are the LC_{90} values
Y and x are the Expected Probits and Log Concentrations respectively

Table 5. Log probit regression analysis of the mortality data of larvae of different mosquito vectors to acetone extracts of seeds of *W. somnifera*

Mosquito Species	Regression Equation	χ^2 (df =3) ***(p value)***	24hr LC_{50} **(LC_{90})** (95% C. limits)	48hr LC_{50} **(LC_{90})** (95% C. limits)
An. stephensi	Y=2.04x +0.35	2.62 ***(.454)***	188.1 **(794.3)** (138.1-256.4)	101.6 **(305.9)** (79.6-129.9)
Ae. aegypti	Y=1.08x +1.54	0.45 ***(.929)***	777.5 **(1448.4)** (625.2-1122.7)	650.2 **(1208.0)** (463.1-987.0)
Cx. quinquefasciatus	Y=1.09x +1.36	0.70 ***(.873)***	822.5 **(1457.3)** (541.4-1275.5)	718.9 **(1287.2)** (501.8-1217.0)

24 and 48hr LC_{50} and LC_{90} values along with their 95% Confidence limits are in mgl^{-1}
χ^2 - Chi Square; df - Degree of Freedom; Values in bold in parentheses are the LC_{90} values
Y and x are the Expected Probits and Log Concs. respectively

Table 6. Log probit regression analysis of the mortality data of larvae of different mosquito vectors to petroleum ether extracts of seeds of *W. somnifera*

Mosquito Species	Regression Equation	χ^2 (df =3) ***(p value)***	24hr LC_{50} **(LC_{90})** (95% C. limits)	48hr LC_{50} **(LC_{90})** (95% C. limits)
An. stephensi	Y=1.87x +0.98	0.46 ***(.928)***	140.3 **(678.9)** (103.5-190.3)	81.6 **(224.8)** (65.2-102.2)
Ae. aegypti	Y=0.81x +2.24	0.24 ***(.971)***	822.9 **(1612.0)**(640.2-1296.0)	765.8 **(1174.9)**(595.7-1211.3)
Cx. quinquefasciatus	Y=1.32x +1.15	1.75 ***(.626)***	778.4 **(1164.2)**(414.0-1463.0)	668.9 **(1086.8)**(371.6-1214.0)

24 and 48hr LC_{50} and LC_{90} values along with their 95% Confidence limits are in mgl^{-1}
χ^2 - Chi Square; df - Degree of Freedom; Values in bold in parentheses are the LC_{90} values
Y and x are the Expected Probits and Log Concs. respectively

Table 7. Comparative efficacy of fruit without seeds and seeds of *W. somnifera* against different mosquito species in different organic solvents

Mosquito species	Solvent used	Fruits without seeds		Seeds	
		24hr LC_{50} **(LC_{90})** (95% C. limits)	48hr LC_{50} **(LC_{90})** (95% C. limits)	24hr LC_{50} **(LC_{90})** (95% C. limits)	48hr LC_{50} **(LC_{90})** (95% C. limits)
An. stephensi	Methanol	88.4 **(349.3)** (66.1-118.2)	54.9 **(215.6)** (41.1-73.5)	245.5 **(2747.0)** (140.2-429.9)	141.6 **(1265.0)** (92.4-226.6)
Ae. aegypti		404.4 (**5029**) (226.3-722.8)	238.6 **(1999)** (157.5-361.6)	769.0 **(6902.0)** (369.4-1601.0)	402.6 **(2839.0)** (256.3-632.5)
Cx. quinque-fasciatus		1030.0 (**2010**) (437.7-2426)	474.7 **(4024)** (292.3-771.1)	1169.0 **(7949.0)** (566.9-2409.0)	609.5 **(5525.0)** (347.0-1070.0)
An. stephensi	Acetone	80.2 **(384.0)** (59.9-107.3)	60.8 **(270.5)** (47.1-78.5)	188.1 **(794.3)** (138.1-256.4)	101.6 **(305.9)** (79.6-129.9)
Ae. aegypti		97.6 **(475.8)** (70.8-134.6)	53.5 **(228.5)** (39.4-72.6)	777.5 **(1448.4)** (625.2-1122.7)	650.2 **(1208.0)** (463.1-987.0)
Cx. quinque-fasciatus		146.6 **(643.7)** (108.1-198.9)	87.7 **(425.6)** (64.9-118.7)	822.5 **(1457.3)** (541.4-1275.5)	718.9 **(1287.2)** (501.8-1217.0)
An. stephensi	Petroleum Ether	30.0 **(127.7)** (22.4-40.2)	22.4 **(71.6)** (17.4-28.7)	140.3 **(678.9)** (103.5-190.3)	81.6 **(224.8)** (65.2-102.2)
Ae. aegypti		44.5 **(225.3)** (32.7-60.7)	27.8 **(176.7)** (19.5-39.8)	822.9 **(1612.0)** (640.2-1296.0)	765.8 **(1174.9)** (595.7-1211.3)
Cx. quinque-fasciatus		54.2 **(197.7)** (41.8-70.4)	33.0 **(107.4)** (24.7-44.1)	778.4 **(1164.2)** (414.0-1463.0)	668.9 **(1086.8)** (371.6-1214.0)

24 and 48hr LC_{50} and LC_{90} values along with their 95% Confidence limits are in mgl^{-1}
Values in bold in parentheses are the LC_{90} values

1.9 A study of factors affecting incidence of malaria in children in desert part of Rajasthan

Principal Investigator: *Dr. S. P. Yadav, Scientist 'E'*
Co-Investigators: *Dr. A. K. Dixit, Scientist 'E' and Mr. R. K. Kalundha, Technical Officer*
Commencement: September, 2008 **Duration**: Three Years **Status**: Completed
Funding: Desert Medicine Research Centre (Intramural)

OBJECTIVES

1. To know the current status of malaria in children in desert part of Rajasthan
2. To know the factors affecting incidence of malaria in children

PROGRESS

A cross-sectional community based study was under taken in the desert. Jaisalmer district was selected as study area on some basic criterion, few of them are mentioned here. Out of 12 desert districts of Rajasthan Jaisalmer was meeting criteria of high API for the last 16 years. To study the incidence of malaria in western Rajasthan, malaria data for six districts was collected from the office of Joint Director, Health and Medical Services, Zone Jodhpur. The average malaria API was classified into five groups i.e. less than 2 API, 2-5, 5-10, 10-20 and more than 20 API. Thus, Jaisalmer was with highest API. Malaria in the district was highly problematic due to mismanagement of water supply to the villages for irrigation and drinking purposes through distributaries of IGNP.

There are 18 PHCs in Jaisalmer district. Due to large area and highest API Ramgarh PHC was selected for the study area. There are 65 villages in Ramgarh PHC. These villages were classified and divided into two groups namely Command and Non-command villages. Command villages (CVs) were defined here as the villages where the water was available for the irrigation and drinking purposes for the last 20 years or more through IGNP and Non-command villages (NCVs) were defined as where water was yet to reach for the same purpose. Using random sampling method, 9 villages from each category of villages were selected namely Seowa, Raghwa, Radial, Sultana, Nagga, Buda, Mocha, Nehdai and Lanera from the CVs and Habur, Kafka, Hairs, Tibansar, Champagne ki Dhani, Markh ka Ganv, Mohammad Khan ki Dhani, Ranau and Tanot were selected from the NCVs. Thus, a total of 18 study villages of both the groups were selected. 30 households were selected randomly from the each selected village for this study. A total of 540 (270 CVs + 270 NCVs) households were surveyed from both the groups of the villages.

The questionnaires were prepared with pre-coded and open ended questions. It was pre-tested in 50 households in the nearest village from Ramgargh PHC which suited with highest criteria. Modifications were made accordingly. The questionnaires were prepared

in English but it was communicated to the informant in Hindi or local dialect *i.e. Marwari* (dialect of people in Thar Desert). Received information was translated in English and recorded in the study schedules. Focus Group Discussions (FGDs) were also held in the selected villages by the investigators with the informants. All the guide lines for FGDs were followed to control quality of data. Pre-tested schedules were used for the data collections on socio-demographic, socio-economic, socio-cultural and health practices, migration and human behavior by door to door survey. Prasad classification was used for the categorization in different social class of respondents i.e. Upper (I), Upper Middle (II), Lower Middle (III), Upper Lower (IV) and Lower (V). FGDs were also held on some events such as marriage, birth day and so on in the study villages. Information such as number of fever cases, collection and examination of blood slide and status of slide in the selected households of the fever cases for the examination of malaria parasite was obtained from health records of PHC. Collected data from the field was computed and analyzed (Table 1).

Table 1. Distribution of children according to their age and sex

Age (Yrs)	Male		Female		Total	
	No.	%	No.	%	No.	%
<1	122	26.6	110	28.3	232	27.5
1-5	100	21.9	93	23.9	193	22.8
5-10	98	21.5	71	18.3	169	20.0
10-15	79	17.3	70	18.0	149	17.6
15+	57	12.5	45	11.6	102	12.1
Total	**456**	**100.0**	**389**	**100.0**	**845**	**100.0**

A total of 845 children were examined in surveyed households. Malaria incidence was significantly higher in children (136.1 per 1,000 population) as compared to adults (49.4 per 1,000 population) and overall it was 71.3 per 1000 population. Nearly seven percent (6.8%) of individuals had two or more malaria episodes to a maximum of five during period of last one year from the date of interview. The children whose both the parents were uneducated were delaying more (81.3%) for the diagnosis and treatment for the febrile children as compared to (37.5%) those who's one or both the parents were educated. Most common reason was long distance of the health facility and lack of transport facility particularly in night. Camel cart and tractor were most of the common transport facility.

It was observed that individual level factors were associated with the malaria incidence in the study area. Children less than five years of age were more sufferers as compared to the other age group of children. Similarly among the adult population less than 40 years

of age were more affected with the disease. It shows that age is one factor affecting at individual level. Self protection measures such as use of bed nets against mosquito bites was also very low (0.7%) which attracts attention (Table 2).

Table 2. Socio-economic characteristics of the studied children

Social Characterstics	Households (children)
Number	540
Mother's education (%)	
Illeterate	51.4(433/842)
Primary	33.2(279/842)
Secondary or higher	15.4(130/842)
Mother's occupation (%)	
farmer/labourer	98.8(825/835)
Father's education (%)	
Illeterate	28.2(231/820)
Primary	42.7(350/820)
Secondary or higher	29.1(239/820)
Father's occupation (%)	
Farmer/labourer	87.5(710/811)
Others	9.1(74/811)
Died/left/at home	3.3(27/811)
Number of family members/household	6.1(2-13)
Number of children/Household	2.8(2-10)
Facilities at Household (%)	
Electricity	17.2(93/540)
Tap water	11.3(61/540)
Radio	4.4(24/540)
TV	1.9(10/538)
Cupboard	2.8(15/539)
Bicycle	3.3(18/540)
Motor-cycle	1.3(7/540)
Fridge	0.4(2/540)
Cattle	99.1(535/540)
Child received any drug in last 2 weeks	1.7(9/540)
Child used bed net last night (%)	0.7(4/540)

More than three fourth (76.3%) households were in dhanies *i.e.* hamlets which were located in the agriculture farms and away from the main village. These dhanies were located 1-10 km from each other dhanies/main village within the demarcated area of the village. Majority (93.1%) of dhanies were not having pucca road and public transport facility. The cows, camels, goats and sheep were the main cattle of households. Many households used household-level prevention against malaria. About two third households were burning cow dung for the prevention of the mosquito bites. However, very few (2.9%) had any window screens or self owned bed nets (3.1%). Distance from the major vector breeding sites and number of adults with indoor jobs were the factors significantly associated with malaria incidence in children. In children of all ages, house distance to the breeding site exerted a profound effect on malaria incidence, particularly within 350m from the breeding site as compared to more than 350m.

IMPORTANT LEADS/OUTCOME FROM THE STUDY

Data may be useful to planners, researchers and implementers in regard to malaria control.

WORK REMAINS TO BE DONE

Study is completed. Final and complete report will be prepared and will be submitted for the publication.

1.10 A study of association between socio-economic factors and transmission of malaria in desert

Principal Investigator: *Dr. S. P. Yadav, Scientist 'E'*
Co-Investigators: *Dr. A. K. Dixit, Scientist 'E' and Mr. R. K. Kalundha, Technical Officer*
Commencement: October, 2007 **Duration**: Three Years **Status**: Completed
Funding: Desert Medicine Research Centre (Intramural)

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

The Thar Desert spreads across the state of Rajasthan and parts of Gujarat in western India covering about 2,59,000 sq km. At present, the Thar Desert of Rajasthan, comprising 12 districts, is spread over a 28,600 km^2 which contributes to 12% of the mainland of the country and 62% of the state. It is also the most populated desert of the world. A cross-sectional community based study was under taken in the desert. Jaisalmer district which was selected as study area for having highest API for last 16 years. The study villages for this study remained the same as were taken in our previous study (1.9)

OBSERVATIONS

Socio-demographic characteristics of the study subjects: Majority (61.7%) of the respondents were belonging to the age group of 25 - 44 years, in which males were 68.5% and females were 31.5%. The confidence interval of mean age was 34.8 ± 11.5 years, Kurtosis -0.1025, Skewness 0.782, sample variance 133.12 median age 34 years, mode age 25 years, smallest age 18 and largest age 65 years. Agriculture (42.5%) was the main occupation of the respondents followed by mining (26.0%), service (11.0%), business (6.6%) and labour (5.2%) respectively. Literacy rate was 60.8% and only 13.7% have done high school and higher education which shows poor education level of the study population. In the study, distribution of Hindu population was 78.1% and non-Hindus were 21.9%. Among the Hindus population, general castes were distributed 49.8%, backward caste 29.1%, schedule caste and schedule tribe 21.1%. The marital status of the population was very high, where 95.9% were married (Table 1).

Malaria illness concept: The major proportion 331 (90.7%) of the respondents considered *'TAV'* as fever in local dialect *i.e.* called *Marwari*. These respondents explained *'TAV'* is raised temperature of the body as compared to normal. They confirm fever by touching

body of febrile person and classified as low, moderate and high based on past experiences. Further, more in depth '*TAV*' was classified as '*HEETAV*' (fever with shivering) or '*EKANTRA TAV*' (fever on alternate days).

Knowledge of malaria: Table 2 depicts the status of knowledge of the respondents about the symptoms of the malaria. In totality, the study subjects reported that important symptoms of malaria were fever (98.9%), headache (94.2%), chills (91.5%), rigors (94.5%), vomiting (92.1%), Arthralgia (86.0%) and back ache (75.1%). There was no significant difference among the male and females respondents with respect to symptoms like Fever, Chills, Rigors, Vomiting and Back Ache. However, significant differences were noted with respect to Headache and Arthralgia. The majority (71.7%) of the respondents told that antimalarial drugs were the appropriate medicine to treat malaria immediately. Simultaneously, 33.4% respondents also stated that traditional medicine was the permanent cure of the disease. Nearly sixty two per cent (62.4%) respondents opined that pregnant women could not be given antimalarial drugs due to its side effects such as abortions could take place spontaneously or congenital defects might occur in new born babies. Besides this, 24.1% study subjects expressed their views that fever is dangerous, if it is not cured; it leads to further complications in the body and ultimately death.

Vector of malaria: All most eighty four per cent (83.7%) respondents said that malaria is spread by female mosquitoes. There were no significant variations with respect to age, sex, education, occupation, or past malaria infection of the respondents. About thirty-five per cent (34.8%) of the respondents stated that malaria can be due to unhygienic living conditions, dirty drinking water and impure and unbalanced diet. This belief was found to vary little with religion and caste.

Causes of malaria: Nearly half (51.3%) of the respondents told that malaria parasite was the cause of disease. 7.3% respondents stated that they didn't know the cause of malaria. Knowledge regarding malaria parasite as the cause of disease was found higher with increased level of education (87.6% in secondary and above as compared to 28.6% in primary school) of the respondents. About twenty-six per cent (25.9%) illiterate respondents opined multiple cause of malaria such as changing environment, unhygienic surroundings around the household, and also through devine curse of God or ancestors.

Prevention of malaria infection: The large proportion (84.6%) of the respondents did not take any preventive measures against the malaria infection. However, very few (1.4%) were using chemoprophylaxis such as chloroquine once in a week as prescribed by their treating physicians. Most of these respondents stated that they received chemoprophylaxis from the Primary Health Centres (PHCs) or from the workers of the National Malaria Control Programme. The males took more (80.0%) antimalarial drugs for the prevention

of malaria infection as compared to females (20.0%). This difference was statistically significant (chi-square test, $p = 0.01$). Further, it was also observed that level of education, occupation, or number of past infections did not influence use of chemoprophylaxis and it was also statistically insignificant. Very few (4.1%) were using bed nets to prevent mosquito bites. None of them were using chemically treated bed nets. Nearly three per cent (2.7%) told that about 18 months back DDT was sprayed in their houses by the government health department. About two percent (1.6%) of the respondents were using chick doors and mesh on windows. Rest of the respondents were using cow dung or Neem (*Azadirachta indica*)/ Akra (*Calotropis procera*) leaf smokes or herbal medicines to prevent the mosquito bites.

Number of past malaria infections: Table 3 gives the picture of the number of past malaria infections. The mean number of past malaria infections was reported 13.7 ± 17.4 in the study population. On average men had significantly more infections than women (14.9 ± 20.3 vs. 6.8 ± 7.9). Related factors such as occupation and education were not found to be significant factors in the number of past infections.

Treatment of last malaria infection: The mean time for the most recent infection among the study subjects was 6.9 ± 4.5 months. Nearly one third (32.9%) respondents told that they themselves have taken antipyretics before seeking treatment form the healthcare facility. Only 12.5% of the study subjects have treated themselves with antimalarial drugs. Sixty five per cent of the respondents had gone to the health care facility for the diagnosis of malaria with the blood smear and its treatment. The mean delay for the blood examination was 7.1 ± 9.8 days. Almost three fourth (75.3%) respondents waited more than two days to go to health facility. Nearly forty eight per cent (47.6%) of these respondents stated that they could not get the transport, 33.4% waited because they were not sure about their health conditions and rest (19.0%) thought that level of disease severity was low. Reasons for delay were not influenced by gender, occupation, education, number of past infections and the marital status of the respondents significantly. Out of 365 respondents 296 (81.1%) have gone for blood examination to confirm malaria. More than sixty four per cent 191(64.5%) respondents were having *P. falciparum* and 132 (44.6%) stated that they were having *P. vivax* type of malaria. Nearly seventy one per cent (70.9%) respondents used government health facility and 7.1% private for their blood smear examination. Females preferred more private blood examination laboratories as compared to males and those respondents who had never malaria infection preferred more than those who had experienced malaria before.

Socio-economic impact of last malaria infection: The mean duration of malarial symptoms was 15.3 ± 10.8 days. The mean number of lost employment was 18.6 ± 27.3 (Table 4). Duration of symptoms and number of days absent from the job was not

found significantly varying between government and private jobs. However, significant difference was observed between those who had *P. falciparum* (26.4 ± 35.8 days) and those who had *P. vivax* malaria (12.4 ± 11.2 days) (*p.* 005). Majority 56.2% respondents who suffered with *P. falciparum* told that they had longer period of weakness as a result of which they could not go to their jobs. The mean cost of blood examination with malaria test and treatment was in Indian Rupees 150 to 400. Majority (71.5%) of the respondents visited government health facility for their diagnosis, blood examination and treatment. However, they stated that all respondent could not avail facility such as check-up by doctors, blood examination by laboratory and medicine from dispensary due to off hours or the holiday. Those who availed private health facility they paid eight times more for their medical checkups, laboratory investigations and medicines. Transport was more expensive as compared to diagnosis and treatment due to long distances and non availability of public transport. Most of the respondents stated that they had to hire vehicles.

INFERENCES

The present study reveals that knowledge about aetiology, symptoms, treatment of malaria was quite satisfactory. Treatment seeking pattern and the compliance of antimalarial drugs of the study population present the scenario of existing health care system of the Thar Desert. A person suffering with malaria living in far flung areas delayed in treatment for longer period as compared to those living along road side. Majority of the respondents (55.1%) believed that pregnant women could not be treated with antimalarial drugs such as chloroquine. Nearly twelve per cent (11.8%) of respondents were using preventive measures against mosquito bites. There is still need for Information, Education and Communication (IEC) for the prevention of malaria through using bed nets to prevent mosquito bites and the benefits of early diagnosis and treatment of the disease. In depth Investigation and role of economics is needed in the control of Malaria.

IMPORTANT LEADS/OUTCOMES FROM THE STUDY

The results of the study may be used by planners, malaria control programme implementers and researchers.

Table 1. Socio-demographic characteristics of the study subjects

Characteristics	Frequency or Mean
Age *(Yrs)*	
<=24	70 (19.2%)
25-34	132 (36.2%)
35-44	93 (25.5 %)
45-54	43 (11.7%)
>=55	27 (7.4%)
Study population	34.8 (SD 11.5)
Male	34.6 (SD 11.3)
female	35.0 (SD 12.0)
Sex	
Male	250 (68.5%)
Female	115 (31.5%)
Education	
Illiterate	143 (39.2%)
Literate	222 (60.8%)
Occupation	
Agriculture	155 (42.5%)
Mining	95 (26.0%)
Service	40 (11.0%)
Business	24 (6.6%)
Labour	19 (5.2%)
Others	32 (8.8%)
Religion	
Hindus	285 (78.1%)
Non-Hindus	80 (21.9%)
Caste among Hindus	
G C	142 (49.8%)
OBC	83 (29.1%)
SC/ST	60 (21.1%)
Marital status	
Married	350 (95.9%)
Unmarried	10 (2.7%)
Widow/Widower	5 (1.4%)
Household	
No. of adults	2.9 (SD 0.8)
No. of children (<14 yr.)	3.0 (SD 1.3)

Table 2. Symptoms of malaria as reported by respondents (n=365)

Symptoms	Male (%)	Female (%)	Total	Percentage	χ^2	P Value
Fever	246 (98.4)	115 (100.0)	361	98.9	1.86	0.17
Headache	240 (96.0)	104 (90.4)	344	94.2	4.49	0.033
Chills	228 (91.2)	103 (89.6)	334	91.5	0.249	0.61
Rigors	235 (94.0)	110 (95.7)	345	94.5	0.41	0.51
Vomiting	232 (92.8)	104 (90.4)	336	92.1	0.60	0.43
Arthralgia	208 (83.2)	106 (92.2)	314	86.0	5.27	0.021
Back Ache	189 (75.6)	85 (73.9)	274	75.1	0.11	0.72

Table 3. Distribution of past malaria infection (n=365)

Group		No. of Past Infection	
		Mean	**SD**
Study population		13.7	17.4
Sex	Male	14.9	20.3
	Female	6.8	7.9
Occupation	Agriculture	11.3	13.2
	Non-agriculture	14.5	20.1
Education	Illiterate	12.3	19.1
	Literate	15.8	16.7

Table 4. Employment lost (days) due to malaria infection (n=365)

Group		Employment Lost (Days)	
		Mean	**SD**
Study population		18.6	27.3
Clinic visited	Public	17.5	28.7
	Private	22.1	24.3
Parasite species	*P. falciparum*	26.4	35.8
	P. vivax	12.4	11.2

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

Principal Investigator: *Dr. S. P. Yadav, Scientist 'E'*
Co-Investigators: *Dr. A. K. Dixit Scientist 'E'* and *Mr. R. K. Kalundha, Technical Officer*
Commencement: November, 2007 **Duration**: Four Years **Status**: Completed
Funding: Desert Medicine Research Centre (Intramural)

OBJECTIVES

1. To study 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

Tuberculosis (TB) is one of the most globally serious public health problems. About one third of the global population has been infected with *Mycobacterium tuberculosis*. With the increasing prevalence of HIV infection, the problem of TB is likely to be compounded in the years to come. India alone accounts for one-third global burden of TB and every year more than 1.8 million new cases appear in the country. Approximately 4,00,000 people die from TB every year in India, more than 1,000 every day and 100 million work-days are lost. The current focus of the Revised National Tuberculosis Control Programme (RNTCP) of Government of India uses Directly Observed Treatment Short Course (DOTS) to achieve and maintain a cure rate of over 85% and augmentation of case finding activities to detect at least 70% of estimated cases. A person with untreated sputum smear positive can infect 10 to 14 persons in the community during one year period. In order to control TB, it is mandatory to eradicate its source and decrease the chances of transmission from one individual to another. It is, therefore, important to diagnose and treat the infected individual as early as possible. Studies have shown in different parts of the country that a person delays in diagnosis and treatment for about 50 to 180 days. One study of DMRC reported that nearly eighty percent persons delay in diagnosis and treatment of tuberculosis in the desert part of Rajasthan. Therefore, there is a need for interventions that encourage symptomatic individuals to seek medical help at the earliest. Early diagnosis and treatment will give many fold benefits such as patient's suffering will be minimized and transmission of disease in the community will be checked. Major achievement will be to the DOTS programme. Burden of new cases will be reduced and goals may be achieved. Therefore, this study was under taken to find out the suitable interventional methods by which delay in diagnosis and treatment can be minimized. Jodhpur district is the zonal office for the operational purposes for the DOTS programmes in desert region. Jodhpur

was found as a suitable district to carry out this study among all the 12 desert districts of the Rajasthan. All the PHCs of the Jodhpur district were categorized in order to find out the minimum to maximum prevalence of disease. Chamau PHC was found on top. On the same criteria 8 villages of the PHC were selected randomly for the study. These villages were divided in two groups, one was study group and other was control group. Thus the four villages of each group were studied. In-Charge of PHC, staff deputed for those villages, school teachers, Sarpanch and village leader were contacted and explained the aims and objective of the study to build confidence in population to participate in the study without any doubt and fear.

Table 1 depicts the frequency of reporting at health facility for diagnosis and treatment of the study and control group of people. The percentage of symptomatic cases reported to DOTS after intervention was 83.5% as compared to without intervention (26.9%). Thus the intervention impact was 3.1 times more,, which is significant.

Table 1. Month-wise distribution of symptomatic cases going to DOTS

Month	Study Group (n=750)		Control Group (n=750)	
	No.	%	No.	%
February, 11	20	2.7	16	2.1
March	29	3.9	10	1.3
April	23	3.1	15	2.0
May	14	1.9	8	1.1
June	17	2.3	6	0.8
July	25	3.3	4	0.5
August	36	4.8	11	1.5
September	44	5.9	14	1.9
October	62	8.3	20	2.7
November	110	14.7	25	3.3
December	119	15.9	32	4.3
January, 12	127	16.9	41	5.5
All	**626**	**83.5**	**202**	**26.9**

Table 2 shows the status of sputum of the study and control groups of people. The percentage of positive sputum rate was high among the control group of people (31.2 Vs 25.4) as compared to study group.

Table 2. Status of sputum

Sputum Status	Study Group		Control Group	
	Number	%	Number	%
+ ve	159	25.4	63	31.2
- ve	467	74.6	139	68.8

Table 3 gives the picture of type of PTB cases of the study and control groups. There is 2.5 times more reporting in category I type of cases in study group as compared to control group. The results of the study are as per expectations. It was observed that the suitable intervention applied for bringing the symptomatic cases for the early diagnosis and treatment of the PTB cases is increasing more than three times (3.1) as compared to self reporting. Among the reporting cases, new PTB cases are 2.5 times more in study group as compared to control group. Reduction in duration of delay for early diagnosis and treatment, suitability of the intervention and its cost effectiveness are being analyzed. Final report will be prepared and submitted in future.

Table 3. Type of PTB cases

Type of cases	Study Group		Control Group	
	N	%	N	%
Cat. I	138	86.8	22	34.9
Cat. III	9	5.7	19	30.2
Relapse Case	7	4.4	13	20.6
Reocc. Case	5	3.1	9	14.3

IMPORTANT LEADS/OUTCOMES FROM THE STUDY

Study is completed. The intervention may be useful for early detection and motivation of new PTB cases and bringing them for diagnosis and treatment under DOTS.

1.12 A study of treatment seeking behaviour for malaria and its management in children in desert part of Rajasthan, India

Principal Investigator: *Dr. S. P. Yadav, Scientist 'E'*
Co-Investigators: *Dr. A. K. Dixit, Scientist 'E' and Mr. R. K. Kalundha, Technical Officer*
Commencement: November, 2009 **Duration**: Three Years **Status**: Ongoing
Funding: Desert Medicine Research Centre (Intramural)

OBJECTIVES

1. To identify different community belief and practices on the basis of which fever could be recognized and classified
2. To explore factors involved in selection of different treatment options in Indian Thar Desert
3. To compare with other studies in different parts of the country as well as world

RATIONALE

To reduce the morbidity and mortality of malaria, the World Health Organization (WHO) has developed a strategy which includes the early diagnosis and treatment of malaria, as one of its main components. It is recommended that antimalarial drugs be given at home to all febrile children. As many deaths occur within 48 hours of onset of symptoms, this strategy will have optimal impact if treatment is given early. Generally, it is the mothers who identify fever in their children particularly in children whose age is less than five years and provide presumptive treatment, but there are few data on these initial steps. Most studies focus on people reporting at health centres and dispensaries, which represent a highly selected proportion of the infected population as most febrile children will not be brought to consultations.

PROGRESS

Selection of the study area was the basis of highest API of the district and highest API of the PHC within the district. Thus, Jaisalmer district was having highest API among all the 12 districts of desert part of Rajasthan. Similarly, Ramgarh PHC was also having the same character as compared to the 18 PHCs within the Jaisalmer district. Following this criteria, Ramgarh PHC was selected in Jaisalmer district for the study area. All the 65 villages in Ramgarh PHCs were classified and divided into two groups namely Command and Non-command villages. Command villages (CVs) were defined here as villages where the water was available for the irrigation and drinking purposes for the last 20 years or more through IGNP and Non-command villages (NCVs) were defined as where water is yet to reach. Using random sampling, 10 villages from each category of villages were selected namely Hajjidatta ki Dhani, Seowa, Raghwa, Raimala, Sultana, Nagga, Bada,

Mokal, Nehdai and Lanera from the CVs and Kadar ki Dhani, Habur, Kakab, Hamira, Tibansar, Chandane ki Dhani, Markh ka Ganv, Mohammad Khan ki Dhani, Ranau and Tanot from the NCVs. 30 households were selected randomly from the each selected village for this study. At the time of selection of households, head of the household was contacted and told about the study. All the head of the selected households agreed to participate in the study. Thus a total of 600 (300CVs+300NCVs) households were surveyed from both the groups of the villages, however, Data from 562 households was collected. Data was collected during home visit. For every child of the household the mother was asked 'Is this child sick today? If the mother answered yes, she was asked to describe the symptoms and their duration and to name the disease. The mothers were also asked 'By which symptoms and signs do you identify malaria?' and 'How did you treat your child? All the children had their axillary temperature taken for 5 minutes with a mercury thermometer and the arm held firmly. They all had a complete physical examination by a PHC staff, including palpation of the spleen and blood taken by finger prick for thick and thin blood smears. These slides were examined at PHC for malaria. Proportions were compared by using the x^2 test. Verbal consent from the local authorities and the head of the household was also obtained before the interviews and blood sampling. Children found to be sick were immediately treated by the same staff. Mothers defined fever as *Tav* and malaria as *Hitav* (fever with shivering) or *Ekantare tav* (fever on alternate day) in their local dialect *i.e.* Marwari.

Socio-demographic characteristics of the mothers: Out of 562 households 572 mothers interviewed. They were able to give the information about 894 children less than 5 years of age. Majority (83.7%) of the mothers were belonged to 21-40 years of age which is most reproductive age of the mothers. Literacy rate was 33.7%. More than half of the mothers 301(52.6%) were earning their livelihood through agriculture and its related work such as animal husbandry. About eighty four percent (83.6%) were Hindus and among the Hindus 58.2% were general caste (Table 1).

Age and sex of the children: Table 2 depicts the age and sex of the children less than 5 years of age. Majority (26.4%) of the children were 36-48 months of age. Out of 894 children 476 (53.2%) were males and 418 (46.8%) were females. The average age of the children was 30.7 months.

Objective Morbidity: Table 3 reveals that out of 894 children less than 5 years of age, 219 (24.5%) were sick and feverish, 57 (6.4%) were sick but not feverish and rest were healthy according to their mother's perception. Out of 219 sick and feverish children 119 (54.3%) were having temperature <37.5^{0}C followed by 74 (33.8%) having the temperature between 37.5-38.4^{0}C and 26 (11.9%) ≥ 38.5^{0}C respectively. The comparisons of the diagnosis of malaria and treatment given to the children are given in Table 4 and 5

Table 1. Socio- demographic characteristics of the study subjects

Characteristics	Number	Percentage
Age *(Yrs)*		
<21	62	10.9
21-30	257	44.9
31-40	222	38.8
>40	31	5.4
Education		
Illiterate	379	66.3
Literate	193	33.7
Occupation		
Agriculture & its related work	301	52.6
Mining	34	5.9
Service	5	0.9
Business	2	0.3
Labour	127	22.2
Others	98	17.1
Religion		
Hindus	478	83.6
Non-Hindus	94	16.4
Caste among Hindus		
G C	280	58.2
OBC	124	25.8
SC/ST	77	16.0

GC= General Caste; OBC= Other Backward Caste; SC/ST= Schedule Caste/Schedule Tribe

Table 2. Distribution of children according to age and sex

Children age (Months)	Male		Female		Total	
	No.	%	No.	%	No.	%
<12	66	13.8	72	17.2	138	15.4
12-24	75	15.8	67	16.0	142	15.9
24-36	90	18.9	84	20.1	174	19.5
36-48	126	26.5	110	26.3	236	26.4
48-59	119	25.0	85	20.3	204	22.8
Total	476	100.0	418	100.0	894	100.0

Table 3. Comparison between fever reported by mothers and axillary temperature at clinical examination

Mother's perception	Children with temperature						
	Children No. (%)	<37.5^{0}C		37.5-38.4^{0}C		≥ 38.5^{0}C	
		No.	%	No.	%	No.	%
Sick and feverish	219 (100.0)	119	54.3	74	33.8	26	11.9
Sick but not feverish	57 (100.0)	33	57.9	18	31.6	6	8.9
Healthy	618 (100.0)	410	66.3	153	24.8	55	10.5
Total	894 (100.0)	562	62.9	245	27.4	87	9.7

Table 4. Comparison of the diagnosis of malaria by mothers with the presence of a temperature ≥ 37.5^{0C} and positive thick smear result

Mother's diagnosis	Children No. (%)	Temperature ≥ 37.5^{0}C and +ive thick smear result			
		positive		Negative	
		No.	%	No.	%
Positive for malaria	112	46	41.1	66	58.9
Sick but no malaria	164	29	17.7	135	82.3
Healthy	618	52	8.4	566	91.6
Total	894	127	14.2	767	85.8

Table 5. Treatment given according to different case definitions

Case definition	Chil-dren	No treatment		Traditional treatment		Chloroquine and Antipyretic		Consultation in health Centre	
	No.	No.	(%)	No.	(%)	No.	(%)	No.	(%)
Sick according to mother	276	83	31.5	71	25.7	65	23.6	57	20.7
Sick & fever according mother	219	69	31.5	78	35.6	57	26.0	15	6.8
Malaria according to mother	112	21	18.8	43	38.4	26	23.2	22	19.6
Temp. ≥ 37.5^{0C}	203	24	11.8	45	22.2	20	9.9	14	6.9
Temp. ≥ 37.5^{0C} and +ive thick smear result	125	28	22.4	32	25.6	40	32.0	25	20.0
Temp. ≥ 37.5^{0C} and parasitic density ≥ 4000/µl	40	9	22.5	10	25.0	12	30.0	9	22.5
Temp. ≥ 38.5^{0C} and +ive thick smear result	26	7	26.9	9	34.6	6	23.1	3	11.5
Temp. ≥ 38.5^{0C} and parasitic density ≥ 4000/µl	14	4	28.6	5	35.7	3	21.4	2	14.3

INFERENCES

Recognition of fever and its presumptive treatment with antimalarial drugs is an essential part of the strategy of the World Health Organization (WHO) to reduce the morbidity due to this disease. Findings of the study indicate that mothers often failed to identify fever in their children and to consult or to provide antimalarial treatment. Without great efforts to improve home care, it is unlikely that the morbidity and mortality due to malaria in young children will be greatly reduced.

IMPORTANT LEADS/OUTCOMES FROM THE STUDY

Results of the study may be useful to the planners, malaria control programme implementers, researchers, malaria control programme evaluators, administrators and others who are associated with malaria control programmes.

1.13 Risk assessment of JE virus emergence in two paddy growing districts of Rajasthan state (Sri-Ganganagar and Hanumangarh)

Principal Investigator: *Dr. P. C. Kanojia, Scientist 'E'*
Co-Investigators: *Dr. N. L. Aseri, M.G. Hospital, Hanumangarh and Dr. M. L. Mathur, Scientist 'F'*
Commencement: December, 2011 **Duration**: One Year **Status**: Ongoing
Funding Agency: Desert Medicine Research Centre (Intramural)

OBJECTIVE

1. To assess potential emergence and public health risk of JE virus in Sri-Ganganagar and Hanumangarh district by carrying out mosquito, vertebrate hosts and human surveillance

RATIONALE

In Rajasthan, Sri-Ganganagar and Hanumangarh districts have undergone a lot of ecological changes in the form of construction of irrigation canals originating from Indira Gandhi Nahar, Gang canal and Bhakara-Sirhind canal. The habitats have been modified for the development of agriculture, including paddy cultivation in the region which has resulted in the vast expansion of water bodies. The rice fields and other water bodies have helped *Cx. tritaeniorhynchus* (a major vector of JE in India) and water frequenting birds (natural host of JE) to establish themselves in the area. The existing ecological conditions, including presence of JE specific neutralizing antibodies in pigs, occurrence of susceptible mosquito vector *viz., Cx. Tritaeniorhynchus* and extensive paddy cultivation and reporting of JE cases from adjoining state of Haryana make Sri-Ganganagar and Hanumangarh district more vulnerable for the emergence of JE virus.

The overall aim of this study is to carry out active surveillance of JEV in both the districts so that potential emergence of JEV could be assessed. The objectives of the study will provide useful information on presence of JE vectors, their population dynamics and seasonal occurrence. In addition, intermediate vertebrate hosts which play vital role in JE natural cycle, will be known. Etiological agent involved in an un-diagnosed acute fever and encephalitis cases will be identified. The results of this study will make crucial information available to detect the potential emergence and public health risk of JEV before it could assume epidemic form and to intervene to reduce that risk substantially.

PROGRESS

Monitoring of JE vectors in Sri-Ganganagar and Hanumangarh district: During 2011, field visit was made to Sri Ganganagar and Hanumangarh district, with a view to monitor

JE vectors population. A total of 266 adult specimens comprising 6 species were collected from different villages/localities *viz*., Tibi, Lakhuwali, Makkasar, (Hanumangarh) and Central Cattle Breeding Farm (Suratgarh, Sri-Ganganagar) Table-1. Among the species collected, three were found to be JE vectors (*Cx. tritaeniorhynchus, Cx. quinquefasciatus* and *An. subpictus*). *Cx. quinquefasciatus* was found to be predominant species whereas population of *Cx. tritaeniorhynchus* was recorded extremely low. Twenty mosquito pools were prepared from above mosquito collections. These pools will be processed for detecting JE virus antigen using IFA and molecular techniques.

Table 1. Mosquito species collected from different localities of Sri-Ganganagar and Hanumangarh district

Species	Tibi		Lakhuwali		Makkasar	Central Cattle Breeding Farm	Total
	Cattle shed dusk	Indoor house day-time	Cattle shed dusk	Cattle shed indoor	Cattle shed dusk	Cattle shed dusk	
Cx. tritaeniorhynchus	-	-	-	-	-	1	1
Cx. quinquefasciatus	12	13	20	64	44	47	200
An. subpictus	-	-	-	-	2	4	6
An. stephensi	3	-	6	7	-		16
An. culicifacies	-	-	6	5	8	20	39
An. splendidus	-	-	-	-		4	4
Total	15	13	32	76	54	76	266

1.14 Field efficacy trials of extracted latex of *Calotropis procera* for its public use as bio-larvicide against dengue vectors

Principal Investigator: *Dr. Manju Singhi, Scientist 'C'*
Co-Investigators: *Dr. Vinod Joshi, Scientist 'F' and Mr. Anil Purohit, Research Assistant*
Research Staff: *Ms. Susmita Chattopadhay, Junior Research Fellow*
Commencement: January, 2011 **Duration**: One Year **Status**: Ongoing
Funding Agency: Indian Council of Medical Research–Translational Research **(**Extramural)

OBJECTIVES

1. To undertake field efficacy testing trials of extracted latex of *Calotropis procera* for control of dengue vector breeding in Jodhpur
2. To optimize the product dose of extracted latex per liter of water volume in breeding habitats of *Aedes* mosquitoes
3. Finalization of its dose for handing over the results into public health actions by Vector Borne Disease Control Programme

PROGRESS

The field efficacy of the extracted latex in +ve breeding containers under different socio-economic settings of Jodhpur city has been undertaken against *Aedes* vector.

Collection & extraction of Latex: Latex from *Calotropis procera* grown in the DMRC campus has been collected in summer and post rainy season of the year 2011-12. The latex has been collected manually from green stems directly into disposable bottles (fig.1). The freshly collected latex is immediately subjected to extraction using AR grade Methanol. The latex volume is measured and extraction solvent is added to it in 1:1 ratio (v/v). After filtration supernatant is collected in small beakers and then dispensed on petri dishes and left for air drying at room temperature. The dried extract is then made into powdered form and stored in air tight bottles for the field application.

Fig. 1. Collection of latex from Calotropis procera

Small scale field efficacy trials in different socio-economic areas and observed efficacy: A field study has been undertaken for screening of + ve breeding containers in all the selected areas of Jodhpur city in all seasons for testing its field efficacy. A total of 600 randomly selected houses have been investigated for the presence of *Aedes* breeding. In the surveyed 600 households 4080 numbers of water filled containers were observed. Out of 4080 water filled containers 305 different containers were found +ve for breeding. The socioeconomic status of people was found to be closely associated with water management and storage habits which influenced vector breeding. The observations have shown that low socio- economic areas contain more water filled uncovered containers as compared to high socio-economic areas. Common breeding containers in low socio economic areas were observed to be uncovered cement tanks, clay pots, plastic containers, under ground tanks, metallic containers whereas in high socio-economic areas clay pots and coolers were found (Table-1 & Fig.2). All the +ve breeding containers were marked for testing efficacy of extracted latex. The optimized dose of extracted latex was applied to all breeding containers found +ve for *Aedes* breeding in all the season as per the WHO protocol.

The larvicide treated containers were monitored after 24 hours for larval mortality and follow up studies have been made for 48 hours. Written consent was obtained from the residents of the household surveyed before carrying out the interventions. The details of the research objectives and its possible utilization for the control of dengue & DHF have been conveyed to the residents of the area. Results have shown 100% mortality in all the field treated containers of the area after 24 hours of exposure in all types of treated containers irrespective of the areas and season (Table 2, Fig. 3).

Fig. 2. Types of +ive breeding containers treated in field

Table 1. Details of +ive breeding containers from different socio-economic areas

Type of Containers	Areas				Total Containers	+ ve breeding Containers
	LSIC	LSOC	HSIC	HSOC		
Cement	166	140	119	68	493	110
Clay	389	403	239	172	1203	70
Plastic	316	290	235	167	1008	39
Metallic	111	136	56	36	339	9
Underground	26	56	81	145	308	22
Overhead	75	108	114	150	447	0
Coolers	95	58	59	70	282	55
Total	1178	1191	903	808	4080	305

LSIC- Low socio-economic area of inside city; LSOC- Low socio-economic area of outside city; HSIC- High socio-economic areas of inside city; HSOC- High socio-economic areas of outside city

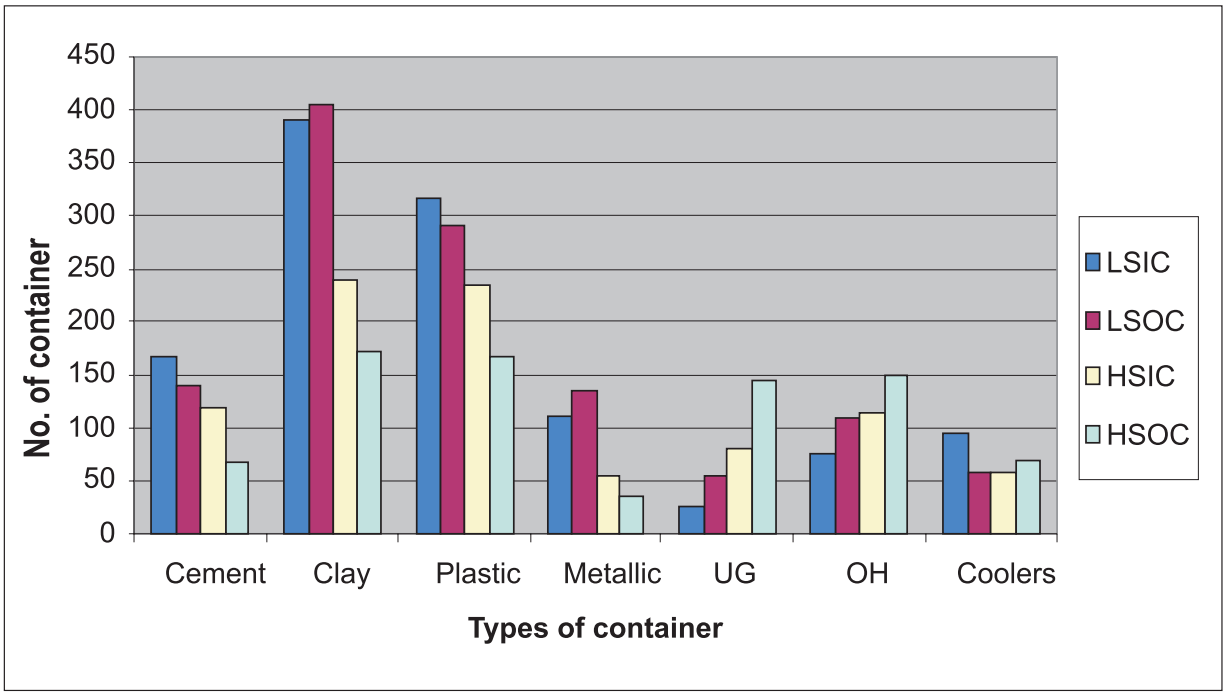


Fig. 3 Type of breeding containers in different socio-economic areas

Table 2. Per cent mortality in Small scale field efficacy testing trials of extracted latex against dengue vector

Season	Area	Total no. of containers	+ve Containers	% +ve	Treated containers	% Mortality
Winter	LSIC	205	12	5.85	12	100
	LSOC	212	19	8.96	19	100
	HSIC	209	4	1.91	4	100
	HSOC	149	8	5.37	8	100
Summer	LSIC	623	43	6.90	43	100
	LSOC	651	44	6.76	44	100
	HSIC	399	12	3.01	12	100
	HSOC	410	10	2.44	10	100
Rainy	LSIC	350	62	17.71	62	100
	LSOC	328	53	16.16	53	100
	HSIC	295	24	8.14	24	100
	HSOC	249	14	5.62	14	100
Total		**4080**	**305**	**7.48**	**305**	100

FINDINGS

1. Developed bio-larvicide has been found effective in all natural breeding containers of different socio-economic areas of Jodhpur, an arid area of Rajasthan.
2. Cement tanks were found to be key containers for low socio-economic areas while coolers and clay pots were observed in high socio-economic areas.
3. No major difference was observed in the feasibility studies of application of latex in all the study settings.

1.15 Polymorphisms in Duffy blood group genes of *Plasmodium vivax* malaria patients and control population

Principal Investigator: *Dr. S. S. Mohanty, Scientist 'C'*
Co-Investigators: *Dr. Karam V. Singh, Scientist 'F', Dr. R. Fotedar, Scientist 'C' and Mr. Pankaj Kumar, Technical Assistant*
Commencement: September, 2010 **Duration:** Two Years **Status:** Ongoing
Funding: Desert Medicine Research Centre (Intramural)

OBJECTIVES

1. Study of four phenotypic variants of Duffy blood group [Fy(a+b+), Fy(a+b-), Fy(a-b+) and Fy(a-b-)] will be calculated within the *Plasmodium vivax* malaria patients and control population
2. Duffy blood group genotyping in *P. vivax* malaria patients and control population from the endemic areas of Rajasthan

PROGRESS

The present study was undertaken to know the distribution of Duffy blood group antigen in the malaria patients of the desert population of Rajasthan and its correlation with malaria infectivity. The study was carried out on the hospital collected samples. Both males and females were included in the study between the ages 1 to 60 years. All the subjects were taken from different families and were not related. Fever cases reported to different hospitals and health Centres of Jodhpur were screened for malaria positive. Those found to be positive were included in the study. Blood samples of all the subjects were screened for the presence of malaria parasite and analysed for the determination of the phenotypic variants of Duffy antigen. For the diagnosis of malaria a thick and thin film was prepared in the same slide from the peripheral blood. Thin film of the slides was fixed with methanol before staining. The blood films were stained with Jaswant Singh Bhattacharjee (JSB) stain and microscopically examined under an oil-immersion lens. A thick smear was regarded as negative on initial examination if no parasites were seen in 100 high power fields. All the *P. vivax* and *P. falciparum* malaria cases were confirmed by the additional rapid malaria diagnostic test. Blood samples were taken from vacutainer collected for clinical studies of the hospitalized patients. The whole blood samples of 200µL were kept in EDTA containing tubes for Duffy blood group testing. Duffy antigen identification was carried out within 24 hours of blood sample collection using the indirect antiglobulin technique. Commercially available antisera anti-Fya, anti-Fyb and anti-human globulin (M/s DiaMed GmbH, 1785 Cressier s/Morat, Switzerland) were used according to manufacturer's instructions.

A total of 119 subjects were enrolled in the study till date. Out of which, 23 confirmed *P.*

vivax malaria cases were enrolled during this year. None of the cases of this year belongs to scheduled tribe group. Among the study population, 78 were found to be control population and 41 were *P. vivax* malaria patients. The results of all the samples are shown in table I. Amongst the total malaria cases, percentage of population with Duffy phenotypes Fy(a+b+) was found to be 75.6. However, the percentage of population with Duffy phenotypes Fy(a+b-) and Fy(a-b+) were found to be 9.75 and 14.63 respectively. When the Duffy phenotype of the *P. vivax* malaria patients (n=41) and healthy population (n=78) were compared, a higher percentage of population with Duffy phenotype Fy(a+b+) were recorded in the malaria patients (75.6%) than the healthy population (58.97%) and inverse to the above was recorded in case of Duffy phenotype Fy(a+b-) and Fy(a-b+) (Table I). The frequencies of FYA and FYB alleles among the malaria patients were 48% and 52% respectively.

Among the study subjects, 54 belonged to the tribal group and 65 to non-tribal group. When the Duffy phenotypes of the tribal population were compared, percentage of population with Duffy phenotype Fy(a+b-), Fy(a-b+) and Fy(a+b+) were 18.51, 11.11 and 70.37 respectively (Table I). However, non-tribal population with Duffy phenotype Fy(a+b+) were 10.37% lower than the tribal population and reverse order was recorded in case of Fy(a-b+). Furthermore, the percentage of both tribal and non-tribal population with Duffy phenotype Fy(a+b+) were higher than the population with Duffy phenotype Fy(a+b-) and Fy(a-b+).

Table 1. Duffy phenotypes and gene frequencies of the various populations of the desert part of India

Name of the Category	Number tested (n)	Duffy phenotypes in %			Gene frequencies	
		Fy(a+b-)	Fy(a-b+)	Fy(a+b+)	*FYA*	*FYB*
***P. vivax* malaria patients**	41	9.75 (n=4)	14.63 (n=6)	75.6 (n=31)	0.48	0.52
Healthy population	78	21.79 (n=17)	19.23 (n=15)	58.97 (n=46)	0.51	0.49
Tribal Population	54	18.51 (n=10)	11.11 (n=6)	70.37 (n=38)	0.54	0.46
Non-tribal population	65	16.92 (n=11)	23.07 (n=15)	60 (n=39)	0.47	0.53

OUTCOME OF THE PRESENT STUDY AND WORK TO BE DONE

The data suggests that the Duffy phenotype Fy(a+b+) was dominant in the *P. vivax* malaria patient than the healthy population. The genotyping of Duffy blood group gene will be done in the *P. vivax* malaria patients and control population of Rajasthan.

1.16 tudy on the age determination of field collected mosquitoes by quantitative reverse transcriptase-PCR (qRT-PCR)

Principal Investigator: *Dr. S. S. Mohanty, Scientist 'C'*
Co-Investigators: *Dr. Karam V. Singh, Scientist 'F' and Dr. S. K. Bansal, Scientist 'F'*
Commencement: September, 2010 **Duration:** Two Years **Status:** Ongoing
Funding Agency: Desert Medicine Research Centre (Intramural)

OBJECTIVES

1. To study candidate age depending expression genes from data generated in female *Drosophilla melanogaster* and *Anopheles gambiae* and to identify orthologue genes of *D. melanogaster* in *An. gambiae*
2. To design specific primer for age depending genes of *An. gambiae* and standardized for the determination of age of known *Anopheles stephensi* and *An. culicifacies*
3. The quantitative assay of pteridine and glucose-6-phosphate dehydrogenase will be standardized for the age of known mosquitoes and will be used for the confirmation of the results of qRT-PCR
4. Technique will be validated with the field collected mosquitoes

PROGRESS

The g6pd activity in *An. stephensi* was found to be inversely proportional to the age of the mosquitoes. Thereafter, the standardization of the technique for the determination of the age of *Aedes aegypti* was done and showed in the present investigation. *Ae. aegypti* used in this study were obtained from a laboratory colony established for more than two years from field-collected mosquitoes. Field collection was conducted in the adjacent areas of Jodhpur and thereafter the mosquitoes were kept in the Insectary. Hundred pupae of *Ae. aegypti* were transferred to an enamel bowl and kept inside the cage. They were left in the cages for the emergence of the adults and un-emerged pupae were discarded on day-3. Adults were fed with 10% glucose solution after emergence. Each of ten males and females in triplicates were taken for the g6pd assays in the intervals of five days. Blood meal was not given to the experimental female mosquitoes. Adults up to one month old were used for the g6pd assays.

Ten males and females were collected in test tubes from the cages. These test tubes were placed in a freezer for 5 minutes for anaesthetizing. Each anaesthetized mosquito was transferred to a 1.5ml microfuge tube. 200 µL of 5mM glycine (pH-8.0) was dispensed to each microfuge tube containing mosquito. Mosquitoes were grinded in microfuge vials by a pestle for homogenization of the tissue. The mosquito sample was transferred to the spectrophotometer cuvettes maintained at 30ºC in a dry bath containing substrates and cofactors. The 3 ml reaction mixture contained 2.7ml of 55mM TrisHcl containing 3.3mM

$MgCl_2$, pH-7.8, 100 µl of 6mM NADP, 100 µl of 100mM glucose-6-phosphate and 100 µl of mosquito tissue. The optical density was read at 340nm for 10 minutes at intervals of 1 minute using a UV-VIS spectrophotometer. Each experiment was conducted in triplicates on different days. Protein contents were estimated by the Bio-Rad protein assay kit in accordance to the manufacturer's instructions. Ten micro liters of mosquito tissue was added to micro-titre plate and, thereafter, 300 µl of diluted Bio-Rad reagent dispensed on it. The reaction was read at 590 nm after 5 min of incubation at room temperature. Eight blanks were prepared for each plate with 10 µl distilled water and 300 µl of Bio-Rad solution. Protein values were calculated for individual mosquitoes from a standard curve of absorbance of known concentration of bovine serum albumin.

The g6pd activity was increased from the larvae to pupae and highest activity was recorded in freshly emerged adults of both the laboratory reared and field collected *Ae. aegypti*. The g6pd activity in the laboratory reared and field collected females was found to be decreased with the age and lowest activity was recorded in the 25 days old mosquitoes (Fig.1). Furthermore, the g6pd activity in the 25 days old females of laboratory reared and field collected were recorded to be 68% and 54% lower than the newly emerged respectively. However, the activity was found to be increased in 30 days old mosquitoes than the 25 days old one. When the g6pd activity was compared between laboratory reared and field collected females, the g6pd activity in field collected females was found to be significantly higher than the laboratory reared females (*t test*, P=0.0002).

The g6pd activity in the laboratory reared and field collected males was found to be decreased with the age and lowest activity was recorded in the 25 days old mosquitoes (Fig 2). Furthermore, the g6pd activity in the 25 days old laboratory reared and field collected males were recorded to be 66% and 67% lower than the newly emerged respectively. However, the activity in 30 days old mosquitoes was found to be higher than the 25 days old mosquitoes. When the g6pd activity was compared between the laboratory reared and field collected males, the g6pd activity in laboratory reared males was found to be significantly higher than the field collected males (*t test*, P=0.006). The enzyme activity was increased from larval stage to pupal stage and highest activity was reported in the freshly emerged males (Fig 2).

RT-PCR assays: Age depending expression genes of *D. melanogaster* has been collected from the micro arrays data. Sixteen genes changed significantly between 5 and 30 days old adults of *D.melanogaster*. Out of the sixteen age depending expression genes of *D.melanogaster*, orthologues of three genes are present in the three mosquito genus, *Anopheles*, *Culex* and *Aedes*. The nucleotide sequences of the three age depending expression genes, Carboxypeptidase, Cytochrome-b5 and Calcium binding were downloaded from the gene bank. The sequences, which show maximum homology between the genuses

were taken from NCBI gene bank. Two types of primers were designed for quantifying the age depending expression genes.

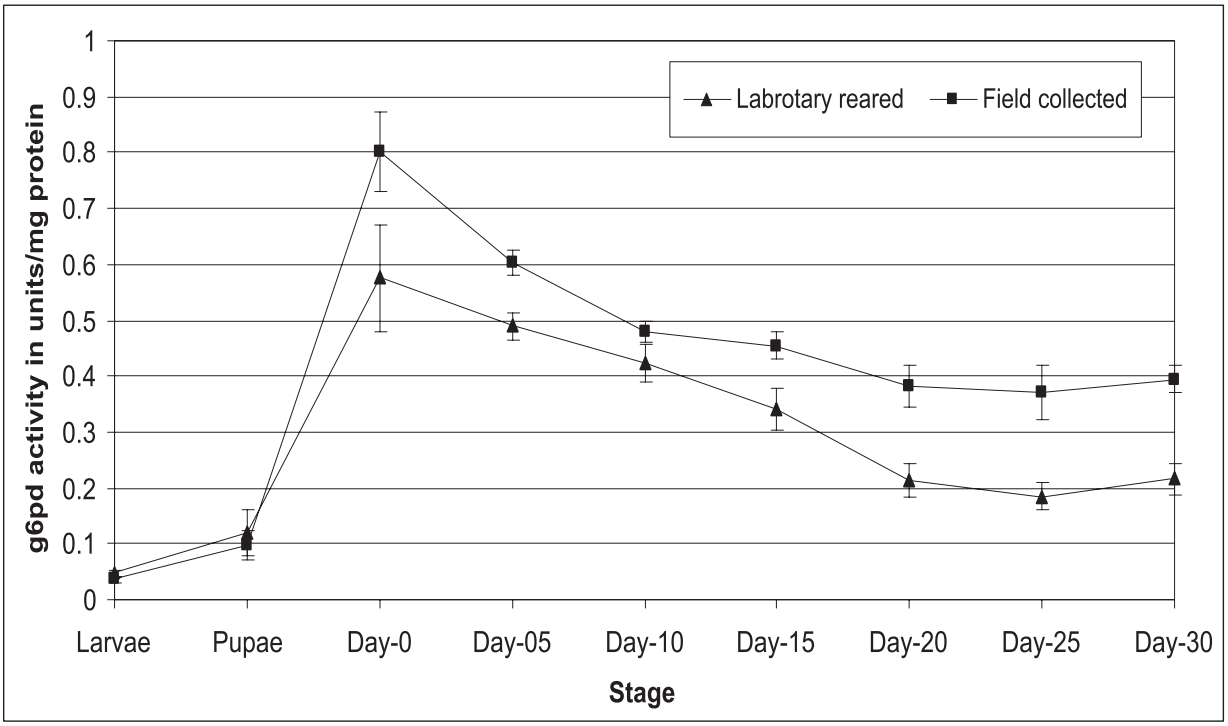


Fig. 1. Glucose-6-phosphate dehydrogenase activity in units/mg protein of laboratory reared and field collected females of *Aedes aegypti*

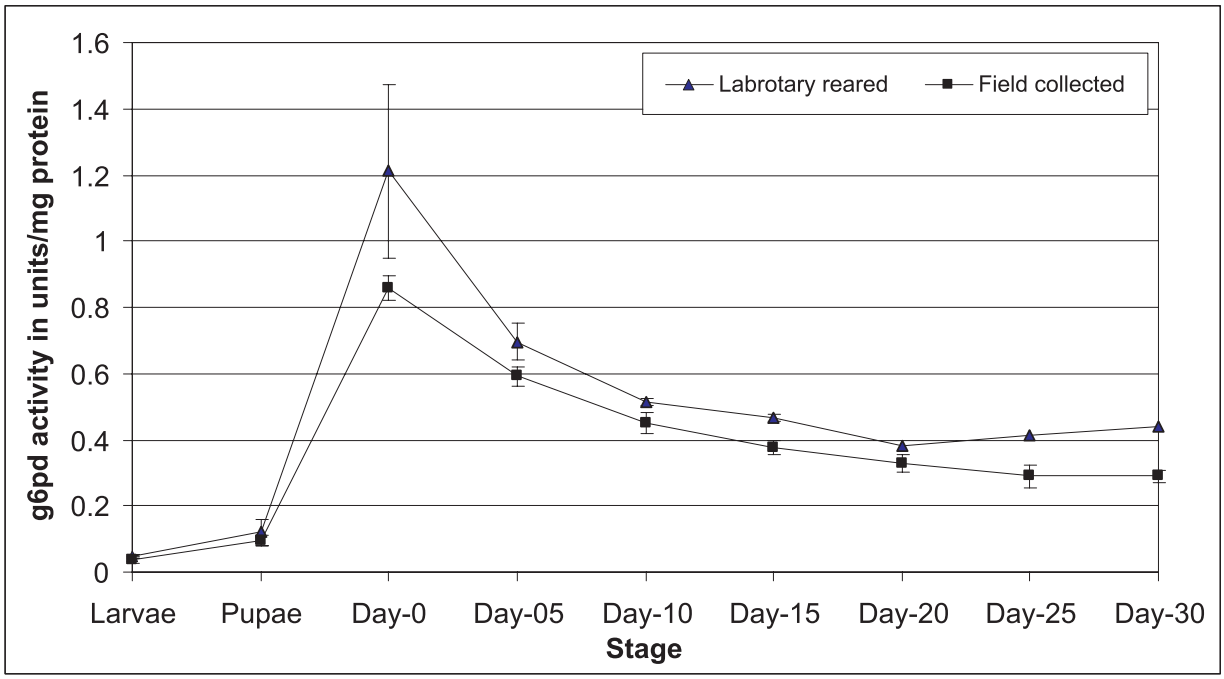


Fig. 2. Glucose-6-phosphate dehydrogenase activity in units/mg protein of laboratory reared and field collected males of *Aedes aegypti*

TaqMan primers designed by the Primer Express Software: The TaqMan primers for each species of mosquito were designed by the Primer Express Software supplied with the RT-PCR, 7500 machine. Three genes, Carboxypeptidase, Cytochrome-b5 and Calcium binding will be used for the age depending expression assays (Table 1, 2 & 3). The nucleotide sequences of the above three age depending expression genes of the three mosquito species were down loaded from the NCBI site. These sequences were put in the Primer Express Software and the primers were designed and selected to avoid the following conditions.

- Should not form primer dimmers.
- No hairpin loop should be formed.
- Tm value should be within the range.

TaqMan has an additional probe and are more specific. The forward and reverse primers of the above three genes with all relevant information are shown in the tables 1-3.

SYBER green primers designed by alignment software: The primers were also designed by the alignment software http://www.ebi.ac.uk /Tools/msa/clustalw2/. These primers are common for an age depending gene of the three genuses of the mosquitoes. The nucleotide sequences of the age depending expression gene of three above species were aligned by the above software. The sequences with maximum homology are taken for the primers and their specificity was checked. The primers of the three age depending expression genes are shown in table-4. These are common to the three genuses of the mosquito species.

House keeping gene: Actin gene has been taken as the house keeping gene for the study. The nucleotide sequence of the actin gene has been taken from the gene bank and published report.

OUTCOME OF THE PRESENT STUDY AND WORK TO BE DONE

The activity of the enzyme g6pd was linearly decreased up to 25 days in males and females of *Ae. aegypti* and it may be used for the prediction of age after trial on wild caught mosquitoes. Three age depending expression genes, Carboxypeptidase, Cytochrome-b5 and Calcium binding gene are identified and primers are designed for the three mosquito genus. The estimation of the transcriptional profiles of the above three genes and their relation with the age of mosquitoes is yet to be established.

Table 1. ist of the primers designed by Primer Express Software for the carboxypeptidase gene of the three species of mosquitoes

Mosquito sp.	Name of the gene	Type	Sequence	Start bp	Stop bp	Tm
Aedes aegypti	Carboxypeptidase	Forward Primer	GGATCGCTTCTGGCGTAAGA	2015	2034	59
		Reverse Primer	CATCGGTTCCCTTACAGGATTT	2073	2052	58
		Probe	ACGCAAGCCGACAGG	2036	2050	69
Anopheles gambiae	Carboxypeptidase	Forward Primer	TCCGCCCAGGATCTTGAC	240	257	58
		Reverse Primer	GCCTCGGAACCAACCAAA	292	275	58
		Probe	TCTGGCAGCTTGATC	259	273	68
Culex Quinque-fasciatus	Carboxypeptidase	Forward Primer	AGCCGGAAACCAAGATCGT	830	848	58
		Reverse Primer	ACTTGCAACTTCCCTTGAGCTT	886	865	58
		Probe	CGCGAAGAACTCCT	850	863	69
Anopheles stephensi	Actin	Forward Primer	CCATTTGCGCCACCTCTCT	1798	1816	60
		Reverse Primer	ATGATAATAGGCGGCGCAAT	1854	1835	58
		Probe	ACCAGACATCGGACGC	1818	1833	68

Table 2. List of the primers designed by Primer Express Software for the Cytochrome-b5 gene of the three species of mosquitoes

Mosquito species	Name of the gene	Type	Sequence	Start bp	Stop bp	Tm
Aedes aegypti	Cytochrome-b5	Forward Primer	CCGCGAGATGATGAAGAAGTT	306	326	58
		Reverse Primer	GCTTCCGCTCGGATTCG	361	345	59
		Probe	AAGGTCGGCGAGCTG	328	342	69
Anopheles gambiae	Cytochrome-b5	Forward Primer	GCCCGGGAGATGATGAAGA	197	217	60
		Reverse Primer	TGTTTTCGTTCCGCTTCGA	257	239	59
		Probe	ATTCAAAGTTGGCGAACTG	219	237	69
Culex quinquefasciatus	Cytochrome-b5	Forward Primer	CGACTGGAGCACCGAACAG	282	300	59
		Reverse Primer	GCGGCACGATCCATGACT	337	320	59
		Probe	AGGACGAAAACTCC	302	315	70
Anopheles stephensi	Actin	Forward Primer	CCATTTGCGCCACCTCTCT	1798	1816	60
		Reverse Primer	ATGATAATAGGCGGCGCAAT	1854	1835	58
		Probe	ACCAGACATCGGACGC	1818	1833	68

Table 3. List of the primers designed by Primer Express Software for the Calcium binding gene of the three species of mosquitoes

Mosquito species	gene	Type	Sequence	Start bp	Stop bp	Tm
Aedes aegypti	Calcium binding gene	Forward Primer	CAAGCGCGGTGGAATCA	465	481	59
		Reverse Primer	CCCATGAACTGGGCGTACAG	521	502	60
		Probe	CCTGCAGAGATACCAGG	483	499	69
Anopheles gambiae	Calcium binding gene	Forward Primer	CGATTTCGATAAGGATGGCAAA	302	323	60
		Reverse Primer	TCTGCTTGACGGCCTGTTT	360	342	58
		Probe	CACCACCGACGAATT	326	340	70
Culex quinque-fasciatus	Calcium binding gene	Forward Primer	CCTGAATGAGTACCGCACCAT	214	234	59
		Reverse Primer	CGGCAAGGGCGGAGAT	272	257	59
		Probe	TGAAGGCTCTGTGGGAC	237	253	69
Anopheles stephensi	Actin	Forward Primer	CCATTTGCGCCACCTCTCT	1798	1816	60
		Reverse Primer	ATGATAATAGGCGGCGCAAT	1854	1835	58
		Probe	ACCAGACATCGGACGC	1818	1833	68

Table 4. List of the primers designed by the alignment software for the Carboxypeptidase, Cytochrome b5 and Calcium binding gene of the three species of mosquitoes

Mosquito species	gene	Type	Sequence
All the mosquito species	Carboxypeptidase	Forward Primer	ACTCCAACTTCCCTGCGACGTTCAAGAT
		Reverse Primer	GGCAGTTCCATCGTCATCGAAAT
All the mosquito species	Calcium binding gene	Forward Primer	AGCGCGTCGAATTCATCGTC
		Reverse Primer	CCGTCGTTGTTGATGTCCAT
Anopheles gambiae	Cytochrome b5	Forward Primer	ATGTCGGAAGTGAAAACGTA
		Reverse Primer	GGTTCCTTCTTGACCGGGAT
Aedes aegypti & *Culex quinquefasciatus*		Forward Primer	TCTACGACGTGACGGAGTTC
		Reverse Primer	CGGTGCTCCAGTCGGGTTCC
Anopheles stephensi	Actin	Forward Primer	ATGGTCGGYATGGGNCAGAAGGACTC
		Reverse Primer	GATTCCATACCCAGGAAGGADGG

2.1 Prevalence of Diabetes mellitus and impaired glucose tolerance in the Raika and other communities with similar life style in Rajasthan

Principal Investigator: *Dr. Bela Shah, Director-In-Charge*
Co-Investigators: *Dr. K. R. Haldiya, Scientist 'F' and Dr. A. K. Dixit, Scientist 'E'*
Commencement: August, 2009 **Duration:** Three Year **Status**: Ongoing
Funding Agency: Indian Council of Medical Research – Task Force (Extramural)

OBJECTIVES

1. To estimate the prevalence of diabetes mellitus and impaired glucose tolerance in Raika community of Rajasthan
2. To compare prevalence of diabetes mellitus and impaired glucose tolerance in Raika community and other communities with similar life style
3. To find the association of diabetes and impaired glucose tolerance with camel milk consumption, if any.

PROGRESS

A total of 2890 households were covered from 67 villages of Jodhpur, Barmer and Pali districts of Rajasthan and 6373 individuals were interviewed and examined. Data of 2847 households and 5500 Raika and non Raika individuals were entered in the computer. The data of 2847 households and 5500 individual's data have been analyzed.

Highlights of Socio Demographic Survey of Households of Raika Community: Total family members in selected households were 15642, of which males and females were 8198 and 7444 respectively. Out of 15642 individuals, 12981 and 2661 individuals were raika and non raika respectively. The average family size of households of Raika community was 5.5±2.3 members (Range 1-19 members) while the same was 5.8±2.4 members in households of non Raika communities (Range 1-14 members). The literacy rate in raika community was 44.5% while in non raika communities, it was 53.3%. Among raika males, main occupations were student (25.9%), animal keeping (22.0), private job (16.3%) and labour (14.9%) while in females, it was house work (39.5%), labour (34.0%) and student (15.8%). Among non raika, the main occupations in males were students (33.1%), agriculture (23.8%), and labour (21.8%) while in females, it was house work (33.4%), student (23.6%) agriculture (23.5%) and labour (17.9%). The average monthly income of households of Raika community was 7424.1± 4872.8 rupees which was lower than monthly income of the households of non raika community (Rs.8299.7±5723.1). The safe drinking water sources in raika and non raika households were 63.9% and 68.9% respectively. The household amenities were higher in non raika households as compared

to raika households. The majority of the raika households had animals (89.9%) but only 10.6% HH had camels as compared to non raika households i.e.77.2% and 2.4% respectively. The camel milk use was higher in raika households (12.5%) as compared to non raika households (1.7%).

Highlights Socio Demographic and clinical profile of 5500 Raika individuals and Non-Raika individuals surveyed: The average age of raika individuals was 40.1±16.8 years while in non raika individuals, it was 39.2±17.3 years. The main occupation of male raika individuals was animal keeping (38.5%) followed by labour (15.3%) and agriculture (37.6%) whereas in male non raika individuals, it was 2.3%, 25.4% and 37.6% respectively. The main occupation of female raika individuals was labour (44.0%) followed by house work (43.9%) and agriculture (9.6%) whereas in female non raika individuals, it was 29.1%, 40.6% and 24.6% respectively. The personal addiction of smoking, tobacco chewing and alcohol in male raika individuals was 41.1%, 24.0% and 3.15% respectively while in male non raika individuals it was 30.5% and 27.4% and 12.2% respectively. The majority of Raika individuals were vegetarian (99.0%) as compared to non raika individuals (75.4%) and their staple diet was bajra and wheat. The majority of Raika individuals used Soyabean oil for cooking vegetables (87.3%) and pure ghee on chapattis (97.2%) whereas non raika individuals, it was 96.7% and 99.9% respectively. Fruits were occasionally consumed by 98.1% raika and 99.8% non raika individuals. The majority of Raika had drunk camel milk in their life time (61.3%) and in last 12 months was only 10.0% while non raika individuals, figures were 10.0% and 1.3% respectively. The majority of individuals drunk camel milk occasionally. The majority of Raika individuals drunk 400ml or more camel milk at a time. A total of 12 known diabetics were detected during the survey, of which 9 belonged to raika and three were non raika communities. Among Raika males, the type of work was migratory (37.6%) followed by hard work (20.5%), sitting (16.0%) and standing (15.8%) whereas in non Raika males, it was hard work (33.1%), standing (34.5%) sitting (25.6%) and migratory (1.9%). Among raika females, the type of work was hard work (20.5%) followed by sitting (21.0%), stationary (18.7%), standing (13.7%), and migratory (2.3%) whereas in non Raika females, it was hard work (26.1%), sitting (36.4%), stationary (7.5%), standing (23.9%) and migratory (0.3%). The average height and weight of raika individuals was 160.2±8.5 cm and 50.1±9.4 kg while in non raika individuals, it was 158.4±9.2 cm and 49.9±10.7 kg respectively. The average abdominal circumference and hip circumference of raika individuals was 70.5±9.8 cm and 84.6±9.8 cm while in non raika individuals, it was 70.1±10.6 cm and 82.0±8.8 kg respectively. The average systolic and diastolic blood pressure of raika individuals was 124.8±15.7 mm. Hg and 70.7±11.3 mm Hg while in non raika individuals; it was 124.1±16.7 mm Hg and 72.3±17.3 mm Hg respectively

A total of 31 (0.6%) diabetics were detected from the surveyed individuals, of which 22 (0.5%) individuals belonged to raika individuals and 9 (0.9%) from non raika individuals. Out of 31 diabetics, 18 drunk camel milk, of which 9 drunk camel milk during last 12 months. The analysis of the data is in progress will be presented in the meeting.

WORK TO BE DONE

Field survey of remaining Raika individuals of Jodhpur, Ajmer, Pali and Barmer will be carried out and report will be submitted to ICMR in the month of July 2012.

2.2 Clinical presentation, treatment outcome and risk factors of coronary artery disease among patients admitted in tertiary care hospital in Jodhpur

Principal Investigator: *Dr. P. K. Anand, Scientist 'C'*
Co-Investigators: *Dr. M. K. Chauhan, Med. Supdt., MG Hospitsl, Jodhpur and Dr. A. K. Dixit, Scientist 'E'*
Commencement: January, 2011 **Duration:** Two Years **Status:** Completed
Funding: Desert Medicine Research Centre (Intramural)

OBJECTIVES

1. To estimate the hospital admission rates of these diseases during period 2000-2009
2. To identify the associated risk factors of coronary artery disease among admitted patients of coronary artery disease

PROGRESS

Data extraction work was carried out by observing the indoor bed head tickets of patients of Coronary Artery Disease (CAD) or Ischemic Heart Disease (IHD) as often used synonymously, admitted in Mahatma Gandhi Hospital, Jodhpur, Rajasthan. IHD patients are classified according to International Classification of Disease IX as ICD IX code 413 for Angina and ICD IX code 410 for Myocardial infarction. We have so far collected data of 1161 admitted patients. The data on age, gender, residence, tobacco use, diet, diagnosis, and clinical presentation, treatment outcome, past disease history and investigations report including biochemistry and electrocardiograph if found available, were recorded on data extraction form. Collected data was entered in epi-info software of computer and analysed later. Analysis of entered data showed hundreds of diagnosis and clinical presentation based on combinations of diseases diagnosed and symptoms complained by admitted patients respectively. For example diseases such as IHD, shock, diabetes will give rise to 4 types of diagnosis while symptoms such as chest pain, vomiting, headache and breathlessness will give rise to 15 types of clinical presentations either alone or in combinations of these symptoms. Therefore, for ease of demonstrating the data of all patients we have made few grouping.

Frequency distribution of manually created groups of data on diagnosis and clinical presentations is presented in the following figures as given below (Fig. 1-4):

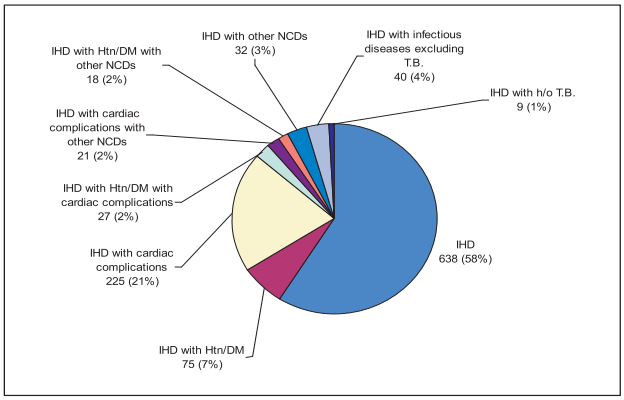


Fig. 1. Frequency distribution of diagnosis of IHD patients ICD IX code 410 (n=1085)

About 58% of these admitted patients were grouped as cases of IHD. Interestingly about 1 % of these patients were diagnosed as IHD who had given history of tuberculosis. Although their percentage is very less, this phenomenon may demonstrate the third dimension of epidemiological transition. As developing countries are experiencing the epidemiological shifting from infectious diseases i.e. diseases of underdeveloped or poverty to non-infectious diseases i.e. diseases of developed or rich communities, the occurrence of tuberculosis and IHD in same patient seems to demonstrate the third dimension of epidemiological transition.

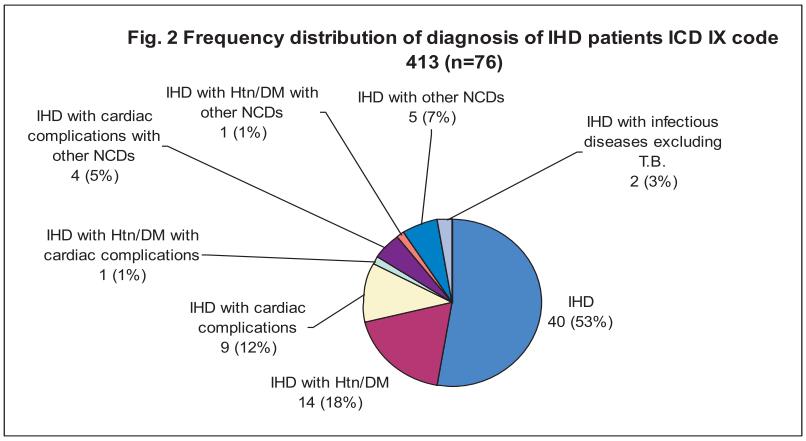


Fig. 2. Frequency distribution of diagnosis of IHD patients ICD IX code 413 (n=76)

Statistical analysis revealed that there was statistically significant difference in proportion of patients grouped as IHD with Htn/DM between ICD IX code 410 and 413 ($p<0.05$). These patients were more likely to be coded as ICD IX code 413. In other words proportion of patients diagnosed as IHD with Hypertension (Htn) and/or diabetes mellitus (DM) was significantly higher amongst angina and related diseases group as compared to myocardial infarction and related diseases group.

The proportion of patients diagnosed as IHD with cardiac complications was higher (20.73%) in ICD IX code 410 as compared with in ICD IX code 413 (11%). However, the difference was at borderline and insignificant (p=0.06). Statistical analysis for other diagnosis groups did not compute because of less number of cases available in 413 code.

Fig. 3 and 4 shows the types of clinical presentations in patients of ICD IX code 410 and 413 respectively. The proportion of patients presented with complaint of chest pain alone was 42% in ICD IX code 413 as compared with 23% in ICD IX code 410 group. The difference between these proportions was statistically significant (p<0.05).

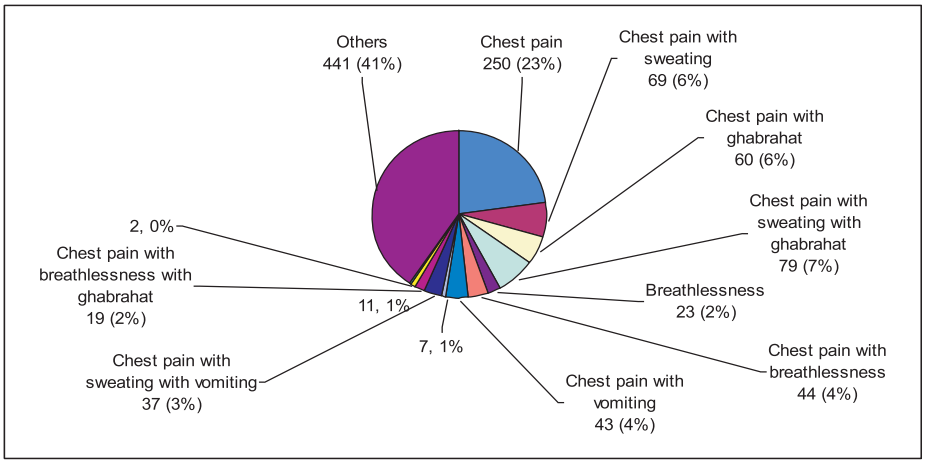


Fig. 3. Clinical presentation of patients of ICD IX code 410 (n=1085)

The difference between other types of clinical presentations could not be computed due to less number of cases available in 413 group.

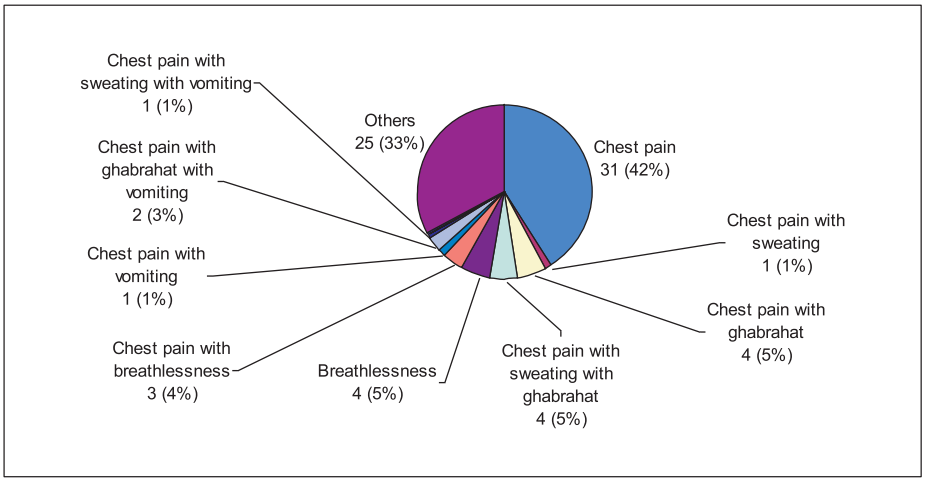


Fig. 4 Clinical presentation of patients of ICD IX code 413 (n=76)

Result on proportion of risk factors in ICD IX code 410 and 413 is presented as a table for both genders in Table 1.

Table 1. Proportion of patients in ICD IX code 410 and 413 exposed to various risk Factors

Risk factors	Proportion of patients reporting the status of behavioural risk factors				
		ICD IX code 410 (n=1085)		ICD IX code 413 (n=76)	
		Frequency	Proportion	Frequency	Proportion
Alcohol use	Information not available	608	56.03	57	75.00
	No	372	34.28	3	3.94
	Yes	97	8.94	16	21.05
	Stopped	8	0.73	0	0.00
Smoking	Information not available	488	44.97	63	82.89
	No	315	29.03	2	2.63
	Yes	255	23.50	10	13.15
	Stopped	27	2.48	1	1.31
Tobacco use	Information not available	854	78.70	71	93.42
	No	156	14.37	0	0.00
	Yes	70	6.45	5	6.57
	Stopped	5	46.08	0	0.00
Opium use	Information not available	858	79.07	70	92.10
	No	189	17.41	2	2.63
	Yes	37	3.41	4	5.26
	Stopped	1	0.09	0	0.00

Proportion of consumers of alcohol and opium was higher in ICD IX code 413 while proportion of smokers was higher in ICD IX code 410.

Table 2. Frequency of treatment outcomes in ICD IX code 410 and 413

Treatment outcome	ICD IX code 413 (n=76)	ICD IX code 410 (n=1085)
Death	2 (2.6%)	157 (14.5%)
Discharge	71 (93.4%)	894 (82.4%)
Left against medical advice	3 (3.9%)	34 (3.1%)

As evident from table 2, proportion of deaths is 14.5% and 2.6% in patients of ICD code 410 and 413 respectively. The difference was statistically significantly higher in ICD IX code 410 ($p<0.05$).

3.1 Study of food and nutrient consumption pattern in women of child bearing age and 6-59 months of age children, with particular reference to Pearl millet consumption pattern and effects of storage, processing, & cooking practices on retention of Iron, Zinc, Phytate and Polyphenols in Nagaur, a desert district of Rajasthan

Principal Investigator: *Dr. Madhu B. Singh, Scientist 'E'*
Co-Investigator: *Dr. J. Lakshminarayana, Scientist 'E'*
Commencement: August, 2010 **Duration**: One Year **Status:** Completed
Funding Agency: Harvest plus, Washington (Extramural)

OBJECTIVES

1. To assess the food and nutrient consumption distributions in a probabilistic sample of women of child-bearing age and under five children by means of 24 hour dietary intake recall in Nagaur district of Rajasthan
2. To assess the importance of pearl millet and other foods as sources of essential nutrients among women of child-bearing age and children less than 5 years of age in Nagaur district of Rajasthan
3. To study the seasonal patterns and time trends in pearl millet production and consumption in Nagaur district of Rajasthan by means of desk review and qualitative research
4. To assess the traditional processing and cooking methods and their effects on iron, zinc, phytate, and polyphenol retention among women of child-bearing age and children under 5 years of age through desk research, quantitative food technology and qualitative research in Nagaur district of Rajasthan

PROGRESS

Project activities included the scientific meetings with Stakeholders i.e. Deputy Directors of ICDS Department & Agriculture Department of Nagaur district, and CDPOs of ICDS department of Nagaur district, collection of background data regarding the pearl millet production, consumption, and other relevant information and use of 30 cluster Sampling, standardization of the approaches i.e. study design, data collection tools, data processing and analysis and training in CSDietary. Preparation of final schedules/questionnaires after pre testing. A cross sectional study was carried out in all ten tehsils / blocks of Nagaur district. Women in the child bearing (15-45 years) age and children under five years of age (6-59 months) were subjects for this study. 30 cluster sampling approach (as propagated by WHO) was adopted in dietary survey keeping in view the operational feasibility. The Sampling unit was kept at household level as in each house, mother and child were available. The Sample size was calculated on the basis of prevalence of iron

deficiency in diet of women in desert area as reported in scientific literature (Singh *et al*., 2009) as 20%, level of confidence of 95% relative precision of 20% and design effect of 2 using formula $(Z\alpha)^2 Q/(L^2) P$, sample size worked out to be 768, adjusted for a 20 percent non response. The sample size was rounded of to 900 from Nagaur district of Rajasthan or 900/30 =30 households per cluster and 3 house hold per cluster were repeated only for dietary intake. In Nagaur, geographically, a cluster consisted of a village. These 30 clusters/villages were selected from 10 tehsils (Sub-districts) of Nagaur district by means of simple random sampling using the Indian census 2001. 30 household in a village were selected systematically.

Inclusion Criteria: Only those households were selected which had women of child bearing age (15-45 years) and children of age between 6 to 59 months.

Exclusion Criteria: If in one household, women of child bearing age had two or more children with age of 6 to 59 months, then only one child at that household was considered for the study.

The study has been done in two parts i.e. collection of data from the eligible women of child bearing (15-45 years) and a child between 6-59 months of age from the selected household and the biochemical analysis of the food collected from the field.

In selected villages a random walk method starting from a central place (usually a temple) in the village and proceeding in at least four different directions was adopted. A household was selected only if eligible women of child bearing (15-45 years) and a child between 6-59 months of age were among the members of the family.

Most of the popular varieties cultivated and consumed were considered for estimation of nutrient contents. A sub sample i.e. 10% of women of selected household were requested to provide raw varieties of pearl millet in LDPE pouches. These were brought to laboratory for estimation of Iron, Zinc, Phytate and polyphenols retention in the laboratory.

At each household level, information on the demographic and socio-economic aspects were collected. At each household level, women, were interviewed for the dietary pattern using 24 hour recall method (Data was collected by the standard technique as followed by NIN (ICMR), Hyderabad) along with Roseland Gibson/harvest manual for 24 hour recall. Dietary intake details were collected for women of child bearing age i.e. (15-45 years) and children between 6-59 months at each household (HHs) level for the day prior to the interview. Information for Food Frequency Questionnaire (FFQ) was also collected from women at each HHs level.

At each village level Focus Group Discussions were conducted for collection of information regarding seasonal pattern and time trends in pearl millet production and consumption,

traditional processing and cooking methods etc. At each village level, focus group discussions were conducted from two types of key informant groups i.e. One group of 6 or more male persons (key informants) mainly key persons from village i.e. Panch, Sarpanch, teacher etc from the village to provide the above mentioned information. Second group consisted of 6 or more knowledgeable women (key informants) including Aganwari workers for providing the information regarding the preparation of different type of recipes from pearl millet, their consumption and preservation etc. They also demonstrated the method of preparation of different recipes made up from the pearl millet which were commonly consumed by the villagers. The Team learnt and standardized five commonly consumed recipes of pearl millet in the field in Nagaur district of Rajasthan.

Dietary intake data were entered by trained data entry operators and project staff into CSDietary software, whose training was given by HarvestPlus. Intake data were then transferred to Stata 10 software for statistical analysis. Data on socio-demographic aspects and other data collected from FFQ and FGDs were computerized in FOX-PRO and used Stata10 software for statistical analysis.

The samples of raw varieties of pearl millet collected from field were taken by the project staff to laboratory of Baroda Pearl Millet Center (Department of Foods and Nutrition, A WHO collaborating Center for health promotion, Faculty of Family and Community Sciences, MS University of Baroda, Vadodara, Gujarat) for testing of iron, zinc, phytate, and polyphenol retention. Pearl Millet project staff also prepared the five standardized recipes of pearl millet in the Baroda laboratory and dried them for testing to be done for above mentioned parameters. At Baroda laboratory, the determination of iron and zinc was done by Atomic Absorption Spectrophotometer, by Diacid mixture of HNO & HCLO in 3:1 ratio (Ryan *et al*., 2001) and phytate from raw Pearl Millet samples using official AOAC, Anion Exchange and quantitative determination of total phenol, method propagated by Lowry, *et al*., 1951.

At each household level information for demographic and socio-economic aspects and food frequency questionnaire were collected from 900 women of child bearing age i.e. 15-45 years. Information for dietary intake was also collected from these women and 900 children belonging to 6-59 months of age. A total of 30 Focus Group Discussions were recorded in which 291 males and 265 women key informants participated.

A total of 5 raw varieties of Pearl Millet i.e**.** 'Desi bajra', 'Pro Agro hybrid', 'MH-169', '118+154 Ghua Seed' and Pioneer' commonly consumed in Nagaur district were collected. The most common standardized recipes prepared from pearl millet in the study villages were *Sogra*, *Rab1* (Pearl millet Grains), *Rab2* (Pearl millet Flour), *Kadhi* and *Khitchri* (*Kheech*). Five samples each of raw varieties and five cooked recipes along with mixed flour of raw varieties of pearl millet were tested for iron, zinc, phytate and polyphenols.

RESULTS

Information collected from a total of 900 women and 900 households has been analyzed. Main socio-demographic characteristics of women participating in the study have been shown in Table 1. Majority of women belong to 18-25 years age group (56.8%). Pregnant women were 5.7% and lactating women were 66.0% of the total. Table 1 revealed that 88% of women were Hindus and 48.2% were illiterate. 90.1% were house wife whereas 4.8% were laborers and 3.3% were working women. Table 2 depicts the socio-demographic characteristics of children. It was observed that 37.1% belong to 6-23 months age group and 62.9% to 24-59 months age group. 42.7% children belong to breastfed & complementary feeding category whereas 43.8% were in weaned category. Table 3 shows that main morbidities observed among children were fever (42.2%) followed by ARI (15.2%) whereas 24.1% mothers suffered from fever and 9.8% from ARI.

Analysis of the household characteristics revealed that household size was 5.67 and 97.7 % head of household were male and 33.6% were illiterate. Main occupation were 'other Laborer' (46.4%) followed by 'service' (11.9%) and 'owner cultivator' (10.4%). In family type, 43.8% were of 'nuclear type' whereas 46.2 percent were 'extended nuclear type'. It was observed that 61.6% of the household had electricity and 65.7% had separate kitchen. Only 23.9% household had latrines. Main water sources were tank/pond (41.1%), tap (35.9%) followed by open well (11.6%). Main source of cooking fuel was firewood (91.2%). Majority of the households were made up of brick/stone wall+ tiled/asbestos/tin roof/ stone slabs (74.0%).

Analysis of frequency intake of high iron foods which contain 6-20mg iron/100g revealed that 83.2% consume Pearl millet daily whereas 54.3% consume wheat. Rice flakes are being consumed once a week (7.9%). As per the nutritive value of Indian foods (ICMR, 1989), Pearl millet contains more iron (8.0 mg) and zinc (3.1 mg) as compared to wheat (Iron- 4.9 mg, zinc 2.2 mg) and rice (iron- 2.8 mg & zinc- 1.4 mg) per 100 gm of edible portion.

Analysis of daily pearl millet intake by children and women of the target population of the district revealed that overall in children of 6-59 months of age, mean consumption was 118.8 g/day and 48.6% were consumers whereas in women it was 167.2 g/day and 100% were consumers of this flour.

Table 1. Main socio-demographic characteristics of women participating in the study

Women characteristics	No.	Percentage
Number examined	900	
Age Years		
18-25	511	56.8
26-35	342	38.0
36-45	47	5.2
Total	900	100.0
Pregnant	51	5.7
Lactating	594	66.0
Religion		
Hindu	792	88.0
Muslim	107	11.9
Sikh	1	0.1
Marital Status		
Married	889	98.8
Widowed/Divorced	11	1.2
Relationship with head of Household		
Wife	621	69.0
Other	279	31.0
Highest level of Schooling**		
Illiterate	434	48.2
Reads & write	140	15.6
1-4 standard	36	4.0
5-8 standard	212	23.6
9-12 standard	58	6.4
College	20	2.2
Occupation		
House wife	811	90.1
Labourers	43	4.8
Service	30	3.3
Owner cultivator	13	1.4
Artisans	3	0.4

**P<0.05

Table 2. Main socio-demographic characteristics of children participating in the study

Child characteristics	Number of children	Percentage
Number examined	**900**	
Age (Months)		
6-23 months	334	37.1
24-59 months	566	62.9
Total	900	100.0
Gender		
Male	487	54.1
Female	413	45.9
Breastfed*		
6-23 months	203	22.6
24-59 months	33	3.7
All	236	26.2
Breastfed & complementary fed	274	30.4
Weaned	390	43.4

* Information derived from Household Demographic Particulars (HDP), ** $p<0.05$

Table 3. Occurrence of illness in past 14 days in women and children

Disease	Number	Percent	Number	Percent
	Children (N=900)		Mothers (N=900)	
NAD	646	71.8	730	81.1
Fever	380	42.2	217	24.1
Diarrhoea	4	0.4	-	-
Dysentery	3	0.3	1	0.1
ARI	137	15.2	88	9.8
Measles	3	0.3	1	0.1
Other	2	0.2	1	0.1

Table 4 showed the energy and nutrient intakes per day of only 790 children of 6-59 months age groups as 110 children were exclusively breastfed. Analysis revealed that mean of energy in 6-23 months age group was 447.5 Kcal indicating 63.9% deficit in comparison to Recommended Dietary Allowances (RDA) ICMR whereas in 24-59 months age group, mean was 895.8 Kcal showing deficit of 47% with reference to RDA, ICMR. Children in 6-23 months age group showed deficit to the extant of 15% in proteins, 19.2% in fat, 69.2% in iron, 85.5% in zinc, 84.5% in vitamin C, 50% in thiamine, niacin, vitamin B12, vitamin A, and beet carotene 82.7% with reference to RDA, ICMR. In case of 24-59 months of age group, deficit was observed in iron (44.4%), zinc (66.4%), vitamin C (58.5%), thiamine

(33.3%), niacin (70.9%) with reference to RDA, ICMR whereas adequate consumption of proteins, fat, calcium and folate. The values for iron intakes are extremely low but this may be consistent with the very high levels of anemia found in the different surveys. Also it does not account for the fact that some children are getting iron from breast milk.

Table 4. Energy and nutrient intakes per day by children by age group

Nutrient	RDA		6-23 months N= 230		24-59 months N= 560		All N= 790	
	EAR	ICMR[3]	Mean	Percentile (25^{th}, 75^{th})	Mean	Percentile (25^{th}, 75^{th})	Mean	Percentile (25^{th}, 75^{th})
Energy (Kcal)	-	124016 (90)	447.5	390 (22.8,608)	895.8	723 (489, 964.5)	763.1	622.5 (392, 885)
Protein (gm)	-	22/30	18.7	16.3 (9.7, 24.9)	35.0	27.7 (18.3,37.7	30.2	23.6 (14.9,35.5)
Lipid (Fat) (gm)	-	25	20.2	17.3 (9.6, 27.7)	36.1	24.2 (14.2, 36.9)	31.4	22.3 (12.7, 33.9)
Calcium (mg)	500	400	558.0	445 (250, 749)	882.7	571.5 (304, 940)	786.3	531.5 (281, 878)
Iron (mg)	Low: 10.8/14.8 Mod: 5.4/7.4	12/18	3.7	1.7 (0.7, 5.4)	10.0	7.7 (5.1, 12.1)	8.1	6.5 (2.6, 10.3)
Zinc (mg)	WHO: 6.9/ 8.7 IZiNCG: 2 / 4	-	1.0	0.0 (0, 1.6)	2.9	2.5 (1.4, 4)	2.4	2.1 (0.4, 3.5)
Vitamin C (mg)	13/22	40	6.2	4.5 (2.2, 7.5)	16.6	8.5 (3.7, 15.5)	13.4	6.8 (3.1, 12.6)
Thiamine (mg)	0.4/0.5	0.6/0.9	0.3	0.3 (0.1, 0.4)	0.6	0.4 (0.3, 0.6)	0.5	0.4 (0.2, 0.6)
Riboflavin (mg)	0.4/0.5	0.7/1.0	0.9	0.7 (0.4, 1.2)	1.4	1.0 (0.5, 1.5)	1.3	0.9 (0.5, 1.4)
Niacin (mg)	5/6	8/11	1.3	0.7 (0.3, 1.9)	3.2	2.7 (1.8, 3.9)	2.7	2.2 (1.0, 3.5)
Folate (mg) DFE[2]	120 160	30/40	46.3	41.0 (22, 61)	86.6	66.5 (44.5, 94.5)	74.7	58 (37, 87)
Vitamin B 12 (mg)	0.7/1.0	0.2-1.0	0.0	0 (0,0)	0.1	0 (0, 0.6)	0.09	0 (0, 0.6)
Beta-carotene (mg)	-	1600	277.5	237 (133, 372)	596.3	373.5 (223, 559.5)	502.6	317.5 (190, 499)

[1] YEARs are shown for children 1-3 years of age/4-5 years of age for comparison to dietary intakes. For iron, EARs are given for low (5%) & moderate (10%) iron bioavailability. For zinc, EARs are given for WHO/FAO (2005) & for IZiNCG (2004) corresponding to low bioavailability. [2]Folate DFE, Dietary Folate Equivalents; Vitamin A RAE, Retinol Activity Equivalents. Indian Council of Medical Research[3] (ICMR), 1989

Table 5. Energy and nutrient intakes per day by women

Nutrient			Women (15-45 years) N= 900	
	EAR	RDA, NIN, ICMR	Mean	Percentile (25th, 75th)
Energy (Kcal)	-	1875 (2175)	1757.7	1658 (1318, 2016)
Protein (gm)	-	50 (65)	60.1	56.2 (45.1, 69.5)
Lipid (Fat) (gm)	-	20 (30)	38.7	33.6 (26, 43.4)
Calcium (mg)	1000	400 (1000)	657.1	532.5 (396.5, 764.5)
Iron (mg)	Low: 29.1 Mod: 14.5	30 (38)	33.9	29.4 (22, 41)
Zinc (mg)	WHO: 8.2 IZiNCG: 7	-	10.9	10.4 (7.4, 13.7)
Vitamin C (mg)	60	40	34.5	19.8 (9.9, 37.3)
Thiamine (mg)	0.9	0.9	1.2	1.1 (0.8, 1.4)
Riboflavin(mg)	0.9	1.1 (1.3)	1.3	1.2 (0.9, 1.6)
Niacin (mg)	11	12 (14)	9.8	9.0 (7.1, 11.9)
Folate (mg) DFE[2]	320	400	146.6	138 (103.5, 181)
Vitamin B 12 (mg)	2.0	1	0.2	0 (0, 6.8)
Beta-carotene (mg)	-	2400	1003.3	641 (482, 895.5)

[1]YEARs are shown for non-pregnant/non-lactating women 15-45 years of age for comparison to dietary intakes. For iron, EARs are given for low (5%) and moderate (10%) iron bioavailability. For zinc, EARs are given for WHO/FAO (2005) and for IZiNCG (2004) corresponding to low bioavailability. [2]Folate DFE, Dietary Folate Equivalents; Vitamin A RAE, Retinol Activity Equivalents. [3]Indian Council of Medical Research (ICMR), 1989

Table 5 shows the energy and nutrient intakes per day by women of 15-45 years age groups. Analysis revealed that mean of energy was 1757.7 Kcal indicating marginal deficit of 6.3 percent in comparison to Recommended Dietary Allowances (RDA) ICMR. Women showed adequate intake of iron, calcium, fat, protein, and riboflavin whereas inadequate intake of vitamin C (13.7% deficit), niacin (18.3% deficit), folate (63.3% deficit), vitamin B 12 (80.0% deficit), and vitamin A with reference to RDA, ICMR.

Analysis revealed that in case children of 6-59 months of age groups, milk was the main dietary source of energy (45.6%) followed by pearl millet 20.6 percent, fat & oils (17.8%) and roots & tubers (8.34%). In case of women of child bearing age groups, milk, fats &

oils and pearl millet were the main dietary source of energy i.e. 29.6, 25.9 and 23.7 percent respectively followed by roots & tubers (10.2%), pulses and vitamin C rich fruits (4.6%).

Analysis of dietary sources of iron by food group among the children and women of the studied population showed that in children of 6-59 months of age groups and women of child bearing age groups, main dietary source of iron was observed to be pearl millet i.e. 41.2% and 53.3% respectively.

Analysis of information collected during 'Focused Group Discussions' revealed that pearl millet was reported to be staple grains followed by wheat. Majority of villagers cultivate the pearl millet and only 25.7 percent purchased from the market. Fifty percent villagers kept the pearl millet stocks from 4 to 6 months and stored mainly in Bori (Sacks) and large metal bins.

In 40% of the villages, purchase of pearl millet was from local market for last year and majority (87%) preferred to grind the pearl millet from market generally purchased pearl millet flour from village flour mill generally one to three times per week and purchased 3 kg of pearl millet in individual buys (46.4%) in villages of Nagaur district. 86.7% villagers used the process of 'Soak, Pound, Dehusk, Sieve along with Pound and Mill'. The seasonal food consumption pattern revealed that the maximum pearl millet was consumed from November to February (33.3%), followed by December to February (30.0%). Results showed that 6.8% population did not consume pearl millet from April to July, followed by May to June (13.3%). Regarding the food consumption of pearl millet, it was observed that combination of *Sogra*, *Rabadi*, *Kheech* and *Kadhi* was consumed by 43.4%, followed by combination of *Sogra*, *Rabadi* and *Kadhi* (16.8%). It was interesting to note that none of the respondents neither preserved the pearl millet products nor purchased them from the market.

Biochemical Analysis of Food: The study revealed that five varieties of pearl millet i.e**.** 'Desi bajra', 'Pro Agro hybrid', 'MH-169 (commonly consumed) and '118+154 Ghua Seed', and Pioneer' (Rarely consumed) were consumed in Nagaur district. The most common recipes prepared from pearl millet in the study villages were *Sogra*, *Rab1* (Pearl millet Grains), *Rab2* (Pearl millet Flour), *Kadhi* and *Khitchri* (*Kheech*). Five samples each of raw varieties and five cooked recipes mentioned above along with mixed flour of three raw varieties of pearl millet commonly consumed by the villagers i.e. Desi bajra, Pro Agro hybrid, and MH-169, were tested for iron, zinc, phytate, and polyphenol retention. It was observed that maximum Iron was found in 'mixed flour (5.99 mg/100g) and MH-169' variety, followed by 'Desi bajra' (4.89 mg/100g). Maximum zinc was observed in 'Mixed Flour' i.e. 3.64 mg/100g, followed by Desi bajra (3.39 mg/100g). Phytate was maximum in 'Desi bajra' (468.2 mg/100g), whereas, total phenols in '118+154 Ghua seeds' (380

mg/100g). In the cooked recipes retention of zinc and iron increased in *rab1* (Pearl millet Grains) preparation i.e. 3.64 to 4.40 mg/100g and 5.99 to 10.5 mg/100g respectively (table 6). Trends indicated that MH-169, Mixed flour and Desi Bajra varieties of pearl millet were observed to be good as the contents of total phenols and phytate were less and Iron was more in comparison to other varieties.

Table 6. Summary of Results of zinc, total iron, phytates and total phenols in raw and cooked Pearl Millet based recipes

Food Sample	Zinc (mg/100g)	Iron (mg/100g)	Phytate (mg/100g)	Total Phenol (mg/100g)
Raw varieties				
118+154 Ghua Seed	3.25	4.60	467.2	380
Desi Bajra, District Nagaur	3.39	4.89	468.2	340
MH-169, District Nagaur	2.34	5.99	322.8	120
Pro Agro hybrid, District Nagaur	2.65	4.90	537.0	370
Pioneer	3.29	NA	468.0	350
Mixed Flour	3.64	5.99	300.0	210
Cooked Recipes				
Rab1 (Pearl millet Grains)	4.40	10.5	200.5	270
Rab2 (Pearl millet Flour)	4.84	5.29	267.4	260
Khitchri (Kheech)	3.59	5.29	200.5	320
Sogra	3.89	9.99 (Iron Tava) 6.31 (Mitti Tava)	234.0	310
Kadhi	3.74	6.39	267.2	220

In biochemical analysis, phytate content shows that we have a really high phy/zn ratio, way above the 15 cut off value. This provides us with sufficient evidence to assume low bioavailability for both iron and zinc. Analysis revealed that retention of phytates and phenols were reduced after cooking in most of the preparations where processes of Soaking, Pounding and Dehiscing were involved such as Rab1 (Pearl millet Grains), Khitchri and Kadhi. Combination of Rab1(Pearl millet Grains), Khitchri and Kadhi are good, where processes of Soaking, Pounding and Dehusking were involved due to which Iron retention was found good.

LEADING TRENDS

- MH-169 variety of pearl millet was observed to be best as the contents of total phenols and phytate were less and iron was more in comparison to other varieties, followed by mixed flour and Desi bajra.
- Among the cooked recipes, combination of Rab1(Pearl millet Grains), Khitchri and Kadhi is best, where processes of Soaking, Pounding and Dehiscing were involved due to which Iron retention was found good.
- Diet of children is deficient in intake of energy (calories), proteins, iron, zinc and calcium in their diet, in comparison to RDA (ICMR), whereas, in women of child bearing age, deficiency was observed mainly in energy (calories) intake.
- Pearl millet is main staple diet (63.0%) of rural areas of the district, followed by wheat (26.7%), revealing that it is a significant source of dietary energy and nutritional security for rural populations.
- Micronutrient enriched millet verities may find beneficial application in the communities those subsisting mainly on pearl millet as staple food in reducing the micronutrient deficiencies in this area. The study suggests further research in this direction.

RECOMMENDATIONS

1. The study indicated that attempts to develop/utilize the pearl millet varieties constituting high content of iron and zinc & low phytate and polyphenols may be made in view to reduce the micronutrient deficiency disorders in the desert population, as pearl millet is the main staple food for their subsistence.
2. The appropriate combinations of the preparations of the food products of pearl millet have to be promoted to enhance the quantity of micronutrients especially, iron and zinc.
3. Multi-location Bio-Efficacy trials should be carried out in different states like Gujarat, Rajasthan and Maharashtra in India, where base line work has already been carried out.
4. For dissemination of the results of the study, there is an urgent need to produce a range of educational materials highlighting the health, nutrition and therapeutic values of different pearl millet varieties & their products aimed at desert population/ consumers (villagers) and ecological values of millets addressing the farmers.
5. In order to study the Varietals' difference versus processing products versus Nutrients, probably requires the support of nodal agencies like ICAR/ICRISAT, who are involved in the development of the pearl millet varieties rich in iron contents. Multi centric field trials of different varieties of fortified pearl millet may be conducted. Joint proposal from ICMR in collaboration with above mentioned agencies may be developed for field trials.

3.2 Assessment of Iodine deficiency disorder, anemia and nutrition intervention in school age children of Jodhpur district of Rajasthan

Principal Investigators: *Dr. Madhu B. Singh, Scientist 'E' and Dr. K. S. Premavalli, Scientist 'F', DFRL, Mysore*
Coordinators*: Dr. Bela Shah, Director-in-Charge, DMRC and Dr. A. S. Bawa, Director, DFRL, Mysore*
Commencement: July, 2008 **Duration:** Three years **Status:** Completed
Funding Agency: *Defence Food Research Laboratory, Mysore (Extramural)*

OBJECTIVES

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

PROGRESS

Project activities included working on sampling plan in detail. According to WHO/ UNICEF/ICCIDD, for school based survey, 30 cluster sampling approach has 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 $(Za)^2 Q / (L^2) P$, 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 were 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 was done from district education office, Jodhpur. Secondly cumulative enrollment was determined. Finally schools were selected using PPS sampling technique as recommended by WHO. In the selected schools, children were selected randomly using Tippets random number table. Equal proportion of boys and girls and proportionate distribution of children from 6-11 years were covered in the selected school. Nutrition intervention has been conducted on 311 school children (15% of calculated sample size i.e. 270 and consideration of 15% non response of 270 i.e. 41).

In this nutrition intervention project 311 school children of 6-11 years age groups belonging to 4 schools of 4 villages i.e. Bichat, Swazi Khurd, Bedevils and Gangani of Jodhpur tehsil of Jodhpur district were registered for supplementation. All registered children were

interviewed /examined for Socio-demographic profile, nutritional deficiency signs and morbidity for last 15 days. All registered children were examined for anemia assessed by hemoglobin levels (Cyanmethaemoglobin technique), and have been classified as per WHO classification. Iodine deficiency disorders were assessed by clinical examination of thyroid gland using the standard method as recommended by the joint WHO/ UNICEF/ ICCIDD consultation. A casual urine sample was collected for estimation of Urinary Iodine Excretion (UIE) levels to asses the Iodine nutriture status. Iodine has been determined by Ammonium Persulphate Digestion on Microplate method (APDM) using standard laboratory technique. UIE level less than 10 mcg/dl have been considered as indicator of iodine deficient nutriture. Children were requested to bring 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 then 15 ppm were classified as with inadequate iodine. Each child was assessed for psychological tests on learning attributes in terms of memory, intelligence, reasoning and attention etc before supplementation.

Four psychological tests were performed to test the IQ level of children of age group 6-11 years. These were Colored Progressive Matrices Test (CPM) grades (For Cognitive development & Intelligence level of children- Grade 1 Excellent, Grade II Above Average, Grade III Average, Grade IV Below Average, Grade V Impaired or mentally retarded), Knox Cubes Test, KCT test grades (Intelligence level and short term memory- Grade 1 Idiot, Grade 2 Imbecile, Grade 3 Moron, Grade 4 Border line seeable minded, Grade 5 Dull, Grade 6 Average, Grade 7 Superior, Grade 8 Very superior and Grade 9 Genius), Techistoscope Test, (TT) points (concentration and short term memory- Score points <5 Below Average, Score points 7–2 i.e. 5-9 Average, Score points >9 Above Average) and Digit Span Test. (DST) points (short term memory- Score points <5 Below Average, Score points 7–2 i.e. 5-9 Average, Score points >9 Above Average). Hands on Training was imparted by Dr. Ravi Kumar Gunthe, Associate Professor, Psychology Department, JNV University, Jodhpur for performing these psychological tests and their interpretation and analysis.

All the registered school children were administered with supplements daily for a period of 180 days. These supplements have been supplied by Defence Food Research Laboratory, Mysore. These registered children were re-examined after a period of 180 days supplementation for all above mentioned parameters to observe the impact of the supplements on reduction of micronutrient deficiency disorders, malnutrition and psychological parameters in school children of Rajasthan. Salt, urine analysis and haemoglobin estimation was done at laboratory by trained technician and project staff at DMRC. Data collected in the field have been computerized under the supervision of Dr. J. Lakshminarayana, biostatistician in this project. Dr. Ranjana Fotedar, Scientist C was also involved in field survey for examination of children before and after supplementation.

Analysis of 311 children registered for the study revealed that out of 311 children, 129 were boys and 182, girls (Table 1). 89.1% children belong to Hindu religion whereas 10.9% belong to Muslim religion. 46% children belong to OBC caste and 25.7% to SC caste. Illiteracy was high among the parents i.e. 46.5% belong to illiterate category whereas 1.6 % have studied up to college level. 43.3% children belong to category of monthly income group of Rs. 285 to Rs. 569 following modified Prasad classification, 1997. The non-response in giving blood samples before and after supplementation was 14.1%.

Table 1. Age and sex wise distribution of school age children covered

Age Group Years	Boys	Girls	Total
6+	25	20	45
7+	14	26	40
8+	22	32	54
9+	29	36	65
10+	20	40	60
11+	19	28	48
POOL (N=311)	**129**	**182**	**311**

Table 2. Age-wise distribution of school age children according to anemia (Hemoglobin Estimation) before the supplementation

Haemoglobin Valuesfi Age group years		On the basis of Haemoglobin Estimation							
		Non-Anemic		Anemic					
		Normal (≥ 12 g/dl)		Mild (10-12 g/dl)		Moderate (7-10 g/dl)		Severe (<7 g/dl)	
	N	N	%	N	%	N	%	N	%
6+	45	8	17.7	25	55.6	12	26.7	0	0.0
7+	40	10	25.0	21	52.5	8	20.0	1	2.5
8+	54	6	11.1	31	57.4	16	29.6	1	1.9
9+	65	14	21.5	41	63.1	10	15.4	0	0.0
10+	60	15	25.0	31	51.7	14	23.3	0	0.0
11+	47	11	23.4	28	59.6	8	17.0	0	0.0
Total	**311**	**64**	**20.6**	**177**	**56.9**	**68**	**21.9**	**2**	**0.6**

Table 2 revealed the distribution of overall school age children according to Hb estimation before supplementation and observed that overall only 20.6% children were in non

anemic category. Overall 56.9% children belong to mild category of anemia whereas 21.9% to moderate and 0.6% to severe category. In case of boys, 53.5% belong to mild category and 0.8% to severe category. Table 3 revealed the distribution of overall school age children according to Hb estimation after supplementation and observed that overall 38.2% children were non anemic category. Overall 55.8% children belong to mild category of anemia whereas only 6.0% to moderate category. No case was observed in severe category.

Table 3. Age-wise distribution of school age children according to anemia (Hemoglobin estimation) after the supplementation

Hemoglobin Valuesfi→ Age group years		On the basis of Haemoglobin estimation							
		Non-Anemic		Anemic					
		Normal (≥ 12 g/dl)		Mild (10-12 g/dl)		Moderate (7-10 g/dl)		Severe (<7 g/dl)	
	N	N	%	N	%	N	%	N	%
6+	38	13	34.2	24	63.2	1	2.6	0	0.0
7+	35	12	34.3	20	57.1	3	8.6	0	0.0
8+	48	20	41.7	26	54.2	2	4.1	0	0.0
9+	54	23	42.6	29	53.7	2	3.7	0	0.0
10+	52	22	42.3	24	46.2	6	11.5	0	0.0
11+	41	13	31.7	26	63.4	2	4.9	0	0.0
Total	**267**	**102**	**38.2***	**149**	**55.8**	**16**	**6.0**	**0**	**0.0**

*P<0.05 Non-anemic Before supplementation Vs Non-anemic After supplementation-Non Response= 44 (14.1%)

Results of table 2 and 3 revealed that supplementation of pearl millet products has reduced anemia from 79.4 to 61.8% i.e. decline of 17.6%. After supplementation, decline was observed in moderate category of anemia i.e. from 21.9% to 6.0% and no case of severe anemia was found after supplementation. Decline in anemia was more in case of girls than boys. The overall percentage of non anemic children increased significantly from 20.6 to 38.2% ($p<0.01$) after supplementation.

Before supplementation, only 174 children gave adequate quantity of urine samples and rest of them either denied to give due to fear or gave inadequate quantity of urine sample. But after supplementation, 245 children gave urine samples which were adequate in quantity for analysis. Analysis revealed that overall 71.3% children were deficient in UIE levels whereas 23.6% were in normal category. After supplementation, 38.8% children

were observed in normal category whereas 30% deficient in UIE level. Results showed that there was a significant decline in UIE deficiency after supplementation ($p<0.01$). The percentage of use of salt inadequate in iodine content i.e. less than 15 ppm was 2%.

Analysis of total children according to Colored Progressive Matrices Test (CPM) grades (For Cognitive development & Intelligence level of children) before and after the supplementation revealed that overall 31.6% children were in III grade (average category) whereas 63.1 in Grade IV before supplementation. After supplementation, 3.2% were observed in II grade (Good) whereas 73.6% in III grade (average category) and only 18.2% in Grade IV. Analysis showed that supplementation has shifted 42% children from IV grade (Below Average) to III grade (Average) which is good impact on the Cognitive development & Intelligence level of children ($p<0.01$). Analysis revealed that overall 84.6% children showed increase in CPM grades after supplementation.

Analysis of distribution of total children according to Knox Cubes Test, KCT test grades (Intelligence level and short term memory) before and after the supplementation revealed that overall 10.6% children were observed in Grade 9 (average IQ level) whereas 11.0% in Grade 5 IQ level before supplementation. After supplementation, children observed in Grade 9 (average IQ level) were 16.7%, 30.9% in grade 7 (superior Q level), 7.1% in grade 8 (Very superior IQ level and 16.0% in grade 9 (genius IQ level). Significant increase in percentage of children in grade 7 to 9 were observed ($p<0.05$) after supplementation.

Distribution of total children according to Techistoscope Test, (TT) points (concentration and short term memory) before and after the supplementation revealed that overall 67.7% children were observed in Scale of 7–2 (average) whereas 25.5% in Scale of >9 (above average) before supplementation. After supplementation, children observed in Scale of 7–2 (average) were 76.6%. Supplementation has shown significant increase in percentage of children in average category ($p<0.01$).

Distribution of total children according to Digit Span Test. (DST) points (short term memory) before and after the supplementation showed that overall 42.2% children were observed in Scale of 7–2 points (average) whereas 0.7% in Scale of >9 points (above average) before supplementation. After supplementation, children observed in Scale of 7–2 points (average) were 98.5% whereas 1.1% in Scale of >9 points (above average). Supplementation has shown significant increase in percentage of children in average category ($p<0.01$) i.e. from 42.2 to 98.5 %. Analysis revealed that overall 68.7% children showed increase in DST points scale after supplementation.

Distribution of children according to Standard Deviation classification for weight for age before and after the supplementation revealed that 76.0% boys and 71.1% girls were observed to be in normal category whereas 23.3% boys and 28.3% girls in the category

of Moderate malnutrition and 0.6 to 0.8% in severe category before supplementation. The percentage of children observed in normal category was 87.4% boys and 83.2% girls after supplementation. Results showed that there is a decline of moderate malnutrition from 26.2 to 14.3% after supplementation. Significant increase in percentage of children belonging to normal category has been observed i.e. from 73.1 to 85 % ($p<0.01$).

Distribution of children according to SD classification for height for age before and after the supplementation revealed that 87.4% boys and 79.1% girls were observed to be in normal category whereas 12.6% boys and 15.4% girls in the category of Moderate malnutrition and 3.3% in severe category before supplementation. The percentage of children observed in normal category were 93.3% boys and 87.6% girls after supplementation. Results showed that overall there is a significant incline of children in normal category from 82.5 to 90.0% after supplementation ($p<0.05$).

Analysis of the distribution of children according to Nutritional deficiency signs before and after the supplementation revealed that the overall percentage of hair dispigmantation, bitot spot, angular stomatitis and Cheliosis were 24.3, 1.3, 0.3, 0.7% before supplementation and 26.8, 0.7, 0.0, 0.7 % after supplementation respectively ($p>0.05$). The overall percentage of UTI, fever, GIT and respiratory complaints were 0.6, 1.9, 1.3, 3.9 respectively before supplementation whereas the overall percentages after supplementation were 0.3, 3.2, 1.3, 3.8 respectively ($p>0.05$).

IMPORTANT FINDINGS

- Demographic profile revealed that majority of children belongs to Hindu religion and income group below Rs. 949/pm whereas illiteracy among parents was high i.e. 46.5%.
- The supplementation of pearl millet products has reduced anemia, on the basis of haemoglobin estimation, from 79.4% to 61.8% i.e. decline of 17.6%. After supplementation, decline was observed in moderate category of anemia i.e. from 21.9% to 6.0% and no case of severe anemia was found after supplementation. Decline in anemia was more in case of girls than boys. The overall percentage of non anemic children increased significantly from 20.6 to 38.2% ($p<0.01$) after supplementation.
- There was a good decline in UIE deficiency from 71.3 to 30% after supplementation. Results showed that there was a significant decline in UIE deficiency from 71.3-31% after supplementation ($p<0.01$).
- For Cognitive development & Intelligence level of children, Colored Progressive Matrices Test (CPM) grades were assessed before and after the supplementation. Analysis showed that supplementation has shifted 42% children from IV grade (Below Average) to III grade (Average) which is good impact on the Cognitive development & Intelligence level of children ($p<0.01$). Analysis revealed that

overall 84.6% children showed increase in CPM grades after supplementation.

- Knox Cubes Test, KCT test grades measured for Intelligence level and short term memory revealed that supplementation has increased the percentage of children (6.1%) in average IQ level. Significant increase in percentage of children in grade 7 to 9 were observed ($p<0.05$) after supplementation.
- Techistoscope Test, (TT) points were used to measure the concentration and short term memory revealed that 8.9% children increased in average scale of 7–2 after supplementation. Supplementation has shown significant increase in percentage of children in average category ($p<0.01$).
- Digit Span Test (DST) points used for short term memory revealed that overall 68.7% children showed increase in DST points scale after supplementation. Supplementation has shown significant increase in percentage of children in average category ($p<0.01$) i.e. from 42.2 to 98.5%.
- SD classification for weight for age showed a decline of moderate malnutrition from 26.2 to 14.3% after supplementation. Significant increase in percentage of normal category has been observed i.e. from 73.1 to 85% ($p<0.01$). SD classification for height for age showed that overall there is a significant incline of children in normal category from 82.5 to 90.0% after supplementation. Slight decline has been observed in prevalence of bitot spot and angular stomatitis though statistically insignificant.

RECOMMENDATIONS

1. The supplementation of pearl millet products had improved haemoglobin levels and declined anemia. Significant increase in non anemic children was observed after supplementation in rural school children residing in desert areas of Rajasthan who are in a constant state of stress due to extreme environmental conditions of desert. The findings suggest that these products can also be included, in the ongoing national programs in the study/arid areas as pearl millet is staple diet of desert area.
2. The supplementation of pearl millet products also had significant positive effect on iodine nutriture and malnutrition.
3. The supplementation of pearl millet products also had significant positive effect on psychological tests performed on learning attributes in terms of memory, intelligence, cognition i.e. reasoning and attention after supplementation.
4. Study suggests large scale field trial to validate/strengthen these findings in desert area.

3.3 Nutrition Monitoring Survey on NNMB pattern in Jodhpur district of Rajasthan

Principal Investigator: *Dr. Madhu B. Singh, Scientist 'E'*
Co-Investigator: *Dr. J. Lakshminarayana, Scientist 'E' and Dr. Ranjana Fotedar, Scientist 'C'*
Commencement: January, 2005 **Duration:** Long Term **Status:** Ongoing
Funding: Desert Medicine Research Centre (Intramural)

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

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 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. 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.

Fifth phase has been completed in which 30 villages were covered from six tehsils of Jodhpur district i.e. Jodhpur, Osian, Pahlavi, Shergarh, Bilbray and Bhopalgarh (five

villages from each tehsil), covering 600 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 FIFTY), 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 2797 individuals of the fifth phase has been done. Table 1 showed age and sex wise distribution of population (1469 males and 1328 females). Analysis revealed that 96 percent of population was Hindus. Nuclear families were significantly more (87.5 %) as compared to 1.3 % joint families (Tables 2). Table 3 revealed that illiteracy is significantly high in females (57.5 %) than males (34.5 %). Higher education is very low in this area i.e. 1.5 percent.

Table 1. Age and sex wise distribution of population covered

Age group	Males	%	Females	%	Total	%
0-5	227	15.5	213	16.1	440	15.8
6-9	147	10.0	135	10.2	282	10.1
10-14	208	14.2	175	13.2	383	13.7
15-17	111	7.6	69	5.2	180	6.4
18– 29	283	19.3	248	18.7	531	19.0
30 - 39	177	12.0	200	15.1	377	13.5
40-49	158	10.8	141	10.6	299	10.7
50 -59	90	6.1	78	5.9	168	6.0
>=60	68	4.6	69	5.2	137	4.9
Total	1469	100.0	1328	100.0	2797	100.0

Table 2. Distribution of households according to type of family

Type of family	N	%
Nuclear	525	87.5
Extended Nuclear	67	11.2
Joint	8	1.3
Pooled	600	100.0

Table 3. Distribution of population according to educational status

Educational status	Males	%	Female	%	Total	%
Illiterate	507	34.5	764	57.5	1276	45.4
Read & Write	12	0.8	13	1.0	24	0.9
1-4 Standard	217	14.8	200	15.1	417	14.9
5-8 Standard	358	24.4	224	16.9	581	20.9
9-12 Standard	251	17.1	60	4.5	311	11.1
College	38	2.6	4	0.3	42	1.5
N. A.	86	5.8	63	4.7	149	5.3
Pooled	1469	100.0	1328	100.0	2797	100.0

X^2=225.393, P<0.01

Main morbidities observed in population were, fever (8.4 %), and acute respiratory infection (5.2%) followed by diarrhea (1.0 %). Both the morbidities i.e. acute respiratory infection and fever were higher in females than males (Table 4). Regarding nutritional deficiency signs, it was observed that discoloration and sparseness of hair, a sign of protein calorie malnutrition was observed to be high i.e. 8.1 percent which was significantly higher in females than males. Marasmus was observed only in females (0.1 %). Angular stomatitis, cheliosis and glossitis were ranging from 0.3 to 2.6%. Vitamin A deficiency i.e. Night blindness and Bitot spot were 0.8 and 2.2%. Dental caries (27.9%) and dental fluorosis (24.9%) observed high in this area. Koilonychia, a sign of anemia, was observed only in females i.e. 0.1%.

Table 4. Distribution of population according to morbidity profile

Morbidity	Males N=1469	%	Females N=1328	%	Total N=2797	%
N.A.D.	1220	83.0	908	68.4	2128	76.1
Fever	97	6.6	138	10.4	235	8.4
Diarrhoea	12	0.8	17	1.3	29	1.0
Dysentery	0	0.03	0	0.0	0	0.0
A. Res. Infection	64	4.4	82	6.2	146	5.2
Measles	3	0.2	1	0.08	4	0.1
GIT	22	1.5	10	0.8	32	1.1

X^2=35.2, P<0.01

Table 5. Distribution of population according to nutritional deficiency signs

Deficiency Signs	Males N=1469	%	Females N=1328	%	Total N=2797	%
Hair Discoloured	91	6.2	136	10.2*	227	8.1
Hair sparseness	0	0.0	1	0.1	1	0.0
Marasmus	0	0.0	1	0.1	1	0.0
Night Blindness	8	0.5	14	1.1	22	0.8
Bitot Spot	22	1.5	40	3.0	62	2.2
Angular stomatitis	31	2.1	38	2.9	69	2.5
Cheilosis	34	2.3	38	2.9	72	2.6
Glossitis	3	0.2	6	0.5	9	0.3
Koilonychia	0	0.0	2	0.2	2	0.1
Gums-Spongy bleeding	8	0.5	18	1.4	26	0.9
Dental Caries	266	18.1	514	38.7**	780	27.9
Dental Fluorosis	245	16.7	454	34.2**	699	24.9
Thyroid gland palpable	0	0.0	1	0.1	1	0.0
Thyroid gland visible	0	0.0	1	0.1	1	0.0
Others	1	0.1	0	0	1	0.0

* P<0.05, ** 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. 70.4% which was higher in SC community (80.6%) in comparison to other communities. The overall prevalence of severe under nutrition was high i.e. 6.4% and found higher in lower income group (14.0%) where as reverse trend was observed in case of mild under nutrition i.e. more in high income group (31.1%). Under nutrition was higher in nuclear families (70.7 %) and observed maximum in semi pucca houses (74.2 %).

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
Nuclear	205	29.3	36.6	26.8	7.3
Extended	31	32.3	38.7	29.0	0.0
Pooled	236	29.6	36.9	27.1	6.4

$\chi^2 = 7.27$ P<0.05 * NCHS Standards

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

Community	N	Nutritional Grades* (Percent)			
		Normal	Mild	Moderate	Severe
S.C	72	19.4	41.7	33.3	5.6
S.T	22	40.9	31.8	18.2	9.1
B.C	124	31.5	34.7	27.4	6.5
Others	18	44.4	38.9	11.1	5.6
Pooled	236	29.6	36.9	27.1	6.4

$\chi^2 = 26.296$ $P<0.05$, * 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 as well as WHO 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.

Underweight (Weight for age) in preschoolers was observed 59.3%, higher than NFHS III (44.0%). The proportion of severe underweight was high (25.9 %) (Fig.1). Underweights were observed higher in males than females though statistically insignificant. Declining trend has been observed in underweight in comparison to Phase one (71.6%), Phase II (59.5%) study but higher than Phase III (58.3 %), and Phase IV (49.8 %) study. Underweight in preschoolers observed was 53.8 percent using WHO standards.

Stunting (Height for age) was 51.7% in preschoolers with the prevalence of severe stunting 28.4%, 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%), DMRC Phase II (62.1%), DMRC Phase III (57.1%) and DMRC Phase IV (59.5%) as shown in Fig. 2. Stunting computed by adopting standard deviation classification using WHO standards was 54.2% in preschoolers with the prevalence of severe stunting 31.3%. Wasting (Weight for Height) computed by adopting standard deviation classification using WHO standards was 25.9% in preschoolers with the prevalence of severe wasting 8.5%.

Fig 1. SD Classification for Weight for Age in Preschoolers

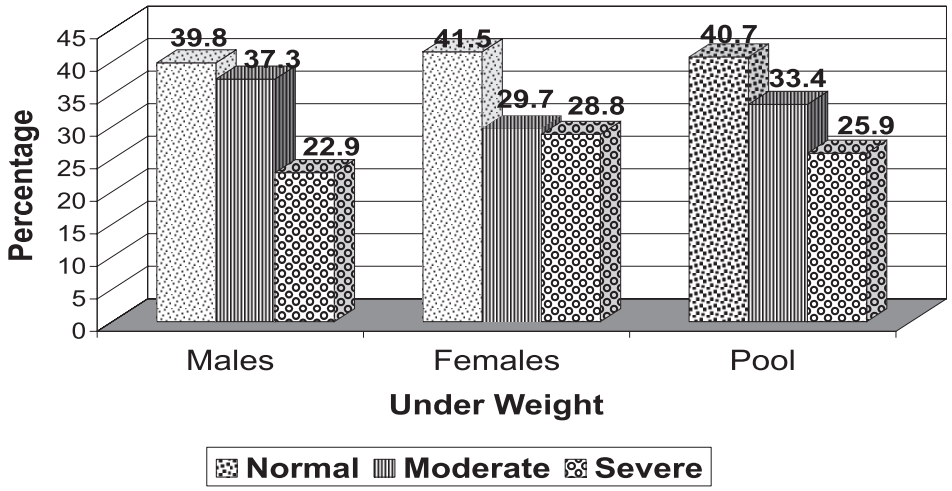
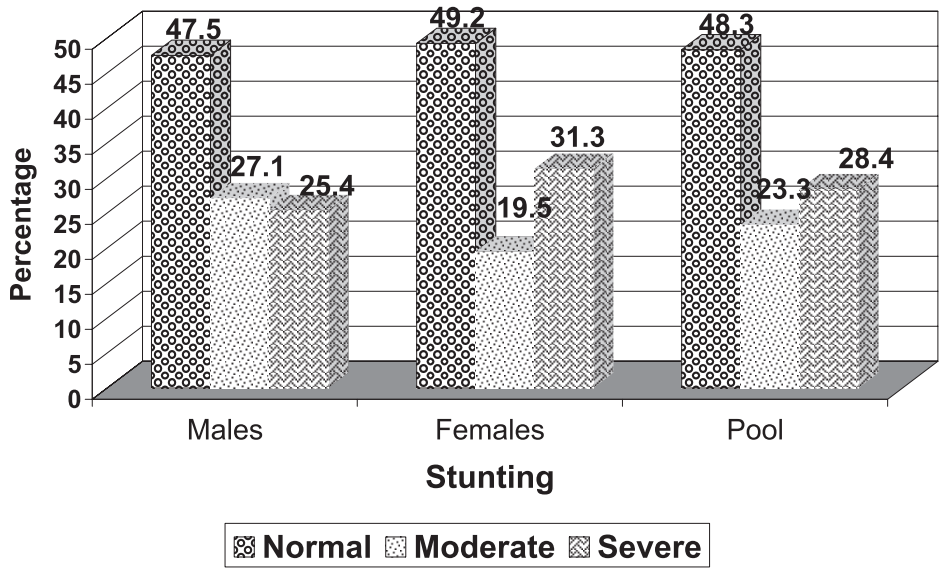


Fig 2. SD classification for Height for Age in Preschoolers



The distribution of adults according to BMI grades have been shown in Tables 8 to 10. At the aggregate level, 54.8% had normal BMI (18.5-25.0), while 32.2% had chronic energy deficiency (Fig. 3). Severe chronic energy deficiency was higher in extended families (8.9%) and maximum in pucca type houses (12.9%) and SC community (12.2 %) though statistically insignificant.

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

Gender	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=215)	0.9	10.7	31.2	19.1	18.6	7.9	11.6
Female (N=538)	0.9	12.6	36.1	20.4	17.3	7.4	5.2
Pooled (N=753)	0.9	12.1	34.7	20.1	17.6	7.6	7.0

$\chi^2 = 3.606$ P<0.01

Fig 3. Body Mass Index in adults

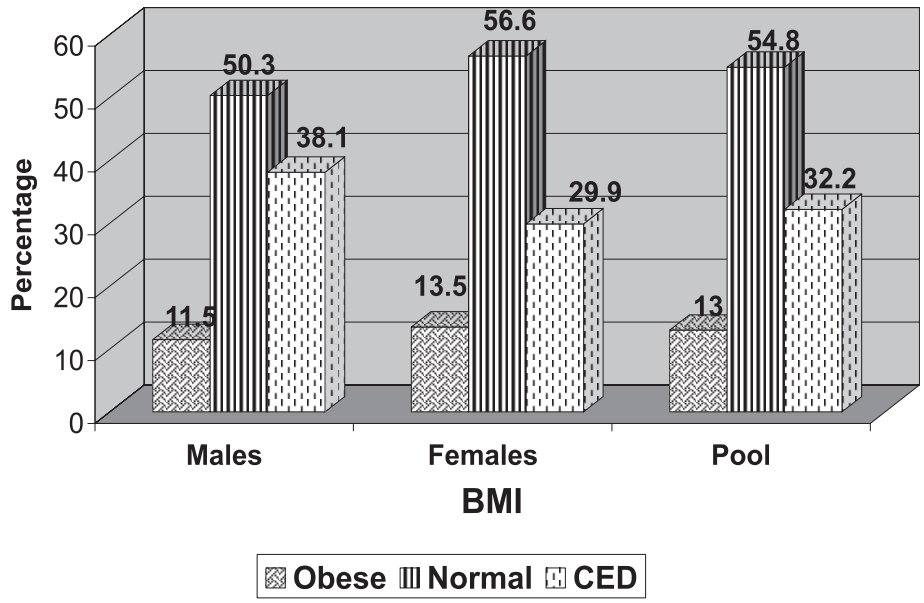


Table 9. Distribution of adults (>=18 years) according to BMI classification and Type of family

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
Nuclear (N=655)	0.9	11.9	35.3	20.2	17.6	7.3	6.9
Extended (N=90)	1.1	13.3	30.0	20.0	17.8	8.9	8.9
Joint (N=8)	0.0	12.5	37.5	12.5	25.0	12.5	0.0
Pooled (N=753)	0.9	12.	34.7	20.1	17.6	7.6	7.0

$\chi^2 = 15.65$ P>0.05

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

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
Pucca (N=31)	0.0	6.5	35.5	19.4	25.8	0.0	12.9
Kutcha (N=590)	0.9	11.0	34.6	19.8	18.6	8.3	6.8
Mixed (N=132)	1.5	18.2	34.8	21.2	11.4	6.1	6.8
Pooled (N=753)	0.9	12.1	34.7	20.1	17.6	7.6	7.0

$\chi^2 = 48.82$ P<0.01

Dietary Factors:

Dietary intake of 1319 individuals was collected from 300 households i.e. 750 adults (381 males and 369 females) and 569 children (292 males and 277 females). Consumption of food stuffs per day was observed marginally low as compared to consumption of Cereals & Millets in males and females respectively. Very low consumption of fats & oils, pulses & legumes and leafy vegetables was extremely low. In children 1-3 year's age group, average energy intake was observed to be 840.53 Kcal. In dietary intake, average energy intakes (calories) were less than RDA in all age groups of children and adults.

Fig 4. Distribution of preschoolers according to SD Classification

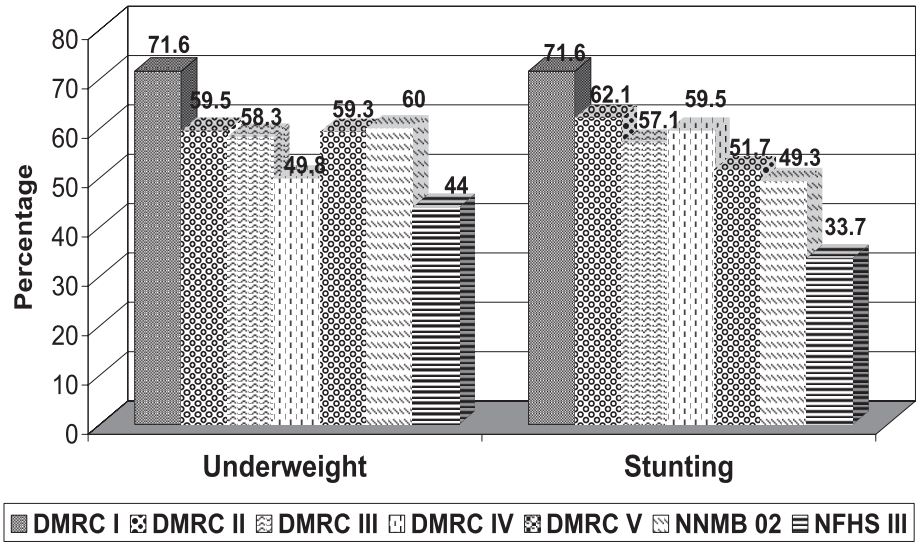
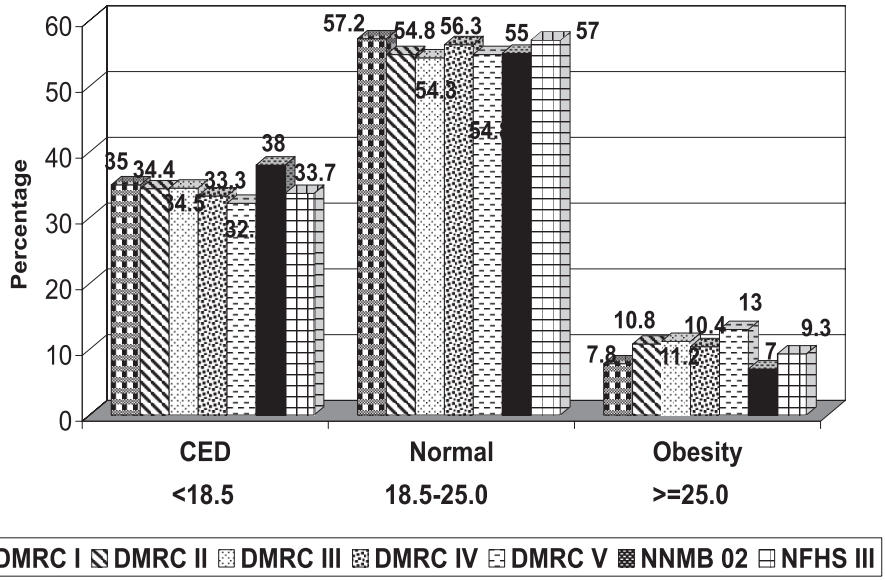


Fig 5. Distribution of Adults by BMI grades



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 along with adults and ranging from 64.7 to 97.5 percent, where as in DMRC phase I (59.0 to 99.2 %), DMRC phase II (53.5 to 85.9 %), DMRC phase III (45.8 to 98.8 %) and DMRC phase IV (37.0 to 97.4). Calorie deficit was observed more in children (15.6 to 35.3 %) than adults (2.5 to 14.8 %) and observed highest in 4-6 years group (32.3 to 35.3 %) followed by 1-3, 10-12 and 7-9 years groups. Deficiency in protein intakes was observed only in 13-15 (13.5 %) followed by 10-12 and 16-17 years age group. Diet of preschoolers (1-6 years) years age group was deficient in fats (10.8 to 34.7 %), 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 and in DMRC Phase II, diet of preschoolers and juvenile adolescents was deficient in fats, and in DMRC Phase III, diet of preschoolers (1-6 years) and adolescents (13-17) years age group was deficient in fats (17.5 to 55.1 % & 14.3 to 37.5 %), whereas in DMRC Phase IV, diet of preschoolers (4-9 years) and adolescents (13-15) years age group was deficient in fats (38.0 to 55.6 % & 5.5 to 28.2 %).

Trends revealed that there is gradual decline in stunting and underweight in preschoolers on comparing between four phases i.e. DMRC Phase I, DMRC Phase II and DMRC Phase III and DMRC Phase IV studies, whereas, slight decline in chronic energy deficiency and incline in obesity in adults was observed (Fig. 4 & 5). 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. Now NNMB surveys will

be undertaken annually in Pali district for next three years on similar sample design and similarly both the districts can be monitored annually and continuously long term at the interval of three years. Data will be collected from 600 households from 30 villages from nine tehsils of Pali district in first phase. 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

IMPORTANT LEADS/ OUTCOMES

The results of the study carried out on representative segment of the population in desert areas as well as non-desert areas would provide information and useful guidelines for food policies and nutritional programs.

3.4 Nutritional status along with morbidity and mortality of neonates and infants in Jodhpur district of Rajasthan – A community based longitudinal study

Principal Investigator: *Dr. Ranjana Fotedar, Scientists 'C'*
Co-Investigator: *Dr. Madhu B. Singh, Scientist 'E'*, *Dr. J. Lakshminarayana, Scientist 'E', Sh. M. S. Chalga, Technical Officer and Mr. Pankaj Kumar, Technical Assistant*
Commencement: September, 2007 **Duration:** Three Years **Status:** Completed
Funding: Desert Medicine Research Centre (Intramural)

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 the types of morbidity, mortality and their causes among neonates and infants

PROGRESS

In the beginning the present project proposal was revised as per the suggestions made by SAC on 19-20th April 2007, thereafter, the present project proposal was initiated. Study design and sampling design were formulated in detail for conducting the fieldwork. Schedules were structured for carrying out the field Survey. Preliminary field visits were made to study the area in order to observe the timings of the availability of the respondents and gathering information relevant to the project.

Jodhpur district has been selected for feasibility reasons. So far new registrations of 300 newborns have been completed from 28 villages namely Jhalamand, Sangaria, Tanawada, Salawas, Nandwan, Saraecha, Sar, Mogar Kalla, Feach, Bacharna, Dhundada, Hanwant Nagar, Piparali, Dedas, Luni, Vishnu Nagar, Shikarpura, Chainpura Bhatan, Karnayali, Modi, Golia Magra, Madopur, Dudia, Nai Basti, Krishna Kheda, Sangasni Gao, Nimbla, Guda Bishnoi belonging to Salawas Community Health Centre of Luni Panchayat Samiti, Jodhpur district. ANM or Anganwadi workers were contacted at village level for reporting the new birth. All 300 newborns (0-7 days) of selected 28 villages have been registered in our study. Details of baby were obtained from mother, who was the respondent for filling up schedules. All neonates and infants were examined clinically for morbidities and nutritional deficiencies. Information regarding mortality of neonates and infants along with clinical examination (such as general infection, respiratory tract infection especially pneumonia, diarrhea and nutritional marasmus) were recorded. Social-cultural / economic causes responsible for mortality were recorded from mother of each child registered in the study in pre-designed schedules.

All the newborns were 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. The feeding practices of newborn were collected for nutritional assessment. Infant feeding practices comprising of both the breast-feeding as well as complementary feeding were also recorded. They have major role in determining the nutritional status of the child.

The socio demographic/economic information along with socio-cultural/ economic causes responsible for mortality was also recorded from mother of each child registered in the study. Each registered child was followed up every month for above-mentioned clinical examination. The registration of 300 neonates (0-7 days) has been completed in July 2009 and then they were followed-up to the age of 12 months (one year)-July 2010.

WORK DONE DURING APRIL 2011 TO MARCH 2012

During this reported period, field work has been completed. The data of 300 neonates and infants collected in this project through out the year is being entered into the computer since Jan. 2011 by data entry operator / cum analyst. Analysis of their follow-ups (1 to 12 follow-ups) is under process.The observations (findings) arising from the analysis of the data collected have been given in the Tables 1 to 5. Table 1 showed that neonates belonging to nuclear families were 28.7% whereas 63.0% to joint families. 48.7% neonates were males and 52.0% females. Majority of mothers of neonates were illiterate i.e. 58.0% whereas higher education was low (0.7%). It was observed that 41.6% mother were married before the legal age of marriage i.e. less than 18 years and 37% mothers had cohabitation before 18 years of age. Regarding the antenatal care, 95% mothers received the antenatal care during pregnancy in which 92% mothers received two doses of Tetanus toxoid and 66.3% mothers received Iron Folic acid tablets.

It was observed that 98.7 percent neonates were delivered full term with normal mode of delivery (98.7%) and 49.7% neonates received timely first suckling. Majority of deliveries occurred at hospital (79.7%). Most of the deliveries were done by paramedical staff (75.7%) followed by TBA (13.3%) and UBA (3.7%). Only 17.3% neonates received BCG vaccine at the time of birth (Tables 3).

Table 3 revealed that 83.7% neonates had normal weight at the time of birth (>=2.5 Kg) whereas the percentage of low birth weight babies was 16.3%. It was observed that 10 percent neonates were sick at the time of survey. Main morbidities observed at the time of birth were fever (3.79%) followed skin infection (Boils) and umbilical infection (2.7%) and congenital abnormalities (1%) (Table 4). Neonatal mortality was 13.3 per thousand and infant mortality was 43.3 per thousand in the studied population (Tables 5).

The infant Mortality Rate was 43.3 per thousand, of whom 76.9% occurred at home and 23.1% in hospital. Main causes of mortality reported were fever (viral), premature delivery-2 (7 months Wt. 1.7 & 1.3 Kg.), stop suckling, congenital abnormality-bi- lateral cleft lip & hard palate, fever, stiffed body, convulsion, stop suckling-3 days before death, pneumonia, high fever, asthma, jaundice, diarrhea, septicemia, immunization not done.

Table 1 Distribution of neonates according to different demographic parameters

Demographic parameters		N	%
Gender of Neonates			
Males		146	48.7
Females		154	51.3
Type of family			
Nuclear		86	28.7
Extended Nuclear		25	8.3
Joint		189	63.0
Mother's Education			
Illiterate		174	58.0
Literate		31	10.2
Primary		68	22.7
Middle		20	6.7
Secondary		5	1.7
College		2	0.7
Mother's Age at marriage (years)			
<=12		27	9.0
12-15		7	2.3
15-18		91	30.3
>=18		175	58.4
Mother's Age at Cohabitation (years)			
12-15		6	2.0
15-18		105	35.0
>=18		189	63.0
Mothers- Antenatal received during last pregnancy			
Yes		285	95.0
No		15	5.0
Mothers- Tetanus Toxoid received or not			
Yes	One dose Received	13	4.3
	Two dose Received	276	92.0
No		11	3.7
Mothers- Iron F.A Tab received or not			
Yes		199	66.3
No		101	33.7

Table 2. Distribution of neonates according to delivery and immunization

Neonates	N	%
Birth terms		
Full Term	296	98.7
Pre Term	4	1.3
Mode of Delivery		
Normal	296	98.7
Caesarian	4	1.3
Place of delivery		
Home	61	20.3
Hospital	239	79.7
Delivery Agency		
UBA	11	3.7
TBA	40	13.3
Relative	6	2.0
Paramedical Staff	227	75.7
Doctors	16	5.3
BCG vaccine		
Yes	52	17.3
No	248	82.7
Timely first sucking received or not		
Yes	149	49.7
No	151	50.3

Table 3. Distribution of neonates according to weight for age

New Born (0-7 days) (N=300)	Nutritional Grades		
	Normal >=2.5 Kg	Mild < 2.5 – 2.0 Kg	Severe < 2 Kg
No. Examined	251	42	7
Percent	83.7	14.0	2.3

Table 4. Distribution of neonates according to Morbidity Profile (30 cases)

Morbidity (N = 300)	Newborns (0-7 days)	Percent
NAD	270	90.0
Fever	11	3.7
Vomiting	2	0.7
Diarrhea	1	0.3
Constipation	2	0.7
Stomach Ache (Colicky)	1	0.3
Respiratory (Common Cold & Cough)	2	0.7
Skin Infection (Boils) & Umbilical Infection	8	2.7
Congenital Abnormality (Bilateral Cleft Lip & Hard Palate) & Urine Problem	3	1.0
Ophthalmic Neonatorum	1	0.3

Table 5. Distribution of neonates according to mortalities rate

Types	Age	No.	Per 1000's
Perinatal	0-7 days	3	10/1000
Neonatal	0-28 days	4	13.3/1000
Post-neonatal (Infant)	29 – 12 months (1 year)	6	20/1000

WORK TO BE CARRIED OUT DURING 2012-2013

The field work of the project has been completed on 300 newborns registered from 28 villages belonging to Salawas CHC of Jodhpur district. The complete analysis of the follow-up (1 to 12 follow-up) is under process. The complete analysis of 300 newborns (0-7 days) will be done. After entering the data in the computer, the complete analysis and report writing of 300 newborns and infants will be done very soon during 2012-13.

EXPECTED OUTCOME

The research out put of this project will help in developing a package of 'Essential Newborn Care' (E.N.C.) which will be useful to state health department functionaries in the second phase of the study.

3.5 Nutritional status along with morbidity and mortality of under five children- a follow up study of earlier registered neonates and Infants up to 5 years

Principal Investigator: *Dr. Ranjana Fotedar, Scientist 'C'*
Co-Investigators: *Dr. Madhu B. Singh, Scientist 'E', Dr. J. Lakshminarayana, Scientist 'E', Sh. M. S. Chalga, Technical Officer and Mr. Pankaj Kumar, Technical Assistant*
Commencement: September, 2010 **Duration**: 5 years **Status:** Ongoing
Funding: Desert Medicine Research Centre (Intramural)

OBJECTIVES

1. To study profile of health and nutritional status of earlier registered infants followed up to 5 years of age group by means of anthropometry and clinical examination for nutritional deficiency signs along with feeding practices and their follow up to 5 years of age group at the interval of 6 months
2. To study the types of morbidity, mortality and their causes of earlier registered infants followed up to 5 years of age group
3. To study the time trend analysis of growth and nutrition of earlier registered infants followed up to 5 years of age group

RATIONALE

This period of childhood, especially the second year of life, is notoriously fraught with risk. The young child is "transitional" as regards to diet, immunity to infections and psychological dependence. This is a period of rapid growth with high nutrient needs, particularly of protein for swiftly increasing muscle tissue. It is the time when several meals a day are required and when foods should be easily masticable and digestible. It is at this time also that the non-immune child comes in contact with a succession, or more often accumulation of bacterial, viral and parasitic infections. Lastly, it is often the occasion for the psychological trauma that occurs as a result of the sudden separation from the mother after a prolonged period of continuous intimate contact and permissive breast-feeding frequently caused by a further pregnancy.

Certain vitamin and mineral deficiency diseases occur with varying frequency in under 5 children in different parts of the world, for example, avitaminosis A, rickets and iron-deficiency anemia. The incidence varies greatly from place to place, depending upon the local dietary and social factors. Thus, rickets in sunnier parts of the world may be related to lack of exposure to available ultraviolet light due to overcrowding in urban dwellings, as in old style walled cities or in slums, or to a deliberate sheltering of young children from the sun for various cultural reasons, e.g. to prevent their acquiring a darker complexion or to escape the "evil eye". However, the principal forms of malnutrition

seen during this transitional period are those now termed "protein calorie malnutrition of early childhood" including kwashiorkor. However, time trend analysis of growth and nutrition is still lacking from desert part of Rajasthan where conditions are very harsh, demanding a great amount of work to be done from this part of the country.

PROGRESS

This is continuation of the earlier project entitled, 'Nutritional status along with morbidity and mortality of neonates and infants in Jodhpur district. It is a follow up study on earlier registered 300 neonates & infants from 28 villages from Luni Panchayat Samiti of Jodhpur District. It will be followed up to 5 year of age group at an interval of 6 months in the above mentioned project. The study will be continued on same 300 subjects as mentioned above.

In the last SAC meeting held on 20-22nd May 2010, SAC Chairman suggested to continue the study on earlier registered 300 neonates & infants in Jodhpur district.

Thus regular survey was initiated from Sept 2010. The survey is going on. So far 300 infants have been completed at the interval of 6 months up to the age of two and half years. They were examined clinically for morbidity, mortality and nutritional deficiency signs, anthropometry (height, weight, arm circumference, chest circumference, head circumference and fat fold triceps following standard WHO techniques) and feeding practices.

WORK TO BE CARRIED OUT DURING 2012-13

The project is on going and the survey would be carried out to cover remaining earlier registered infants followed up to 5 years of age group belonging to 28 villages of Salawas CHC of Jodhpur District. Each earlier registered infant will be followed up every six month for clinical examination of nutritional deficiencies, morbidities, mortality, anthropometry, feeding practices and socio-cultural causes for mortality. Analysis and data entry is under process.

EXPECTED OUTCOME

Results of the study will be helpful in formulation of simple interventional plan for under five children for reduction of under nutrition, morbidity and mortality among under five children and their parents especially mother of under five children and reduce the health problems in the second phase of study. The Research output of this project will also help in developing package for under 5 children, which will be useful to state health department functionaries in the second phase of study.

4.1 A study of predictors of community access to primary health care in desert

Principal Investigator: *Dr. A. K. Dixit, Scientist 'E'*
Co-Investigator: *Dr. S. P. Yadav, Scientist 'E'*
Commencement: April, 2009 **Duration:** 2 Years **Status:** Completed
Funding: Desert Medicine Research Centre (Intramural)

OBJECTIVES

1. Estimation of community access to Primary Health Care
2. Determination of factors influencing the access in desert
3. Investigation into mechanism that how these factors influence the access

PROGRESS

This project of two years duration was approved by the SAC, 2009 and got started in April, 2009. We have already reported regarding curative potential of the Primary Health Centres (PHC's) in desert. This included information gathered from 55 subcentres attached to two Community Health Centres (CHCs) in typical desert district of Jaisalmer on (1) Availability of beds (2) Availability of minor surgical (3) Needed infrastructure (4) Needed equipments (5) Needed medicines (6) Out reach of ANM and (7) Image of subcentre among its users. 15 villages from this area were also surveyed with 30 H/H from each village to know more details at H/H level regarding access to primary health care. The subsequent analysis of this information is reported here.

The analysis of the information gathered from 450 H/H from 15 villages in the study area, provided coverage of 2512 persons (Av. H/H size 5.58),among which there were 1315 males and 1197 females (sex ratio among the covered ~ 52:48). Persons reported fallen sick during last 3 months (preceding the visit) were 397 (16%). The numbers of cases taken to Health Centre were only 75 (18.8%, i.e. less than 1/5th being taken to primary health care). The noted poor access to primary health care was studied for various factors, *viz.* seriousness (with which the health condition was taken in family), difficulties in taking patient to PHC, stay at PHC, physician attention received at PHC, treatment cost at PHC, availability of medicine at PHC, diagnostics done at PHC, condition of patient when discharged from PHC, referral at PHC, nursing quality provided at PHC, subjective assessment of being cured at PHC, expenditures on treatment of minor ailments at PHC and expenditures on treatment of major ailments at PHC. Distributions of these factors are presented below (Table 1).

Table 1. Distribution of persons attending PHC according to the considered factors

Factors	Factor's Category	Freq. of persons attending PHC	%
Felt Seriousness	Low felt need	67	15
	Medium felt need	327	73
	High felt need	11	2
	No response	45	10
	Total	450	100
Reaching PHC	Quite difficult	80	17.78
	Some what difficult	312	69.11
	Not difficult	13	2.67
	No response	45	10.44
	Total	450	100
Stay at PHC	Not comfortable	83	18.44
	Adequate	307	68.22
	Well Comfortable	1	0.22
	No response	59	13.12
	Total	450	100
Physician attention	Poor	210	46.67
	Satisfactory	194	43.11
	Good	1	0.22
	No Response	45	10.00
	Total	450	100
Treatment Cost	Very costly	26	5.78
	Moderate costly	369	82.00
	Low costly	10	2.22
	No Response	45	10.00
	Total	450	100
Medicine availability	Poor	271	60.22
	Satisfactory	133	29.56
	Good	1	0.22
	No Response	45	10.00
	Total	450	100

Diagnostics done at PHC	No diagnostics	122	27.11
	Partial	263	58.45
	All as required	6	1.33
	N.A	59	13.11
	Total	450	100
Condition at discharge	Not satisfactory	151	33.55
	satisfactory	240	53.34
	Good	0	0
	No Response	59	13.11
	Total	450	100
Referral by PHC	No referral	346	76.89
	Referral to DH	40	8.89
	Referral to a specialist	5	1.11
	No Response	59	13.11
	Total	450	100
Nursing quality	Poor	200	44.45
	satisfactory	203	45.11
	Good	2	0.44
	No Response	45	10.00
	Total	450	100
Subjective assessment of being cured	Not hopeful	203	45.11
	hopeful	202	44.89
	Very hopeful	0	0
	No Response	45	10.00
	Total	450	100
Expenditures on treatment of minor ailments	Not Affordable	23	5.11
	Affordable with difficulties	376	83.56
	Well Affordable	6	1.33
	No Response	45	10.00
	Total	450	100
Expenditures on treatment of major ailments	Not Affordable	222	49.33
	Affordable with debt	152	33.77
	Well Affordable	5	1.11
	No Response	71	15.79
	Total	450	100

OBJECTIVES ACHIEVED AND ANALYSIS ENVISAGED FURTHER

We could estimate the access to primary health care in desert (18.8%). We could also assess the potential of subcentres to attract its users as reported earlier. This indicated for more strengthening of the subcentres in terms of infrastructure, staff, equipments and the prompt supply of medicines. As regard to factors affecting access to primary health care prevailing at H/H level, it is revealed from the information presented here that multiple factors act in making decision to take the patient to primary health care at personal level. One could note that only about 2% had strong 'felt need' to take the patient to primary health care, for 87% of people it was difficult to approach to health care, for 68% the stay at PHC was not adequate, which was desired as going back to their places the same day in time was not possible due to distances. 47% of people were of the opinion that proper attention of the physician was not paid. More than 82% of cases, treatment expenditures were involved; though they were rated as moderate cost by the respondents. Availability of medicine at PHC was satisfactory to only 1/3rd of them. More than 1/3rd of them had to have diagnosis from out side. The condition of the patient after treatment at PHC was not satisfactory to 33% of the cases. In spite of this, even referral was not encouraged. In fact, in 77% of cases, referral was not suggested where it could have been. In case of 45% of patients brought to PHC, the nursing rendered was told to be poor. Even almost half of them were found not hopeful of being cured by PHC treatment. When, inquired about the magnitude of expenditure in taking treatment at PHC, 83% could afford it with difficulties in case of minor ailments and in case of major ailments 33% of them had to go for taking debt.

We have now information regarding factors prevailing at H/H level (reported here) and factors prevailing at health care unit (reported earlier); affecting the access to primary health care in desert. Through logistic regression, we intend now to prioritize their influence so as to suggest for appropriate intervention to improve upon the access.

5. LIST OF PAPERS PUBLISHED/ACCEPTED /PATENTS /CHAPTER IN BOOK DURING 2011-12

AWARDS RECEIVED

Centre was awarded trophy for the achievements in medical research carried-out by DMRC-ICMR in '*3rd Vision Rajasthan, 2012'*, organized by Friendz exhibitions and promotion at Birla Auditorium, Jaipur, from 15th to 17 January, 2012.

PUBLICATIONS FOR THE YEAR 2011-12

1. Anand, P.K., Book: Malaria associated factors in Thar desert of Rajasthan, India: The personal, household and environment related factors. January 5, 2011. LAP Lambert Academic Publishing, Germany.
2. Anand, P.K., Swarn, L., Yadav, S. P. and Singh, H.: Disease dynamics, distribution and surveillance of malaria in arid ecology of Jodhpur, Rajasthan, India during 2002-2006. *Journal of Public Health and Epidemiology*, 2011, **3**: 301-307.
3. Bansal, S.K., Singh, Karam V., Sharma, Sapna and Sherwani, M.R.K.: Comparative larvicidal potential of different plant parts of *Withania somnifera* against vector mosquitoes in the semi-arid region of Rajasthan. *J. Environ. Biol*., 2011, **32**: 71-75.
4. Bansal, S.K., Singh, Karam V., Sharma, Sapna and Sherwani, M.R.K.: Laboratory observations on the larvicidal efficacy of three plant species against mosquito vectors of malaria, dengue fever/dengue hemorrhagic fever (DF/DHF) and lymphatic filariasis in the semi-arid desert. *J. Environ. Biol*., 2012, **33**: 617-621.
5. Chalga M.S., Dixit A.K., Shah, Bela and Bhati A.S.: Real Time Health Informatics System for early detection and monitoring of malaria in desert district, Jaisalmer, India. *Journal of Health Informatics in Developing Countries.* 2011, **5**: 286-298.
6. Chalga M.S. and Dixit, A.K.: Development of an ICT based support system for improving health care. *International Journal on Computer Science and Engineering*. 2011; **3**: 1323-1332.
7. Chalga, M.S., Kumar Pankaj and Dixit, A. K.: A retrospective analysis of malaria cases in desert in comparison to national scenario. *Proceeding 4th International Conference Health GIS 2011*, 5-6 August at New Delhi. 2011, 106-110.
8. Chalga, M. S. and Dixit, A. K.: Development of a system for monitoring disease occurrence by networking of different ICT technologies. *Proceeding 2nd International Conference on Open Source for Computer-Aided Translational Medicine*, 22-25th February at Institute of Microbial Technology, Chandigarh, 2012, 94-96.
9. Dixit, A. K. and Ansari, F.: Prediction of individual cell frequencies in the combined 2x2 table under no confounding in stratified case-control studies. *International Journal of Mathematical Sciences*, 2011, **10**: 411-417.

10. Dixit, A. K. and Anand, P. K.: A study of household disease burden and its associated factors in a rural population of a desert district in India. *South African Journal of Epidemiology and infections,* 2011, **26**: 165-168.

11. Dixit, A. K.: Differential diagnosis: Cases with equal posteriori likelihoods. *J. Clin. Diagn. Res*., 2011, **5**: 418.

12. Joshi V, Chalga, M. S. and Angel, B.: Development of a software module for forecasting Malaria outbreak based on an equation derived from real-time parameters. *International Journal of Computer Applications*. 2011, **35**: 12-15.

13. Joshi, V., Angel, B., Purohit, A., Singh, H., Bohra, N. and Angel, A., Chauhan, R., Singhi, M. and Mathur, A.: Cases of pandemic influenza A (H1N1) 2009 in western Rajasthan, India. *Indian Journal of Medical Research,* 2012, **135**: 437-438.

14. Mathur M. L., Gaur, J., Sharma, R. and Haldiya, K. R. Antidiabetic properties of a spice plant *Nigella sativa. J. Endocrinol. Metabol*., 2011; **1**: 1-8.

15. Mohanty, S. S. Singh, Karaj. V., Footed, R., Lakshminarayana, J., Parihar, R. S.: Prevalence of duffy blood groups among the population of the desert region of India. *Journal of Rural and Tropical Public Health*, 2011, **10**: 53-56.

16. Mohanty, S. S., Singh, Karam V. and Bansal, S. K.: Changes in glucose-6- phosphate dehydrogenase activity in Indian desert malaria vector *Anopheles stephensi* during aging. *Acta Tropica*, 2012, **123**: 132-135.

17. Singh, Madhu B; Sharma, S. K.; Siri, Nair, Pandey, R. M., Kapil, Umesh and Singh, C. Status of Iodine content of salt in four regions of India. *Indian Journal of Pediatrics*, 2011, **78**: 684-687.

18. Siri, Nair, Singh, Madhu B., Sharma, S. K., Pandey, R. M. and Kapil, Umesh: A multicentric study on validation of spot testing kit. *Indian Journal of Pediatrics*, 2012 (In Press)

19. Yadav, S. P., Anand, P. K. and Singh, H.: Awareness and practices about silicosis among the sand stone quarry workers in desert ecology of Jodhpur, Rajasthan, India. *J. Human Ecol.,* 2011, **33**: 191-196.

20. Yadav, S. P. A study of social status of people with disabilities due to leprosy in desert part of Rajasthan, India. *J. Commun. Dis.*, 2011, **43**: 201-207.

21. Yadav, S. P. and Kumar, P.: A study of knowledge, treatment seeking behaviour, and socio-economic impact of malaria in desert part of Rajasthan, India. *Southern African Journal of Epidemiology and Infection*, 2012, **27**: (In Press)

6. WORKSHOPS/ CONFERENCES/ SYMPOSIA/ SCIENTIFIC MEETINGS ATTENDED BY SCIENTISTS DURING 2011-12

Dr. K. R. Haldiya, Scientist 'F'

- 'Epidemiology on identification of public health research priorities in India' on 13th August 2011 at NIE Chennai.
- ICMR-INSERM Workshop on Gene Environment Interactions, Epi-genetics Nutrition and Drugs in Diabetes held on October 16-18, 2011 in Jodhpur as a part of Centenary Celebrations of ICMR.

Dr. Vinod Joshi, Scientist 'F'

- Scientific advisory group meeting of Division of NCD on 13-14th February, 2012, at ICMR headquarters and presented work of DMRC.
- Brain storming Session on Japanese Encephalitis, Visceral leishmaniasis and cutaneous leishmaniasis in Vector Science Forum on 23.02.2012 at Institute of Pathology, New Delhi.

Dr. Karam V. Singh, Scientist 'F'

- 8th Joint Conference of ISMOCD and IAE, organized by Regional Medical Research Centre, Bhubaneswar, from 15-17th April, 2011 and presented a paper entitled, '*Natural Insecticides: their mode of action and future prospects in vector control*'.
- 23rd National Symposium on Chronobiology and Seminar on Diversity and Physiology of Desert Fauna, organized by Department of Zoology, JNV University, Jodhpur, from 1-3rd March, 2012 and presented a paper entitled, '*Succession of Anopheles species in outdoor breeding spots of Jodhpur, Rajasthan*'.

Dr. S. K. Bansal, Scientist 'F'

- 23rd National Symposium on Chronobiology and Seminar on Diversity and Physiology of desert Fauna organized by Department of Zoology, JNV University, Jodhpur from 1- 3rd March, 2012 and presented a paper entitled, *'Comparative larvicidal efficacy of Cleome viscosa L. (Family: Capparaceae) against different mosquito vectors in the semi-arid region (Rajasthan)'*

Dr. S. P. Yadav, Scientist 'E'

- 8th Joint Conference of ISMOCD and IAE, organized by Regional Medical Research Centre, Bhubaneswar, from 15-17th April, 2011 and presented a paper entitled, '*A study of awareness and health seeking practices of women about malaria who experienced of their children less than one year of age suffered with fever in the desert part of the Rajasthan, India*'.

- Tribal Health Forum meeting held at Regional Medical Research Centre for North-Eastern Region, Dibrugarh from 9-10th August 2011.

Dr. Madhu B. Singh, Scientist 'E'

- International symposium on 'Recent trends in Processing and Safety of Specialty and Operational Foods' at Defence Food Research Laboratory, Mysore from 23-25th November 2011, and delivered an invited lecture on '*Role of electrolyte supplementation on Mineral & Nutritional profile in arid situation*'.
- 8th Joint Conference of ISMOCD and IAE, organized by Regional Medical Research Centre, Bhubaneswar, from 15-17th April, 2011 and presented a paper entitled, *'Studies on the nutritional status and morbidities among rural population in desert area of Rajasthan'*.
- Organized a Conference under the project on '*Study of food and nutrient consumption pattern of Pearl millet in women of child bearing age and pre-school children, with particular reference to consumption pattern and effects of storage, processing, & cooking practices and retention of micro-nutrients like Iron, Zinc, Phytate and Polyphenols*' funded by HarvestPlus, Washington from 14-15th July, 2011 at DMRC, Jodhpur
- Organized an invited lecture of Prof. V. K. Paul, an eminent biomedical scientist on '*Malnutrition*' as a part of 'Centenary Celebrations of ICMR', on 14th July, 2011 at DMRC, Jodhpur, under the Chairmanship of Dr V. M. Katoch, Secretary, Department of Health Research & Director General, Indian Council of Medical Research.
- ICMR-INSERM Workshop on Gene Environment Interactions, Epi-genetics Nutrition and Drugs in Diabetes held on October 16-18, 2011 in Jodhpur as a part of Centenary Celebrations of ICMR.
- Projected the work of DMRC in general and Nutrition in particular through exhibits in '3rd Vision Rajasthan, 2012' organized by Friendz exhibitions and promotion at Birla Auditorium, Jaipur, from 15-17th January, 2012.

Dr. A. K. Dixit , Scientist 'E'

- 29th Annual National Conference of Indian Society for Medical Statistics at Chennai from 3-5th November, 2011 and presented a paper entitled, 'A study of behavior of OR_{MH} under stratified random sampling of controls in case- control studies.

Dr. Manju Singhi, Scientist 'C'

- workshop on frontiers on structural Bio-informatics organized by Bio-Medical Informatics Centre, AIIMS, New Delhi, from 23-25 June, 2011.
- projected the Research achievements and Public Health contribution of group on dengue & malaria in '3rd Vision Rajasthan, 2012', organized by Friendz exhibitions and promotion at Birla Auditorium, Jaipur, Rajasthan , 15-17 January, 2012.

Dr. S. S. Mohanty, Scientist 'C'

- Presented a research paper entitled, '*Polymorphisms in Duffy blood group genes of Plasmodium vivax malaria patients and control population'* in the Tribal Health Forum meeting held at Regional Medical Research Centre for North-Eastern Region, Dibrugarh from 9-10th August 2011.
- Research Methodology Workshop for ICMR Scientists at National Institute of Epidemiology, Chennai from 31st October to 11th November 2011.

Dr. Manjeet Singh Chalga, 'TO'

- 2nd International Conference on Open Source for Computer-Aided Translational Medicine by Institute of Microbial Technology, CSIR, India, from 22-25th February, 2012 at Chandigarh and presented a paper entitled, *'Development of a System for Monitoring Disease Occurrence by Networking of different ICT Technologies'*.
- 4th International Conference Health GIS 2011 organized by ESRI India from 5-6th August, 2011 at New Delhi and presented a paper entitled, '*A retrospective analysis of Malaria cases in Desert in comparison to National Scenario*'.
- 32nd Asia-Pacific Advanced Network Meeting' organized by ERNET India from 21-26th August, 2011 at New Delhi and presented a paper entitled, *'Networking of ICT Technologies for Improvement in the Health Care'*.

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14. Dr. P. K. Anand, Scientist - C
15. Dr. S. S. Mohanty, Scientist - C

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JUNIOR HINDI TRANSLATOR

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3. Smt. Chandra Kala
4. Sh. Yash Pal Singh

LOWER DIVISION CLERK

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3. Sh. Manohar Singh Seervi

HINDI TYPIST

1. Sh. Jaideep Gaur

DRIVERS

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2. Sh. Mohd. Gaffar
3. Sh. Ishwar Khetani
4. Sh. Manohar Singh
5. Sh. Rana Ram
6 Sh. Naveen Kumar

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2. Sh. Sridhar Bohra, Laboratory Attendant
3. Sh. Lal Chand Bandra, Laboratory Attendant
4. Sh. Raghu Nath Singh Bisht, Animal Attendant
5. Sh. Babu Lal Bunker, Animal Attendant
6. Sh. Mahaveer Prasad, Animal Attendant
7. Sh. Satya Prakash, Animal Attendant
8. Sh. Mahesh Chand Sharma, Attendant
9. Sh. Jodha Ram, Attendant

10. Smt Laxmi Kanta, Attendant
11. Sh. Khushal Singh, Attendant
12. Sh. Ram Lal, Peon
13. Sh. Ladhu Ram, Peon
14. Smt. Sua Devi, Sweeper
15. Smt. Soni Devi, Peon (Under Suspension)

TRAINEE

1. Sh. Bhawar Manohar Singh

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1. *Prevalence of Diabetes mellitus and impaired Glucose tolerance in the Raika & other communities with similar life style in Rajasthan- PI: Dr. Bela Shah, Director-in-Charge*

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    2. Mr. Rishi Pal, Field Investigator
    3. Ms. Suman Rathor, Lab Technician
    4. Mr. Ravinder, Lab Technician
    5. Mr. Suresh Kumar, Driver

2. *Translational Research for development and testing of ICMR-DMRC module of Dengue control for Rajasthan- PI: Dr. Vinod Joshi, Scientist 'F'*

    1. Mrs Annette angel, Field Investigator (Ph.D. Student)
    2. Mr. Ajay Vyas, Field Investigator
    3. Mr. Gajender Singh, Field Investigator
    4. Mr. Narender Vyas, Field Investigator
    5. Mr. Chandan Singh, Field Attendant

3. *Mapping of risk of dengue hemorrhagic fever (DHF) through dengue virus typing in Aedes mosquitoes in different settings of Rajasthan- PI: Dr. Vinod Joshi, Scientist 'F'*

    1. Dr. Bennet Angel, Scientist B
    2. Mr. Dhan Raj Choudhary, Insect Collector

4. *Development of molecular markers for the identification of Biological forms of Anopheles stephensi prevalent in arid areas of Rajasthan- PI: Dr. Karam V. Singh, Scientist 'F'*

    1. Mr. Robin Marwal, Junior Research Fellow
    2. Ms. Anusha Mishra, Research Assistant

5. *Study of food and nutrient consumption by women of child bearing age and 6-59 months of age, with particular reference to pearl millet consumption patterns and the effects of storage, processing, and cooking practices on the retention of Iron, Zinc, Phytate and Polyphenols in Nagaur, a desert district of Rajasthan- PI: Dr. Madhu B. Singh, Scientist 'E'*

    1. Dr. Neetu Parihar, Research Assistant
    2. Ms. Priyanka Malviya, Research Assistant
    3. Mr. Rakesh Kumar, Field Investigator

4. Mr. Sudhir Pratap, Data Entry Operator
5. Mr. Prashant, Field Attendant
6. Mr. Goverdhan, Field Attendant

6. *Assessment of Iodine deficiency disorder, Anemia and Nutrition Intervention in school age children of Jodhpur district of Rajasthan funded by Defense Food Research Laboratory, Mysore- PI: Dr. Madhu B. Singh, Scientist 'E'*

1. Ms. Suman Senacha, Research Assistant
2. Mr. Lalit Rangi, Technician

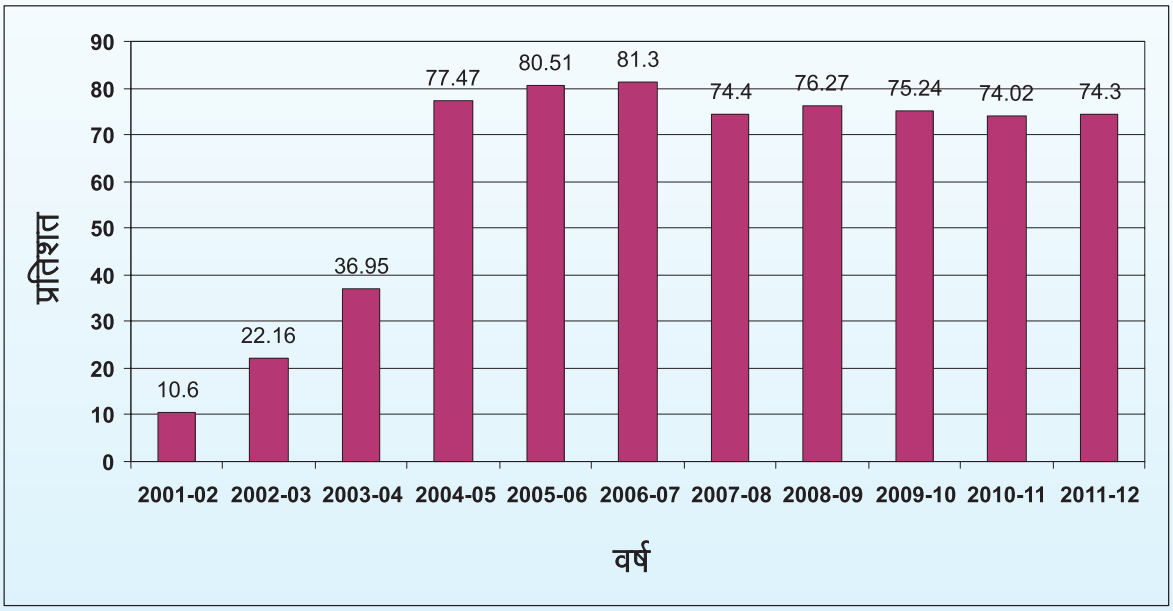
10 केन्द्र में राजभाषा को प्रोत्साहन

1. त्रैमासिक पत्रिका

द्विभाषी त्रैमासिक पत्रिका "चेतना" केन्द्र की अनुसंधानिक एवं अन्य गतिविधियों को जन–साधारण तक संप्रषित करने का सशक्त माध्यम है। साथ ही, इस पत्रिका में विभिन्न बीमारियों व उनके निवारण की उपयोगी जानकारी से हमारे पाठक लाभान्वित होते हैं। इन लेखों में केन्द्र के वैज्ञानिकों का योगदान तो होता ही है, हमारे पाठक भी अपने बहुमूल्य लेखों से पत्रिका का संवर्धन कर रहे हैं। अब तक (वर्ष 2002 से) इस पत्रिका के 37 अंक प्रकाशित हो चुके हैं। प्रतिवेदन में प्रकाषित अंकों की एक झलक :

2. हिन्दी में पत्राचार

वैज्ञानिक क्षेत्र में प्रगति करने के साथ–साथ केन्द्र ने राजभाषा हिन्दी में अधिकाधिक कार्य करने पर विशेष ध्यान दिया है। गृह मंत्रालय, राजभाषा विभाग द्वारा समय–समय पर जारी वार्षिक कार्यक्रम में निर्धारित लक्ष्यों की ओर ध्यान देते हुए केन्द्र में हिन्दी में पत्राचार के लक्ष्यों को प्राप्त करने का प्रयास किया जाता रहा है। पिछले कुछ वर्षों में प्राप्त प्रतिषतता की झलक निम्न दण्ड तालिका में दर्शाई गई है :–



3. हिन्दी सप्ताह

हिन्दी में कार्य करने के उत्साह को और अधिक बनाए रखने हेतु मरुस्थलीय आयुर्विज्ञान अनुसंधान केन्द्र में गत वर्शों की भांति इस वर्ष भी दिनांक 14 से 21 सितम्बर, 2011 तक हिन्दी सप्ताह का आयोजन किया गया। हिन्दी सप्ताह के कार्यक्रमों की शुरुआत 'हिन्दी में वैज्ञानिक व्याख्यान' प्रतियोगिता से की गई। सप्ताह के दौरान विभिन्न प्रतियोगिताओं, यथा – श्रुतिलेख प्रतियोगिता, हिन्दी टिप्पणी व पत्र लेखन प्रतियोगिता, कार्यालय शब्दावली ज्ञान, निबन्ध लेखन प्रतियोगिता, सुलेख प्रतियोगिता का आयोजन किया गया। सप्ताह का समापन प्रश्न मंच जैसे रोचक कार्यक्रम के साथ हुआ। सप्ताह के दौरान, प्रतिदिन आयोजित, विभिन्न प्रतियोगिताओं में केन्द्र के सभी वैज्ञानिकों, अधिकारियों एवं कर्मचारियों ने अति उत्साह के साथ भाग लिया, एवं पुरस्कार जीते।

4. हिन्दी कार्यशाला

केन्द्र में राजभाषा के प्रयोग को बढ़ाने के लिए नियमित रूप से हिन्दी कार्यशालाओं का आयोजन किया जाता है। दिनांक 14–21 सितम्बर, 2011 को आयोजित हिन्दी सप्ताह के दौरान दिनांक 20 सितम्बर, 2011 को हिन्दी कार्यषाला का आयोजन किया गया जिसमें केंद्रीय रुक्ष क्षेत्र अनुसंधान केन्द्र (काजरी), जोधपुर के प्रशासनिक अधिकारी श्री सुजीत कुमार सिंह को आमंत्रित किया गया। श्री सुजीत कुमार सिंह ने अपने संबोधन में कहा कि हमें हिन्दी में कार्य करने के लिए उन अंग्रेजी शब्दों को जो बहुतायत से समझे जाते है, बाधक नहीं मानना चाहिए। जैसे 'एरियर' षब्द सभी की समझ में आ जाता है। ऐसे शब्दों की हिन्दी करने में यदि हमें दिक्कत महसूस होती हो तो हम उन्हें देवनागरी लिपि में भी लिख सकते हैं। श्री सुजीत कुमार ने हिन्दी में पत्र व टिप्पणी लिखने की पद्धति पर भी प्रकाष डाला।

विशेष :

युनिकोड़ प्रशिक्षण हेतु केन्द्र का अग्रणी कदम

केन्द्र ने नगर राजभाषा कार्यान्वयन समिति (नराकास), जोधपुर के सदस्य केन्द्रीय कार्यालयों को युनिकोड़ प्रशिक्षण देने में अग्रणी कदम उठाते हुए; नराकास की एक बैठक में इस मुद्दे के उठने पर सहमति दी। तदनुसार, केन्द्र में दिनांक 14 अक्टूबर, 2 नवम्बर तथा 3 नवम्बर, 2011 को तीन अर्द्धदिवसीय युनिकोड़ प्रशिक्षण कार्यक्रमों का आयोजन किया गया।

इन प्रशिक्षण कार्यक्रमों में जोधपुर नगर के 9 विभिन्न कार्यालयों, एफ सी आई अरावली जिप्सम, जोधपुर, केन्द्रीय अन्वेशण ब्यूरो, भारतीय खाद्य निगम, भारतीय खेल प्रधिकरण, भारतीय वनस्पति सर्वेक्षण, इंडियन ऑयल कॉरपोरेशन लिमिटेड, समादेशक कार्य अभियंता (सेना), मंडल रेल प्रबंधक कार्यालय एवं मरुस्थलीय आयुर्विज्ञान अनुसंधान केन्द्र के 50 प्रतिभागियों ने प्रतिभागिता की।

केन्द्र के तकनीकी अधिकारी एवं आई. टी. प्रोफेशनल श्री मंजीत सिंह ने सभी प्रतिभागियों को युनिकोड़ पर विस्तृत जानकारी दी। इसके बाद उन्हें कम्प्यूटरों पर स्वयं प्रैक्टिस करने का अवसर दिया गया और सभी प्रतिभागियों की लिखित परीक्षा ली गई। इस परीक्षा में उनकी उपलब्धि के आधार पर प्रतिभागियों को प्रमाण पत्र भी प्रदान किए गए।

प्रशिक्षण कार्यक्रम के अंतिम दिन मंडल रेल प्रबंधक कार्यालय के राजभाषा अधिकारी एवं नगर राजभाषा कार्यान्वयन समिति के सचिव श्री रेवती लाल मीणा को आमंत्रित किया गया।

राजभाषा के प्रति वैज्ञानिकों का योगदान
पर्यावरण संरक्षण पर कार्यशाला में प्रतिभागिता

केन्द्र के वैज्ञानिक 'एफ' एवं कार्यालय प्रधान डॉ. विनोद जोशी को दिनांक 5 जून, 2011 को विश्व पर्यावरण दिवस के अवसर पर मेहरानगढ़ पर्यावरण विकास समिति, जोधपुर द्वारा आयोजित कार्यशाला में आमंत्रित किया गया।

इस अवसर पर डॉ. जोशी ने अपने विचार हिन्दी में व्यक्त

केन्द्र के वैज्ञानिक द्वारा वैज्ञानिक राजभाषा सम्मेलन में प्रतिभागिता

केन्द्र के वैज्ञानिक 'सी' डॉ. प्रवीण कुमार आनन्द ने रक्षा प्रयोगशाला, जोधपुर में दिनांक 23–24 जनवरी, 2012 को 'उन्नत सामग्री का विकास – रक्षा एवं जनसाधारण में इसकी उपयोगिता' विषय पर आयोजित राजभाषा वैज्ञानिक संगोष्ठी में भाग लिया। डॉ. आनन्द ने "तृतीयक देखभाल अस्पताल जोधपुर में भर्ती कोरीनरी धमनी की बीमारी के मरीजों के

करते हुए कहा कि पर्यावरण की रक्षा करना और उसको संतुलित बनाए रखना एक बुद्धिजीवी समाज की आकांक्षा होती है। पर्यावरण को संतुलित बनाए रखना बहुत ही बड़ा कार्य है। इन कार्यशालाओं के माध्यम से हम विचार कर सकते हैं कि हमारी पर्यावरणीय समस्याएं क्या हैं ? हम उन पर्यावरणीय समस्याओं का किस तरह समाधान कर सकते हैं ?

बीच नैदानिक प्रस्तुति–उपचार परिणाम और जोखिम कारकों का अध्ययन" विषय पर अपनी प्रस्तुति दी। इस अध्ययन में एन्जाइना ग्रूप के मरीजों और मायोकार्डियल इंफार्कषन ग्रूप के मरीजों में कोरेनरी धमनी रोग साथ रक्तचाप तथा/या मधुमेह के रूप में नैदानिक प्रस्तुति के लिए सांख्यिकी रूप से महत्वपूर्ण अंतर पाया गया है।

11. DMRC ACTIVITIES 2011-12: A PICTORIAL VIEW

Centre's participation in a meeting at Swashthya Bhawan, Jaipur, 19-04-2011. Dr. O. P. Gupta, Director, Public health invited Mr. Manjeet Chalga, DMRC, to present the data on real time health informatics management system.

Meeting of the Scientific Review Group on Medical Sociology and Biostatistics, 28-29 April, 2011 under the chairmanship of Prof. A. K. Kalla, Emeritus Prof., Deptt. Anthropology, Delhi University Delhi.

Conference organized on July 14-15, 2011 under the project on 'Food and nutrient consumption pattern with reference to pearl millet in district Nagaur (Rajasthan)' The conference was inaugurated by Dr. V. M. Katoch, Secretary, DHR and Director General, ICMR, New Delhi, as Chief Guest.

Independence Day celebration in the Centre on 15th August, 2011. Dr. K. R. Haldiya, Scientist 'F' of the Centre hoisted the National Flag.

ICMR-INSERM Workshop on Gene Environment Interactions, Epi-genetics Nutrition and Drugs in Diabetes held on October 16-18, 2011 in Jodhpur as a part of Centenary Celebrations of ICMR.

Meeting of the Scientific Review Group on 'Medical Sociology and Biostatistics' held on 20th & 21st October, 2011 under the Chairmanship of Prof. A. K. Kalla, Emeritus Prof. (UGC), Department of Anthropology, Delhi University, Delhi.

Vigilance Awareness Week was observed in DMRC from 31st October to 5th November, 2011. Dr. K. R. Haldiya, Scientist 'F' administered the oath to all the staff of the Centre.

Meeting of Scientific Review Group on 'Nutrition, Biochemistry & Non-Communicable Diseases' on 1st & 2nd December, 2011 under the Chairmanship of Dr. Kamla Krishnaswamy, Former Director, NIN, Hyderabad.

Meeting of Scientific Review Group on 'Infectious & Vector Borne Diseases' on 9th & 10th December, 2011 under the Chairmanship of Prof. R. C. Mahajan, Emeritus Professor, PGI, Chandigarh.

Meeting on January 12-13, 2012 of Directors, Divisional Heads, Administrative and Accounts Officers of ICMR Institutes/ Centres, Chaired by Dr. V. M. Katoch, Secretary, Department of Health Research and Director General, ICMR, New Delhi.

DMRC participation in "Exhibition III Vision Rajasthan 2012" at Birla Auditorium, Jaipur from 15th to 17th January, 2012, organized by M/s Friendz Exhibition & Promotion Pvt. Ltd., inaugurated by Sh. Mahesh Joshi, Hon. Minister of Health & Family Welfare, Govt. of Rajasthan. DMRC was awarded trophy for participation in the exhibition.

On completion of 'Golden 100 years of ICMR', silver coins as memento were distributed to all the employees of the Centre on 27th January, 2012. Financial Advisor Sh. Sanjeev Dutta; Senior DDG & IAS Sh. Arun Baroka; Ex-Officer-in-Charge Dr. R. C. Sharma and Director-in-Charge Dr. Bela Shah were present on this occasion.

Meeting of the Review Group on 'Medical Sociology and Biostatistics' held on 15th & 16th March, 2012 under the Chairmanship of Professor A. K. Kalla, Emeritus Prof. (UGC), Department of Anthropology, Delhi University, Delhi.

Republic Day celebration on 26th January, 2012. Dr. Bela Shah, Director-in-Charge of the Centre hoisted the national flag. Dr. R. C. Sharma, Ex-Officer-in-Charge of the Centre was also present on the occasion.

Meeting of Scientific Advisory Committee was held on 23-24th March, 2012 under the chairmanship of Prof. N. K. Mehra, Head, Department of Transplants Immunology & Immunogenetics, AIIMS, New Delhi.