

Studies on Dengue and Dengue Hemorrhagic Fever in Rajasthan, India – Vinod Joshi and Manju Singhi

Commencement: **February, 2003**

Duration: **Five Years**

Status: **Ongoing**

Objectives (Research)

1. To undertake situation analysis with respect to entomological, virological, epidemiological and serological aspects of dengue /DHF in study areas. Establishment of inter-relation among pupal density of *Aedes* species, naturally infected adult mosquitoes and cases of dengue in a given setting to develop entomological indicators of disease: Development of predictors and determinates of dengue in Rajasthan.
2. To study quantum of transovarial transmission of virus taking place in nature among generations of susceptible *Aedes* mosquitoes. Study of its epidemiological significance in retention of disease during inter epidemic periods.
3. To study mechanism of DHF in study areas in context to Halstead's hypothesis and role of cytotoxic factor and/or genomic changes: Determination of regional risk factors of DHF.

Objectives (Capacity Building)

1. Creation of a databank to provide knowledge of occurrence and distribution of *Aedes* species in representative paradigms (study areas) of Rajasthan. Development of an updating mechanism of above database in existing computer soft wares to know status of entomological and virological parameters at a given time for a given setting. Computer simulations of expected dengue prone areas, validation of the simulated results and their ultimate utilization for developing early warning system.
2. Establishment of a Regional Dengue/DHF Reference Centre in the host institute. The proposed laboratory will have its own generated data in the demonstrable form, to guide in developing a suitable control programme. In addition, necessary reference facilities will be initiated in the host institute to help health implementing authorities in developing expertise of identifying vector species and their region specific habitats.

Progress of the Work

Present project has been pursued to accomplish two major categories of objectives;

- To carry out research pertaining to the aspects crucial in Dengue control in Rajasthan.
- To develop capacity strengthening component of the host institute for undertaking research and imparting training on dengue.

All the research objectives such as development of a surveillance design for situation analysis of dengue vectors, studying quantum of transovarial transmission of dengue virus across mosquito generations and determining risk factors of DHF in Rajasthan, India have been achieved. Similarly, capacity building objectives of grant for developing specialized dengue laboratory and developing trained manpower have also been achieved.

To accomplish above, we have undertaken comprehensive survey of 20 villages and 5 towns in five physiographic regions of Rajasthan, India. These study areas were representative of all the existing eco-social paradigms of this dengue endemic, north-western, border state. Each village and urban locality has been studied thrice in the year i.e. during summer, rainy and winter seasons. The investigations undertaken have been performed as first study and then as follow up with the gap of at least 10 days between two study points. During the first year the entire area was covered while during the second year repeat studies were made to confirm the trend of observations.

Based on search of 6581 human dwellings, 37,525 human population and 62,747 domestic water containers in study settings; the following salient observations on entomological, virological and serological aspects of dengue have been made.

- In desert and semi-desert areas of Rajasthan, socio-economic criteria can serve as the basis of stratification of an area for dengue surveillance.
- Mosquito infectivity emerging through vertical passage of virus across mosquito generations infect marks the first incidence of the disease in a house.
- Presence of vertically infected mosquitoes from tree holes was observed consistently during the study period. These studies need to be confirmed further for ascertaining their possible role in causing DHF or supplementing to endemic DF.

Details of research question, work accomplished and inferences drawn

Research question 1: Through study of dengue as a system consisting of *Aedes* vectors, dengue virus and affected human population, in association with regional ecological conditions of a given setting, can we develop predictors and determinants of dengue infection for different settings of Rajasthan so as to workout surveillance design?

Work accomplished:

To address the above contention, investigations were made with respect to entomological parameters, natural infectivity of mosquito in nature and number of persons infected of dengue in a particular sampled house. Details are provided below:

A. Entomological Observations

Prevalence of Adult *Aedes aegypti* in urban and rural settings of 5 study zones across 3 seasons

Investigations were undertaken in the 5 urban settings of 5 zones. In all 500 houses during summer season, 500 in rainy season and 612 houses during winter season of the year were studied. On the other hand in the rural settings of the study zones, 1088 houses in the summer season, 1525 in rains and 2193 in winter-spring season were investigated. *Aedes aegypti* was found to be only species

available in urban areas of all the study settings, whereas in peri-urban foci *Ae. albopictus* and *Ae. vittatus* were also collected. The study Area I represented Desert Region, Area II, Hilly Region, Area III as Foot Hills Region, Area IV Forest & River Region and Area V as Saline River Region. Following observations emerged:

- During summer season, among urban settings, in Area I maximum AHI was observed as 24.0 followed by 13.0 in Area II; Area III, 15.0; Area IV, 3.0 and 6.0, in Area V. Inter-regional differences of AHI of different areas in summer season were statistically significant.
- No adult *Aedes aegypti* (AHI 0.0) was found in rural areas of Area I during summer season. In area II, 10.0; Area III, 12.5; Area IV, 18.5 and in Area V, 9.0 AHI was observed.
- During rainy season also, among urban settings, maximum AHI (20.0) was observed in Area I only followed by 6.0 in Area II, 3.0 in Area III, 5.0 in Area IV and 1.0 in Area V. Inter-regional differences of AHI were found to be statistically significant.
- No adult *Ae. aegypti* was found during rainy season in rural settings of Area I (AHI 0.0). However, in Area II, 10.3; in Area III, 14.5; in Area IV, 89.1 and in Area V, 5.5 AHI was observed.
- During winter-spring season also urban AHI was highest (25.0) in area I followed by 7.8, 23.3, 11.6 and 17.6 in Area II, III, IV and V respectively. Data indicates that among urban settings of all study areas during all the seasons, Area I had highest number of houses positive for the presence of *Aedes aegypti*.
- In rural areas, during winter-spring season AHI was observed 0.0 in area I. However, during this season highest AHI (10.1) was observed in area IV followed by area III (7.3) and area V (6.3).

Prevalence of Larval *Aedes aegypti* in urban and rural settings of 5 study zones across 3 seasons

A total number of 14,785 domestic water containers among urban settings were studied. Percentage of houses found positive for breeding has been recorded as Breeding Index (BI). Similarly, among rural settings, 25,813 domestic containers were screened in summer, 11,756 during rainy season and 10,393 containers during winter-spring season.

- During summer season, among urban settings, BI was maximum (37.0) in Area I, followed by 3.0 in Area II, 15.0 in Area III and 5.0 in Area IV and 19.0 in Area V. Among rural settings of Area I, during summer season, BI observed was 60.0. In Area II, 9.0, Area III, 94.0, Area IV, 44.0 and Area V, 19.0 BI was observed (figures 1-5).
- During rainy season, BI among urban settings of Area I was only 5.0, whereas BI was 16.0 in Area II, 14.0 in Area III, 16.0 in Area IV and 4.0 in Area V. During rainy season, in rural settings of Area I, BI was 2.5; Area II, 63.0; Area III, 23.0; Area IV, 85.0 and Area V, 97.0 BI was observed (figures 1-5).

- During winter-spring season also, among urban settings, Area I showed BI 12.0 as against 0.9 in Area II, 48.3 in Area III, 5.8 in Area IV and 43.3 in Area V (figures 1-5). In rural areas, only 0.1 BI was observed in area I Maximum BI were observed during winter-spring in area III(25.7) and area IV (25.2).

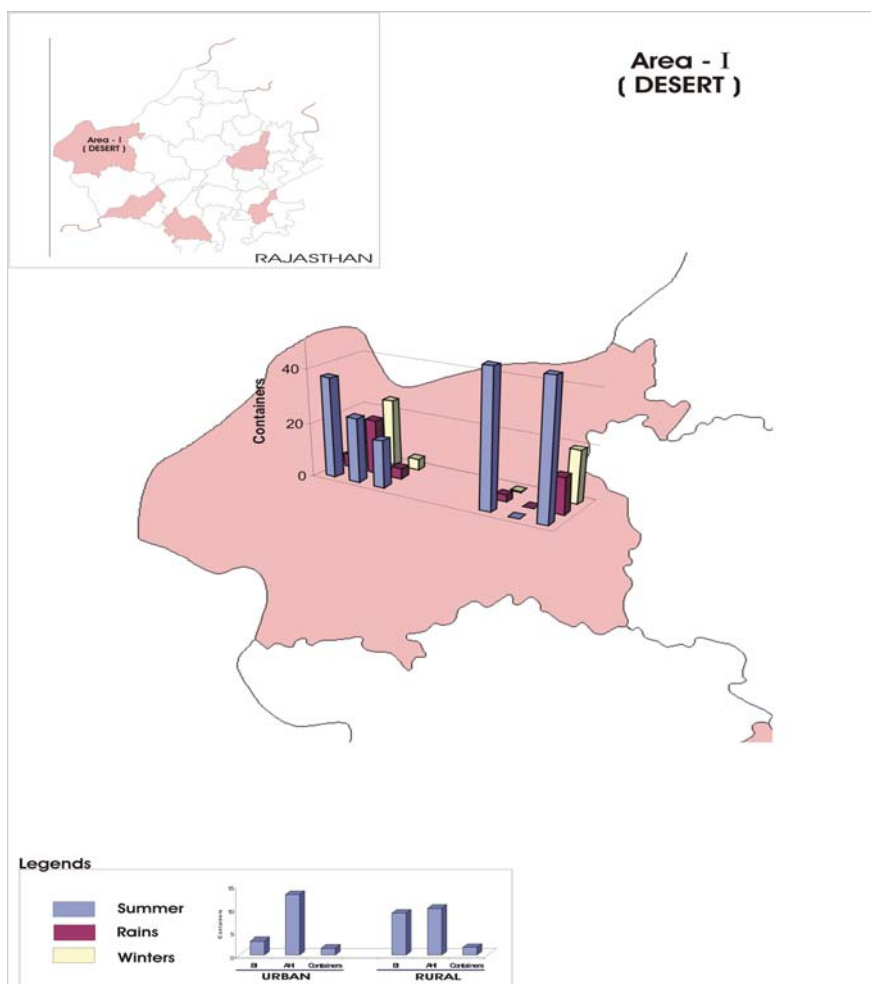


Fig. 1. AHI, & BI and containers among urban and rural settings in Area I

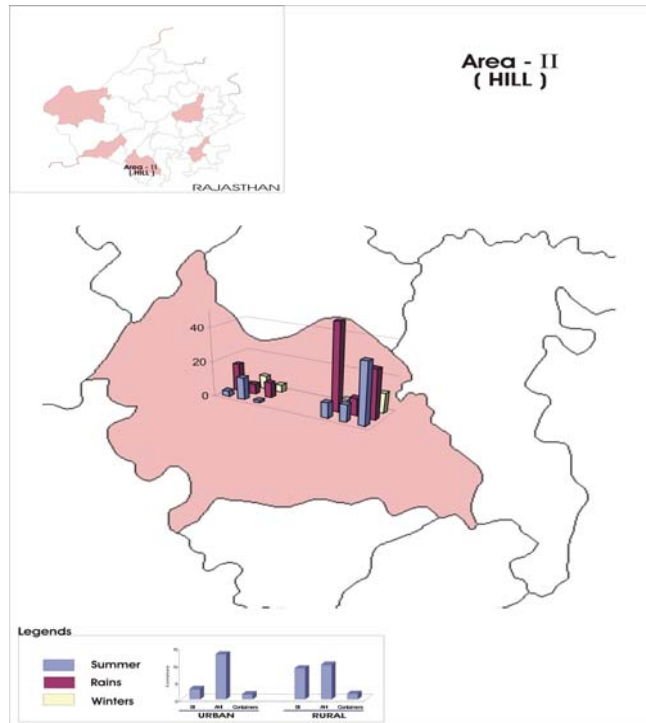


Fig. 2. AHI, BI and containers among urban and rural settings in Area II

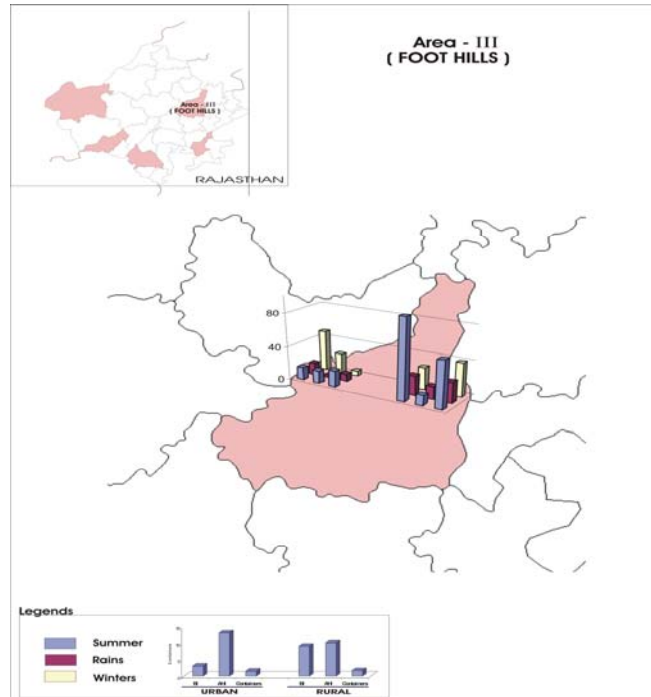


Fig. 3. AHI, BI and containers among urban and rural settings in Area III

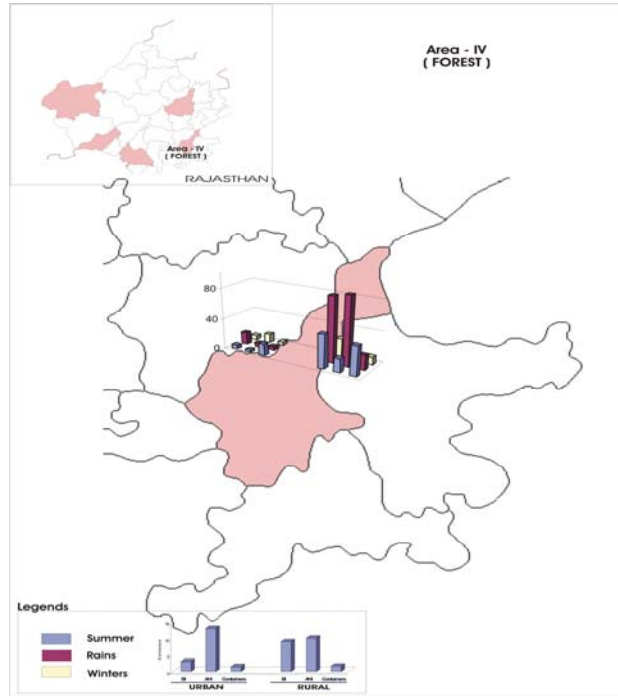


Fig. 4. AHI, BI and containers among urban and rural settings in Area IV

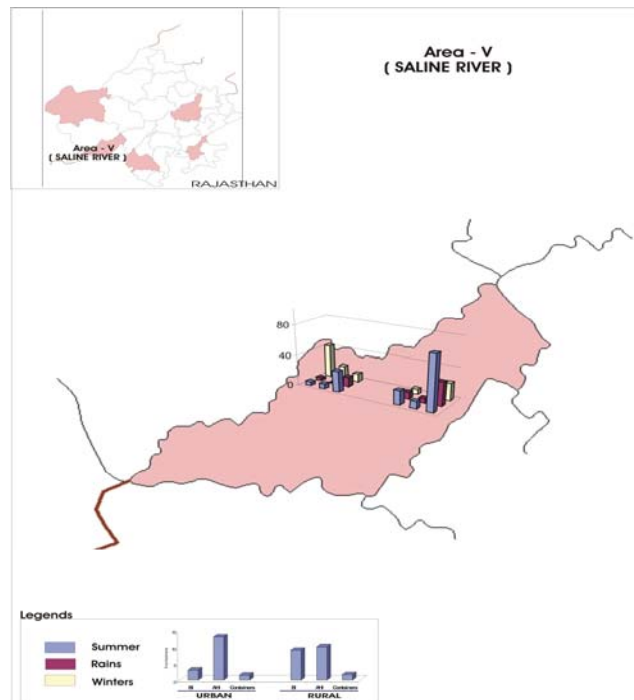


Fig. 5. AHI, BI and containers among urban and rural settings in Area V

B. Virological Observations

Virological observations undertaken in the present study pertain to investigations on natural infectivity of mosquitoes and serological detection of antibodies to dengue among suspected cases in sampled home of study settings. As the entomological studies have focused on area-wise, similar resolution for the virological investigations have also been attempted during the study. The details are given below:

Mosquito Infections

Indirect Fluorescence Antibody Test (IFAT) was performed to detect natural infection of dengue virus among field collected *Aedes aegypti*. A total number of 1133 individual mosquitoes were tested for IFAT of which 27 (2.38%) showed natural infection of dengue virus. While rural settings showed 2.7% mosquitoes infected, urban settings showed only 1.7% mosquito infectivity. Two important points emerged from the above observations.

- Natural infection of dengue virus among *Aedes aegypti* was confined to only to urban settings of Area I and rural settings of Area II & III. However, in areas IV & V both urban as well as rural settings showed infection among mosquitoes.
- Secondly inter-seasonal analysis of data suggested that winter-spring was the most preferred season of mosquito infectivity in all the study areas whereas no mosquito infections were encountered during other two seasons in areas I, II, III, & IV however, Area IV emerged to be the most consistent one for mosquito infectivity from where, infected mosquitoes were observed during all the three seasons.

Human infections

- Maximum numbers of human dengue cases (22) were observed during summer season followed by 14 cases in rainy season and nil during winter-spring season. Presence of disease during summer was maximum in the Area I (12 cases) followed by Area II (7 cases), Area III (2 cases) and least in Area V (1 case). No case of dengue fever was observed during summer season in Area IV.
- During rainy season whereas no case of dengue was observed in Area I and Area II, maximum number of cases during this season were encountered in Area IV (7 cases) followed by 6 cases in Area V and least number of cases (1 case) in area III.
- During winter-spring season none of the areas had any suspected or laboratory diagnosed case of dengue.

Table 1. Mosquito and human infections in study areas

Area	setting	City/ Village screened	Seasonal observations on mosquito & human infections											
			February-March				April-June				July-January			
			Mosquito		human		mosquito		human		mosquito		human	
			Ex.	% +ive	Case	% sero +ive	Ex.	% +ive	Case	% sero +ive	Ex.	% +ive	Ex.	% sero +ive
Area I (Arid)	Urban	1	89	2.2	0	0	50	0	22	21.5	45	0.0	0	0.0
	Rural	4	5	0.0	0	0	0	0	4	2.0	1	0.0	4	0.0
Area II (Hilly)	Urban	1	7	0.0	0	0	24	0	13	15.5	10	0.0	0	0.0
	Rural	4	31	3.2	0	0	40	0	0	0.0	69	0.0	0	0.0
Area III (Semi- arid)	Urban	1	50	0.0	0	0	26	0	7	3.0	4	0.0	6	1.5
	Rural	4	74	6.7	0	0	39	2.5	0	0.0	120	0.0	6	0.0
Area IV (Forest & River)	Urban	1	39	10.2	0	0	7	0	0	0.0	13	0.0	7	100
	Rural	4	36	0.0	0	0	72	1.3	0	0.0	87	6.8	0	0.0
Area V (Semi- arid)	Urban	1	75	2.6	0	0	7	0	1	2.1	1	0.0	13	12.7
	Rural	4	36	11.1	0	0	35	2.8	0	0.0	41	0.0	0	0.0
Total	Urban	5	442	4.0	0	0.0	300	1.0	47	2.6	331	1.8	29	1.0
	Rural	20	182	0.0	0	0.0	186	6.6	4	2.0	318	6.8	10	0.0

Ex= Examined

Inferences

- In different eco-social settings of Rajasthan, desert, semi-desert and non-desert areas need to be stratified on the different criteria as relevant for each one of them. A common surveillance strategy may not be suitable.
- The observations suggest that for urban areas, in arid and semi-arid settings, socio-economic criteria of the inhabitants emerges to be the suitable criteria for surveillance to develop status report and predict prospective risk of disease transmission. For non-desert areas such as area II & IV, ecological criteria of availability of favorable environmental temperature and relative humidity may be applied for stratification of urban settings. Representative areas with different conditions of ambient temperature and relative humidity could be adopted for undertaking surveillance.
- However, for rural areas of all the study settings (Desert as well as non-desert), many variations were not observed among different settings and our studies suggest that a uniform criterion of socio-economic nature may be used for stratification of any rural settings of Rajasthan.
- Study of key containers has revealed Cement tanks as the most preferred habitat of mosquito breeding hence vector control measures could be achieved by focusing in these habitats. A

Carefully designed strategy targeting Cement tanks may lead to commendable results of vector and virus control.

- Correlations of virological investigations among mosquitoes and human cases have highlighted different seasons for their corresponding seasonal peaks of activity.
- While Mosquito infections have emerged lowest during winter-spring season in all the desert and semi-arid areas, this period highlighted least mosquito breeding. This observation could lead to prioritization of time of controlling vector breeding.
- The observations suggest that residual mosquito population available during winter period retain extrinsic virus activity in the desert and semi-desert areas and this period, therefore, prevents interphase of disease cycle when virus contained by mosquitoes transmits to humans during forthcoming period. Observations suggest that spring season in desert and semi-arid areas, therefore, could be the best period for the effective containment of dengue virus, preventing its onset during summer season.
- On the other hand, non-desert area such as area IV and other similar settings in Rajasthan need to be targeted for extrinsic and intrinsic virus control during rainy season (Table 1).

Research question 2: Since dengue appears as a seasonal morbidity in community, during inter epidemic periods, where virus remains in nature?

Work accomplished

Indirect Fluorescence Antibody Test (IFAT) was performed to detect natural infection of dengue virus among field collected *Aedes aegypti*. A total number of 1133 individual mosquitoes were tested for IFAT. Simultaneously, active dengue cases present in sampled houses were analysed for detection of antibodies against dengue using MAC-ELISA test. Maximum number of cases (22) were observed during summer season followed by 14 cases in rainy season and no case during winter-spring season. On other hand, for presence of dengue infection among mosquitoes, winter-spring was the most preferred season of all the study areas (Table 1).

Inferences:

- Winter-Spring is the period when least mosquito breeding has been observed, where as this period had shown maximum infected mosquitoes. The data suggest that residual mosquito population available during this period represent peak of extrinsic virus activity and therefore this seasons serves as the interphase of virus cycle leading to human infections during subsequent summer season.
- Spring season in desert and semi-arid areas could be the best period for the effective containment of dengue virus.

Research question 3. What are the risk factors of DHF in Rajasthan in context of Halstead's hypothesis, in context of concept of development of cytotoxic factor and/or in context of genomic constitution of affected population?

Work accomplished

Most of the work to answer this question has been accomplished. The observations made so far indicate following important trends:

- Since most of mosquito infections have been observed during winter-spring seasons and rainy seasons in all the study settings, disease appearance regulations from mosquitoes to human, seems to be controlled by increase in infected mosquito population through vertical passage of virus as an action of increased ambient temperature during spring summer months.
- A study of virus typing from mosquito and human sera is in progress. The mosquito populations of peri-urban and urban areas have been screened to detect presence of virus.
- A study of monthly trend of natural infectivity of mosquito species *Aedes aegypti*, *Aedes albopictus* and *Aedes vittatus* as carried through vertical transmission, was undertaken in urban and peri-urban areas of Jodhpur, India from the month of December, 2005 to October, 2006. Of the total 104 individual mosquitoes from urban areas and 229 from park area examined for IFAT, infectivity of mosquitoes in urban areas was observed during months March to May and August to November. Whereas from peri-urban areas representing park, mosquito infections through vertical transmission were observed all across the year except the months of January-February and May-June.

Inferences

- More proportion of infected mosquitoes have been observed from peri-urban areas. Frequency of appearance of infected ones across different months is also more pronounced from peri-urban foci.
- The initial observations suggest a possible role of extrinsic dengue viruses observed in peri-urban foci, to cause the DHF and/or amplify DF.

Development and demonstration of a surveillance design for control of dengue vectors in Jodhpur, using GIS – Vinod Joshi, Himmat Singh, P. K. Dam, Anil Purohit and Manju Singhi, Dr. J. S. Paribar* and Dr. Y. R. Joshi**

Commencement: **October, 2006**

Duration: **Two Years**

Status: **Ongoing**

Objectives

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

Progress of Work

The work accomplished during reported period included following steps:

- 1. Selection of study areas as per the design**
- 2. Undertaking the work and extrapolation of observations**
- 3. Undertaking follow up studies for seasonal variation trends**
- 3. Reporting the existing situation of vectors and viruses of dengue**
- 4. Suggestions for the future strategy as a model programme**

1. Selection of study areas as per the design

Being predominantly a domestic breeding and feeding species, area selection for surveillance of dengue vectors was based on the socio-economic characteristics of different localities of Jodhpur. Since it has been shown through our earlier studies that such a sampling criteria would be useful, in present study the same criteria was employed to present a translation research study. Accordingly, following localities with their socio-cultural attributes were chosen (Figure 1):

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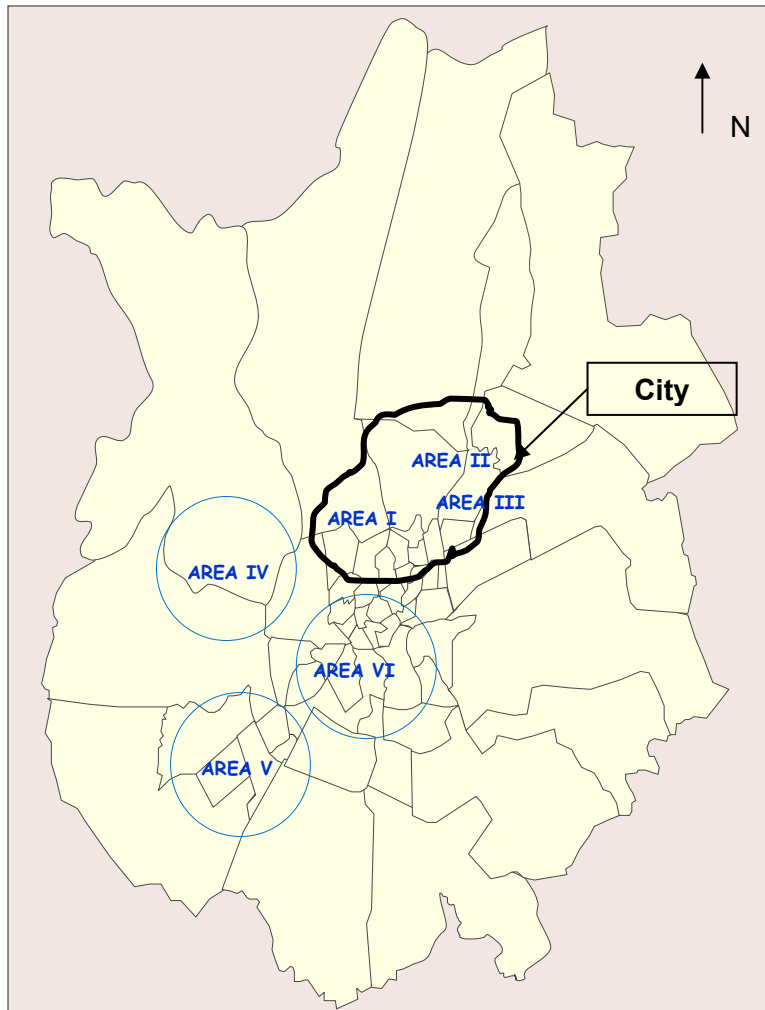


Figure 1. Study areas for dengue surveillance and their geographical location in Jodhpur

Area I represents interior most and old locality of city of Jodhpur. The houses are closely agglomerated and constructed in the way that two houses are communicated with each other through windows on the common walls between two houses. Government water supply to houses is once in two days with low pressure at 1st or 2nd floors. People maintain large number of domestic containers in houses to ensure availability of essential volume of water. In addition, owing to religious beliefs peri-domestic tanks, filled periodically by the people, are also maintained for providing drinking water to cows. Although socio-economy is of middle level but from point of view of domestic and peri-domestic water storage pattern, this area is almost similar to low socio-economic area.

Area II is similar locality as Area I from housing pattern and geographical situation standpoint. However, unlike Area I, this area does not include peri-domestic water tanks for cows. Unlike Area I, this setting involves people of low socio-economic category.

Area III also represents socio-economic status of Area I, with similar housing pattern as of Area I and Area II but unlike Area I this also does not involve peri-domestic water tanks maintained for cows and unlike Area II, it involves people of middle socio-economic group.

Area IV represents outer city location with sparse housing pattern, no peri-domestic water containers and less number of domestic containers. Although Government water supply is of same frequency, but water storage pattern is more organized in these areas in the form of overhead and underground tanks. Socio-economically the area represents middle level.

Area V presents a poor socio-economic status; housing pattern is of Area I & III only. This locality is situated in outskirts of city and is of semi-urban location. Unlike Area I this setting also does not involve peri-domestic tanks for cows. Owing to poor socio-economy, domestic water storage pattern is unorganized type indicated by large number of ad-hoc water containers.

Area VI is of outer city location, represents high socio-economic status, independent housing pattern, water storage pattern is organized with least number of containers for domestic water storage.

2. Project implementation

2-A. First Study (October-November, 2006)

1. Collection, maintenance and identification of adult and larval mosquitoes:

Adult *Aedes* mosquitoes were collected by the team of trained investigators with the help of a suction tube and torch. Adult mosquitoes were transported from field to laboratory in Barraud cages and were maintained at 25-30 °C and 70% relative humidity. Larval mosquitoes were collected from domestic and peri-domestic water tanks and were brought to laboratory for further studies. Larvae collected were maintained in house-wise samples on larval feed of dog biscuit powder. These were reared into adults under laboratory conditions of 70% relative humidity and temperature of 25-30 °C. Field collected as well as laboratory reared adult *Aedes* were identified for the species using standard taxonomic keys.

2. Detection and typing of dengue virus from field collected mosquitoes:

Head squashes of individual mosquitoes were subjected to Indirect Fluorescence Antibody Test (IFAT) using standard protocol. Mid guts of mosquitoes found positive for IFAT were dissolved in Phosphate Buffer Saline (PBS) and were reacted with monoclonal antibodies of DEN-1, DEN-2, DEN-3 and DEN-4 and processed for IFAT. Monoclonal antibodies commercially procured from Fitzgerald, USA were used. Fluorescence microscope model, BH2 RFL1 PM 10 ADS made by OLYMPUS, Japan, was used for IFAT. To confirm results of IFAT, gut samples showing positive IFAT towards a DEN type were subjected to Western blotting using same MAB primary antibodies and following standard methods. Basic Electrophoresis equipment and Blotter made by BIO RAD, USA were used.

Human sera were obtained as part samples from the Central laboratory of Dr. S. N. Medical College, Jodhpur. The samples were stored at -75 °C and were subjected to Western Blotting for typing DEN viruses in human sera. Results of mosquito and human sera were corroborated to arrive at conclusion of a particular virus type in a locality.

3. Collection of blood reports of dengue cases from hospital:

The cases as diagnosed by the central laboratory of Dr. S. N. Medical College, Jodhpur were recorded and extrapolated over the study settings to study association of entomological and virological observations with occurrence of disease.

Observations

- Associations of entomological observations with socio-economic attributes of study settings such as location at inside city and characteristics of agglomerations of human dwellings showed more AHI (Adult House Index) (Areas I, II & III) as compared to areas of locations in outskirts and having spread housing pattern. However, area V even being located in outskirt, represented similar pattern of housing as of inside city houses, and it showed higher AHI (26.0).
- Similarly breeding of mosquitoes were also more pronounced among houses of inner city locations as compared to those in the outer areas, except for Areas V which even being situated outside the city, showed BI of 92.0%.
- The observations showed that poor to middle socio-economic status, agglomeration of houses and presence of peri-domestic water containers were the attributes, which supported breeding and adult stages of *Ae. aegypti*, where as Area IV and VI without these attributes showed much less indices of *Ae. aegypti*. The present studies support the socio-economic criteria to be the appropriate basis of stratification for dengue surveillance. (Table 1).
- Virological observations made on field collected adult mosquitoes showed high percentage of naturally infectivity. Mosquito infections were observed maximum (65.0%) in Area V followed by Area I (58.3%) and the least in Area II (25.0%). In area IV no mosquito infectivity was recorded.
- Virus typing was done inoculating monoclonal antibodies in mosquito's heads and suspensions. The results of IFAT showed that while DEN-3 was the most common virus type circulating in mosquito samples of all the study areas, in Area I, in addition to DEN-2, DEN-3 strain was also detected in mosquito samples. (Table 2).
- Suspected cases of dengue (clinical symptoms) were recorded maximum in Area I (39 cases) followed by Area II (19 cases) and least cases were in Area III, IV and V. The maximum confirmed cases of dengue were in Area I (Table 2). Inner city contributed maximum (62%) of the total cases.

Table 1: Association of entomological Indices, virological and socio-economic attributes of study settings (October-November, 2006)

Study Area	Socio-economic Attributes	Houses Examined	AHI	Container Examined	%+ive Containers	Mosquito tested for IFA	No of IFA +ive Mosquito	% +ive of IFA test
Area I (Navchowkia)	Inner city unplanned agglomeration of houses, with peri-domestic water tanks	49	40.8	341	77.5	24	14	58.3
Area II (Bamba)	Inner city, unplanned, agglomeration of houses, no peri-domestic tanks	50	14.0	266	64.0	12	3	25.0
Area III (Moti-Chowk)	Inner city, non-agglomeration of houses, no peri-domestic tanks	50	40.0	286	78.0	34	18	52.9
Area IV (Chopasni Housing Board)	Outer city, planned, no agglomeration, no peri-domestic tanks	47	2.12	203	19.14	2	0	0.0
Area V (Masuria)	Outer city, unplanned agglomeration of house, peri-domestic water tanks	50	26.0	272	92.0	20	13	65.0
Area VI (Shastri Nagar)	Outer city, planned, no agglomeration, no peri-domestic tanks	52	7.69	217	26.92	9	5	55.0

2-B. Follow up Studies (January-February, 2007)

A follow up study of same houses, which were screened during September-November, 2006, was made during January-February and relative observations are shown in Table 2.

Table 2: Association of entomological Indices, virological and socio-economic attributes of study settings (January - February, 2007)

Study Area	Houses Examined	AHI	Container Examined	BI	Mosquito tested for IFA	No of IFA +ive Mosquito	% +ive of IFA test
Area I	49	2.0	127	6.1	1	0	0
Area II	50	0.0	164	12.0	0	0	0
Area III	50	2.0	241	28.0	1	0	0
Area IV	47	0.0	197	8.5	0	0	0
Area V	50	8.0	196	8.0	4	0	0
Area VI	52	1.9	146	5.7	1	0	0

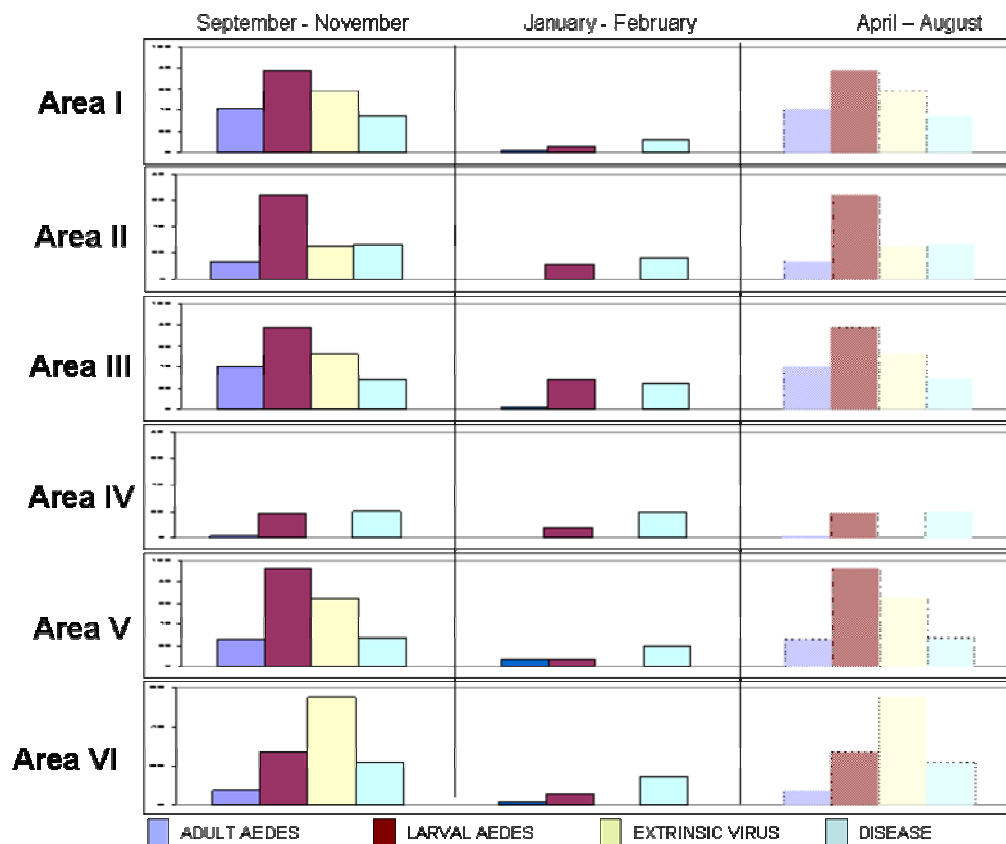


Figure 2. Observed Amplification, Reduction and anticipated re-emergence of mosquito-virus-disease nexus of dengue in different settings across different seasons

Observations

Follow up studies were made in months of January-February screening same cohort of houses which were surveyed during October to November, 2006. Following observations were made:

- Adult House Indices (AHI) of *Ae. aegypti* were reduced from a range of 2-40% (Table 1) of post rainy season (October-November) to as low as 0-2% during winter season (January-February) (Table 2, Figure 2).
- Percentage of premises positive for *Aedes* breeding, which ranged from 19-92% during post rainy season (Table 1) were reduced to a range of 4-12% during winter season. Key breeding containers were cement tanks.
- Along with reduced positivity of houses for presence of adult *Ae. aegypti*, their natural infectivity by dengue virus also showed a reduction from 65% infectivity in October- November, 2006 to 0 % during January- February, 2007. (Table 1-2, Figure 2).

- Present studies revealed the important epidemiological clues that decreasing trend of vector-virus and disease combination from post rainy season October to winter in disease affected areas of Jodhpur, may be utilized as appropriate period for intervention to prevent rebuilding of disease system in the forthcoming season.

Work remains to be done

An intervention programme is likely to be launched in collaboration with local Municipal Corporation. The physical elimination of larval forms as well as bio larvicidal application of *Calotropis procera* will be attempted in one selected area during the month of March-April to prevent disease onset in forthcoming season.

Development of molecular and genetic markers of virus transmission competence of dengue vector species in Rajasthan- *Vinod Joshi and Manju Singhi*

Commencement: **April, 2005**

Duration: **Three Years**

Status: **Ongoing**

Objectives

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

Rationale

Dengue fever associated with Dengue Hemorrhagic Fever (DHF) is an emerging public health problem in many parts of India including north western state, Rajasthan. At present, presence of species such as *Aedes aegypti* and *Aedes albopictus* and their abundance in a particular area, are being used as transmission risk factors. However, owing to wide biological variations among mosquitoes in different settings, vector biological indices do not appear to be accurate and dependable markers of risk of transmission of disease in an endemic setting. Through available biotechnological methods, proposed project envisages to develop molecular markers of transmission potential of dengue vectors species viz; *Aedes aegypti* and *Ae. albopictus* in different ecotypes of Rajasthan. The protein responsible for imparting virus transmission/refractory competence to available mosquito species will be implicated and areas wherever such proteins are reported in the state, will be marked as transmission risk zones using Geographical Information System (GIS). In addition, the gene controlling appearance of these proteins in wild caught mosquitoes will also be studied for frequency of its occurrence across generations of mosquitoes to predict seasonality of dengue onset in different ecotypes of Rajasthan. The study ultimately intends to understand molecular epidemiology of DF & DHF in Rajasthan.

Progress of the work

The present study is aimed to study molecular diversity of mid gut proteins of *Aedes aegypti*, *Aedes albopictus* and *Aedes vittatus* mosquitoes with reference to their competence to transmit dengue viruses in a particular area. The species have been collected during three seasons namely Rainy, Winter and Summer seasons. So far, the adult and larval *Aedes* mosquitoes have been collected from rural and urban settings of areas viz; Jodhpur, Kota, Jaipur, Udaipur and Bharatpur. Unfed adult mosquitoes were fed on fowls in the laboratory and fed specimens along with blood fed specimens collected from the field were caged for egg laying. Larvae/pupae collected from the field were reared in laboratory into their adult forms. F₁ generation of field caught and laboratory-reared adults were

dissected in normal saline and the mid guts of individual mosquitoes were taken out and processed for further studies employing SDS-PAGE. On other hand, head portion of each individual mosquito was processed for the examination of presence of dengue antigen using IFAT. Details of tests performed and interpretations made are mentioned below:

SDS Poly Acrylamide Gel Electrophoresis (PAGE)

A total number of 788 field collected and laboratory reared *Aedes* mosquitoes were dissected in normal saline and were subjected to SDS PAGE for their protein assay. Proteins bands were studied. Species wise details of assayed mosquitoes is given below:

Extrinsic replication of dengue virus and present work

The thinking behind present work is that whether membrane proteins of inner wall, lining of lumen of vector mosquitoes can serve as the molecular markers of vector competence of a particular mosquito species to transmit or refract dengue viruses. Since in the replication of dengue virus, the virus molecule along with its envelope enters mid gut cells of mosquitoes, the membrane proteins forming inner wall of mid gut are bound to play the crucial role for internalization of virus particle into the cells. To study this contention, mid guts of field collected and laboratory reared adult *Aedes* mosquitoes have been subjected to SDS PAGE to resolve their proteins. Presence of specific mid gut protein has been studied with simultaneous enquiry into head squashes of assayed mosquitoes and presence or absence of virus in the head squashes (using IFAT). Susceptible or refractive status of specimens was recorded respectively. Details of investigations have been provided below:

Proteomics

To establish possible relation between mosquito mid gut membrane proteins and their vector competence for dengue viruses, assays of mid guts of three species viz; *Ae. aegypti*, *Ae. albopictus* and *Ae. vittatus* was done. Electrophoresis using SDS PAGE standard protocols has been followed to study molecular diversity of mid gut proteins of these species in urban and rural settings of five study areas namely Jodhpur, Bharatpur, Jaipur, Kota and Udaipur districts of Rajasthan.

Since our earlier studies have shown that a non-transmitting or refractory species i.e., *Ae. vittatus*, invariably contains 200 kDa proteins and that transmitting species *Ae. aegypti* and maintaining species *Ae. albopictus* contains occasional presence of this protein, in present studies we have focused on studying prevalence of 200 kDa protein in midgut tissue samples of individual mosquitoes collected from field of above study settings. Numeric details are shown in Table 1 whereas figure 1 shows the protein assays of mid guts of different species with reference to marker bands as reference protein.

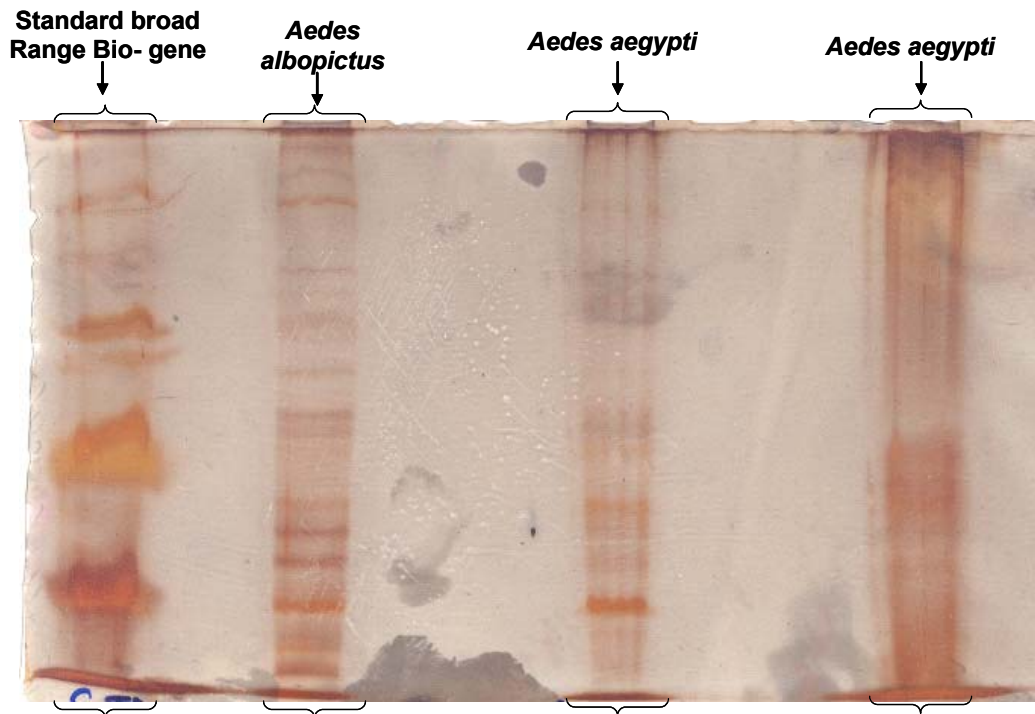


Figure 1. SDS PAGE assays of proteins of mid guts of *Ae. aegypti* and *Ae. albopictus*

Virus detection from mosquitoes assayed for proteins

It was perceived that species such as *Ae. aegypti* which is a proven vector for dengue viruses, if contains virus, would facilitate successful multiplication of virus within its body so that virus ingested and penetrated into mid gut cells reaches to salivary glands making the host infective. In such case if a mosquito is infected, it's head squash will show presence of virus. To study this association, field collected mosquitoes were first subjected for their mid gut assay for SDS PAGE and head squashes of same specimen was examined using IFAT (Indirect Fluorescence Antibody Test).

Interpretation of Experimental results

Table 1 shows association of percentage of mosquitoes showing presence of 200 kDa protein with number of them showing presence of virus in IFAT. Similarly, Table 1 also shows percentage of mosquito species not showing presence of 200 kDa protein and number out of these pools showing presence of virus.

Table 1. Area-wise details of association of mid gut 200 kDa protein with IFAT

URBAN AREAS							RURAL AREAS				
Area	Species	Total no. assayed	% with 200 kDa protein	IFA T +ve	% Without 200 kDa protein	IFAT +ve	Total no. assayed	% with 200 kDa protein	IFAT +ve	% without 200 kDa protein	IFAT +ve
Jodhpur	<i>Ae.aegypti</i>	221	54.76	3	45.23	44	2	0	0	100	0
	<i>Ae.vittatus</i>	28	100	0	0.0	0	41	44.82	0	55.17	8
	<i>Ae.albopictus</i>	129	35.0	5	65	21	0	0.0	0	-	-
Kota	<i>Ae.aegypti</i>	97	68.42	0	31.57	7	95	76.47	0	23.52	4
	<i>Ae.vittatus</i>	ND	-	-	-	-	-	-	-	-	-
	<i>Ae.albopictus</i>	ND	-	-	-	-	-	-	-	-	-
Jaipur	<i>Ae.aegypti</i>	45	4.76	0	95.23	1	66	100	4	-	8
	<i>Ae.vittatus</i>	ND	-	-	-	-	1	-	-	-	-
	<i>Ae.albopictus</i>	ND	-	-	-	-	-	-	-	-	-
Udaipur	<i>Ae.aegypti</i>	14	0.0	-	100	2	10	0	-	-	1
	<i>Ae.vittatus</i>	ND	-	-	-	-	-	-	-	-	-
	<i>Ae.albopictus</i>	ND	-	-	-	-	-	-	-	-	-
Bharatpur	<i>Ae.aegypti</i>	39	100	0	-	-	0	0	-	-	-
	<i>Ae.vittatus</i>	ND	-	-	-	-	-	-	-	-	-
	<i>Ae.albopictus</i>	ND	-	-	-	-	-	-	-	-	-
Total	<i>Ae. aegypti</i>	416	53.28	3	46.71	54	173	80.3	4	19.69	13
	<i>Ae. vittatus</i>	28	100	0	0	0	42	44.82	0	55.17	8
	<i>Ae. albopictus</i>	129	35.0	5	65.0	21	0	0	0	0	0

Observations and Inferences

The work undertaken so far has led to following observations and inferences thereon:

Mid gut Proteins versus mosquito infectivity - *Aedes aegypti* in Urban areas

- Of total 416 *Ae. aegypti* assayed in urban areas of five districts, 53.28% have shown presence of 200 kDa protein in their mid guts.

- The percentage of presence of this protein was maximum, 100 % in samples of Bharatpur district, followed by 68.4% in samples of Kota district, 54.7 % in Jodhpur district and least 4.7% in Jaipur district. As our studies are addressing the contention that presence of 200 kDa protein is inversely proportional to competence of species to transmit dengue viruses, present molecular epidemiological observations suggest that fauna of Jaipur district could be most susceptible for dengue transmission, followed by Jodhpur and Kota districts. Those of Bharatpur are least susceptible.
- Enquiry of presence of virus in mosquito head squashes of specimens showing presence of 200 kDa protein confirmed the above inverse association of this protein with presence of virus. We observed no mosquito infected from 39 individual mosquitoes showing presence of 200 kDa from Bharatpur district and also no mosquito infectivity among sample of 97 mosquitoes from urban areas of Kota, which showed 68.4 % of them with presence of 200 kDa. On the other hand from same area i.e., Kota, a lot of 31.5% mosquitoes showing absence of 200 kDa, 7 of them were +ve for IFAT. Similarly, in sample of Jodhpur district where 54.7 % of 221 mosquitoes assayed showed presence of 200 kDa, only 3 mosquitoes were IFAT +ve, whereas 45% of them not displaying 200 kDa showed as many as 44 mosquitoes +ve for virus test. However, Jaipur district sample of 45 mosquitoes, only 4.7 % of which showed the presence of 200 kDa should have shown more mosquitoes +ve for IFAT, but none of them were +ve, may be because they did not carry any virus.

Mid gut Proteins versus mosquito infectivity - *Aedes aegypti* in Rural areas

- Among rural settings of all the study areas, total number of 173 *Ae. aegypti* have been assayed for SDS PAGE of which 80.30 % have shown presence of 200 kDa protein.
- Samples of Jaipur district which showed only 4.7 % presence of 200 kDa in their urban areas, showed a reverse picture in their samples of rural areas. In these settings all, 100 % of the mosquitoes showed presence of 200 kDa. From this pool 4 mosquitoes showed IFAT +ve.
- In sample of 95 *Ae. aegypti* from the rural areas of Kota district, 76.4% assayed showed presence of 200 kDa and none of the mosquitoes from this lot was found +ve for the presence of virus. On the other hand, 23.5% of mosquitoes which showed absence of 200 kDa, 4 mosquitoes from this lot were IFAT positive.

Mid gut Proteins versus mosquito infectivity - *Aedes albopictus* in Urban areas

- *Aedes albopictus* mosquitoes were collected from tree holes as among urban containers this species was not encountered. So far the samples from urban areas of Jodhpur district have been collected. Of 129 samples assayed from urban areas of Jodhpur district, 35.0% showed presence of 200 kDa protein. Only 5 mosquitoes out of this lot showed IFAT +ve. On the other hand, 65.0 % of these mosquitoes showed absence of 200 kDa and out of this lot, as many as 21 mosquitoes showed presence of virus. It appears that tree hole breeding *Aedes*

albopictus are retaining high proportion of natural infectivity to play the role of a maintenance species in desert settings of Jodhpur. Association of absence of 200 kDa with their infectivity has been well observed in the present studies.

Mid gut Proteins versus mosquito infectivity - *Aedes vittatus* in Urban and rural areas

- Of the 28 *Ae. Vittatus* assayed for SDS PAGE in urban areas of Jodhpur district, 100 % of them showed presence of 200 kDa protein. None of these samples showed any +ve IFAT.
- Among rural settings of Jodhpur district, 41 *Aedes vittatus* assayed for proteins, only 44.8% of them showed presence of 200 kDa and none of them was +ve for IFAT. However, 55.1 % of this sample did not show presence of 200 kDa and out of this lot as many as 8 mosquitoes showed presence of dengue virus.

Leads achieved

The present work is ongoing one and observations reported are of interim nature. Nevertheless, data generated so far highlights following molecular epidemiological leads in molecular epidemiology of transmission of dengue:

- Presence or absence of a particular species may be an index to evaluate broad idea of transmission risk of dengue in a setting. However, even presence of refractory species such as *Ae. vittatus* may not deny the risk of transmission nor presence of vector species such as *Ae. Aegypti* is necessarily be indicator of transmission risk of dengue. It appears that molecular capability of an individual mosquito to allow or disallow virus replication within itself irrespective of what species it is, is required to be known to develop markers of transmission vulnerability of an area for dengue. Our observations suggests that *Ae. aegypti*, a known vector could be non vector for possessing molecular specificity of a non vector and that *Ae. vittatus*, a non vector, could be vector with molecular virtue of a vector.
- It has been observed consistently that non-vector or refractory *Aedes* species such as *Aedes vittatus* have shown presence of a High Molecular weight Protein in the mid-guts of 23 individual mosquitoes assayed for electrophoresis. Since mid-gut proteins are being pursued as markers of vector competence by few workers, our present finding initiates a new direction that in addition to study of vector competence of mosquitoes in relation to the presence of certain proteins, we may also enquire whether absence of a particular protein makes a species vector.
- Molecular epidemiological studies mentioned in present report, are sufficient only to highlight epidemiological associations between mosquito mid gut protein and virus presence. However, cause and effect relationship between a midgut protein and vector competence needs further in-vitro studies to prove the contention.
- On confirmation, the studies may lead to development of 200 kDa as the marker of virus refractiveness and its absence as marker of vector competence.

Work remains to be done

With initial leads of association of 200kDa midgut protein with naturally infected mosquitoes, issue of development of molecular marker of vector competence is becoming worth pursuing. We propose to carry out in-vivo studies of oral feed of virus to different mosquito species and study presence or absence of virus in their head squashes. We also envisage to undertake in-vitro studies on presence or absence of 200kDa in cell lines and then studying internalization of dengue virus employing IFAT.

Study of role of zoonotic cycle of dengue virus in maintaining / amplifying endemic DF and causing DHF in different settings of Rajasthan, India - *Vinod Joshi and Manju Singhi*

Commencement: **December, 2005**

Duration: **Two Years**

Status: **Ongoing**

Objectives

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

Rationale

Dengue Hemorrhagic Fever (DHF) is a major public health problem in the countries of south-east Asia. According to existing theories, DHF is the stage believed to be the result of failure of immune response of a human host towards a new dengue serotype when responding host is already immune against another dengue strain to which it was exposed previously (Halstead, 2002). At present it is being believed that active dengue cases infected of a strain different than previous strain to which a population is already exposed, are the sources of introducing second strain to cause DHF.

Dengue viruses are the members of flaviviruses which are highly antigenic. Owing to strong antigenicity, net viremia available in human blood is around 10^5 /ml of blood only. We hypothesize that with this stock of virus which is a challenged stock by immune system of humans, an infected human may not be able to provide sufficient unchallenged dengue antigen to a previously exposed case of DF to cause DHF. We believe that to cause DHF humans may be a dead end as they are in the case of another arbo-virus Japanese encephalitis. It is perhaps for this reason that in Delhi appearance of DEN-2 in 1996 and now appearance of DEN-4 in 2006 have not led to an epidemic of DHF. In Africa also with a gap of 10 years two strains of dengue infected the population, no DHF could be reported. We believe that for providing viraemically strong virus stock, a mosquito passaged virus in peri-urban foci, can only cause DHF. We thus need to type and sequence the virus of DHF patients and match sequences with available peri-urban viruses within mosquitoes to prove this hypothesis.

In support of above hypothesis it is mentioned here that dengue viruses were primarily the viruses of mosquitoes before they adapted to lower primates. We believe that crude and undisturbed ecological niches of peri-urban or sylvatic foci of dengue maintained within mosquitoes of zoos/parks or within larval mosquito pools breeding in permanent and protected reservoirs in city setup do not get weakened titre for less feeding opportunity available with the tree hole breeders to

feed upon challenging systems of humans. If strains contained by mosquitoes of peri-urban foci happens to be different than urban strain, mixing of two strains may straight be causal for the DHF. In such case, proposed research will pave a very effective prevention strategy against DHF. To pursue this contention, present studies have been pursued. The results are expected to bring the global impact on mechanism of Dengue Hemorrhagic Fever and its subsequent strategy.

Progress of the work

Study areas

Present understanding of etiology of Dengue Hemorrhagic Fever (DHF) is based on cross reactivity of two dengue strains appearing in a common host with a gap of few months/years of infectivity from each other. Keeping this in view, we have presumed that endemic dengue has a uniform infectivity by one DEN strain and that zoonotic or sylvatic foci of virus between tree hole breeding *Aedes albopictus* and monkeys or through vertically transmitted virus within generations of *Aedes albopictus*, introduce another DEN strain to endemic population. To study above concept we have selected following two areas:

1. Dengue endemic localities of Jodhpur town
2. Peri-urban or sylvatic foci of dengue in Mandore garden

Mosquito rearing and Virus isolations

Field work pertaining to collection of breeding stages of *Aedes* from household containers and tree holes of peri-urban foci has been made from April 2006 till December, 2006 (Table 1). Virus isolation attempts were made using Indirect Fluorescence Antibody Test (IFAT).

Table 1. Details of virus detection from *Aedes* mosquitoes from urban and peri-urban settings of Jodhpur

Month	Urban Settings					Peri-urban Settings								
	<i>Ae. aegypti</i>			<i>Ae. albopictus</i>	<i>Ae. vittatus</i>	<i>Ae. aegypti</i>			<i>Ae. albopictus</i>			<i>Ae. vittatus</i>		
	Ex	+ve	%			Ex	+ve	%	Ex	+ve	%	Ex	+ve	%
April	x	x	x	x	x	x	x	x	44	4	9.0	x	x	x
May	11	2	18.0	x	x	x	x	x	0	0	0.0	x	x	x
June	X	x	x	x	x	x	x	x	9	0	0.0	x	x	x
July	12	+	+	x	x	1	0	0.0	18	+	+	4	0	0.0
August	3	3	100	x	x	23	2	8.6	27	0	0.0	47	0	0.0
September	4	3	74.0	x	x	x	x	x	6	2	33.3	x	x	x
October	29	11	37.9	x	x	x	x	x	x	x	x	x	x	x
November	X	x	x	x	x	x	x	x	x	x	x	x	x	x

x = Not done + = Virus positive

Table 2. Distribution of DF cases in Jodhpur city as per hospital records in months of October, 2006

Study area	Suspected Cases	Cases Positive	Percent Positive
Navchoukiya*	49	39	79.1
Bamba	34	19	55.8
Moti Chowk	23	9	39.1
Chopasni Housing Board	40	14	35.0
Masuria	36	17	47.2
Sharstri Nagar	48	16	41.6
Total	230	114	49.5

* Area of occurrence of DHF case

Table 3. Cases of DF & DHF and virus typing studies in mosquito samples of study areas during October, 2006

Study area	Positive cases	Virus typing in mosquito samples			
		DEN-1	DEN-2	DEN-3	DEN-4
Navchoukiya*	39	x	+	+	x
Bamba	19	x	x	+	x
Moti Chowk	9	x	x	+	x
Chopasni Housing Board	14	x	x	+	x
Masuria	17	x	x	+	x
Sharstri Nagar	16	x	x	+	x
Total	114	x	+	+	x

* Area of occurrence of DHF case, x = Not done + = Virus positive

Observations & Inferences

- Observations as depicted in Table. 1 highlights that peri-urban foci of mosquitoes present three *Aedes species* viz; *Ae. aegypti*, *Ae. albopictus* and *Ae. vittatus*, whereas in urban areas, only *Ae. aegypti* has been encountered.
- Virus detection in both the area have shown that in peri-urban foci, virus activities within mosquitoes have been observed during April, 2006, where as in urban areas, commencement of extrinsic virus activities have been observed during the month of May, 2006. Data suggest that extrinsic virus activities are preceded in peri-urban foci over its commencement within mosquitoes of urban areas.
- During months of July-September, both urban and peri-urban foci have shown maximization of mosquito contained virus activities. Months of August to September, 2006 have shown built up

of mosquito virus activities in urban as well as peri-urban foci (Table 1) resulting to outbreak of human infection (Table 2) during subsequent month of October, 2006 in the town.

- Data generated so far supports the contention that peri-urban virus activities in tree hole breeders supplement urban activities of virus among mosquitoes and humans.
- Virus typing investigations indicate DEN-3 to be the most widespread virus strain in all the localities of Jodhpur.
- Study further highlights an important observation that in the area of occurrence of DHF, mosquito fauna have shown DEN-2 and DEN-3 virus types. The investigations provide initial lead that mosquito contained virus having different strain than circulating one, appear to be causal for DHF.
- Serum sample of DHF case was subjected to SDS-PAGE, gel was blotted on nitro cellulose membrane and titrated with monoclonal antibodies raised against DEN-1, DEN-2, DEN-3 and DEN-4 as the primary antibodies. Results of Western Blotting showed presence of DEN-2 band in the serum sample. The results provide initial lead that DEN-2 strain is the viral strain causal for DHF in the study areas and that this strain DEN-2 appears to be introduced from mosquito stocks containing DEN-2, as no other human case of DHF was observed from study setting.
- Further work is in progress to locate source of mosquitoes containing DEN-2 virus from peri-urban foci of garden.

Work remains to be done

We are pursuing typing of virus among tree hole breeding *Aedes albopictus*, subsequently we propose to sequence the genome of viral strain causing DHF in humans and match it with genome of *Aedes albopictus* contained virus.

Confirmation of “Introduction, Transmission and Aggravation” conceptuality of Desert Malaria: Verification of research undertaken and softwares developed – Vinod Joshi, M. S. Chalga, Himmat Singh

Commencement: **December, 2005**

Duration: **Two Years**

Status: **Ongoing**

Objectives

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

Rationale

Under influence of desert ecological condition of Rajasthan Malaria has been reduced to its seasonal existence during post monsoonal period of August-October. The work undertaken so far by our group has shown that in desert settings of Rajasthan, Malaria appears every year afresh, it completes its one or two transmission cycles with the help of local vector fauna and then disappears. In certain pocket where more imported cases of malaria and high vector density combines, outbreaks of disease are resulted. The research undertaken in 26 villages of irrigated, semi-irrigated and non-irrigated villages of Jaisalmer and Jodhpur districts had shown that malaria is introduced into desert every year through in-migrated natives. Once introduced through in-migration, the transmission of malaria is controlled by a combined interactive resultant of mosquito density, human population density, cattle population density and carriers of active infection. The inference drawn on the basis of research undertaken so far need to be validated through repeat studies adopting few villages.

Progress of the work

Three villages were adopted in Jaisalmer district, Rajasthan, during pre-monsoonal season (June, 2005) and followed up till August, 2005, to accomplish the stated objectives. The work undertaken during last year in the present project has been focused to address following contentions:

Contention 1.

That malaria in desert conditions is **Introduced afresh** every year otherwise there is no consistent malaria situation in most of the desert villages.

Observations

Only occasional cases of malaria were observed in above adopted villages during months of June-August, 2005 (Table). Similar situation of malaria existed during the year 2006 also. The data collected during 2005 were analyzed with respect to whether cases appeared during start of season were migrated or indigenous. Results are shown in the table below:

Months	Villages	Adult density*		Cattle population	Human population	No. of cases	Predicted status
		<i>An. culicifacies</i>	<i>An. stephensi</i>				
June	Tejpala	9(7.01*)	1(0.77)	29000	1151	1**	Sporadic
	Sheikhaser	0(0.0)	0 (0.0)	14000	2200	0	Sporadic
	Bada	20 (20.6)	0(0.0)	11000	1240	3**	Sporadic
	Naga	0(0.0)	0 (0.0)	8000	900	0	Sporadic
July	Tejpala	1(7.5)	0 (0.0)	29000	1151	0	Sporadic
	Sheikhaser	ND	ND	14000	2200	ND	Sporadic
	Bada	2 (12)	0 (0.0)	11000	1240	0	Sporadic
	Naga	0 (0.0)	0 (0.0)	8000	900	3**	Sporadic
August	Tejpala	23 (25.0)	20 (21.8)	29000	1151	0	Sporadic
	Sheikhaser	6 (6.5)	0 (0.0)	14000	2200	0	Sporadic
	Bada	ND	ND	11000	1240	0	Sporadic
	Naga	0 (0.0)	0 (0.0)	8000	900	0	Sporadic

*Average density of month **In migrated cases

Contention 2.

That the introduction of malaria in desert can be prevented by checking in-migrating natives. The demonstration of the prevention can be shown in adopted villages through introduction of check posts.

Observations

Since there were not many cases during 2006, cases occurred during 2005 were analyzed with respect whether these were in-migrated or not. Since outbreaks of malaria have not erupted during 2006, this step need to be confirmed when disease is witnessed. During the study there were not many cases in rainy season.

Contention 3.

- That once introduced, the prospective transmission of malaria is the resultant interaction of following:
- *An. stephensi* – Human population density – Active malaria patients
- *An. culicifacies* – Cattle population – Active malaria patients
- That through our epidemiological understanding of above relationship, we can develop a mathematical Equation predictive of malaria transmission in a setting.

Following equation of quantitative relationship among above parameters has been developed.

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

Where TI = Transmission Index; Ac = Active Cases; PMHst = density *stephensi*; PMHcu = density *culicifacies*; Hd = Human density (Human/ Sq.km); Cd = Cattle density (Cattle/ Sq.km)

Contention 4.

- That we can convert this equation into **A software** for predicting malaria transmission.

Observations

- A working software has been developed

Contention 5.

- That this **Software could be tested** in simulated and real life conditions.

Observations

- There were not many cases of malaria in Jaisalmer district during 2006. For testing of the software, substantial no. of cases are required which have not occurred in the last two years.
- A computer software has been developed which can process simulated conditions. The software can process simultaneously varying values of all the parameters of proposed Transmission Equation viz; Active Cases (AC), Per Man Hour Density *An. stephensi* (PMHs), Human Density (Hd), Per Man Hour Density *An. culicifacies* (PMHc) and Cattle Density (Cd) in the mathematical

relationship of equation. At the same time software can process varying values of any one parameter fixing other parameters. Few examples of different simulations as processed by the software are given below:

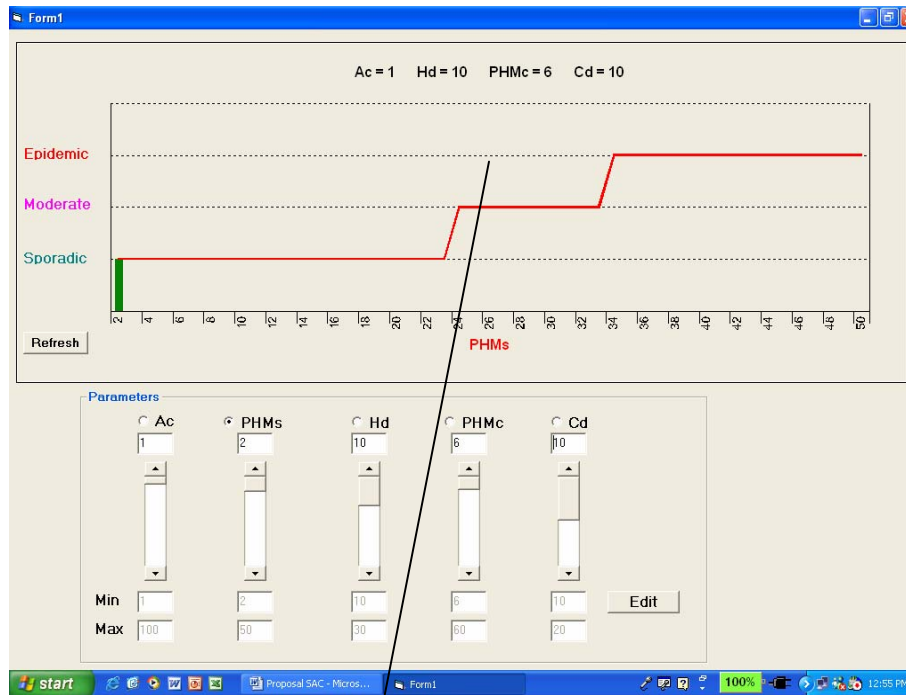


Fig.1. Simulated prediction of malaria magnitude at **AC*=1**; PMHs=2; Hd=10; PMHc=6; cd=10

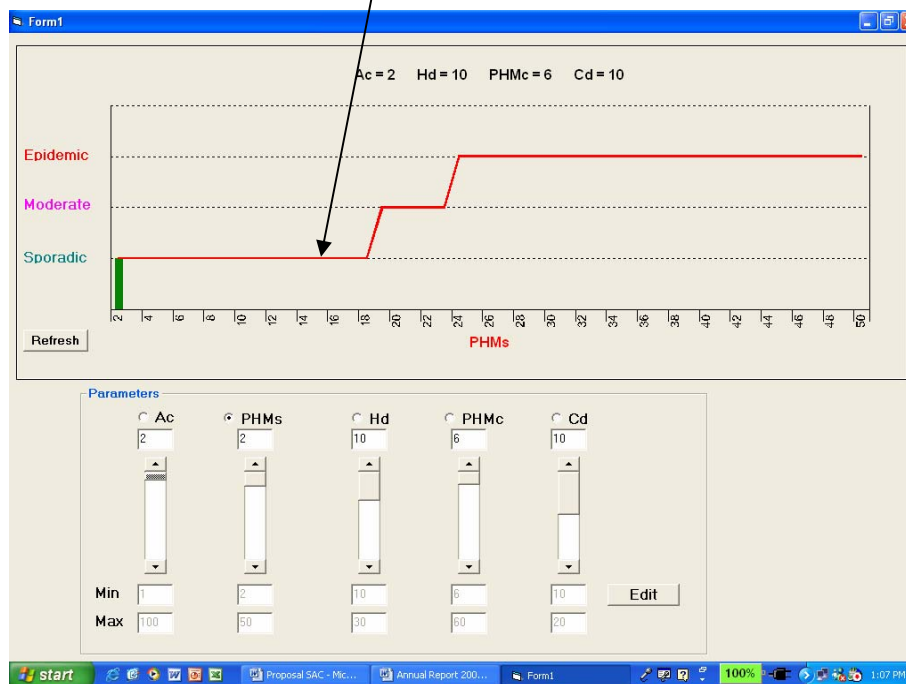


Fig.2. Simulated prediction of malaria magnitude at **AC=2***; PMHs=2; Hd=10; PMHc=6; cd=10

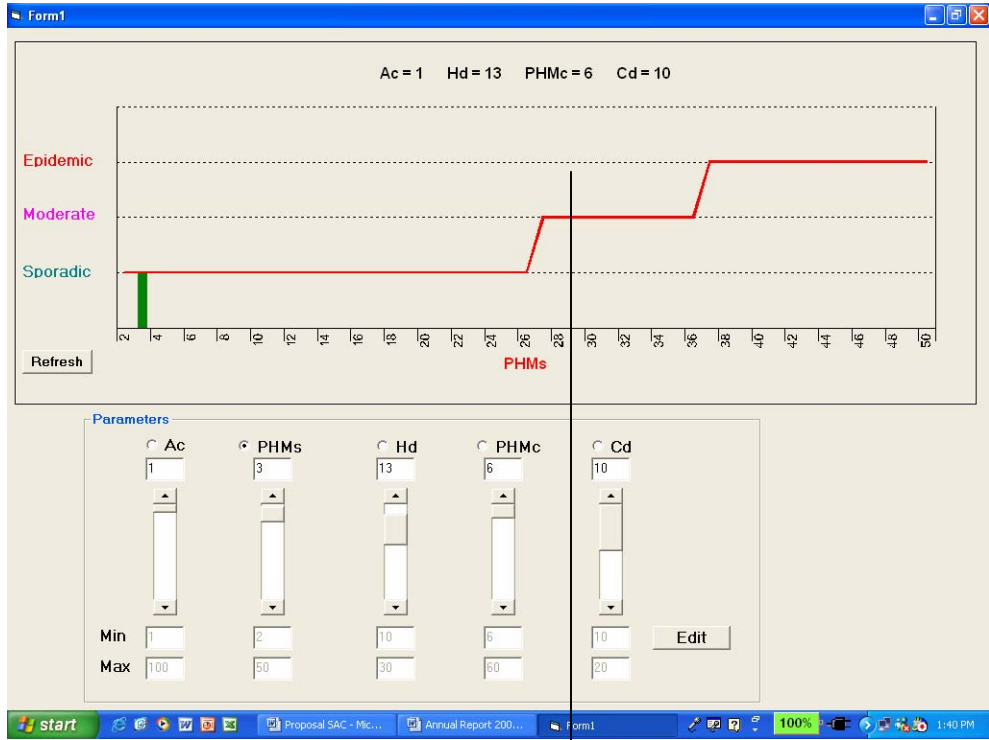


Fig.3. Simulated prediction of malaria magnitude at AC=1; **PMHs=3***; Hd=10; PMHc=6; cd=10

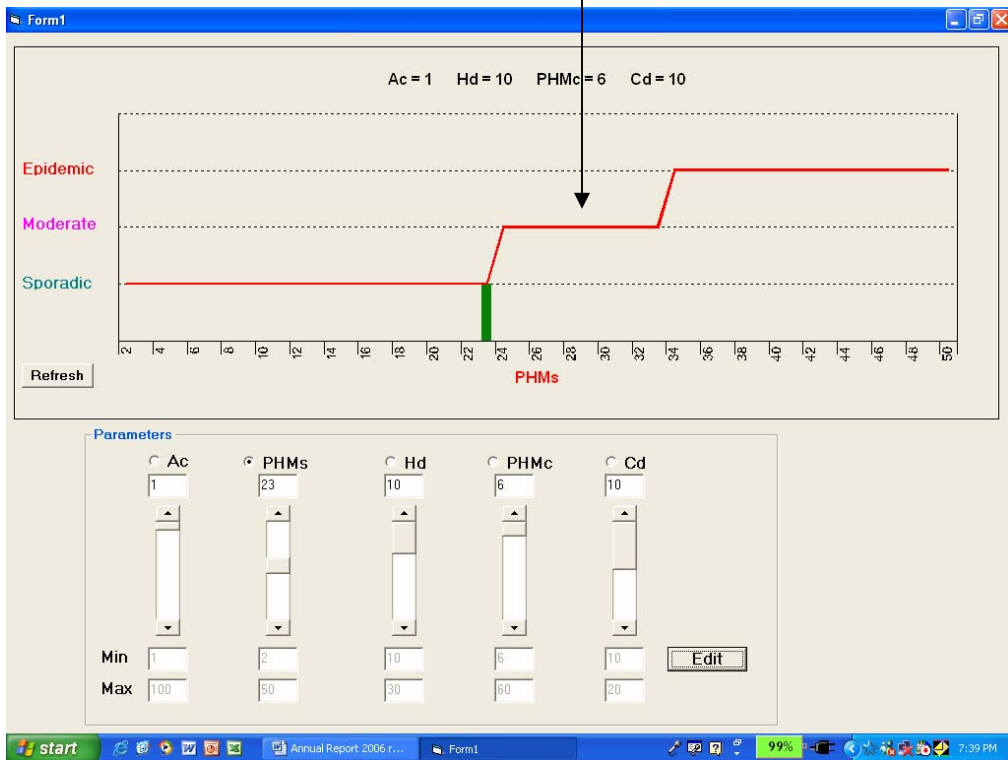


Fig.4. Simulated prediction of malaria magnitude at AC=1; **PMHs=23***; Hd=10; PMHc=6; cd=10

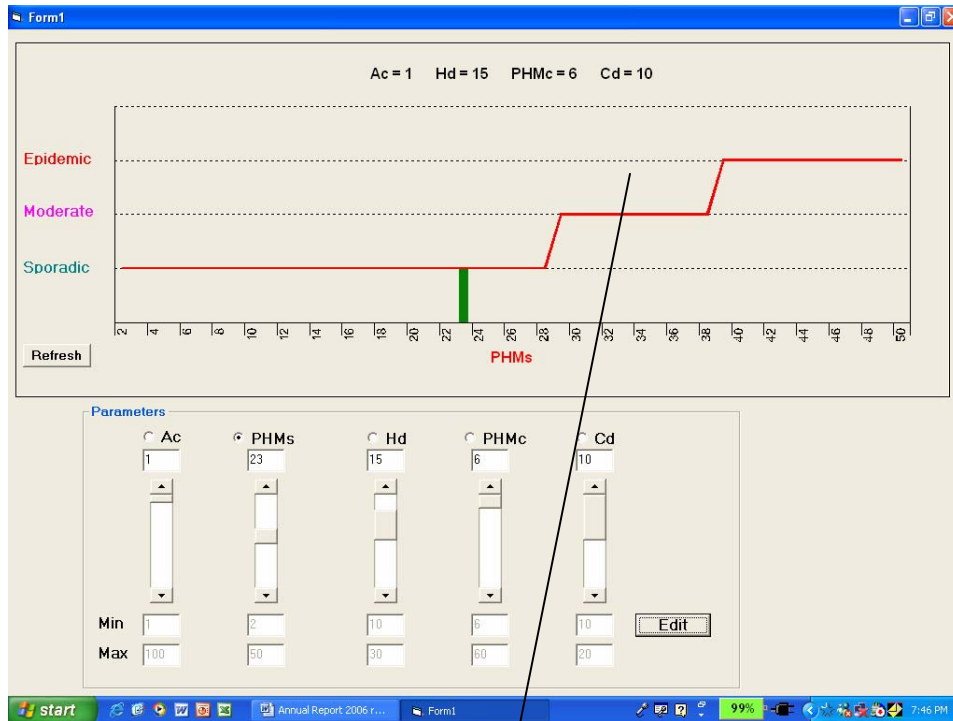


Fig.5. Simulated prediction of malaria magnitude at **AC=1**; PMHs=23; **Hd=15***; PMHc=6; cd=10

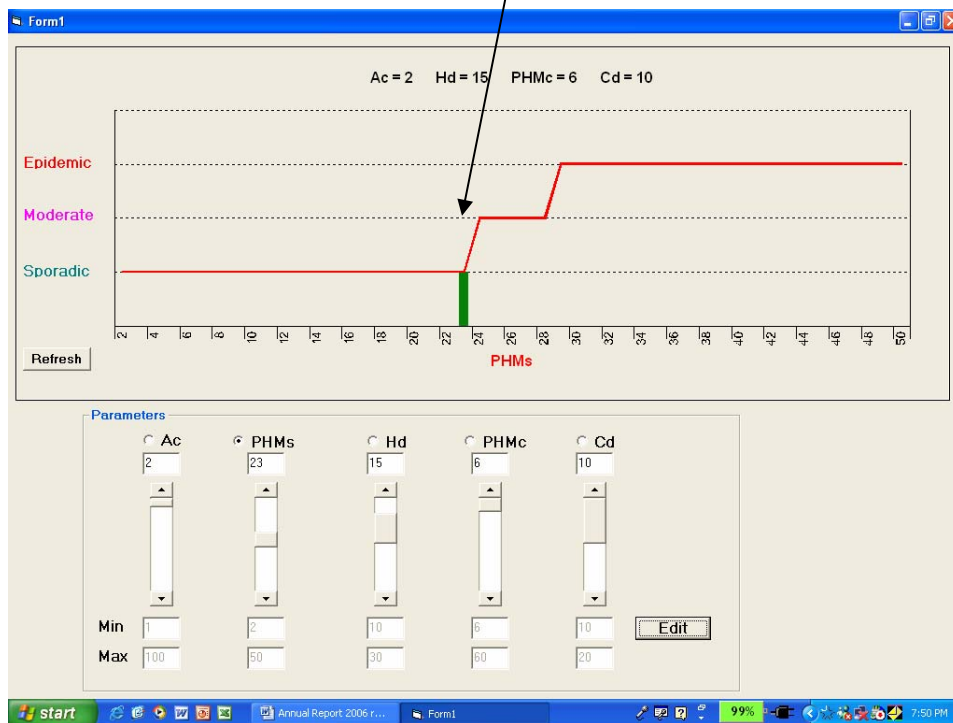


Fig.6. Simulated prediction of malaria magnitude at **AC=2***; PMHs=23; HD=15; PMHc=6; cd=10
 AC= Active Cases PMHs= PMH *An stephensi* HD= Human Density PMHc= PMH *An. culicifacies* Cd= Cattle Density
 *Highlighted letter in each figure denotes the parameter whose value is changed than previous figure

- This has been observed that computer model for developed for simulated parameters of Transmission Equation can process any value of mosquito, human and/or cattle density in multiplication of Active cases of malaria at a given point of time. Based on few exemplary situations the important predictions given by computer model are mentioned below:
- Figure 1 shows 1 Active Case of malaria as may be available in the beginning of malaria season in the area. For corresponding value of PMH density of *An. stephensi* equal to 2, human density as 10, PMH density of *An. culicifacies* as 6 and human density as 10 persons, the resultant malaria situation in a village would be predominantly sporadic only.
- In Figure 2, value of Active cases has increased from 1 in figure 1 to 2 in figure 2. By keeping the values of all other parameters fixed as in figure 1, we observe that there is increase in probability of epidemic occurrence as shown by tangent arrow between figure 1 and 2.
- In figure 3, by fixing back value of AC as 1, PMH density of *An. stephensi* has been increased from 2 in figure 1 & 2 to 3 in figure 3. As shown by the vertical arrow between figures 2 & 3, there is not much change in probability of an epidemic occurrence.
- In figure 4 we have fixed the values of all other parameters as they were in figure 3, but have increased the PMH density of *An. stephensi* from 3 to 23. We find that when AC was 1, even substantial increase in mosquito density has not influence beginning point of epidemic in line graph of figure 4.
- In figure 5, we have increased value of human density from 10 of figure 4 to 15 in figure 5. We observe that point epidemic emergence in the graph has been delayed. This shows that at a given value of active malaria case and mosquito density, when humans as bait population to *An. stephensi* increase, probability of malaria epidemic reduces.
- Figure 6 shows that at the values of mosquito and man and animal bait density as fixed in figure 5, an increase in number of active cases from 1 to 2 makes substantial pre-ponement of epidemic situation as compared to situation when active case was one.

Inferences

- The malaria transmission equation or mathematical model $TI = Ac (PMH_{st} - Hd + PMH_{cu} - Cd)$ Where TI = Transmission Index; Ac = Active Cases; PMH_{st} = density *stephensi*; PMH_{cu} = density *culicifacies*; Hd = Human density/Sq.Km; Cd = Cattle density/Sq.Km, has been developed based on understanding of PI about the possible inter relationship which is likely to exist among ultimate participating components of malaria as the disease. For example, whatever is the ecological set up of study area from ambient temperature, relative humidity, rainfall, insecticide spray or insecticide resistance, what will matter is the density of vector species viz; PMH *stephensi* and PMH *culicifacies*. Hence these parameters have been included in the model. However, the role of a particular density of a vector species in malaria transmission will depend on bait of preference of a species. For *An. stephensi* human bait and for *An.*

culicifacies, cattle bait. Therefore, Hd and Cd have been included in inversely proportional relationship to their corresponding baits densities. Over and above all, these all indices will matter in malaria transmission when an active case is present. If case is not present values of all these indices from transmission point of view will be zero. Therefore, Active cases have been place in the position of multiplying parameter in the equation.

- The simulation model developed is capable of processing varying values of all the parameters simultaneously as well as fixing all the values and varying value of one particular parameter.
- The software developed appears to process any existing value of vector, parasite and bait parameter very sensitively. Under real life conditions the values of any or all the parameters will be within range of simulated values (1-99), therefore, it can be validated under the field conditions for its possible application as the forecasting mechanism.

Work remains to be done

The software developed need to be tested by the independent agencies for its validity. The model is presented for its validation, after which it could be proposed for patenting and then be proposed for the public health use.

Studies on *Calotropis procera* as larvicidal and repellent plant against vectors of Dengue and DHF in Rajasthan, India - Manju Singhi, Vinod Joshi, P.K. Dam and Sunita Kumbhat*

Commencement: July 2003

Duration: Three and half years

Status: completed

Objectives

1. To isolate and identify active larvicidal ingredient from crude latex of *Calotropis procera*
2. To undertake operational research with respect to community application of latex for larval control of *Ae. aegypti*

Rationale

Dengue fever associated with Dengue Haemorrhagic Fever is emerging as one of the major infectious diseases in many parts of India including North-western , Rajasthan. In absence of any specific chemotherapy against this viral infection and the confirmed reports of virus being maintained through transovarial transmission in *Aedes aegypti* and *Aedes albopictus* , vector control measures, attempting source reduction (larval control), appears to be the most effective method of interrupting transmission of dengue in an endemic setting. *Calotropis procera*, a wild shrub, is an abundant desert plant available throughout the year in all the parts of Rajasthan.

Our earlier studies on this plant had shown that the methanol, acetonitrile and aqueous extract of latex of this shrub gives promising results as a larvicide. It has also been observed that not only larvicidal effects but presence of latex in the water also repels the gravid females of *Ae. aegypti* from egg laying

For application of larvicidal property of latex of this plant (*C. procera*) as a tool against dengue vectors, it was thought necessary to isolate the larvicidal ingredient from the crude latex and develop the resultant compound as a bio larvicide. Present study describes the results of isolation attempts of larvicidal component from the latex and also presents results of preliminary trials of application of latex in the disease endemic localities of Jodhpur.

Progress of the Work

Identification & Isolation of larvicidal ingredient:

Active ingredients present in the latex and responsible for larvicidal and ovicidal actions were isolated using latest available Chemical tools. Resolutions of latex were accomplished using High Performance Liquid Chromatography (HPLC), Spectro Photometry (UV Mode).

Details of Chemical Resolution of latex by HPLC are given below:

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Fig.1. HPLC curves as obtained in distilled water (control) and aqueous solution of latex (experimental)

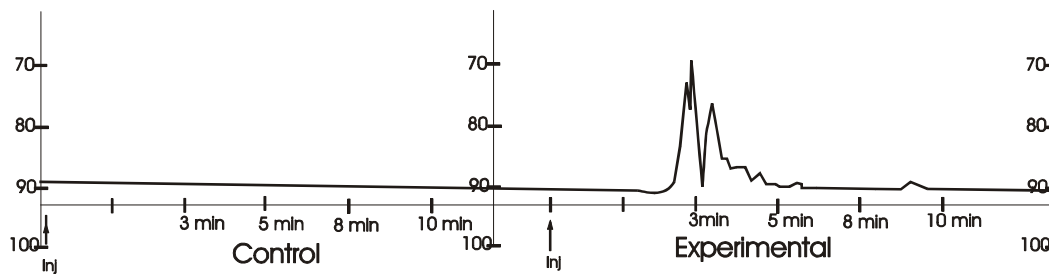


Fig.2. HPLC curves as obtained in methanol (control) and methanol extracted latex (experimental)

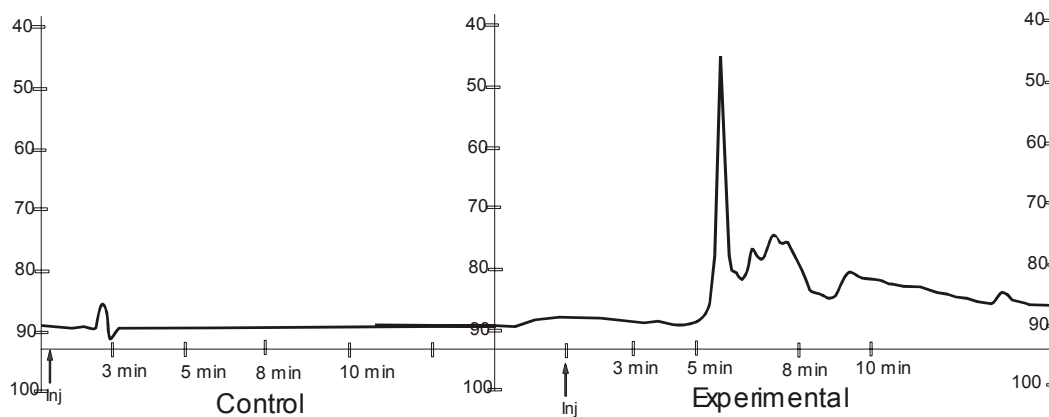


Fig.3. HPLC curves as obtained in acetonitrile (control) and acetonitrile extracted latex (experimental)

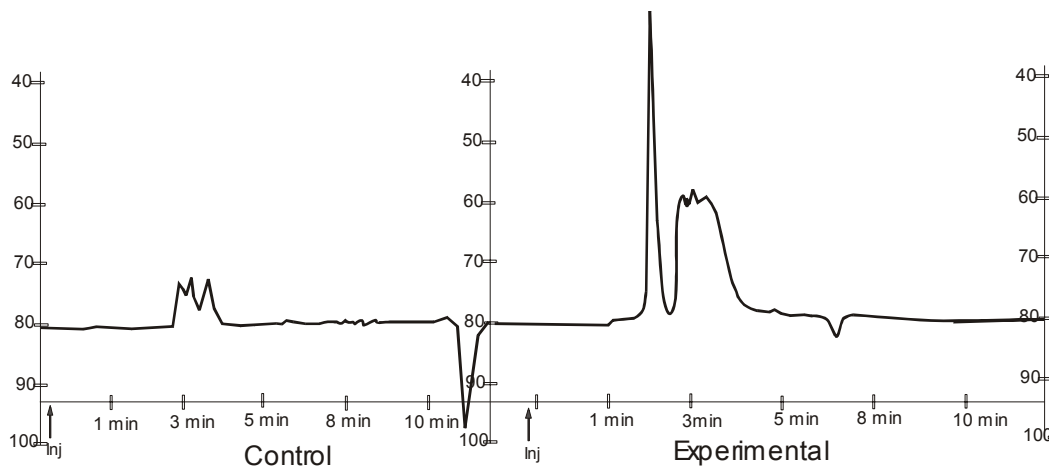


Figure- 1 shows HPLC curves as obtained in the aqueous solution of latex (experimental) and in distilled water (control). The graph obtained clearly shows two prominent curves (Curve 1 & 2) and 5 overlapping and suppressed curves (Curves 3-7). In all, a group of 7 compounds in crude aqueous solution of latex has been obtained.

Figure- 2 shows HPLC runs of control and experimental solutions of *C. proceru* in methanol extract. While one small curve was obtained in control solution (methanol), methanol extract showed one prominent and three small curves (Fig. 2). It appears that curve 1 of aqueous extract (Figure 1) disappeared in methanol extract and curves 3-7 of aqueous extract became more prominent as 1 and 2-4 curves (Fig. 2). The observations show that by extracting latex in methanol, we obtain selection of a group of 4 compounds as compared to a mixture of 7 compounds in aqueous solution.

Figure -3 shows HPLC runs of control (acetonitrile) and acetonitrile extract of latex. The acetonitrile extract of latex displayed the single compound peak as compared to its control when resolved by HPLC. This fraction showed 231 nm wavelength when studied employing spectrophotometry. Confirmation of this fraction as larvicide was made performing efficacy trials against larvae of dengue vectors.

The present study was aimed to identify and isolate the compound responsible for larvicidal action in latex of *C. proceru*. Through present work the objectives studied have been accomplished.

Feasibility trials of application of latex in domestic/community containers of Jodhpur

Concentrations of crude latex of *C. proceru*, which were tested in laboratory as effective larvicide, were tried under field conditions for their acceptability by the public and efficacy of latex in natural habitats. The trials gave the similar results as obtained under laboratory conditions. Under natural conditions, when latex was applied in domestic habitats, acceptability of people was less. In the outdoor utensils, application of larvicide was most acceptable by the inhabitants of the area. The observations are mentioned in the Table 1 & 2 .

Table 1. Field efficacy trials of latex in domestic water containers (Pitchers)

Larvae exposed (Nos.)	Latex Concentration (%)	Time exposure (hrs.)	Larval mortality (%)
50	1.0	24	100
40	1.0	24	100
32	1.0	24	100
60	1.0	24	100

Table 2. Field efficacy trials of latex in outdoor water containers (Earthen pots)

Larvae exposed (Nos.)	Latex concentration (%)	Exposure time (hrs)	Larval mortality (%)
30	1.0	24	100
35	1.0	24	100
65	1.0	24	100
50	1.0	24	100

Inferences

1. Development of a bio larvicide is the ultimate need to control transmission and sustenance of dengue virus in disease endemic areas. Development of such a product through indigenous plant stock will be a cost effective and acceptable product. We have identified and isolated larvicidal ingredient from *C. procera*.
2. Acetonitrile extracted, HPLC fraction of latex gave larvicidal efficacy showing two peaks against one peak in control. This resolution indicated that in larvicidal fraction of acetonitrile extract of latex, only one peak was present. Further analysis of this curve by Spectrophotometry showed that this curve represents 232 nm wavelength. This observation indicated that the peak obtained in the HPLC belongs to the compound, which is an alcohol derivative.
3. The available literature comprehending different compounds of latex has been reviewed. The search showed that in latex of *C. procera*, the alcohol derivative present is Calotropin. Chemical resolution of latex of *C. procera* has been done by other workers (Duke, 1992) but none so far has reported any compound to be a bio larvicide.
4. Our study reports Calotropin as a new Bio larvicide against dengue vectors. The patent claim for the product has been filed, in the category of “Known compound for new use”.
5. The crude latex of *C. procera* gave similar results of larvicidal efficacy as under laboratory conditions. However, acceptability of people towards domestic larval habitats was very low as compared to peri- domestic containers.

Work remains to be done

The SAC, DMRC, suggested that ongoing phase of work on *C. procera* be accomplished in two phases; Isolation/identification of larvicidal compound and study of mode of action of the identified compound. Since the isolation of larvicidal compound has been completed, study of its mode of action at intra cellular level in mosquito cell lines and in-vivo will be pursued as an independent project.

Mapping of insecticide resistance in vectors of malaria in Rajasthan - Karam V. Singh and S. K. Bansal

Commencement: **October, 2005**

Duration: **Two Years**

Status: **Ongoing**

Objectives

1. To determine the current status of insecticide susceptibility/ resistance among malaria vectors against conventional and newer compounds in different parts of Rajasthan, and
2. To determine the biochemical mechanisms involved in the development of resistance.

Rationale

Several years of intensive use of organic insecticides to control disease vectors has led to the precipitation of insecticide resistance among them. Resistance management has evolved as a favoured approach to prevent, delay or reduce the impact of insecticide resistance. To develop this new strategy a thorough knowledge of the current status of resistance-developed and the mechanism involved are essential. Factors that induce resistance are numerous and the mechanism adopted by a particular vector species depends on the prevailing pressure and mode of action of insecticide in use. The information collected on the current status of resistance and the mechanism(s) involved would help in formulating probable means for the management of insecticide resistance and state insecticide policy as well.

Progress of the work

During the report period, the studies have been carried-out in four districts namely Jodhpur, Pali, Sirohi and Udaipur, however, for the comparison of the data the studies undertaken in Barmer and Jaisalmer districts during last year have also been considered. First three districts *viz.* Jodhpur, Pali and Sirohi, have the insecticide spray history of DDT, whereas, in Udaipur district the spray history of DDT, BHC and Deltamethrin.

In Jodhpur district the studies were carried out in Ramdeo colony, which has spray history of DDT and this has not been sprayed since 5 years. In Sirohi district, which also has the spray history of DDT, the studies were conducted in two villages namely Dhanari and Dheldhar. Village Dhanari is under regular DDT spray since last four years, whereas, village Dheldhar has been sprayed with DDT after a gap of 5 years. In Udaipur district, the first village Bilak Keekavat has the spray history as DDT-BHC-DDT-Deltamethrin and no spray since last two years, whereas, Barapal village followed the spray history as DDT-BHC-DDT with no spray since last one year. In Pali district, which has the spray history of DDT only, the studies were carried out in village Gadwara and this village has not been sprayed since last four years.

During the studies two vector species viz. *Anopheles culicifacies* and *An. stephensi* were tested. Four insecticides i.e. DDT, Malathion, Propoxur and Deltamethrin (Synthetic Pyrethroid) were used to determine the susceptibility status of these vector species against the candidate insecticides in different situations.

For determining the susceptibility status of vector species dawn collection was made in selected villages using suction tube method. The collected individuals were identified species-wise, abdominal conditions recorded and transported to centre's insectary in Burraud cages. Tests were conducted as per standard WHO method for determining the insecticide susceptibility of adult mosquitoes. The individuals of each species were exposed to the diagnostic doses of DDT (4.0%), Malathion (5.0%), Propoxur (0.1%) and Deltamethrin (0.05%) for 1.0 hour and the observations were made after 24 hours. The WHO criterion was followed for considering the vector species susceptible (mortality >98 %), resistant (mortality <80 %) and intermediate resistant (mortality 80 – 98 %). Abbott's formula was applied wherever required to correct the observed mortality.

Table 1. Insecticide Susceptibility status of *Anopheles culicifacies* in different Districts

Candidate Insecticides (Diagnostic doses)	Exposure Time (hrs.)	Percent Mortality and Susceptibility Status					
		Jodhpur	Pali	Sirohi		Udaipur	
		Ramdeo Colony	Gadwara	Dhanari	Dheldhar	Bilak Keekavat	Barapal
DDT (4.0%)	1.0	78.7 R*	68.8 R	57.0 R	76.9 R	58.3 R	65.0 R
Malathion (5.0%)	1.0	90.0 IR	80.0 IR	88.9 IR	100.0 S	75.0 R	85.0 IR
Propoxur (0.1%)	1.0	86.6 IR	75.0 R	-	-	70.0 R	80.0 IR
Deltamethrin (0.05%)	1.0	100.0 S	100.0 S	100.0 S	100.0 S	100.0 S	100.0 S

***R** – Resistant, **IR** – Intermediate Resistant, **S** – Susceptible.

Table 1 depicts the results of the susceptibility tests carried out with *An. culicifacies* against candidate insecticides. The results revealed that in all the districts, the species has developed resistance against DDT, whereas, against deltamethrin, which is a synthetic pyrethroid, the species was found totally susceptible. Against Malathion and Propoxur the species exhibited intermediate resistance and at some place total resistance too, but in one village the species was found susceptible to Malathion.

The susceptibility status of *An. culicifacies* and *An. stephensi* was also correlated with the duration of spray since last spray (Table 2) and it was observed in case of both the species that with the increase in duration since last spray, the susceptibility status of the vector species also increases.

Table 2. Showing mortalities of vector species in respect of duration since last spray of insecticide

Vector Species	Percent Mortalities in different backgrounds of DDT Spray			
	Regular Spray	Before 1.0 Yr.	Before 2.0 Yrs	>2.0 Yrs
<i>An. stephensi</i>	55.8	55.6	72.5	60.0*
<i>An. culicifacies</i>	57.1	65.0	58.3**	78.7

* To be verified; **Under Synthetic Pyrethroid (SP) spray.

During the studies attempts were also made to correlate the history of the spray with the susceptibility status of the tested species and the species-wise results have been tabulated in Tables 3 and 4. In case of *An. stephensi* four types of backgrounds of insecticide spray *viz.*: DDT (regular spray), DDT (spray before 5 yrs), DDT-Malathion-DDT (spray before 2 yrs) and DDT-BHC-DDT-SP (Regular spray) have been studied. It was observed that in the area, where synthetic pyrethroid has been used since last 2 years species has shown maximum resistance against DDT (25.0%), followed by DDT-Malathion-DDT (52.8%), DDT regular use (55.8%) and DDT sprayed before >5 years (60.0%) backgrounds (Table 3). The resistance against other insecticides also followed the same pattern.

Table 3. Showing mortalities of *An. stephensi* against tested compounds in different spray backgrounds

Insecticides Tested	Percent mortalities in different spray backgrounds (%)			
	DDT (Regular Spray)	DDT (Before >5.0 Yrs)	DDT-Malathion-DDT (Before 2.0 Yrs)	DDT-BHC-DDT-SP (Regular spray)
DDT	55.8	60.0	52.8	25.0
Malathion	77.5	80.0	73.3	54.5
Deltamethrin	100.0	100.0	100.0	100.0

Table 4. Showing mortalities of *An. culicifacies* against tested compounds in different spray backgrounds

Insecticides Tested	Percent mortalities in different spray backgrounds (%)					
	DDT (Regular Spray)	DDT (After 5.0 Yrs)	DDT (Before 4 .0 Yrs)	DDT (Before >5.0 Yrs)	DDT-BHC-DDT (Before 1.0 yrs)	DDT-BHC-DDT-SP (Before 2.0 yrs.)
DDT	57.1	76.2	68.8	78.7	65.0	58.3
Malathion	88.9	100.0	80.0	90.0	85.0	75.0
Propoxur	-	-	75.0	86.6	80.0	70.0
Deltamethrin	100.0	100.0	100.0	100.0	100.0	100.0

An. culicifacies, which has been compared among six spray backgrounds (Table 4), exhibited highest DDT resistance in case of regular DDT spray (57.1%), followed by DDT-BHC-DDT-SP (58.3%), DDT-BHC-DDT (65.0%), DDT sprayed before 4 years (68.8%), DDT after 5 years gap (76.2%) and DDT without spray since >5 years (78.7%). In case of Malathion resistance the maximum resistance was observed in case of DDT-BHC-DDT-SP background and minimum in DDT sprayed before >5 years. The Propoxur resistance was also found high in the spray background DDT-BHC-DDT-SP. The varied response of the vector species to different backgrounds of the insecticide spray can be explained after determining the biochemical mechanism(s) involved in the development of the insecticide resistance against different compounds.

Work remained to be carried-out

The studies will be conducted in the remaining areas covering more insecticide spray combinations. The biochemical mechanisms involved in the development of insecticide resistance in the study areas will also be determined.

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

Commencement: **September, 2005**

Duration: **Two Years**

Status: **Ongoing**

Objectives

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

Rationale

Vector control through alternative means is an essential and effective tool for controlling transmission of vector-borne diseases especially in areas where vector have become resistant to the insecticides. Moreover, increasing awareness of the environmental hazards of these synthetic insecticides and development of resistance has forced the scientists to look for other alternatives of vector control. Hence, biologically active plant materials with complex chemical substances of different composition found in various parts of the plants with selective anti-larval and anti-adult properties are being paid attention to replace the synthetic one's. In the process a number of plant species with insecticidal properties have been considered including *Solanum xanthocarpum*, the Indian nightshade commonly known as 'baigan kateli'. It is found throughout the country but more abundantly in the arid areas and used widely for a variety of ailments in public health. Extracts prepared from different parts of this plant species will be screened for their larvicidal potential against larvae of different mosquito vectors present in this area.

Progress of the Work

Susceptibility tests were carried out with larvae of different mosquito vectors viz. *Anopheles culicifacies*, *Anopheles stephensi*, *Aedes aegypti* and *Culex quinquefasciatus*. For this purpose adults and larvae of all the mosquito species were collected from different areas of Jodhpur city and reared in the laboratory for further generations under controlled conditions of temperature ($28\pm 2^\circ\text{C}$) and humidity ($75\pm 5\%$). *S. xanthocarpum* has been selected for undertaking the proposed study. The different parts of this plant differ in their active constituents when extracted in different solvents. Samples of roots, leaves, fruits and seeds of this plant were chopped and shade dried between $30-40^\circ\text{C}$ for 10-15 days. Dried plant material was powdered separately and dissolved in different

solvents and stock solutions and duration and serial dilutions were made as per requirement. Third or early fourth instar larvae of these mosquito species were tested as per standard WHO method for determining the baseline data on their susceptibility status. Experiments were carried out in 500 ml beakers containing 249 ml of water by using 20-25 larvae of each mosquito species. Mortality was noted after 24 hr and corrected by using Abbott's formula. Average of four observations was taken and data subjected to log probit regression analysis.

Observations on the results of the larval susceptibility to methanol extracts of epicarp of six months old fruits are given in Table 1&2. With all the mosquito species mortality was dose dependent i.e. mortality increased with increase in concentration. 24 & 48hr LC₅₀ and LC₉₀ values along with their fiducial limits, regression equation and chi-square were calculated. 24 & 48hr LC₅₀ values as observed for *An. culicifacies*, *An. stephensi*, *Ae. aegypti* and *Cx. quinquefasciatus* were 79.6, 131.4, 273.4 & 384.9 and 60.0, 67.3, 108.7 & 150.0 mg/l respectively. Comparative susceptibility calculated on the basis of 24hr LC₅₀ value showed that larvae of *An. culicifacies* are 1.65, 3.43 and 4.84 times more susceptible than larvae of *An. stephensi*, *Ae. aegypti* and *Cx. quinquefasciatus* respectively. 48hr LC₅₀ value was also found to be much less as compared to the 24hr LC₅₀ showing that efficacy was much more pronounced after 48hrs as compared to 24hrs.

Experiments were also carried out after one year of storage of the fruits in order to see the efficacy of the fruit epicarp. 24 & 48hr LC₅₀ values as observed for *An. culicifacies*, *An. stephensi*, *Ae. aegypti* and *Cx. quinquefasciatus* were 103.3, 146.3, 316.9 & 393.0 and 62.3, 80.4, 126.0 & 222.2 mg/l respectively. Efficacy was further decreased as revealed by the 24 and 48hr LC₅₀ values (Table 2).

Observations on the results of the larval susceptibility to methanol extracts of fresh and six months old whole fruits are given in Table 3&4. 24 & 48hr LC₅₀ and LC₉₀ values along with their fiducial limits, regression equation and chi-square were calculated. 24 hr LC₅₀ and LC₉₀ values as observed for *An. culicifacies*, *An. stephensi*, *Ae. aegypti* and *Cx. quinquefasciatus* were 51.6, 52.2, 118.3 & 157.1 and 91.7, 186.9, 290.9 & 450.6 mg/l respectively. The 48hr LC₅₀ values calculated on the whole fruits after six months were 66.5, 87.9, 144.1 & 220.4 mg/l respectively which are about half of the 24hr LC₅₀ values. Comparative susceptibility calculated on the basis of 24 LC₅₀ value showed that larvae of *An. culicifacies* are 2.04, 3.17 and 4.91 times more susceptible than larvae of *An. stephensi*, *Ae. aegypti* and *Cx. quinquefasciatus* respectively.

Susceptibility tests on larvae were also carried out with methanol and petroleum ether extracts of six months old seeds of this plant with larvae of all the four mosquito species and the results have been given in Table 5&6. With all the mosquito species concentrations tried were 25 to 400 mg/l of the extract. 24 hr LC₅₀ and LC₉₀ values as determined for *An. culicifacies*, *An. stephensi*, *Ae. aegypti* and *Cx. quinquefasciatus* were 66.9, 73.7, 123.8 & 154.9 and 131.7, 195.6, 377.6 & 520.0 mg/l respectively which revealed that larvae of *Anophelines* were more susceptible than that of the culicines. The 48hr LC₅₀ values determined for the seed extracts were 79.3, 95.6, 131.1 & 222.7 mg/l respectively, which are about two to three times less than the 24hr LC₅₀ values. Larvae of *An. culicifacies* were found 1.49, 2.87 and 3.95 times more susceptible than *An. stephensi*, *Ae. aegypti* and *Cx. quinquefasciatus* respectively.

Table 1. Log probit regression analysis of the mortality data of larvae of different mosquito vectors to methanol extracts of fruit epicarp of *S. xanthocarpum* after six months of storage

Mosquito species	Regression Equation	Chi-Square (DF)	24hr LC ₅₀ (Fiducial limits)	24hr LC ₉₀ (Fiducial Limits)
<i>Anopheles culicifacies</i>	Y=2.83x-0.52	0.72 (2)	79.6 (60.0) (70.3-114.1)	253.8 (157.4-409.2)
<i>Anopheles stephensi</i>	Y=2.63x-0.57	1.92 (2)	131.4 (67.3) (100.7-171.2)	402.9 (217.3-746.9)
<i>Aedes aegypti</i>	Y=2.17x-0.29	3.32 (2)	273.4 (108.7) (196.0-381.4)	1063.0 (490.5-2305.0)
<i>Culex quinquefasciatus</i>	Y=1.82x+0.28	3.13 (2)	384.9 (150.0) (256.8-576.9)	1936.0 (730.0-3140.0)

Values in bold are the 48hr LC₅₀ for that species

Table 2. Log probit regression analysis of the mortality data of larvae of different mosquito vectors to methanol extracts of fruit epicarp of *S. xanthocarpum* after one year of storage

Mosquito species	Regression Equation	Chi-Square (DF)	24hr LC ₅₀ (Fiducial limits)	24hr LC ₉₀ (Fiducial Limits)
<i>Anopheles culicifacies</i>	Y=2.31x-0.56	0.81 (2)	103.3 (62.3) (62.2-111.5)	297.6 (167.2-563.3)
<i>Anopheles stephensi</i>	Y=2.28x-0.06	1.28 (2)	146.3 (80.4) (115.2-185.8)	533.1 (303.0-938.0)
<i>Aedes aegypti</i>	Y=1.70x-0.74	5.71 (2)	316.9 (126.0) (216.2-466.5)	1091.0 (600.0-2358.0)
<i>Culex quinquefasciatus</i>	Y=3.01x-2.81	0.99 (2)	393.0 (222.2) (308.4-500.7)	1046.0 (593.1-1844.0)

Values in bold are the 48hr LC₅₀ for that species

Table 3. Log probit regression analysis of the mortality data of larvae of different mosquito vectors to methanol extracts of whole fruits of *S. xanthocarpum*

Mosquito species	Regression Equation	Chi-Square (DF)	24hr LC ₅₀ (Fiducial limits)	24hr LC ₉₀ (Fiducial Limits)
<i>Anopheles culicifacies</i>	Y=2.62x+0.51	0.25 (2)	51.6 (40.1-66.4)	159.0 (93.3-270.6)
<i>Anopheles stephensi</i>	Y=2.41x+0.86	1.26 (2)	52.2 (40.4-67.4)	177.3 (101.6-309.4)
<i>Aedes aegypti</i>	Y=4.35x-4.01	2.69 (2)	118.3 (102.5-136.4)	233.1 (171.4-317.0)
<i>Culex quinquefasciatus</i>	Y =2.43x-0.33	1.42 (2)	157.1 (121.3-203.4)	529.4 (283.1-990.0)

Table 4. Log probit regression analysis of the mortality data of larvae of different mosquito vectors to methanol extracts of whole fruits of *S. xanthocarpum* after six months

Mosquito species	Regression Equation	Chi-Square (DF)	24hr LC ₅₀ (Fiducial limits)	24hr LC ₉₀ (Fiducial Limits)
<i>Anopheles culicifacies</i>	Y=2.08x+0.92	1.17 (2)	91.7 (66.5) (59.4-141.9)	379.0 (152.3-942.4)
<i>Anopheles stephensi</i>	Y=3.06x-1.94	4.50 (2)	186.9 (87.9) (151.1-231.2)	490.3 (296.1-811.7)
<i>Aedes aegypti</i>	Y=2.70x-1.66	0.44 (2)	290.9 (144.1) (161.3-263.7)	865.8 (486.8-1539.0)
<i>Culex quinquefasciatus</i>	Y =2.06x-0.47	0.25 (2)	450.6 (220.4) (302.6-671.0)	1881.0 (676.9-3228.0)

Values in bold are the 48hr LC₅₀ for that species

Table 5. Log probit regression analysis of the mortality data of larvae of different mosquito vectors to methanol extracts of seeds of *S. xanthocarpum*

Mosquito species	Regression Equation	Chi-Square (DF)	24hr LC ₅₀ (Fiducial limits)	24hr LC ₉₀ (Fiducial Limits)
<i>Anopheles culicifacies</i>	Y=2.77x-0.05	0.95 (2)	66.9 (52.4-85.4)	194.2 (121.0-311.7)
<i>Anopheles stephensi</i>	Y=2.48x+0.35	0.99 (2)	73.7 (58.3-93.3)	241.1 (146.1-397.7)
<i>Aedes aegypti</i>	Y=2.73x-0.72	0.68 (2)	123.8 (98.6-155.4)	364.0 (224.6-590.0)
<i>Culex quinquefasciatus</i>	Y=2.51x-0.50	3.11 (2)	154.9 (122.0-196.7)	501.2 (280.3-896.0)

Table 6. Log probit regression analysis of the mortality data of larvae of different mosquito vectors to petroleum ether extracts of seeds of *S. xanthocarpum* after six months

Mosquito species	Regression Equation	Chi-Square (DF)	24hr LC ₅₀ (Fiducial limits)	24hr LC ₉₀ (Fiducial Limits)
<i>Anopheles culicifacies</i>	Y=2.54x-0.39	2.07 (2)	131.7 (79.3) (102.2-169.7)	419.9 (214.6-821.3)
<i>Anopheles stephensi</i>	Y=1.89x+0.80	1.89 (2)	195.6 (95.6) (124.3-220.8)	787.4 (356.1-1741.0)
<i>Aedes aegypti</i>	Y=2.01x-0.19	0.94 (2)	377.6 (131.1) (252.2-565.4)	1633.0 (601.7-2434.0)
<i>Culex quinquefasciatus</i>	Y=1.73x-0.30	0.28 (2)	520.0 (222.7) (304.7-887.8)	2860.0 (782.7-3450.0)

Values in bold are the 48hr LC₅₀ for that species

Longitudinal studies for Disease dynamics in desert ecosystem of Rajasthan: Study of socio-cultural, socio-economic and epidemiological aspects of malaria-SP Yadav, AK Dixit and RK Kalundha

Commencement: **January, 2005**

Duration: **Two years**

Status: **Concluded**

Objectives

1. Study of health seeking behaviour of desert population and determination of socio-economic, socio-cultural and educational basis of behaviour.
2. Study of migration of human population as risk factor for import and spread of desert diseases with special reference to malaria.
3. Study of inter-relationship of sociological, biological and epidemiological components for malaria.

Rationale

Desert is a system of peculiar adaptive abilities. At a glance, it appears as a stable system having acquired its own characteristics for centuries. To develop a long term and reliable strategy for monitoring and controlling desert diseases, a complete profile of desert is needed through launching continuous longitudinal studies in its selected representative areas. Proposed study has been aimed to generate baseline data needed to understand human and its interaction with environment to maintain viable health status. The basic health relevant information on socio-cultural, socio-economic and education status of human population in desert areas will generate a basis for understanding transition of any disease in desert with special reference to vector-borne diseases, for example malaria.

Progress of the work

During the year 2005-06 total households in all the 60 villages of Ramgarh PHC were covered for base line data and presented in SAC meeting which was held on 23rd & 24th June, 2006. Data of 573 malaria patients related with treatment seeking behaviour was analysed and reported during the year 2006-07. Fever was used as a proxy for malaria. Data was collected on how the disease was diagnosed including self diagnosis (an individual diagnosing himself as having malaria without direct involvement of another person), where the treatment was sought first and reasons for choosing a particular care provider. Information was also sought from the second and third places where the patient sought treatment for the same episode, in case they did not recover from the first malaria episode. The respondents who could not read or write were considered illiterate while those who could write their own name were taken as just literate. A socio-economic status (SES) index was used to examine whether there are systematic differences in seeking for diagnosis and treatment by socio-economic group and the specific choice of care provider, with reasons for non-use of government health facilities. To construct a relative index of socio-economic status, household-level information on assets and household income were combined. Least poor, poor, very poor and poorest were classified based on ownership of radio,

bicycle, motorcycle, television, jeep, tractor and cost of food. The cost of food was used as proxy to income as it is difficult to collect reliable information on household income.

Table 1 depicts the socio-demographic characteristics of the respondents. About two third 382 (66.7%) respondents were male and 344 (60.0%) in the age group of 20 to 39 years of age. Nearly three fourth 436 (76.1%) were Hindus. Among the Hindus, 51.1% were SC/ST followed by OBC (33.9%) and FC (Forward Caste) (14.9%). Sixty four per cent were illiterate and the occupation of the respondent was mainly agriculture (33.5%), animal keeping (26.5%), labour (23.4%) and artisan job (9.1%). A few (5.8%) were in service. Majority (56.9%) were earning <5000/= rupees per month with the collective efforts of the total household members. The average family size was 6.2. Most of the households owned a radio set, camel cart and cycle. However, very few owned a fridge, television set, motorcycle, jeep and tractor. More than ninety (91.7%) per cent respondents were having household 'tanka' for the storage of water with a capacity of 5000 to 10,000 liters. 'Tanka' is man charged like well structure. Some 'tankas' are covered while others are opened with cooling effects less than 30°C in summer when out side temperature is more than 40°C and reverse in winter when inside temperature is more than 20°C when out side temperature is less than 4°C. In this way, the 'tankas' are found always suitable for the breeding of the *An. stephensi* which is the dominating species of the area and responsible for desert malaria. All most all the houses were storing water generally in pots for their drinking purposes as well as domestic uses apart from the 'tanka'. Around half of the respondents (51.2%) were having 5-7 pots, 33.4% were having more than 7 pots and 15.4 % were having less than 5 pots for storing of water and all the pots were kept together. More than ninety per cent (93.1%) were earthen pots with the capacity of 10 to 40 liters. The respondents were distributed according to socio-economic status. Utensils with capacity of 25 liters or more used by 16.7% of households were not made empty before filling them a fresh.

The word used for malaria in the local language, "EKANTRA TAV", covered a broad symptom complex of disease which consistently corresponds to the clinical case definition of malaria. Majority 461 (80.5%) of the respondents reported that malaria was an important health problem which had various causes, including poor diet 63 (11.0%), environmental conditions 86 (15.0%), and the bites of mosquitoes 317 (55.3%). The symptoms associated with malaria were quite varied and ranged from generally 'feeling unwell' to a specific fever diagnosis of 'a rise in body temperature with chill and rigors on alternate day 394 (68.8%)'. Malaria was perceived as a relatively mild illness and much less severe as compared to measles, difficulty in breathing and tuberculosis 412 (71.9%). The impact of malaria on the health was clearly visible as 88.3% of the malaria patients reported their working capacity being reduced during indisposition. This led the rural folk to follow certain taboos as more than half (54.2%) of the respondents avoided fried foods but preferred 'Rabadi' (local preparation made from millet flour and yogurt), 'Khichadi' (a semi-liquid preparation from the mixture of rice and pulses), 'Thuli' (a semi-liquid preparation from wheat daliya) and 'Mateera' (fruits of a cucurbitaceous plant akin to watermelon). Table 2 depicts time laps between occurrence of malaria fever and initiation of action for its management. It was found that 72.1% cases start their action on third day or onwards to manage the malaria fever. Treatment seeking behaviour and health facility utilization by desert population is an important factor for the policy implications in the national malaria control programme. Factors which influence which treatment sources people should seek when symptoms occur include socio-cultural factors like beliefs and household decision to seek care, social networks, gender and economic status.

Table 3 shows self diagnosis 423 (73.8%) was more common among the respondents. This was followed by diagnosis through laboratory tests, 12 (2.1%) community health workers such as MPW/ANM 42 (7.3%), family members 27 (4.7%) and traditional healers 69 (12.1%). Treatment at

home was extremely common among them. Out of 573 cases of febrile illness, 443 (77.3%) were treated at home with local herbal remedies as the first treatment such as Akda (*Calotropis procera*) milk mixing with sand and making small round balls which can be swallowed easily or with water in morning and evening for three days or a glass of Neem (*Melia azadirachta indica*) leaf juice in the morning before taking anything for 3 to 7 days or patients put into earth (sand dune) pit and cover up to neck for at least three hours for worming up the body, extreme sweating etc. when ever ekantra fever (malaria) comes. If it was failed to cure the malaria fever, medicines were purchased and doses and courses based on self knowledge about anti malarial drugs or on the advised of the family members/friends/person having the experiences of malaria in past/shopkeeper/dealers from local medical stores where he/she purchased and the poorest households were more likely to use shops/medical general stores for the malarial fever.

There was not statistically significant difference across economic status. Also the least poor were more likely to use the general hospitals for the malarial fever. Generally, poor household patients believe that they would be cured as earliest as possible in private health sector as compared to government health sector and they do not loose their income which is based on the daily work and they also thought that government health facilities, supply of drugs were not of good quality and attention was also poor. If it also could not cure fever at last the formal health sector such as government health facility etc was sought. Commercially available chloroquine preparations were perceived as more effective than either antipyretics or herbal remedies for the treatment of malaria, and injections were regarded as more effective than oral medications 408 (71.2%). Self diagnosis was practiced more (86.9%) by the poorer households; on the contrary richer households were more (90.3%) likely to use the health facility such as Government and private health facility. The richer households complained more (91.2%) about poor staff attitude and lack of drugs as their reasons for not attending the Government health facilities. Even attendance at a health centre did not ensure adequate treatment because of the common practice of sharing medicines among family members and few of them kept medicines for next malaria episodes. Improper and incomplete treatment of malaria led to drug resistance among the patients and as their level of knowledge about the treatment of malaria they lost their faith in malarial treatment at the government health facility. There were clear cut indications that introduction of developmental activities in the public sectors have made a shift in the utilization of Public services, increasing the use of other treatment sources such as private health facilities. Many reasons have also been advanced for households' and individuals' treatment seeking behaviour for the treatment of malaria. These include knowledge and duration of sickness, the anticipated cost of treatment, the patient's judgment of the intensity of sickness, accessibility to health facilities, level of endemicity in the population and demographic characteristics. There was found an association between treatment behaviour and outcome of malaria in children in the study households.

Inferences

The study indicates the importance of socio-cultural factors in malaria treatment. This also indicates that socio-cultural factors influencing the treatment seeking behaviour in high and low economic group of study population. Misconceptions about malaria have been found in study population. It was found that 72.1% cases start their action on third day or onwards to manage the malaria fever. Low socio-economic group of population was taking double time to avail health facility between the occurrence of the malaria and diagnosis and treatment as compared to high socio-economic group of population. Distribution pattern of malaria cases among different age groups of inhabitants of different socio-cultural groups of study population was also found different.

Important leads/outcomes from the study

The study reveals the importance of socio-cultural factors in malaria transmission, prevention and treatment seeking behaviour. Based on this, intervention could be planned for malaria control.

Work remains to be done

Follow-up and objective specific data collection could be done periodically in future.

Table 1. Socio-demographic characteristics

Characteristics	Number	Percentage
Age (Years)		
<20	107	18.7
20-29	153	26.7
30-39	191	33.3
40-49	69	12.0
>50	53	9.2
Sex		
Male	382	66.7
Female	191	33.3
Religion		
Hindu	436	76.1
Other than Hindu	137	23.9
Caste		
GC	65	14.9
OBC	148	33.9
SC & ST	223	51.1
Education		
Illiterate	367	64.0
Literate	105	18.4
Primary	63	11.0
Middle and above	38	6.6
Occupation		
Agriculture	192	33.5
Animal keeping	152	26.5
Labour	134	23.4
Artisans job	52	9.1
Service	33	5.8
Others	10	1.7
Household Income per month (Rs.)		
<5000	326	56.9
5000-10000	211	36.8
>10000	36	6.3

Table 2. Time taken for first step action for treatment of malaria fever

Period	Number	Percentage
1 st day	35	6.1
2 nd day	125	21.8
3 rd day	174	30.4
> 3 day	239	41.7
Total	573	100.0

Table 3. Distribution of subjects according to utilization of type of diagnosis for malaria

Type of diagnosis	Number	Percentage
Self diagnosis	423	73.8
Diagnosis through laboratory	12	2.1
Diagnosis through MPW/ANM	42	7.3
Diagnosis through family members	27	4.7
Diagnosis through traditional healers	69	12.1
Total	573	100.0

A study of the potential interventional variables associated with delay in diagnosis/treatment of pulmonary tuberculosis (PTB) cases in the Thar Desert of Rajasthan. S.P. Yadav and M.L. Mathur

Commencement: **April, 2004**

Duration: **3 years**

Status: **Ongoing**

Objectives

1. To measure the time lapse between disease onset and diagnosis/or starting treatment of PTB under DOTS.
2. To find out the causes for delay in diagnosis/treatment for PBT

Progress of the work

During reported period 156 new cases more were included in the study . were reported at District Tuberculosis Centre (DTC), Jodhpur and CHC, Balesar for medical check up and laboratory investigation for the confirmation of the tuberculosis. The sputum samples, three times (same day, next day morning and next to next day) of all the subjects were examined. Disease history was collected. Sputum slides were prepared and microscopy was done to see the presence of *Mycobacterium tuberculosis*. Thus, the data of 443 new PTB patients were summarized and reported. New positive case was defined as whose sputum was first time examined and found positive for *Mycobacterium tuberculosis* and old positive case whose sputum was examined and found positive more than one time at least 8 weeks interval after taking ATT. Out of 443 new cases 387 (87.4%) cases were delayed in their diagnosis and treatment. Table 1 depicts socio-demographic characteristics of the delayed study subjects. About twelve percent (12.1%) subjects were < 20 years of age, 23.3% between 20-30 years, 37.5% between 30-39 years, 18.3% between 40-49 years of the age group and 8.8% were > 50 years of age. About fifty eight per cent (58.4%) were male and 41.6% female. About seventy five (75.2%) subjects were Hindus, 24.8% were other than Hindus. Among the Hindus 20.3% were general caste, 30.2% OBC, 49.5% SC/ST, 48.8% Illiterate, 25.3% Literate, 17.4% Primary, 8.5% Middle School and above, 5.2% at home, 11.6% house wife, 14.7% service, 52.2% labour and 16.3% were doing other jobs. Nearly thirty five per cent (35.1%) were living in urban area and 64.9% were belonging to the rural area. About fifty six per cent (56.1%) were smokers, 26.4% alcoholics and 17.1% opium addicts.

Majority (70.5%) belonged to low socio-economic group, followed by Middle income group (23.0%) and high income group (6.5%). Source of referral was one of the important factors for suspected case reporting to examination and treatment. It was found that majority (42.9%) of the cases were reported to DTC and PHC by own or on the suggestions of the TB patients who had taken experiences of diagnosis and treatment of the disease or by the motivation of their relatives and friends. About thirty nine (38.5%) subjects were referred by government hospitals and 18.6%

referred by private practitioners (Table 2). The study subjects (76.2%) were complaining cough with expectoration followed by fever (66.5%), chest pain (49.6%), breathlessness (38.2%), loss of weight (33.3%), haemoptysis (14.5%) and loss of appetite (24.5%) Table 3. The period of delay is depicted according to category wise in table 4. Illiterate as compared to literate, smokers as compared to non-smoker, rural cases as compared to urban cases and low income group as compared to high income group were delayed more in diagnosis/or treatment. Table 5 shows the causes for the delay in diagnosis/treatment of the tuberculosis patient. Most dominating cause of delay in diagnosis /treatment was lack of awareness among the patients about causation of disease, signs and symptoms of the disease, preventive and curative measures as a result failed to recognize symptoms of disease by the patient (76.2%) and did not report to the doctors for the diagnosis and treatment.

Inferences

There is a substantial proportion of patients (87.4%) of Pulmonary Tuberculosis (PTB) who delay the diagnosis and treatment of the disease.

Out of this 87.4% of patients delaying, nearly 91.2% makes delay of more than 1 month which is a dangerous period of delay for infection such as PTB.

Lack of awareness was the major cause of delay in 76.2% of patients who delayed.

Socio-Demographic characteristics suggest that patients belonging to illiterate, low income and SC/ST & OBC are more vulnerable groups in the population.

Suitable interventional method needs to be find out to reduce the delay of diagnosis and treatment under DOTS.

Important leads/outcomes from the study

The study revealed the duration and causes of delay in diagnosis and treatment of the tuberculosis in the desert part of Rajasthan. Suitable intervention programme would be identified which may help in the reduction in delay in diagnosis and treatment under national tuberculosis control programme.

Table 1. Socio-Demographic characteristics of the study subjects

Characteristics	Number	Percentage
Age		
<20	47	12.1
20-29	90	23.3
30-39	145	37.5
40-49	71	18.3
>50	34	8.8
Sex		
Male	226	58.4
Female	161	41.6
Religion		
Hindu	291	75.2
Other than Hindu	96	24.8
Caste		
General Caste	59	20.3
OBC	88	30.2
SC & ST	144	49.5
Education		
Illiterate	189	48.8
Literate	98	25.3
Primary	67	17.4
Middle School & above	33	8.5
Occupation		
At home	20	5.2
House wife	45	11.6
Service	57	14.7
Agr., Agri. Lab & Labour etc	202	52.2
Others	63	16.3
Place of Living		
Urban	136	35.1
Rural	251	64.9
Personal Habits		
Smokers	217	56.1
Alcoholics	102	26.4
Opium addicts	66	17.1

Table 2. Distribution of subjects according to economic status and referral source

Characteristics	Number	Percentage
Economic Status		
Low Income Group	273	70.5
Middle Income Group	89	23.0
High Income Group	25	6.5
Source of referral		
Self/sufferers/ relatives	166	42.9
Private Practitioners	72	18.6
Government Hospital	149	38.5

Table 3. Complaints of the study subjects at the time of Survey

Complaints	Number	Percentage
Fever	234	66.5
Cough with expectoration	295	76.2
Haemoptysis	56	14.5
Loss of appetite	95	24.5
Loss of weight	129	33.3
Chest Pain	192	44.6
Breathlessness	148	38.2

Table 4. Category wise Period of delay

Parameter	<1 Month	1-2 Month	2-3 Month	>3 Month	Total
Educational status					
Illiterate	8 (4.2)	12 (6.3)	26 (13.8)	143 (75.7)	189 (100.0)
Literate	30 (15.2)	98 (49.5)	57 (28.8)	13 (6.6)	198 (100.0)
Economic Status					
Low	9 (3.3)	61 (22.4)	56 (20.5)	147 (53.8)	273 (100.0)
Middle	11 (12.6)	40 (44.9)	30 (33.7)	8 (9.0)	89 (100.0)
High	15 (60.0)	7 (28.0)	2 (8.0)	1 (4.0)	25 (100.0)
Place of living					
Rural	25 (10.0)	57 (22.7)	79 (31.5)	90 (35.9)	251 (100.0)
Urban	57 (41.9)	39 (28.7)	28 (20.9)	12 (8.8)	136 (100.0)
Source of Referral					
Self/Sufferers/relatives	7 (4.2)	10 (6.0)	13 (7.8)	136 (81.9)	166 (100.0)
Private practitioners	2 (2.8)	9 (12.5)	43 (59.7)	18 (25.0)	72 (100.0)
Government Hospital	31 (20.8)	96 (64.4)	18 (12.1)	4 (2.7)	149 (100.0)
Personal Habits					
Smoker	17 (7.8)	35 (16.1)	73 (33.6)	92 (42.4)	217 (100.0)
Non-smoker	35 (20.6)	67 (39.4)	48 (28.2)	20 (11.8)	170 (100.0)

Table 5. Reasons for delay in diagnosis/treatment

Reasons	Number	Percentage
Lack of Knowledge	295	76.2
Others	92	23.8
Total	387	100.0

Sub-clinical vitamin-A deficiency among children (6-71 months) of Assam and Rajasthan –
R. C. Sharma, K. R. Haldiya and Madhu B. Singh

Commencement: June 2005

Completion: September 2006

Status: Concluded

Objectives

1. To study the magnitude and distribution of the problem of sub-clinical vitamin-A deficiency among children of representative districts of Rajasthan.
2. Association of distribution of magnitude with agro climatic status of study areas.
3. Determination of current status of vitamin-A deficiency and prioritization of preventive/curative measures for supplementation.

Progress of Work

A multicentric task force study as conceptualized by the ICMR, New Delhi and jointly sponsored by ICMR and UNICEF was undertaken by DMRC as one the centre's. Study involved following steps:

1. Selection of study areas on the basis of agro-climatic characteristics

- To select the representative area sample for estimating distribution of prevalence of sub-clinical deficiency of vitamin-A, the state of Rajasthan was divided into different agro-climatic zones so that a representative sampling of the magnitude of disease could be developed. To represent the above agro-climatic conditions, 7 districts viz; Jaisalmer (Arid western plains), Ajmer (Semi-arid Eastern plains), Sri Ganganagar (Irrigated north-western plains), Sikar (Transitional plains of Inland Drainage), Kota (Humid south-eastern plains), Bharatpur (Flood prone eastern plains) and Dungarpur (Humid southern plains) have been selected for collecting blood samples for vitamin-A deficiency (Figure 1).
- Area wise blood samples of children in the age group of 6-71 months have been collected with recording of corresponding demographic details in the pre tested schedules.
- The blood samples have been sent to Department of Bio Chemistry Hamdard University, Delhi, for quantitative estimation of vitamin-A.

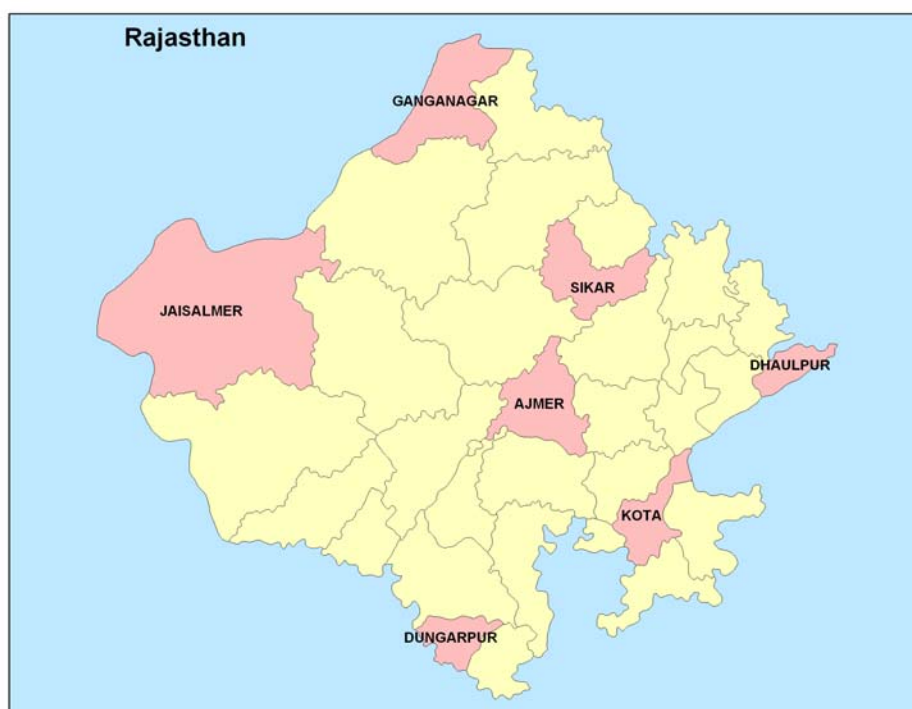


Figure 1: Map Showing study areas of Sub-clinical vitamin-A deficiency in Rajasthan

- The District wise sub-clinical vitamin-A levels are shown in Table 1. Data indicates that deficiency in serum retinol (< 20 mg/dl) was observed maximum in Bharatpur (25.5%) and lowest in Jaisalmer district (12.2%). The overall profile of serum retinol deficiency in the examined population of Rajasthan was found to be 17.4 % who were found to be deficient below 20mg/dl.

Table 1. Prevalence of vitamin-A deficiency (sub-clinical) among children in districts of Rajasthan (preliminary Analysis)

Parameter	Jaisalmer	Sikar	Sri-Ganganagar	Dungarpur	Kota	Bharatpur	Ajmer	Total
No. of samples analyzed	353	393	407	426	423	416	416	2834
Serum retinol <20mg/dl	43 (12.2)	76 (19.3)	64 (15.7)	45 (10.6)	103 (24.6)	106 (25.5)	55 (13.2)	492 (17.2)

Possible outcome and Utilization

Present study is very important to highlight the non-symptomatic cases of vitamin-A deficiency that may emerge to be clinical cases of in prospective time. Since the study pertains to the children in the age of (6-76), present study will serve to be of great predictive significance and timely preventive measures thereon.

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

Commencement: **January, 2005**

Duration: **Long Term**

Status: **Ongoing**

Objectives

- 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.
- Aim at doing comparisons with other states data so as to assess the percentage of variation among the states.

Progress of the work

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

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

A total of 30 villages were covered from six tehsils of Jodhpur district i.e. Jodhpur, Bilara, Osian, Phalodi, Shergarh and Bhopalgarh (five villages from each tehsil), covering 600 households. Second phase has been started in which 50 percent new villages and 50 percent old villages had been selected randomly in view to cover variation. In the second phase data has been collected from 10 villages from Jodhpur and Osia tehsil covering 200 households. The entire selected household were examined for Socio-demographic and Socio- economic aspects. All the members in the household

have been examined for nutritional deficiency signs, anthropometric measurements (Height, weight, arm circumference and FFT), Dietary intake (24 hours recall method) and examination of Nutritional Morbidities in preceding 15 days. Information on dietary intakes of the individuals were recorded in alternate houses i.e. 10 households from each village are covered.

Analysis of 600 households covering 3540 individuals of the first phase has been done so far. Table 1 showed age and sex wise distribution of population (1859 males and 1681 females). Analysis revealed that 94.1 percent of populations were Hindus and 4.7 percent were Muslims. Nuclear families were more (59.0 %) as compare to joint families i.e. 25.2 percent (Tables 1 & 2). Table 3 revealed that illiteracy is high in females (54.3 %) than males (29.5 %). Higher education is very low in this area.

Table 1. Age and sex wise distribution of population covered

Age group	Males	%	Females	%	Total	%
0-5	293	15.8	260	15.5	553	15.6
6-9	212	11.4	168	10.0	380	10.7
10-14	248	13.4	218	12.9	466	13.2
15-17	133	7.1	108	6.4	241	6.8
18-29	353	19.0	351	20.9	704	19.9
30-39	204	11.0	194	11.5	398	11.2
40-49	157	8.4	161	9.6	318	9.0
50-59	114	6.1	109	6.5	223	6.3
>=60	145	7.8	112	6.7	257	7.3
Total	1859	100.0	1681	100.0	3540	100.0

Table 2. Distribution of households according to type of family

Type of family	N	%
Nuclear	354	59.0
Extended Nuclear	95	15.8
Joint	151	25.2
Pooled	600	100.0

Table 3. Distribution of population according to educational status

Educational status	Males	%	Female	%	Total	%
Illiterate	548	29.5	913	54.3	1461	41.2
Read & Write	26	1.4	22	1.3	48	1.4
1-4 Standard	420	22.6	337	20.0	757	21.4
5-8 Standard	322	17.3	141	8.4	463	13.1
9-12 Standard	278	15.0	58	3.5	336	9.5
College	64	3.4	7	0.4	71	2.0
N. A.	201	10.8	203	12.1	404	11.4
Pooled	1859	100.0	1681	100.0	3540	100.0

Main morbidities observed in population were acute respiratory infection (5.4 %) and fever (4.5 %) followed by diarrhea (1.1 %). Both the morbidities i.e. acute respiratory infection and fever were higher in females than males (Table 4). Regarding nutritional deficiency signs, it is observed that discoloration and sparseness of hair, a sign of protein calorie malnutrition was observed to be high i.e. 6.9 percent which was higher in females than males. Marasmus was observed only in females (0.2 %). Angular stomatitis, chelosis and glossitis were ranging from 0.4 to 1.6 percent. Vitamin A deficiency (Bitot Spot) was 0.3 percent, higher in males than females. Dental caries (31.2 %) and dental fluorosis (25.2 %) observed high in this area. Females suffered more from dental caries and dental fluorosis than males (Table 5). Thyroid palpable and visible were 0.6 percent. Koilonychia, a sign of anemia, was observed higher in females (0.2 %).

Table 4. Distribution of population according to Morbidity

Morbidity	Males	%	Females	%	Total	%
N.A.D.	1672	89.9	1469	87.4	3141	88.7
Fever	66	3.6	94	5.6	160	4.5
Diarrhoea	21	1.1	17	1.0	38	1.1
Dysentery	4	0.2	2	0.1	6	0.2
A. Res. Infection	93	5.0	99	5.9	192	5.4
Measles	0	0.0	0	0.0	0	0.0

Table 5. Distribution of population according to Nutritional deficiency signs

Deficiency Signs	Males N=1859	%	Females N= 1681	%	Total N=3540	%
Hair Discolour	96	5.2	116	6.9	212	6.0
Hair Sparseness	7	0.4	24	1.4	31	0.9
Emaciation	6	0.3	7	0.4	13	0.4
Marasmus	0	0.0	3	0.2	3	0.1
Bitot spots	9	0.5	2	0.1	11	0.3
Angular Stomatitis	6	0.3	7	0.4	13	0.4
Chelosis	29	1.6	29	1.7	58	1.6
Glossitis	10	0.5	9	0.5	19	0.5
Koilonychia	1	0.05	3	0.2	4	0.1
Gums-spongy Bleeding	33	1.8	60	3.6	93	2.6
Dental Caries	471	25.3	632	37.6	1103	31.2
Dental Fluorosis	400	21.5	491	29.2	891	25.2
Thyroid Palp.	6	0.3	2	0.1	8	0.2
Thyroid Visible	12	0.6	1	0.05	13	0.4

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. 81.4 percent which was higher in SC community (86.4 %) in comparison to other communities (76.5 %). The overall prevalence of

severe under nutrition was high i.e. 38.2 percent. Under nutrition was higher in nuclear and joint families (81.9 %) and observed maximum in semi-pucca houses (88.2 %) followed by pucca houses. Overall under nutrition was highest in low-income group (36.8 %) and decline with the rise in income (Fig. 1).

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

Type of Family	N	Nutritional Grades*			
		Normal	Mild	Moderate	Severe
Nuclear	310	18.1	29.7	18.7	33.5
Extended	83	21.7	25.3	14.5	38.5
Joint	160	18.1	23.8	11.3	46.8
Pooled	553	18.6	27.3	15.9	38.2

* NCHS Standards

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

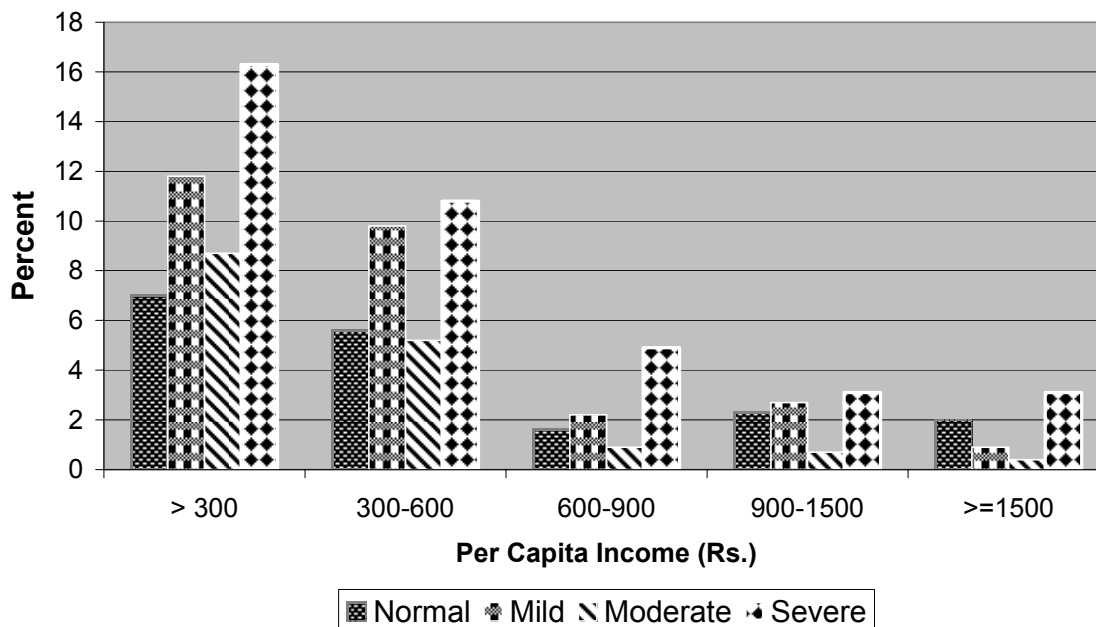


Table 7. Distribution of 1-5 years children according to Gomez distribution and type of house

Type of House	N	Nutritional Grades			
		Normal	Mild	Moderate	Severe
Kutchra	110	19.1	28.2	15.5	37.2
Semi Pucca	144	11.8	26.4	19.4	42.4
Pucca	299	21.7	27.4	14.4	36.5
Pooled	553	18.6	27.3	15.9	38.2

* NCHS Standards

The extent of different types of malnutrition viz. stunting (Height for age) and under nutrition (Weight for age) were computed by adopting standard deviation classification using NCHS standards. All the children with any of the above anthropometric measurement less than Median - 2SD of NCHS values were considered as undernourished. Prevalence of under nutrition computed using Gomez classification and SD classification differ, as the cut off values are different. Stunting (Height for age) was 71.6 percent in preschoolers with the prevalence of severe stunting 56.8 % which needs attention. Stunting was higher in females. Its higher than NNMB (57.7 %) and NFHS II (36.1 % - up to 3 years) Fig. 2.

Underweight (Weight for age) in preschoolers observed 71.6 %, higher than NNMB (62.4 %) and NFHS II (50.6 %). The proportion of severe underweight was high (50.3 %) Fig. 3.

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

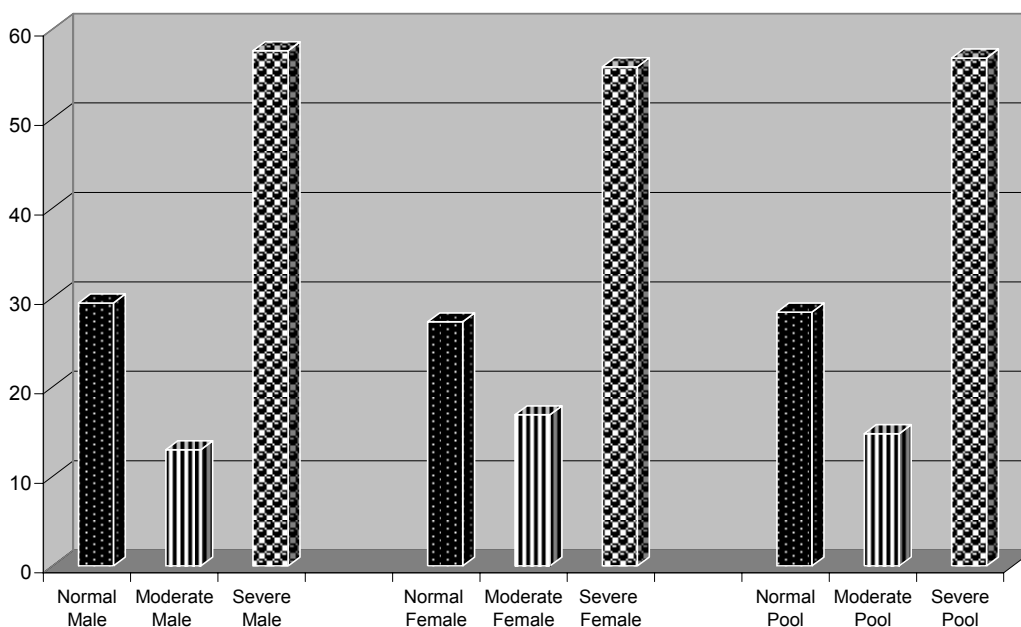
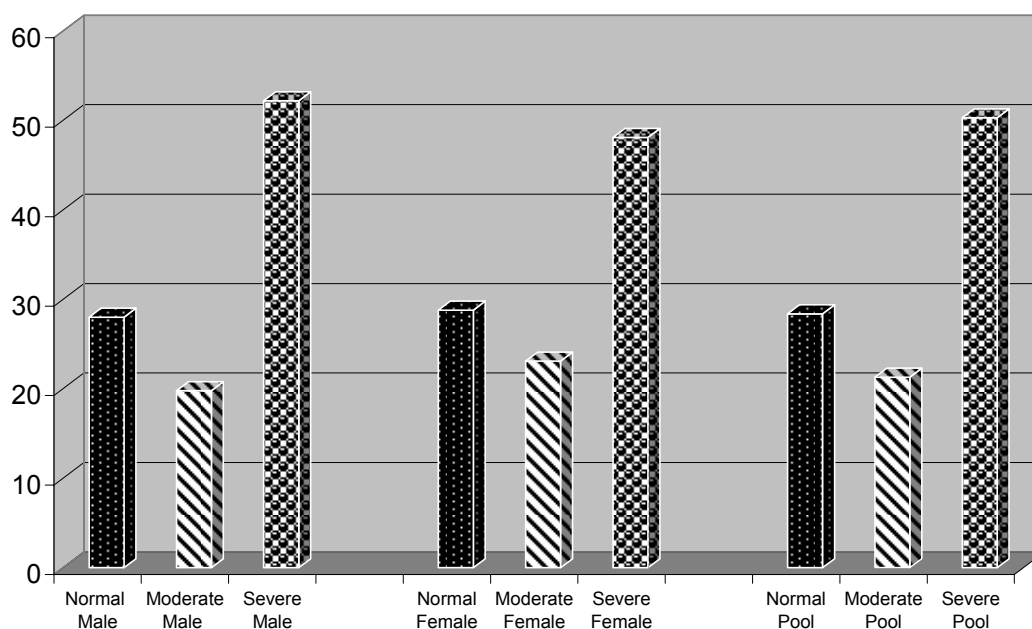


Fig. 3. Standard Deviation Classification for Weight for age in preschoolers



The distribution of adults according to BMI grades have been shown in Tables 8 to 10. At the aggregate level, 57.2 percent had normal BMI (18.5-25.0), while 35.0 percent had chronic energy deficiency (Fig. 4). Severe chronic energy deficiency was higher in extended nuclear families (82.1 %) than nuclear families (6.5 %) and maximum in Semi pucca houses (7.9 %). Chronic Energy deficiency declined with increase of per capita income i.e. 13.3 percent in low income (<300 Rs.) and 2.7 percent in high income group (≥ 1500 Rs.).

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

Type of Family	Obese II ≥ 30	Obese I 25-30	Normal 20-25	Low wt Normal 18.5-20	CED I 17-18.5	CED II 16-17	CED III <16
Male N=449	0.4	4.9	36.7	19.7	19.6	11.6	7.1
Female N=699	2.1	7.1	36.5	21.2	18.2	7.6	7.3
Pooled N=1148	1.5	6.3	36.7	20.5	18.7	9.1	7.2

Fig. 4. Body Mass Index in adults

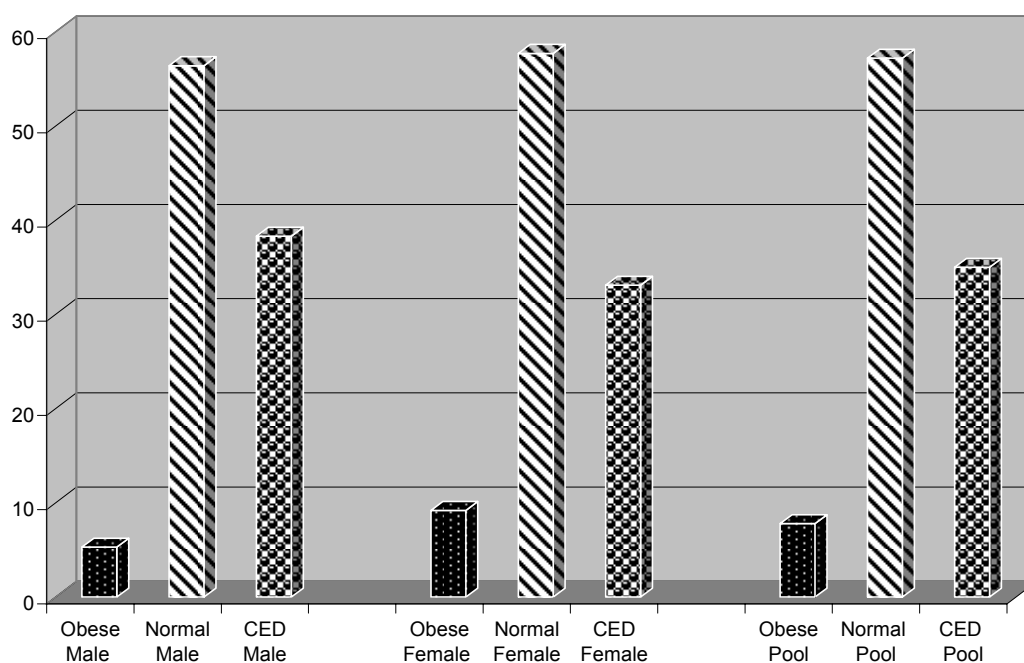


Table 9. Distribution of adults (>=18 years) according to BMI classification and Type of family

Type of Family	Obese II >=30	Obese I 25-30	Normal 20-25	Low wt Normal 18.5-20	CED I 17-18.5	CED II 16-17	CED III <16
Nuclear N=589	1.0	5.3	36.8	19.7	21.2	9.5	6.5
Extended N=220	1.4	6.8	33.6	24.1	15.4	10.5	8.2
Joint N=339	2.3	7.7	38.1	19.8	16.5	7.7	7.9
Pooled N=1148	1.5	6.3	36.7	20.5	18.7	9.1	7.2

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

Type of House	Obese II >=30	Obese I 25-30	Normal 20-25	Low wt Normal 18.5-20	CED I 17-18.5	CED II 16-17	CED III <16
Pucca N=178	0.6	2.2	36.5	27.0	20.2	7.9	5.6
Kutchra N=257	0.8	7.8	31.5	19.4	21.8	12.1	6.6
Mixed N=713	1.9	6.7	38.4	19.4	17.3	8.4	7.9
Pooled N=1148	1.5	6.3	36.7	20.5	18.7	9.1	7.2

Dietary analysis of 300 households covering 1565 individuals (824 males and 741 females) revealed low consumption of Cereals & Millets (80 %), very low consumption of Pulses & Legumes (40.5 %), Roots & Tubers (58.4 %), Fats & Edible Oil (45 %) of RDA. Green Leafy Vegetables consumption was extremely low i.e. 8.5 percent of RDI only.

Average energy intakes (Calories) was less than RDI in all age groups i.e. 1-3, 4-5, 7-9, 10-12, 13-15, 16-17 years and adults ranging from 59.0 to 99.2 percent. Calorie deficit was observed more in girls and highest in 4-6 years group followed by 1-3 and 7-9 years groups.

Deficiency in protein intakes was observed in 16-17 years age group. Diet of preschoolers and juvenile adolescents was deficient in fats (19.6 to 34 %) and maximum in 1-3 years age groups. In all age groups Vitamin A intakes in their diet was below the RDI, ranging from 16.8 to 39.8 percent. Preschoolers suffered from Thymine and Riboflavin deficiencies in their diet. Diet of all individuals in all age groups was deficient in intakes of Vitamin C, maximum in 1-3 years age group (26.1 % of RDI).

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 also to assess the impact of the nutritional programs currently in progress and for future planning in the state of Rajasthan.

Nutritional Status along with micronutrient deficiency disorders and morbidity in pregnant and lactating women in desert areas of Rajasthan - Madhu B. Singh, Ranjana Fotedar and J. Lakshminarayana

Commencement: **July, 2004**

Duration: **2 years**

Status: **Concluded**

Objectives

1. To what extent the micronutrients deficiency disorders i.e. Iron deficiency Anemia, Vitamin A deficiency disorder and Iodine deficiency disorder are prevalent in pregnant and lactating women
2. To assess the nutritional status of pregnant and lactating women by means of anthropometry, dietary intake as well as through clinical examination
3. To study the morbidity profile of diseases among the rural pregnant and lactating women of the desert areas of Rajasthan and
4. To develop nutrition package for pregnant and lactating women of the desert areas based on the findings of the study, during the second phase of the study.

Progress of the work

Project prelude activities included development of protocol, formulation of detailed sampling design and preliminary field visits to the studied area.

Studies were conducted in twenty-eight villages belonging to Luni Panchayat samithi of Jodhpur tehsil, Jodhpur district. A total of 1193 households have been covered. At each household level information for demographic and socio-economic aspects have been collected. Pregnant and lactating women found in these villages along with control group of non pregnant and non lactating women have been interviewed / examined for the nutritional deficiency signs, dietary pattern (24 hours recall method) along with morbidity survey and Micronutrient deficiency disorders i.e. Anemia, Iodine and Vitamin A. Anemia was assessed by clinical signs (platinichia and koilonichia). Hemoglobin levels have been estimated using Cyanmethaemoglobin technique and classified according to WHO classification. Iodine deficiency disorders have been assessed by clinical examination of thyroid gland using the standard method as recommended by the joint WHO / UNICEF / ICCIDD. Casual urine sample were collected for estimation of Urinary Iodine Excretion (UIE) levels to assess the Iodine nutriture status. Iodine was determined by WET digestion method using standard laboratory technique. UIE level less than 100 mcg/l have been considered as indicator of iodine deficient nutriture. Twenty gram of salt consumed in household was collected from 60 percent of women selected randomly in autoseal LDPE pouches. Iodine content of salt sample have been estimated using standard iodometric titration method. Salt samples having iodine content less than 15 ppm classified as with inadequate iodine.

Vitamin A deficiency was assessed by administering pretested semi structured questionnaire on the presence of symptoms of night blindness. The women were asked specific questions regarding their seeing at the time of sunset and later in dim light, had any problems in cooking during dusk period due to lack of proper vision and any change in their activity pattern due to vision problems in the dusk. The women with positive response to these questions have been classified as suffering from VAD.

Analysis of 1193 individuals have been done out of which 384 were pregnant, 400 lactating women and 409 NPNL women (control). The inclusion criteria for the pregnant mothers were those mothers whose pregnancy was more than 24 weeks of gestation period. Those mothers were not registered / excluded whose gestation period was less than 24 weeks and with null pregnancy. The inclusion criteria for the lactating mothers were those mothers who were breast feeding their child and the cut off level for breast feeding was up to 6 months period. The inclusion criteria for the control group was those who were 15 years and above women (unmarried) or mothers who were more than 15 years, just married living together not attained their family status by becoming pregnant because till this age, the girls did not stay with their husband even though they were married.

Table 1 show distribution of women according to anemia on the basis of haemoglobin estimation. Analysis revealed that anemia was observed to be maximum among pregnant women and lactating women and least in control group. Severe anemia observed 3 fold higher in pregnant and lactating women (10.5 to 14.0 %) in comparison to control group (4.1 %).

Table 1. Distribution women according to anemia (Haemoglobin Estimation)

Women	Non-anemic		Anemic					
	Normal (≥ 11 g/dl) * (≥ 12 g/dl) **		Mild (10-11 g/dl) * (10-12 g/dl)**		Moderate (7-10 g/dl)		Severe (<7 g/dl)	
	N	%	N	%	N	%	N	%
Pregnant* N = 371	81	21.8	44	11.9	194	52.3	52	14.0
Lactating** N = 382	64	16.8	121	31.7	157	41.1	40	10.5
Controls** N=391 (NPNL)	111	28.4	163	41.7	101	25.8	16	4.1
Total N=1144	256	22.4	328	28.7	452	39.5	108	9.4

Education and income appeared to play important role in distribution of anemia (Table 2 & 3). Anemia declined as education and income raised.

Table 2. Education wise distribution of anemia (Hemoglobin Estimation)

Hemoglobin Estimation	Pregnant		Lactating		Controls	
	Anemic N=290		Anemic N=318		Anemic N=280	
	N	%	N	%	N	%
Illiterates	220	75.9	258	81.1	99	35.4
Primary	49	16.9	39	12.3	56	20.0
Middle	16	5.5	12	3.8	55	19.6
Secondary & above	5	1.7	9	2.8	70	25.0

Table 3. Income wise distribution of anemia (Hemoglobin Estimation)

Hemoglobin Estimation Income	Pregnant		Lactating		Controls	
	Anemic N = 290		Anemic N = 318		Anemic N = 280	
	N	%	N	%	N	%
Low Income Group	111	38.3	137	43.1	105	37.5
Middle Income Group	78	26.9	89	28.0	62	22.1
High Income Group	101	34.8	92	28.9	113	40.4

Consumption of iron folic acid tablets by anemic pregnant and lactating women observed to be low (39.0 to 48.0 %). Abortions, child deaths, still births and premature births observed to be higher in anemic pregnant and lactating women in comparison to normal women (Table 4).

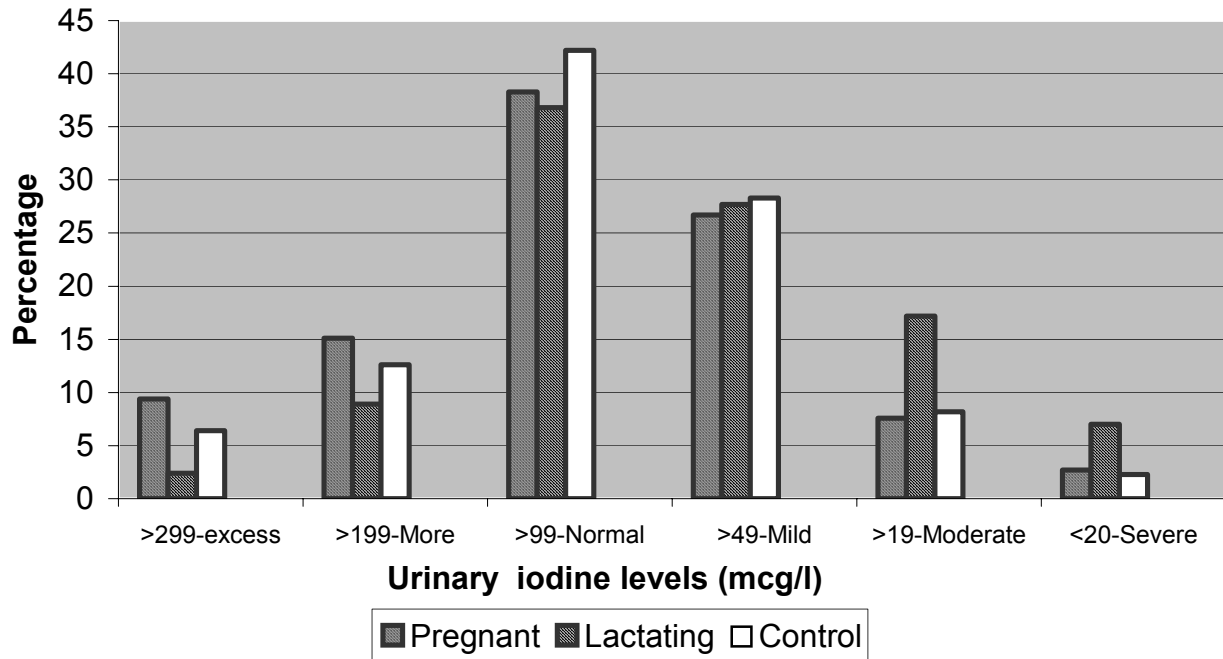
Table 4. Distribution of women according to anemia (Hemoglobin Estimation) and Obstetric history

Hemoglobin Estimation (N=753) Obstetric History	Pregnant (371)				Lactating (382)			
	Normal		Anemic		Normal		Anemic	
	N=81	%	N=290	%	N=64	%	N=318	%
Abortions	4	1.1	33	8.9	11	2.9	28	7.3
Premature Births	0	0.0	8	2.2	2	0.5	7	1.8
Still Births	2	0.5	12	3.2	1	0.3	18	4.7
Child Deaths	12	3.2	45	12.1	12	3.1	34	8.9

Epidemiological criteria, as prescribed by WHO, for assessing iodine nutrition is based on median urinary iodine concentrations / levels as shown in figure 1. Analysis of 1049 urine samples have been done so far. Trend revealed that the median urinary iodine values were observed less in

lactating women (85 mcg/l) according to WHO cut off points (100 mcg/l) where as pregnant and control observed to be just on the marginal values. Nearly 34 to 42 percent pregnant and lactating women suffered from mild to moderate iodine deficiency disorder whereas in control group it was only 34.6 percent. Severe iodine deficiency disorder was observed almost twice in lactating (14.2 %) than control group (7.3 %). Iodine deficiency disorders (UIE levels) showed increasing trend with decline of income and educational status.

Fig. 1 Distribution of Women according to UIE level



Iodine content of 719 salt samples have been estimated using standard iodometric titration method (Table 5). Overall high proportion of women (80.8 %) consumed salt having inadequate iodine content i.e. less than 15 ppm. Consumption of salt deficient in iodine content observed higher in low income (38.9 to 44.9 %) and illiterate women (nearly 80 %) than high income and higher educated women.

Table 5. Iodine content in salt intake of women in different physiological groups

Iodine content in salt→ Women ↓	Normal ≥15 ppm		Mild 10 - <15 ppm		Moderate 5- <10 ppm		Severe < 5 ppm	
	N	%	N	%	N	%	N	%
Pregnant N=256	58	22.7	31	12.1	62	24.2	105	41.0
Lactating N=222	26	11.7	29	13.1	55	24.8	112	50.4
Controls N=241	54	22.4	28	11.6	66	27.4	93	38.6
Total N=719	138	19.2	88	12.2	183	25.5	310	43.1

Sickness at the time of survey was highest in lactating women (9.2 %) followed pregnant (6.5 %). Main morbidities observed to be aches (3.8 %) and gastroenterological complaints (2.8 %). Vitamin A deficiency based on Night blindness observed higher in pregnant women (8.8 %) than controls (0.9 %). Thyroid enlargement observed in 3.1 % pregnant women whereas in controls it was 0.5 %.

Analysis of dietary intake revealed that consumption of cereals and fat observed to be low in pregnant and lactating women i.e. 76 to 84 % and 80 % of Recommended Dietary Allowances (RDA), ICMR. Consumption of pulses and legumes was very low i.e. 47 to 65 % of RDA and very low consumption of leafy vegetables i.e. 12 and 7 percent of RDA in pregnant and lactating women but the consumption of milk and milk foods was found to be adequate. Analyzing the Average intake of nutrients (per day in diet) showed high deficiency of protein and calories in pregnant and lactating women (20 to 40 %) along with high deficiency of Iron and Folic Acid i.e. 36.3 and 57 % and Vitamin A deficiency (20.1 % in lactating women).

Important Leads / Outcome

Results revealed the magnitude and distribution of three micronutrients deficiency disorders i.e. Iron, Vitamin A and Iodine along with the nature and type of nutritional deficiencies and their morbidities. Secondly it also revealed dietary factors i.e. consumption of food stuffs and average intake of nutrients per day in the studied women. Trend indicated iron deficiency anemia to be very high and observed to be maximum among pregnant and lactating women (80.7 %) whereas 71.6 per cent in control group. Severe anemia observed 3 fold higher in pregnant and lactating women (10.5 to 14.0 %) in comparison to control group (4.1 %). Abortions, child deaths, still births and premature births observed to be higher in anemic pregnant and lactating women in comparison to normal women. Vitamin A deficiency based on Night blindness observed higher in pregnant women (8.8 %) than controls (0.9 %). Overall high proportion of women (80.8 %) consumed salt having inadequate iodine content i.e. less than 15 ppm. Nearly 34 to 42 percent pregnant and lactating women suffered from mild to moderate iodine deficiency disorder whereas in control group it was only 34.6 percent. Consumption of pulses and legumes was very low i.e. 47 to 65 % of RDA and very low consumption of leafy vegetables i.e. 12 and 7 percent of RDA in pregnant and lactating women. Analyzing the Average intake of nutrients (per day in diet) showed high deficiency of protein and calories in pregnant and lactating women along with high deficiency of Iron and Folic Acid and Vitamin A deficiency. These results helped in developing the database for micronutrient deficiency disorders along with nutritional deficiencies and morbidity in pregnant and lactating women in desert areas. These results will be helpful in formulating nutritional intervention packages for this region by introducing the adequacy i.e. bioavailability - of iron, and vitamin A etc. in usual diets which can be improved by altering meal pattern to favour enhancers or lower inhibitors or both during the second phase of the study. According to WHO, at present, Iron and Vitamin A supplementation are the most common strategy currently used to control these deficiencies in developing countries for the time being. This is likely to remain the case until either significant improvements are made in the diets of entire populations or food fortification is achieved.

Meta Analysis of Micronutrient Deficiency Disorders and their Mapping in Western Rajasthan - *J. Lakshminarayana, Madhu B .Singh and Ranjana Fotedar*

Commencement: August. 2006

Duration: One Year

Status: Ongoing

Objectives

1. Meta Analysis of available published or unpublished information of three important Micronutrient Deficiency Disorders namely Vitamin A, Iron Deficiency Anemia(IDA) and Iodine Deficiency Disorders.
2. To study regional/ecological variations of these three micronutrient Deficiency Disorders in Rajasthan by systematic reviews and
3. To map out the districts for three micronutrient deficiencies in the western region.

Progress of the work

Data base has been created by searching relevant literature through Pub Med, MEDLARS, EMBASE Chochrane on micronutrients and survey reports of NFHS-I and II, ICMR multi-centric studies, compendium on Micronutrients published by ICMR, NNMB technical report on Micronutrient Deficiency's, and using FOX-PRO package for searched literature. There are a total of 90 studies on various Micronutrients Deficiency Disorders (MDD's) published from Rajasthan. Some of them are case control studies and cohort studies. Systematic search was done for unpublished MD thesis from medical college libraries of Jodhpur and Jaipur. Reports from NGOs covering data reported from Jaipur, Kota, Bharatpur, Ajmer, Udaipur, Jodhpur, Jaisalmer, Barmer and Bikaner districts has been collected. The Literature related to all the intervention studies, prevalence, case control and cohort studies on MDD's in Rajasthan and reports published from DMRC has been collected for re-analysis and creation of database. DMRC has done field based studies in population groups related to Vit. A, Iron Deficiency Anemia, but all are based on clinical signs and symptoms from different ecological zones. Intervention studies on MDD's are vary few and these are to be given attention & priority among different vulnerable population groups in the communities. In India many intervention studies were taken up by ICMR institutes, NGO's and Govt. organizations. The collected data will be re-analysed using EPIMETA Software.

The data for meta analysis was collected through literature search for the Vitamin A Deficiency (VAD). Systematic review of these meta analysis was carried out. Data consist of 13 publications on the effect of Vitamin A among school children The duration was considered to be for a period of 10 years. Binomial model Has been used to estimate the results and rank correlation proposed by Begg and linear regression method proposed by Egger & Trim and Fill method to evaluate bias. The publication bias was adjusted by using "Trim & Fill method. The analysis was done using EPI META package.

The studies with at least 10 subjects per intervention group were included. 84 articles were evaluated, out of which only 20 met the criteria for the meta analysis. Multiple interventions of study samples were identified in 13 of the articles. Each intervention was counted as a separate study, yielding 25 cohorts totaling 1892 subjects. Of these subjects, 51% received interventions and 49% served as controls. Cohorts reported between 1992 and 1999 accounted for 53% of the sample; 56% of all cohorts were based in hospital and 26% involved micronutrient deficiencies. There are two type of interventions reported one is on Behavioral interventions accounted for 11 cohorts (885 subjects), and educational interventions for 8 cohorts (392 subjects), and combined interventions for 19 cohorts (645 subjects). Homogeneity of groupings and effect sizes (ESs) were calculated for each type of intervention. Overall, the data were not homogeneous, so conclusions could not be derived from the entire body of data

The prevalence of Iron Deficiency Anemia (IDA) in Rajasthan in different age groups has been depicted in Table 1 and vitamin A deficiencies among different age groups table 2 are school and community based figures from rural & urban areas. The sub clinical vitamin A of beta carotene among infants was also low reported from Udaipur in a community based study by Jain and Tahiri. The percent prevalence of iodine deficiency disorders among Boys and Girls of 6-12 years of age 18% among boys and 39% among Girls where as 98% were graded as Iodine deficient in as per ICMR study Table 3. The Urinary Iodine Excretion levels based on Biochemical analysis among 6-12 years age group 30% of children are iodine deficient among children of Bikaner district as per reported by Bhardwaj Table 4 and NFHS study, The district wise average intake of nutrients (per cu/day) in Rajasthan. The nutrient intakes are very low in many districts as compared with Recommended Dietary Allowances (RDA) Values of ICMR Table 5. The percent prevalence of clinical nutritional deficiencies the values are not significantly deferring from states combined data when compared with Rajasthan shown in urban rural data as per ICMR multicentric study Table 6, The prevalence of Iron Deficiency anemia based on Hemoglobin levels based on WHO cut off values, in Rajasthan among pregnant and lactating women Table 7. A total of 94% of pregnant women and 90% adolescent girls are anemic where as DMRC reported 80% pregnant women and 85% lactating women are reported to be anemic among covered population. The re-analysis of the data revealed that the educational intervention and combined intervention table 8 and the cohorts were non-homogeneous ($P < 0.001$ and $P < 0.01$, respectively); however, the behavioral intervention cohort was homogeneous ($Q = 32.48$, d.f. = 35, $P=0.36$). The overall ES for behavioral interventions was 0.06 (95% confidence interval [CI] = 0.03–0.08); there were no significant differences among the behavioral & educational interventions which had an overall ES of 0.10 (95% CI = 0.05–0.16). The overall ES of the combined interventions was 0.08 (95% CI = 0.04–0.14). While stratifying the combined interventions the behavioral had the largest impact (ES = 0.38). Meta Analysis of studies of interventions improved the heterogeneity among the studies.

Table 1. Iron Deficiency Anemia (IDA) in Rajasthan

Pop. Groups	Year	No. of persons	Age (years)	% Prev.	Reference
Infants	2000	1852	6-35 Months	82.3	NFHS-II
Adolescent Girls –	2000	329	15-18	50.5 <12 gms	NFHS-II
Bikaner CB	2001	300		90.3	ICMR
Pregnant & Lactating Women					
Jaipur	1983	171	15-45	88.9	Gupta et al
Jodhpur	1989	40P 40L	>=18	100 P <10.8 100 L <12.4	Rewtani & Verma
Udaipur	1992	302	>=18	98.3	ICMR
Bikaner	2001	510	>18	94.9	ICMR
State Pooled	2000	586	>=18	51.4	NFHS

CB-Community Based, SB- School Based Study

Table 2. Vitamin – A Deficiency (VAD) in Rajasthan

District	Year	No. of persons	Age	% Prev.	Reference
Udaipur	1973	566 – Tribal 495 – NT	1 – 5	2.4 - BS 4.8 -	Gupta & Bhandari
Udaipur	1975	839	5 – 17	9.6 - BS	Bhandari et al
Bikaner SB R & U	1977	1000 - U 1000 – R	6 – 12	5.9 – R - BS 1.8 – U - BS	Gupta & Saxena
Jodhpur U & R - C.B	1986	1877	1 – 5	3.62 - NB 6.8 - BS	Indira Bai
State Level	1989	2573	0 – 6	1.24 - NB 0.62 - BS	Madan Mohan
State Level	2000	1774	2 - <5	0.1 - NB	MICS
Bikaner - CB	2001	10730 7686	<6 2 - <6	1.1 - BS 0.96 - NB	ICMR
State Level	2000			14.7 - NB	NFHS - II
Jodhpur	1998	400	1-5	4.4 -R	DMRC, Singh et al
Jodhpur	2001	800	6-12	3.0 -R	DMRC Singh et al

CB-Community based study SB- school based study R-Rural U-Urban

Sub Clinical Vitamin A Status among Infants

	Year	Individuals studied	Age	Vit – A μ g/100 ml	B-Carotene μ g/100 ml	Ref.
Udaipur	1981	60	1 – 5	38.4 \pm 7.4	146.2 \pm 45.8	Jain
CB		20	Exophthalmia	14.6 \pm 5.4	65.5 \pm 35.0	Tahiri

Table 3. Iodine Deficiency Disorders (IDD)

District	Year	Subjects	Age Group	% Prevalence
Bikaner – R SB	1997	466 – Boys 61 - Girls	6 – 12 Bhandari	18.0 – Boys 39.3 – Girls 20.5 – Pooled
Bikaner – CB	2001	9827	6 – 12 ICMR	98.3 Grade – I 0.01 Grade – II 9.86 Pooled

SB – School Based, CB – Community Based

Table 4. Urinary Iodine Excretion levels

Children	Year	No. of individuals	Age Group (years)	UIE (mcg/l)	% Children.
Bikaner SB – R	1997	400	6 – 12 Bhardwaj	< 20 20 – 49.0 50 – 99.0 ≥ 100.0	3.0 9.0 18.0 70.0
Bikaner – SB	2000	400	6 – 12 U-Kapil	< 20 20 – 50.0 50 – 100.0 ≥ 100.0	3.0 9.0 18.0 70.0
Bikaner – CB	2001	1824	6 - <12 ICMR	118.0	
Jodhpur-R DMRC	2006	1193	≥ 18+ Singh et al.	< 20 20 – 50.0 50 – 100.0 ≥ 100.0	6.3 13.3 37.3 43.1

SB- School Based CB- Community Based R-Rural

Table 5. District wise average intake of nutrients (per cu/day) in Rajasthan

State/District	Vit – A	Vit – C	Iron
Rajasthan Combined	400	46	31
Rural	395	45	32
Urban	423	51	25
R.D.A	600	40	28
Ajmer	139.4	33.2	25.0
Alwar	319.0	57.5	30.9
Banswara	131.1	17.0	23.3
Baran	406.2	57.5	29.7
Barmer	191.1	39.7	41.5
Bharatpur	319.0	49.2	44.1
Bhilwara	598.2	85.4	35.1
Bikaner	177.4	38.3	39.4
Bundi	469.0	73.2	30.8
Chittorgarh	370.8	61.1	28.9
Churu	205.0	49.8	43.5
Dausa	342.5	50.2	31.0
Dholpur	207.9	61.5	28.2
Dungarpur	86.7	16.8	20.9
Ganganagar	326.3	63.4	35.3
Hanumangarh	397.7	33.2	29.7
Jaipur	117.7	35.1	30.2
Jaisalmer	157.2	35.1	26.2
Jalore	170.5	32.3	29.1
Jhalawar	73.7	34.3	18.6
Jhunjhunu	198.1	49.6	26.1
Jodhpur	189.5	36.0	34.9
Kota	332.3	50.9	26.3
Nagaur	183.6	36.8	32.2
Pali	235.9	32.1	28.2
Rajsamand	145.3	33.4	20.3
Sikar	127.5	39.8	30.5
Sirohi	142.2	37.4	25.4
Sawaimadhopur	217.4	31.5	31.0
Tonk	383.4	60.1	35.5
Udaipur	263.2	41.2	20.2
POOLED	355.4	56.7	23.0

Source: Micronutrient Profile (ICMR)

Table 6. Percent prevalence of Nutritional Deficiency Signs

	B. Spots	Cor. Xeros.	Cor. Opec.	Keratom.	Goiter
Rajasthan – Bikaner					
Combined	0.22	0.16	0.02	0.09	0.05
Rural	0.25	0.18	0.02	0.11	0.03
Urban	0.09	0.07	0.02	0.01	0.06
States Pooled					
Combined	0.21	0.11	0.05	0.05	0.65
Rural	0.23	0.12	0.05	0.05	0.74
Urban	0.11	0.07	0.04	0.03	0.26

Table 7. Prevalence of Anemia (Hb levels) in Rajasthan

	N	Non-anemic	Anemic <11	Mild >10-11.9	Mod. 7-10	Severe <7
Children 6-35 months Rajasthan		11.7	82.3	20.1	52.7	9.5
Adolescent Girls	300	29 (9.7)	271 (90.3)	56 (18.7)	142 (47.3)	73 (24.3)
Pregnant Women	510	26 (5.1)	484 (94.9)	34 (6.7)	255 (50.0)	195 (38.2)
Married women (pooled)		51.5	48.5	32.3	14.1	2.1
Pregnant	175	29 (3.8)	140 (22.2)	25 (3.3)	93 (12.1)	22 (2.9)
Lactating	286	34 (4.4)	244 (36.3)	98 (12.8)	121 (15.8)	25 (3.3)
Control	332	75 (9.8)	244 (41.6)	143 (18.7)	92 (12.0)	9 (1.1)

Table 8. Showing Calculations of Estimates for meta analysis Odds Ratios of different studies

Study Ref.	a	b	C	d	Sample size (n)	Var.	Weight	O.R.	Weight x OR	Q
1	2	14	33	22	71	0.15	6.51	0.1	0.62	2.88
2	1	3	22	19	43	0.80	1.25	0.17	0.22	0.43
3	12	39	44	24	109	0.09	11.7	0.23	2.64	3.34
4	7	6	11	13	37	0.56	7.18	0.37	2.68	1.07
	7	6	11	13	37	0.56	7.94	2.37	18.83	20.61
13	46	23	97	115	281	0.13	100.21		75.85	34.745

Appendix: Calculation of various parameters for case control studies and cohort studies for conducting meta analysis are as follows:

- Estimate the variance of the ORs for each study
- Variance $i = n_i / (b_i * c_i)$ for $i=1,2,3,\dots$
- Calculate the weights for each study
- Weight = $1/Var$
- Calculate the product of the weights * ORs
- Product = ORs * W
- Calculate the sum of the weights
- Calculate the sum of the product of Weights & ORs and $OR_h = \text{Sum of products wt/sum of the wt}$
- Calculate $Q = \text{Sum } W(\text{Log ORs} - \text{Log } OR_h)^2$
- Test the hypothesis by Chi-square tables with $(n-1)$ d.f.
- Estimate the variance of the ORs for each study
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- Calculate $Q = \text{Sum } W(\text{Log ORs} - \text{Log } OR_h)^2$
- Test the hypothesis by Chi-square tables with $(n-1)$ d.f.

Work remaining to be carried out

The systematic data collection through internet search etc., is being continued and medical colleges of Udaipur, Ajmer, Bikaner and kota has to be visited for secondary data collection and the reanalysis are to be done once data collection is over by METASTAT package.

Household distribution of disease burden in community health management: A study in desert - A. K. Dixit and P. K. Anand.

Commencement: **Oct. 2004**

Duration: **2 Years**

Status: **Concluded**

Objectives

1. Identification of the distribution of disease burden at household level
2. Identification of household level risk factors of disease burden
3. Locating the pockets of population needing prioritized health care attention

Rationale

Burden of disease as measured by DALYs is now widely accepted and adopted for reporting and comparing population health. DALYs account for the suffering period also for the morbidities and simultaneously takes in to account the mortality. The global burden of disease (GBD) study by WHO relies on national level secondary data. A need was felt however to measure Burden Of Disease (BOD) at local level to strengthen local health policy, thought to be effective in management of BOD at community level through identifying the pockets of people needing prioritized health care attention. This would provide useful interventions to address disease burden in the community. A study was therefore designed to read disease burden at household (H/H) level, which is basic unit in the community in which health of an individual is a function of all the other members of the H/H. The disease burden at H/H level is a newer concept used to account for overall burden of diseases found at H/H. It sums BOD of all diseases identified among the members of the H/H. The study aimed to look for physical, economical and social characteristics of the H/H and to correlate them with total disease burden (DB) at H/H in order to determine household level risk factors of disease burden.

Progress of the work done

898 H/H from 30 villages in Jodhpur district were surveyed following standard sampling procedures. Associations of village level characteristics with disease burden found at village level provided clues of villages carrying bigger load of disease burden. It has been observed that availability of a health facility in the village reduces total disease burden at village level by 42.37%, Just from primary level to provision of secondary level education in the village hits the disease burden by 64.82% and development activities in terms of road and electricity reduced the burden by 71.78%. This analysis provides clue to catch hold of the type of villages where the problem may lie. To identify H/H with in such villages, study of the associations of H/H characteristics with H/H DB was done, apart from obtaining the distribution of H/H DB and age, sex distribution of the disease burden in the study population. The associations of H/H characteristics with H/H level disease burden were measured in terms of OR (Odds Ratio), using median of the distribution of H/H level disease burden as cut off point to decide upon the H/H, carrying more disease burden within each defined class of H/H characteristics. OR is thus computed with respect to each

characteristic taking in to account its presence, and its absence as the presence of other characteristics in that class. Besides this, share of each characteristic in average DB expressed in percentage (DB%) was also computed.

Observations

Table 1 gives the distribution of disease burden at H/H level, which meets our objective 1. The distribution, is found to be highly skewed. It is quite interesting to note from here that 90% of DB% is shared by 15% of H/H. Identification of such H/H is our objective 3, which is attempted through studying the association of H/H characteristics with H/H DB.

Table 2, which gives age and sex wise distribution of disease burden percentage (DB %) reveals the increase in DB% with the increase in age. DB% of men when compared with that of women indicates that up to the age of 45, women are better off than men; however in older ages, whereas women carry about 65% of DB%, men carry only 45% of DB%.

Tables 3 through 13 adhere to our objective 3. From Table 3, it is observed that DB% shared by mixed houses is of the tune of 39% with OR (3.96).

Small living area is also observed to be considerably associated with H/H level DB%, as depicted in table 4. 56% of average DB% is shared by the condition of living area being less than 100 sq. ft. with OR (1.95).

It is quite interesting to note from table 5, that not much difference exists with respect to provision of electricity in reducing H/H DB%. However having no electricity is associated with 52% sharing of DB% and OR=1.12.

Cattle keeping in villages are common in desert. If the number of cattle is less (1-5), usually they are kept within the premise of the H/H, else at a place located well outside the H/H premise. Keeping cattle within premise of H/H is found to be associated with H/H DB, as is observed from table 6 that 66% of DB% is shared by this factor with OR=1.12.

Investigations in to highest level of education in a family reveals from table 7 that illiteracy shares ¼th (24%) of DB% with OR=1.44. Even primary or secondary level of highest education of a member in the family does not serve the purpose better, as still it is associated with DB% in H/H with OR=1.55 and 1.12 respectively.

Socio economic condition of poor and middle class families enjoys almost equal share in DB% of H/H and are equitable factors with their share of 44 and 46% of DB% along with OR=1.10 and 1.16 respectively; as is observed from table 8. Only high-income group is found to be associated with low DB% (9.7%).

Drinking water, its storage and interval of change etc have also been the factors of our concern for their influence in increased H/H DB%. It is observed from table 9 that water from wells is the highest risk factor in this category with its share of 18% and OR=2.20. Even tap water inside house is not found safe and is associated with H/H DB with 20% share along with OR=1.33. Storage of drinking water in Matki or in Tanka share equal risk. However, as is observed from table 10, neither of them qualifies to be a risk factor.

Interval of change of drinking water is observed to be a risk factor from table 11, if this change is every 3 days or more. More than 50% of DB, then is shared with OR >1. Thus, emptying the drinking water storage pots at least every 3 days and refill them for further use is the safer course of action. In desert, due to scarcity of drinking water there is practice to store water in Tanka, build outside the house. Practice to change water in these tankas differs from H/H to H/H. It has been of our concern to look for the effect of this interval of change in terms of H/H DB. From table 12, it is observed that highest risk is, when water in them is changed in 6 months time, with DB share of 35% and OR=1.47.

Naturally, poor sanitation gives rise to the risk of H/H DB. It is observed from table 13 that 59% of share owes to this risk factor with OR=1.07.

Though these observations are found by and large logical, at places they at present are hard to be explained. Data analyzed separately for DOB of infectious and non-infectious diseases may better explain. This is being done.

Outcome

Investigations into various H/H characteristics influencing the risk of H/H level disease burden, it is brought under observation that apart from the age, which enhances much burden among women of older age, there are many other H/H level factors contributing to H/H DB. Study of these factors as per the tables shown suggests for certain important issues in reducing H/H level disease burden.

Mixed house structure is found to enhance the risk of H/H DB. Therefore their completion should be encouraged. Living area should exceed 100 sq ft, as below that, it becomes the risk factor for H/H DB. Cattle keeping in houses are common in desert. This is also a risk factor. Cattle keeping outside the house should be encouraged as people do it when the cattle are good in number. Educating one or two persons in the family does not serve the purpose in reducing DB% at H/H level. In fact, each member of the H/H should be encouraged to be educated. Only enhancing the socio-economic status from poor to middle does not bring down the H/H DB, as perhaps it is the life style of the people, which associates much better with DB. Therefore the use of income in enhancing better living standard should be encouraged. Drinking water is important. People should ensure that the supply line in their houses is not cracked or broken.. Storage pots inside the house be made empty at least every 3 days and refilled, while tankas outside house be cleaned at least every six months. Sanitation is also found to play its role in reducing DB. Good sanitation should be encouraged.

The factors discussed above, identify a H/H which may carry bigger load of disease burden. The pockets of people needing prioritized health care attention would be identified by combining the village and H/H level factors associated with H/H DB. This would be the realization of our objective number 3. For this, supplementary analysis of the collected data is to be done to prioritize the risk factors identified, looking into their interactive behavior.

Important leads

With the identification of village and H/H level risk factors of H/H level disease burden, suitable interventions may be planned to reduce the disease burden in the community.

Table 1. Distribution of H/H according to DB%

DB%	No. of H/H	H/H %
< 10	764	85.08
10-20	61	6.80
20-30	27	3.01
30-40	15	1.67
40-50	10	1.11
50-60	8	0.89
60+	13	1.44
Total	898	100

Table 2. Distribution of DB% according to age and sex.

Age groups	Male		Female	
	Individuals carrying health problems	DB%	Individuals carrying health problems	DB%
Less than 1	7	0.12	3	0.02
1-4	7	1.39	3	0.01
5-14	21	11.55	16	4.72
15-24	18	1.65	7	0.75
25-34	37	11.37	40	4.44
35-44	43	9.85	51	7.04
45-54	56	18.79	67	19.36
55 & above	100	45.28	94	63.66
Total	289	100	281	100

Table 3. Distribution of DB% as per the type of house.

Type of House	No. of H/H	DB%	OR
Kutchha	353	33.85	1.09
Pucca	4.2	27.11	1.08
Mix	143	39.04	3.96
Total	898	100	

Table 4. Distribution of DB% as per the living area

Living Area	No. of H/H	DB%	OR
<100 sqft.	36	56.02	1.95
100-200 sqft.	498	19.79	0.77
200 sqft.	364	24.19	1.20
Total	898	100	

Table 5. Distribution of DB% as per the provision of electricity.

Provision of Electricity	No. of H/H	DB%	OR
Yes	410	47.82	0.89
No	488	52.18	1.12
Total	898	100	

Table 6. Distribution of DB% according to pattern of cattle keeping.

Cattle Kept	No. of H/H	DB%	OR
1-5	850	66.79	1.12
5-10	20	26.33	0.92
10 & more	28	6.88	1.32
Total	898	100	

Table 7. Distribution of DB% according to highest education level acquired in the H/H.

Highest education level acquired	No. of H/H	DB%	OR
Illiterate	146	23.73	1.44
Literate	94	6.56	0.66
Primary	220	18.23	1.55
Secondary	201	13.64	0.71
Middle	100	20.25	1.12
Secondary	83	11.07	0.92
Sr. Secondary	54	6.52	0.48
Graduation			
Total	898	100	

Table 8. Distribution of DB% according to Socio-economic status.

Socio-eco status	No. of H/H	DB%	OR
Poor	706	43.91	1.11
Middle	124	46.31	1.17
High	68	9.78	0.25
Total	898	100	

Table 9. Distribution of DB% according to source of drinking water.

Source of drinking water	No. of H/H	DB%	OR
PHED supply from village tank and stored rain water	485	22.97	0.85
Tap water in house	176	20.42	1.33
Hand pump	42	10.19	0.93
Pond	146	11.06	0.89
Tanker	36	16.99	0.93
Well	13	18.37	2.20
Total	898	100	

Table 10. Distribution of DB% according to drinking water storage type.

Storage type	No. of H/H	DB%	OR
Matki	682	48.99	0.99
Matki and Tanka	216	91.01	1.00
Total	898	140	

Table 11. Distribution of DB% according to interval of change of water storage in the house (Matki).

Interval of drinking water change	No. of H/H	DB%	OR
Daily	723	32.69	0.93
every 3 days	119	46.87	1.08
every week	56	20.44	1.01
Total	898	100	

Table 12. Distribution of DB% according to interval of change of tanka water located outside the house.

Interval of change of tanka water	No. of H/H	DB%	OR
every month	17	2.92	0.51
every 3 month	26	17.15	2.31
every 6 month	43	21.31	0.85
every year	94	35.04	0.93
more than a year	36	23.58	0.89
Total	216*	100	

*H/H where tankas outside the house were located.

Table 13. Distribution of DB% according condition of sanitation.

Condition of sanitation	No. of H/H	DB%	OR
Poor	682	58.41	1.07
Good	216	41.59	0.93
Total	898	100	

Rapid culture of *Mycobacterium tuberculosis*: A Preliminary Study - Murli L. Mathur

Objective

To study impact of supplementation of LJ Medium to shorten the time required for culture of *Mycobacterium tuberculosis*

Rationale

Culture of *Mycobacterium tuberculosis* from clinical material is the “GOLD STANDARD” for diagnosis of tuberculosis (TB). However culture requires a long time ranging from 2-8 weeks. It is therefore not commonly used as a method for diagnosis of TB. Considering the emerging problem of drug resistance in TB, there is need to shorten the duration of time required for culture of *Mycobacterium tuberculosis* from sputum samples, so that it can be used for rapid drug susceptibility testing aimed at early diagnosis of Treatment Failure and MDR TB at secondary or tertiary health facilities in Revised National Tuberculosis Control Program.

Progress of the work

Enrichment of LJ medium was done with hemolysed human blood and unhemolysed goat blood separately in varying concentrations. LJ Medium without blood enrichment was taken as control. Controls as well as enriched media were inoculated at the same time, with an inoculum of *Mycobacterium tuberculosis* H37RV of strength McFarland standard no. 1.0. Time taken in appearance of macroscopic colonies of MTB was 2 days on LJ medium enriched with 4% hemolysed human blood as compared to 5 days on controls. Similarly time taken in appearance of macroscopic colonies of MTB was 2 days on LJ medium enriched with 1% unhemolysed goat blood as compared to 5 days on controls. However numbers of colonies appearing on second day were higher on media enriched with human blood (5-40 colonies) as compared to goat blood (2-5 colonies).

Attempt was also made to reduce the time taken in culture of MTB by enriching the media with some amino-acids and other nutrients. Enrichment of LJ medium was done in varying concentrations of supplements. LJ Medium without any enrichment was taken as control. Controls as well as enriched media were inoculated at the same time, with equal volume of the same inoculum of *Mycobacterium tuberculosis* H37RV of strength McFarland standard no. 0.5. Time taken in appearance of macroscopic colonies of MTB was 5 days on enriched LJ medium as compared to 10th to 12th day on controls. The numbers of colonies appearing were also higher on enriched media. Thus the time taken in growing MTB on LJ medium could be reduced to 40%. Using the enriched liquid media and microscopic detection of colonies may further shorten the time required and is therefore proposed for further studies.

Important Leads and Outcome

It was observed that time taken in growing MTB on LJ medium could be reduced to 40% by supplementing it with blood, amino acids and other nutrients. Using the enriched liquid media and microscopic detection of colonies may further shorten the time required.

Dermatoglyphic Patterns in *Diabetes mellitus* Patients And Non- Diabetics : A Preliminary Study - Pradeep Kumar Dam, Vinod Joshi, R. C. Sharma , Anil Purohit and Himmat Singh

Objectives

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

Rationale

Dermatoglyphics are the characteristics of the ridged skin on the fingertips palms, toes and soles of primates (including human beings) and some other mammals. They consist of the alignment of the sweat glands pores and are shaped in the first trimester of gestation. Dermal ridges formed during gestational weeks 12-19 is fixed permanently before the midpoint of pregnancy with no change thereafter, so that troublesome allowances for change with age is ruled out (Holt & Penrose 1968, Mulvihill & Smith, 1969, Babler 1979). Unusual dermatoglyphics are reported as disease marker in Diabetes Mellitus (Verbov, 1973, Ziegler *et al.*, 1993, Shield *et al.*, 1995, Kahn *et al.*, 2001) Dermatoglyphics matures at specific times in the foetus when critical growth in the brain is taking place. Size of a fetal fingertip as compared to its neighbor in the same hand is influenced by factors that stimulate or inhibit growth along the developmental axis extending from brain to lower limb (Heimer, 1994) [Fig.1]. The ridge count on each fingertip provides a measure of fingertip growth activity during early foetal life (Loesch, 1983). Programming (i.e. long term changes in physical structure and metabolism) that occurs by environmental influences because the organs and systems in body matures during periods of rapid growth before and immediately after birth; once during such critical windows of times, if maturation is not achieved, failure of such maturation, to some extent is irrecoverable (Dubos *et al.*, 1966, Winick & Noble 1966, Blackwell *et al.*, 1968, Hahn 1984, Barker *et al.*, 1993). A baby of low birth weight and size carries an increased risk of insulin resistance in later life in T²DM i.e. (type 2 Diabetes Mellitus) indicating long term effects of reduced growth of the endocrine pancreas and other tissues *in utero* (Hales *et al.*, 1991, Barker *et al.*, 1993, Phipps *et al.*, 1993, Philips *et al.*, 1994 Valdez *et al.*, 1994 Leger *et al.*, 1997, Mckeigue *et al.*, 1998).

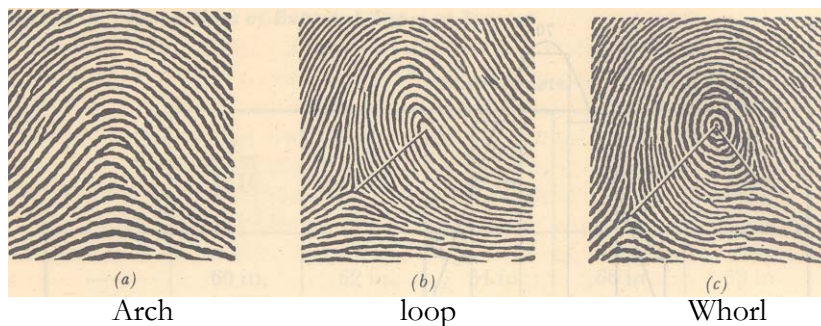
As adverse foetal exposure were to occur before 20th week of gestation, then this effect may be found in the fingerprints of the offspring. Dermatoglyphic features of 290 children and 180 adults with diabetes mellitus revealed a high TRC i.e. Total ridge count value that was more frequent in both girls and boys with diabetes than in controls. (Barta *et al.*,1997). The high concordance observed in monozygotic twins (up to 96%) implies a substantial genetic component to T₂DM (Barnett *et al.*, 1981, Kapiro *et al.*, 1992, Medici *et al.*, 1999). 40% of siblings of T₂DM patients develop diabetes in population, that have a population prevalence of only 6% (Kobberling and Tallil, 1982). Compared with unrelated non-diabetic twins of the same ages, non-diabetic co-twins of diabetic twins had higher glucose levels (P<0.03) (Newman *et al.*, 1987). Segregation analysis suggested a major gene component to disease susceptibility in addition to polygenic component (Ferrel and lyenger 1993, Stern *et al.*, 1996).

Present study would elucidate whether dermatoglyphics pattern can be used as one of the disease marker for the diabetes. Results of a preliminary study undertaken to achieve above, are presented in the report.

Further continuance of the present study would be helpful to formulate counseling messages based on dermatoglyphic pattern prevalent among young generation and their possible simulation to determine the younger people's likelihood to develop diabetes in their later age. Counseling can be imparted through a separate study to the parents of younger generation.

Work Undertaken

Seventeen (17) respondents with Type-1 diabetes from different caste groups and seventeen from age, sex and caste matched non-diabetics controls were interviewed in the PBM hospital, Bikaner. In addition to this, twenty four adult respondents with Type-2 diabetes and 24 age, sex and caste matched non-diabetic controls were interviewed at their households for the family pedigree (Figure-2) and personal history of onset and treatment patterns of diabetes, along with dietary habits. Non-diabetic controls were diagnosed by using *in-vitro* urine reagent strips. Prints were taken of respondents palms and finger tips of right and left hand separately with the help of an ink pad. Then individual prints were scanned and stored in computer for their detailed character study of respective design and ridges



There are three basic types of fingerprint patterns which illustrate how ridge counts are made, viz;

- a) Arch: No triradius, hence ridge-count is zero
- b) Loop (one triradius on the left where 3 ridges meet): To count ridges, a line has to be drawn from the point of the triradius to the centre of the loop. If the ridge count is 13, it means the number of ridges cut by the line is 13.
- c) Whorl (two triradii): The following ridge-counting protocol was used: ridge count= (ridges crossing the longer line only); therefore, the ridge-count values in these examples are A=0, B=13, and C=17 [Adapted from Holt, and penrose 1968]

Dermatoglyphics ridge counting and other parameters quantification in the palm is the most inexpensive and non-evasive method of determining association, as a possible biosensor of certain diseases/ disorders. Bilateral inked impressions of palm and fingertips of subjects are here recorded by the printer's ink followed by ridge-counts.

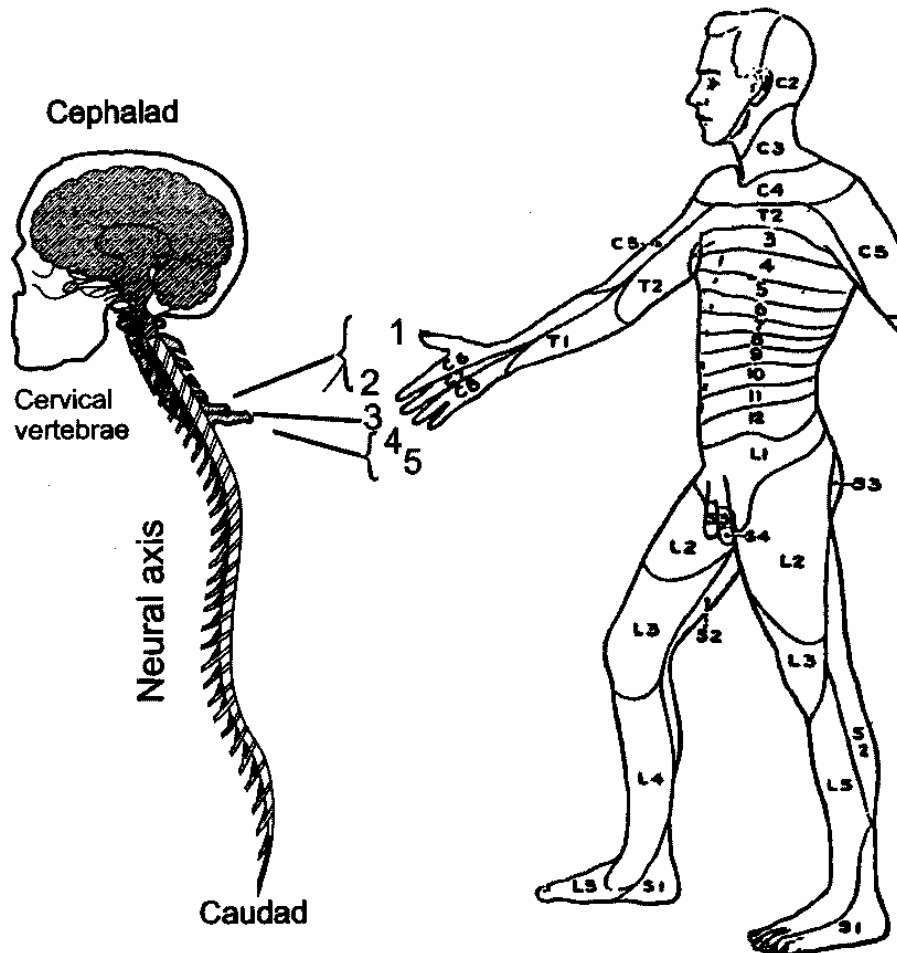


Figure 1. Schematic illustration of the innervations of the fingers, demonstrating anatomic correlations with sixth through the eighth cervical spinal cord segments (C6-C8)

Observation

Out of all the fingerprints collected from $25+17=42$ diabetic cases, present study delineates finger print patterns of 24 cases of type 2, diabetes mellitus and 24 controls. Sex difference for digital pattern was observed by Cummins and Midlo (1961). Frequency of Simian Creases in Caucasian males being approximately twice than that in normal females (Menser & Purvis-Smith, 1972). In present study, difference of ridge counts and palm patterns on right and left hands of 24 male and female Type 2 diabetics and 24 matched controls are shown in Table 1.

Following characteristics were revealed through palm and fingertip print analysis among the diabetics as compared to non-diabetic controls:

- Lower a-b ridge count (i.e. the number of ridges occurring in the second interdigital area between the two A and B digital triradii,
- Lower 3rd finger (Middle) finger ridge count,
- Difference in frequency of occurring true pattern in a) 4th interdigital area b) Thenar area.

Difference of fingertip patterns among male and female diabetics are illustrated in Table 2.

Among women T2 DM cases, middle finger (III) holds 100% occurrence of Loops in both right and left hands, whereas among males, 35.7% whorls and 57.1% Loops, in addition to 7.1% Arch is present in 3rd digit of left hand, accompanied by 66.7% Loops, 25% Whorls and 8.3% Arch in 3rd digit of right hand. This indicates sexual dimorphism in pattern distribution among diabetics.

In 3rd finger of both right and left hands of diabetic cases Loops are 100%, whereas among non diabetic controls, Loops are 100% in 3rd finger of right hand only: 3rd finger of left hand is having 80% Loops and 20% Whorls. Occurrence of Whorls is observed lesser in both the hands of diabetic cases as compared to the controls. No arch is present among controls.

Further, difference of finger tips patterns on ten hand digits among 5 out of 17 type 1 diatetic cases and 5 out of 17 age, sex and caste matched controls are shown in table 3, that reveals existence of arches only in the left hand of the cases, as compared to controlshaving no arch in their fingertips. Third finger of cases shows 100% occurrence of loops only, in both right and left hands, wheras among controls this trend is available in right hand only.

Inferences

To use dermatoglyphics as predictor for diabetes, a larger number of samples in terms of control study pertaining to diabetes need to be undertaken. Genetic factors controlling immune responses are inherited from the father and predispose a particular foetus to these disorders. While dermatoglyphics alone are not specifically abnormal parameters, they remain useful markers of constitutional difference within population. Such studies illustrates the value and necessity of family studies.

Work remaining to be done

Association of fingerprints with diabetes will be made for a sample of about 150 cases and 150 sex matched controls. Molecular basis of all variations will be resolved through appropriate laboratory studies.

Table 1. Percentage of presence of unusual patterns and ridge counts among men and women diabetic patients and matched controls

Cases/ Control	MEN								WOMEN							
	a-b ridge count on palm		3 rd finger ridge count		% Presence of IVth inter-digital pattern		% Presence of Pattern in Thenar area		a-b ridge count on palm		3 rd finger ridge count		% Presence of IVth inter-digital pattern		% Presence of Pattern in Thenar area	
	R	L	R	L	R	L	R	L	R	L	R	L	R	L	R	L
Cases	35.69	37.46	14.30	17.83	61.53	53.84	7.69	7.69	37.18	39.45	13.09	13.5	81.81	63.63	18.18	27.27
Control	42.92	42.15	19.30	20.0	100	84.61	69.23	46.15	50.72	50.36	20.45	20.54	90.90	90.90	63.63	54.54

Table 2. Difference of finger tip patterns on ten hand digits among 24 male and female diabetics

	Left Hand					Right Hand				
	Thumb (I)	Forefinger (II)	Middle finger (III)	Ring finger (IV)	Little finger (V)	Thumb (I)	Forefinger (II)	Middle finger (III)	Ring finger (IV)	Little finger (V)
Male cases										
Arches	-	7.1	7.1	-	-	-	6.7	8.3	-	-
Loops	58.3	35.7	57.1	25.0	69.2	46.1	46.7	66.7	25.0	69.2
Whorls	16.7	50.0	35.7	75.0	30.7	46.1	33.3	25.0	75.0	30.8
Twin Loops	25.0	7.1	-	-	-	7.7	13.3	-	-	-
Total	100	99.9	99.9	100	99.9	99.9	100	100	100	100
Female cases										
Arches	-	7.1	-	-	-	-	6.25	-	-	90.9
Loops	45.4	71.4	100	54.5	72.7	54.5	43.7	100	87.5	9.1
Whorls	18.2	21.4	-	45.4	27.3	18.2	50.0	-	12.5	-
Twin Loops	36.4	-	-	-	-	27.3	-	-	-	-
Total	100	100	100	100	100	100	100	100	100	100

Table 3. Difference of finger tip patterns on ten hand digits among Type 1 diabetic cases and matching controls

	Left Hand					Right Hand				
	Thumb (I)	Forefinger (II)	Middle finger (III)	Ring finger (IV)	Little finger (V)	Thumb (I)	Forefinger (II)	Middle finger (III)	Ring finger (IV)	Little finger (V)
cases										
Arches	-	20	-	-	-	-	-	-	-	-
Loops	60	80	100	40	80	75	75	100	75	100
Whorls	-	-	-	40	20	25	25	-	25	-
Twin Loops	40	-	-	20	-	-	-	-	-	-
Total	100	100	100	100	100	100	100	100	100	100
Controls										
Arches	-	-	-	-	-	-	-	-	-	-
Loops	40	60	80	40	80	40	40	100	40	40
Whorls	40	40	20	60	20	40	60	-	60	60
Twin Loops	20	-	-	-	-	20	-	-	-	-
Total	100	100	100	100	100	100	100	100	100	100

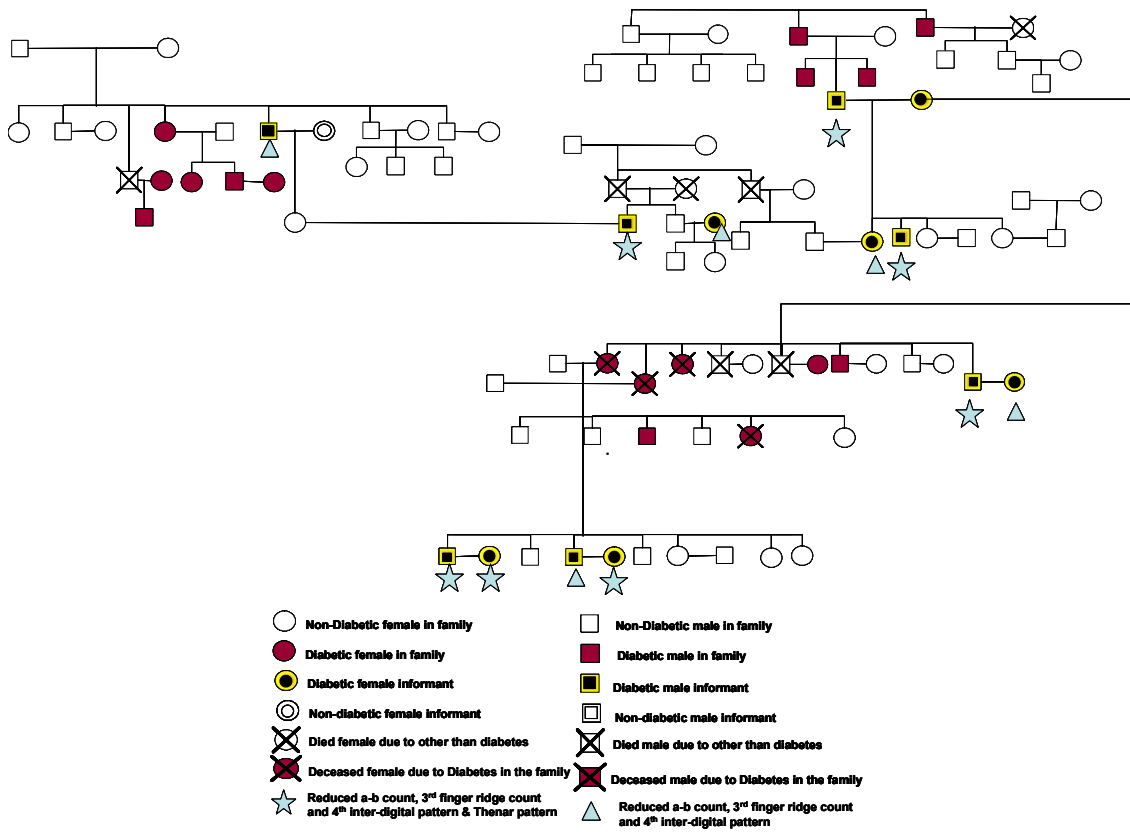


Figure 2. Intra-a-b ridge count, 3rd finger ridge count and 4th inter-digital pattern

Research Capacity Strengthening Activities

A. Infrastructure

A.1. Foundation stone of Laboratories and allied buildings has been laid by Prof. N. K. Ganguly, Director General, ICMR on 5th July 2006.

A.2. New Equipment

- Ultra Centrifuge
- Ultra Cryo Microtome
- PCR
- HPLC
- Electrophoresis
- 2-D Electrophoresis
- Gel Documentation System
- Cryo Deep freeze
- Tissue Embedding System devise
- Tissue staining system

B. Extra Mural grants generated

Project	Source	PI	Grant
• Sub-clinical vitamin-A level	UNICEF, India	Dr. RC Sharma	10.7 lac
• NCD Info-base surveillance	ICMR, India	Dr. R. C. Sharma	1.7 lac
• Risk factors of Desert Malaria	Govt. of Rajasthan	Dr. RC Sharma	4.0 lac
• Studies on dengue & DHF	TDR, WHO, Geneva	Dr. Vinod Joshi	8.5 lac
• Molecular markers of dengue	DST, Govt. of India	Dr. Vinod Joshi	20.0 lac

C. Ph. D. Programmes

Study	Student
• Development of Surveillance Design for Dengue	Ms Keerti Sharma
• Development of molecular markers of vector competence	Ms. Bennet Angel
• Role of peri-urban dengue viruses in causing DHF	Mrs.Shubha Dadhich
• Proteomics of environmental bio allergens	Ms.Manisha Dadhich
• Functionality of iso-electric proteomics in dengue	Mr. Amit Tak
• Studies on the role of ovitraps in control of dengue and malaria	Ms. Rina Kumawat
• Determination of insecticide resistance and biochemical mechanisms	Mr. Suresh Kumar

D. Training Imparted – Short term

Dissertation	Students	Subject
• Detection of dengue antigen using IFA	5	M.Sc. (Bio-tech)
• Detection of DEN antibodies using ELISA	4	B. E. (Bio-tech)
• SDS PAGE of mosquito mid-guts	5	M.Sc. (Bio-tech)
• SDS PAGE & 2-D Electrophoresis	2	M.Sc. (Bio-tech)
• Effect of plant extracts on M. tuberculosis	7	M.Sc. (Bio-tech)

E. Training Imparted – National Level

- Organized a NVBDCP sponsored National Level Training Programme for Entomological Assistants during 2006-07. Three batches of 20 participants starting from 29th January to 9th February, 19th February to 2nd March and 12th to 23rd March, 2007 respectively. The training was attended by the nominated candidates working in different State Health Directorates on the posts of District Malaria Officers (DMO), Assistant District Malaria Officers (ADMO), Assistant Malaria Officers (AMO), Anti Malaria Officers, Assistant Entomologists/Malariologists, Malaria/Filaria/Health Officers.

F. Training Imparted in software/Hardware to B. Tech/MCA Students

Software developed

- Library Management
- Survey Questionnaire developer and analyzer
- Store Management

Student

Gaurav Tripathi, B.E.(IT), SKIT, Jaipur
Ashish Pandya, MCA, APEX IMS, Jaipur
Niranjan Bohra, B.E. (C.S), JIET, Jodhpur
Girish Patel, B.E. (C..S), JIET, Jodhpur

G. LAN, Internet and website developed and library facility was further upgraded

H. Training Received

Title	Personnel
• National Workshop on Micro-array Data Analysis Sri Ramachandra Medical University, Chennai	Dr. K. R. Haldiya
• DPH Course at NIE, Chennai	Dr. Praveen Anand
• Training in Genetic Epidemiology, Mysore	Dr. P. K. Dam
• Surveillance of Communicable Diseases Basel, Switzerland	Dr. Himmat Singh
• Biochemistry test analysis, AIIMS, New Delhi	Dr. Himmat Singh & Mr. Rohit Joshi
• Training in collection and maintenance of laboratory animals, NIN Hyderabad	Sh. Raghunath Bisht & Sh. Babu Lal
• Training in collection and maintenance laboratory animals, NIRR, Mumbai	Sh. Mahaveer & Sh. Satya Prakash

द्वितीय वार्षिक प्रतिवेदन

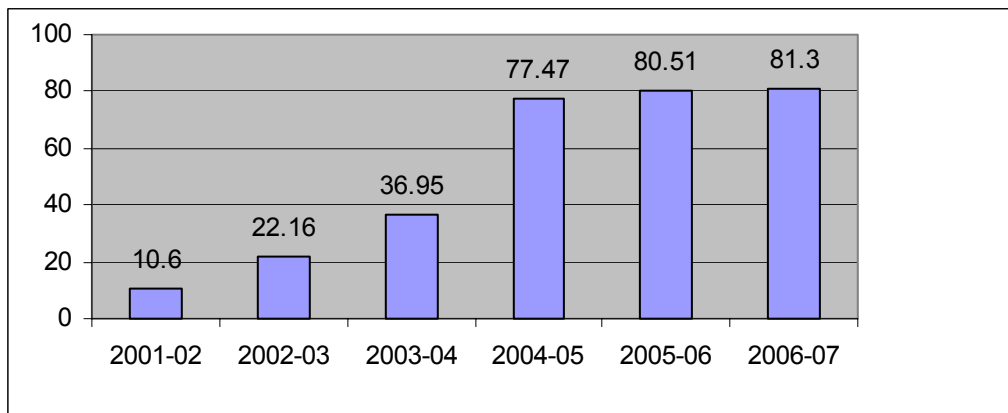
1- प्रकाशित लेखों की संख्या

हिन्दी त्रैमासिक पत्रिका "चेतना" के माध्यम से केन्द्र में हो रहे अनुसंधानिक एवं अन्य गतिविधियों की झलक देखने को मिलती है। विभिन्न बीमारियों, विशेषकर मरुस्थल में व्याप्त बीमारियां व उनके निवारण से संबंधित लेखों से पाठकों को उपयोगी जानकारी मिलती रहती हैं। केन्द्र के वैज्ञानिकों के अतिरिक्त हमारे पाठकों ने भी इस पत्रिका को अपने लेखों से समृद्ध किया है। इस पत्रिका के अब तक 20 अंक (वर्ष 2002 से) प्रकाशित हो चुके हैं।



2- प्रकाशित लेखों की संख्या

अपनी वैज्ञानिक गतिविधियों में प्रगति करने के साथ-साथ केन्द्र ने राजभाषा हिन्दी में अधिकाधिक कार्य करने पर ध्यान दिया है। राजभाषा विभाग द्वारा समय-समय पर जारी वार्षिक कार्यक्रम में निर्धारित लक्ष्यों की प्राप्ति की ओर विशेष ध्यान देते हुए केन्द्र में हिन्दी में पत्राचार, वर्ष 2006-07 में बढ़कर 81.3 : रहा, जिसकी झलक निम्न दण्ड तालिका में दर्शाई गई है :-



3- दलनऱ दकऱ गलुनहऱ ँऱ मरदऱ V दकऱ; 7 दसऱ फऱ, , द वकऱ ङकऱ ऱऱऱे ऱऱऱे लदकऱ



केन्द्र में राजभाषा से जुड़ी गतिविधियों में निरन्तर वृद्धि हुई है जिसके फलस्वरूप आज केन्द्र को नगर राजभाषा कार्यान्वयन समिति द्वारा एक अग्रणी सक्रिय संस्था के रूप में गिना जाता है। राजभाषा के प्रति समर्पित भावना व केन्द्र के कामकाज में अधिकाधिक हिन्दी के प्रयोग के कारण आज केन्द्र को लगातार तीसरी बार उल्लेखनीय सफलता मिली है। दिनांक 31 अक्टूबर, 2006 को नगर राजभाषा कार्यान्वयन समिति, जोधपुर द्वारा हिन्दी में उत्कृष्ट कार्य हेतु केन्द्र को प्रथम पुरस्कार के रूप में चल-वैजयंती व प्रशस्ति पत्र प्रदान किए गए। इससे पहले केन्द्र को इसी समिति द्वारा वर्ष 2003-2004 के लिए प्रथम पुरस्कार तथा वर्ष 2004-2005 के लिए द्वितीय पुरस्कार के रूप में चल-वैजयंती व प्रशस्ति पत्र प्रदान किए गए। यह उपलब्धि केन्द्र के सभी कर्मचारियों के समन्वित प्रयासों का प्रतिफल है। सभी कर्मचारी राजभाषा के अनुपालन में अपना भरपूर योगदान दे रहे हैं।

4- गलुनहऱ दकऱ; 7 ककऱ



कर्मचारियों का राजभाषा हिन्दी में कार्य करने का उत्साह बढ़ाने, मार्गदर्शन करने व इस कार्य में उनकी झिझक दूर करने के उद्देश्य से समय-समय पर हिन्दी कार्यशालाओं का आयोजन किया जाता रहा है। इस वर्ष भी दिनांक 29 जून, 2006 तथा 20 सितम्बर, 2006 को इन कार्यशालाओं का आयोजन किया गया। ऐसी कार्यशालाएं कर्मचारियों को राजभाषा में और अधिक कार्य करने को प्रेरित करती हैं।

केन्द्र के वैज्ञानिकों में हिन्दी के प्रयोग को बढ़ावा देने हेतु केन्द्र में 'डीएमआरसी राजभाषा व्याख्यानमाला' प्रारम्भ की गई है जिसमें राजभाषा को समर्पित देश के मूर्धन्य वैज्ञानिकों के विचारों को हिन्दी में सुना जाएगा। इस माला की प्रथम मणिका के रूप में प्रोफेसर रामगोपाल, पूर्व निदेशक, रक्षा प्रयोगशाला ने गत 29 मार्च, 2007 को "21वीं सदी में पेय जल की चुनौतियां एवं समाधान : मरु पर्यावरण के परिप्रेक्ष्य में" विषय पर एक व्याख्यान प्रस्तुत किया।

5- fgUhh | Irkg



पिछले वर्ष की भांति इस वर्ष भी दिनांक 14–21 सितम्बर, 2006 को हिन्दी सप्ताह का आयोजन किया गया। इस दौरान प्रतिदिन विभिन्न प्रतियोगिताओं का आयोजन किया गया जैसे वाद–विवाद प्रतियोगिता, कविता पाठ प्रतियोगिता, निबंध लेखन प्रतियोगिता, श्रुतिलेख प्रतियोगिता, वाचन प्रतियोगिता, प्रश्न मंच प्रतियोगिता तथा अंताक्षरी प्रतियोगिता। इसमें केन्द्र के सभी अधिकारियों व कर्मचारियों ने मिल जुलकर भाग लिया। दिनांक 21 सितम्बर, 2006 को इन प्रतियोगिताओं के विजेताओं तथा राजभाषा विभाग, गृह मंत्रालय की प्रोत्साहन योजना के अनुसार सरकारी कामकाज में हिन्दी का अधिक प्रयोग करने वाले कर्मचारियों का चयन कर उद्घोषणा की गई। इन सभी विजेताओं को केन्द्र के वार्षिक दिवस पर सम्मानित किया जाएगा।

fo'kšk %

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दिनांक 24 दिसम्बर, 2006 को भारतीय आयुर्विज्ञान अनुसंधान परिषद के महानिदेशक आचार्य निर्मल कुमार गॉगुली के कर–कमलों द्वारा परिषद के सभी संस्थानों के निदेशकों की विशेष बैठक में केन्द्र की वैबसाइट का शुभारंभ किया गया। इस वैबसाइट में केन्द्र का संक्षिप्त इतिहास, उद्देश्य सामयिक कार्यक्षेत्र, जन–स्वास्थ्य में योगदान, वर्तमान घटनाएं और सोच दर्शाई गई हैं। साथ ही वार्षिक प्रतिवेदन, वैज्ञानिक प्रकाशन, रोजगार के अवसर, निविदा सूचना तथा राजभाषा हिन्दी के कार्यक्षेत्र में केन्द्र की उपलब्धियां भी उपलब्ध हैं। इस वैबसाइट को यथाशीघ्र द्विभाषी रूप में उपलब्ध कराने की कोशिश की जा रही है।

dslnz dh jktHkk"kk dk; k&lo; u | fefr ds | nL;

1. डॉ. रमेश चन्द्र शर्मा – अध्यक्ष
2. डॉ. शेषपाल यादव – सदस्य
3. डॉ. मधुबाला सिंह – सदस्य
4. डॉ. आशुतोष कुमार दीक्षित – सदस्य
5. श्री सुभाष चन्द्र शर्मा – सदस्य
6. श्री राजकुमार कालुंधा – सदस्य
7. श्री रोहित प्रसाद जोशी – सदस्य
8. श्रीमती कंचन बाला – सचिव

Papers published/Accepted

1. Bansal, SK and Singh, Karam V. Laboratory evaluation for comparative insecticidal activity of some synthetic pyrethroids against vector mosquitoes in arid region. *J. Environ. Biol.*, 2006, 27: 251-55.
2. Bansal, SK and Singh, Karam V. Relative susceptibility of some common mosquito vector larvae to synthetic insecticidal compounds in northwestern Rajasthan. *J. Environ. Biol.*, 2007, 28: (In Press)
3. Joshi V, Sharma R C, Sharma Y, Adha S, Singh H and Singhi M, 2006. Introduction, Transmission and Aggravation of Malaria in a desert ecosystem of Rajasthan, India, *J Vect. Borne Dis*, 43 :179–185
4. Mathur ML. Potential utility of Mycobacterium w vaccine in control of tuberculosis. *Current Respiratory Medicine Reviews* 2006; 2: 183-188.
5. Purohit SD, Purohit V, Mathur ML. A Clinical scoring system as useful as FNAC in diagnosis of tuberculous lymphadenitis in HIV positive patients. *Curr. HIV Res.* 2006; 4: 459-462.
6. Sachdev R, Mathur ML, Haldiya KR and Saiyed HN. Work related health problems in Salt Workers of Rajasthan, India. *Ind. J. Occ. Environ. Med.*, 2006; 10: 62-64.
7. Singh, Madhu B; Fotedar, R; Lakshminarayana, J; Anand, P.K. Studies on the nutritional status of under five children in drought affected desert area of Western Rajasthan, India. *Pub. Health Nutrition*, UK. 2006; 9 (8), 961- 967.
8. Singh, Madhu B; Lakshminarayana, J; Fotedar, R; Anand, P.K. Childhood illnesses and malnutrition in under five children in drought affected desert area of western Rajasthan, India. *J. Commun. Dis.*, 2006; 38 (1): 88-96.
9. Yadav SP, Mathur ML and Dixit AK. Knowledge and attitude towards tuberculosis among the sandstone quarry workers in desert part of Rajasthan, India. *Ind. J. Tub.*, 2006; 53(4): 187-195
10. Yadav, SP, Sharma, RC and Joshi, V: Treatment seeking behavior of malaria patients in Desert part of Rajasthan, India. *J. Commun. Dis.*, 2007; 39 (1) (In press).

Workshops / Conferences / Symposia / Scientific meetings attended / participated / organized by scientists

Dr. R.C. Sharma, Deputy Director (SG) & Officer-in-Charge

- Organized a workshop on "Reservation in Services" on 21st & 22nd April, 2006 with the faculty support from Institute Secretariat Training & Management, Govt. of India, Department of Personnel & Training, New Delhi.
- Organized "ICMR Directors Conference" on 23rd & 24th December, 2006 .
- Organized three "National Level Training for Entomological Assistants" from 8th to 19th January, 2007, 19th to 2nd March, 2007 and 12th to 23rd March, 2007 sponsored by National Vector borne Diseases Control Programme, Ministry of Health & FW, Govt. of India, New Delhi.
- Attended DMRC- NIMR Traveling Workshop on Vector Borne Diseases from 24th to 28th April, 2006.
- Attended " Scientific Advisory Group" meeting held on 6th & 7th June, 2006 at ICMR Hqrs., New Delhi.
- Attended Project Advisory Committee meeting on " Sub-Clinical Vit. A deficiency" held on 9th & 10th June, 2006 at ICMR Hqrs. New Delhi.
- Attended Project Advisory Group meeting on " Camel Milk and Diabetes" held on 26th September, 2006 at ICMR Hqrs., New Delhi.
- Attended VCRC Scientific Advisory Committee meeting held on 11th & 12th October, 2006 at Pondicherry.
- Attended "National Symposium on Tribal Health" at Jabalpur held on 19th & 20th October, 2006.
- Attended CJIL Scientific Advisory Committee meeting held on 6th & 7th November, 2006 at Agra.
- Attended " International Meeting on Dengue" held on 10th November, 2006 at NIV, Pune.
- Attended " Heart Research- 2007 meet" at Bikaner held on 17th & 18th February, 2007.
- Attended " Indian Space Programme" meeting at RRSSC, Jodhpur held on 21st February, 2007.

- Attended a one-day workshop on " Health Sector Policy Reform Options Database" held on 23rd March, 2007 at New Delhi.

Dr. K.R. Haldiya Deputy Director (SG)

- Participated in "First National Workshop on Micro-array Data Analysis" held at Chennai from 13th February to 14th February 2007 which was organized by Department of Bio Informatics, Sri Ramachandra Medical University.

Dr. Vinod Joshi, Deputy Director (SG)

- Delivered lecture on "Post flood emergence of infectious disease" in Brain storming session on "Flood in *Thar* & Challenges of Natural Disaster: Prevention, Relief and Rehabilitation" held at AFRI, Jodhpur on September, 19th 2006.
- Delivered lecture on "Emergence of infectious disease during flood at CAZRI, Jodhpur.
- Attended meeting of Scientist of Jodhpur with President, Indian Space Research Organization, Govt. of India, at RSSC, Jodhpur
- Award of "*Jodhpur Gaurav Alankaran*" was conferred for contribution in "Dengue & Guinea worm Disease Research" by Jodhpur Municipal Corporation, on 29th March, 2007

Dr. M. L. Mathur, Deputy Director (SG)

- Attended International Symposium on New Frontiers in Tuberculosis Research at International Centre for Genetic Engineering and Biotechnology, New Delhi from 4th Dec to 6th Dec 2006 and presented paper entitled "Anti-tuberculous activity of two plant extracts".
- Attended 6th RAJCON and First Midterm CHESTCON CONFERENCE organized by Department of Respiratory Diseases and Tuberculosis, J. L. N. Medical College, Ajmer on 2-3 December, 2006 and presented paper entitled "Glutaraldehyde test for diagnosis of pulmonary tuberculosis."

Dr. Karam V. Singh, Deputy Director

- Attended a 'National Symposium on Tribal Health' held from 19th to 20th October, 2006 at, Regional Medical Research Center for Tribal, Jabalpur, and presented paper

on 'Perception of a tribal dominated population towards use of impregnated bed net for the control of malaria in Dungarpur district, Rajasthan'.

- Attended a meeting of ERMED-India Consortium on 9th January, 2007 at Resource Centre, Nirman Bhawan, New Delhi, in connection with starting electronic journal consortium for national level institutions conducting R&D in the field of medical sciences.
- Coordinated three NVBDCP sponsored training courses for 'Entomological Assistants', starting from 29th January to 9th February, 19th February to 2nd March and 12th to 23rd March, 2007 respectively.

Dr. S. K. Bansal, Deputy Director

- Associated with teaching and field exercises of three NVBDCP sponsored training courses for 'Entomological Assistants', starting from 29th January to 9th February, 19th February to 2nd March and 12th to 23rd March, 2007 respectively.

Dr. S. P. Yadav, Assistant Director

- Attended 24th Annual Conference of Indian Society for Medical Statistics at PSG Institute of Medical Sciences & Research, Coimbatore from 1-3 December, 2006 and presented a paper entitled "Treatment seeking behavior for malaria in pre-school children: implication for home management in rural part of desert in Rajasthan, India".
- Attended National Workshop on reviving Rajasthan's camel husbandry for income generation and sustainable land use at Jaisalmer, Rajasthan from November, 6th-8th, 2006 and delivered lecture on uses of camel in health sector on 6th November, 2006 and chaired session on 7th November, 2006 on solving the grazing problem: "How to feed camels, so that they can be productive".

Dr. Madhu B. Singh, Assistant Director

- Attended and presented paper on 'Drought and its impact on Nutritional Status of rural population of desert areas of Western Rajasthan, India' in I World Congress of Public Health Nutrition and - VII Congreso de la SENC held at Barcelona International Convention Centre (Centre de Convencions International de Barcelona- CCIB), Barcelona, Spain from 28- 30th September, 2006.
- Attended 'Pre congress workshop on Nutritional anemias' Organized by Michael B. Zimmermann (ETH Zurich) and Klaus Kraemer (SIGHT AND LIFE, Basel on 27th September, 2006 held at Hotel Hilton, Barcelona, Spain.
- Delivered a invited lecture on 'Management of Nutrition during Natural Disaster' in brain storming Session on 'Floods in Thar and Challenges of natural

Disaster: Prevention, Relief and Rehabilitation' held on September 9, 2006 at AFRI, Jodhpur.

- Attended and presented paper on 'Studies on the nutritional status of rural population in desert area of Rajasthan' in National Symposium on Tribal Health held on 19th & 20th October, 2006 at, Regional Medical Research Center For tribals, Jabalpur.
- Attended training in new method for estimation of Urinary Iodine Excretion (UIE) levels from Prof. M. G. Karmarkar, Regional advisor, ICCIDD at All India Institute of Medical Sciences, Delhi from 4/12/06 to 8/12/06 in ICCIDD laboratory of Dr. Chandrakant S. Pandav, Regional Coordinator, ICCIDD (South Asia), and Prof. and Head, Centre for Community Medicine, AIIMS, Delhi.

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Officer-in-Charge

Dr. R. C. Sharma, M. Sc., D. Sc.

A. Scientists

1. Dr. K. R. Haldiya, M.D, Deputy Director (SG)
2. Dr. Vinod Joshi, M.Sc. Ph.D, Deputy Director (SG)
3. Dr. M. L. Mathur, M.D, Deputy Director (SG)
4. Dr. Raman Sachdev, M.D, Deputy Director
5. Dr. Karam V. Singh, M.Sc., Ph.D, Deputy Director
6. Dr. S. K. Bansal, M Sc., Ph.D, Deputy Director
7. Dr. S. P. Yadav, M.A., Ph.D, Assistant Director
8. Dr. Madhu B. Singh, M Sc., Ph.D, Assistant Director
9. Dr. A. K. Dixit, M.Sc., Ph.D, Assistant Director
10. Dr. J. Lakshminarayan, M.Sc., Ph.D, Assistant Director
11. Dr. (Mrs.) Ranjana Fotedar, M.B.B.S. Senior Research Officer
12. Dr. (Mrs.) Manju Singhi, M.Sc., Ph.D, Research Officer
13. Dr. P. K. Anand, M.B.S.S., Research Officer

B. Technical staff

1. Dr. P. K. Dam, M.Sc., Ph.D, Technical Officer
2. Sh. Raj Kumar Kalunda, M.A., Technical Officer
3. Sh. Manjeet Singh, AMIE, M.Tech., Technical Officer
4. Dr. Himmat Singh, M.Sc., Ph.D, Research Assistant
5. Sh. Anil Purohit, M.A., Research Assistant
6. Sh. Rajneesh Kumar, Laboratory Technician
7. Sh. S. K. Dhawal, Laboratory Technician
8. Sh. Ramesh Chandra, Laboratory Technician
9. Sh. Pooranmal Meena, Laboratory Technician
10. Sh. Colvin Sunil Singh, Laboratory Technician
11. Sh. Rohit Prasad Joshi, Laboratory Technician
12. Sh. Rajendra Chouhan, Laboratory Assistant
13. Sh. Nivesh Bhardwaj, Laboratory Assistant

C. Ministerial staff

1. Sh. S. C. Sharma, Administrative Officer
2. Sh. Anup Sarin, Accounts Officer
3. Sh. S. K. Lotan, Section Officer
4. Sh. Narender Bajaj, Section Officer
5. Smt. Neelam Devi, Assistant
6. Sh. Dharampal, Assistant
7. Sh. Rajendra Singh, Assistant
8. Sh. Joginder Singh, Stenographer
9. Km. Kanchan Bala, Junior Hindi Translator
10. Sh. Shamshad Ali, U.D.C.
11. Smt. Chandra Kala, U.D.C.
12. Sh. Mahesh Chand Pargi, U.D.C.
13. Sh. Yesh Pal Singh, U.D.C.
14. Sh. Ram Niwas, L.D.C
15. Sh. Nand Kishore, L.D.C
16. Sh. Jaideep Gaur, Hindi Typist

D. Supporting Staff

1. Sh. Babu Lal, Driver
2. Sh. Mangu Singh, Driver
3. Sh. Raghunath Singh, Driver
4. Sh. Mohammed Gaffar, Driver
5. Sh. Ishwar Khetani, Driver
6. Sh. Manohar Singh, Driver
7. Sh. Rana Ram, Driver
8. Sh. Banwari Lal, Laboratory Attendant
9. Sh. Shridhar Bohra, Laboratory Attendant
10. Sh. Lal Chand Bandra, Laboratory Attendant
11. Sh. Raghunath Bisht, Animal Attendant
12. Sh. Babu Lal Bunker, Animal Attendant
13. Sh. Mahaveer Prasad, Animal Attendant
14. Sh. Satya Prakash, Animal Attendant
15. Sh. Mahesh Chand Sharma, Attendant
16. Sh. Jodha Ram, Attendant
17. Smt. Laxmi Kanta, Attendant
18. Sh. Khushal Singh, Attendant
19. Sh. Ram Lal, Peon
20. Smt. Sohni Devi, Peon
21. Sh. Ladu Ram, Peon
22. Smt. Sua Devi, Sweeper.

E. Project staff

A. Development of Molecular and Genetic Markers of Vector Competence

1. Ms. Keerti Sharma – Junior Research Fellow
2. Ms. Bennet Angel – Junior Research Fellow

B. Environmental Health Studies in Jodhpur

1. Dr. Harcharan Singh – Research Associate
2. Sh. Achlesh Kumar Sharma – Lab. Cum-Field Assistant

C. Vitamin-A Project

1. Dr. Komal, Staff Nurse
2. Sh. Sunil Beniwal, Staff Nurse.

Foundation Stone Laying of DMRC Building, July 2006



Construction Work in Progress



SAC Meeting June, 2006



Directors Conference, December, 2006



Inauguration of Website, December, 2006

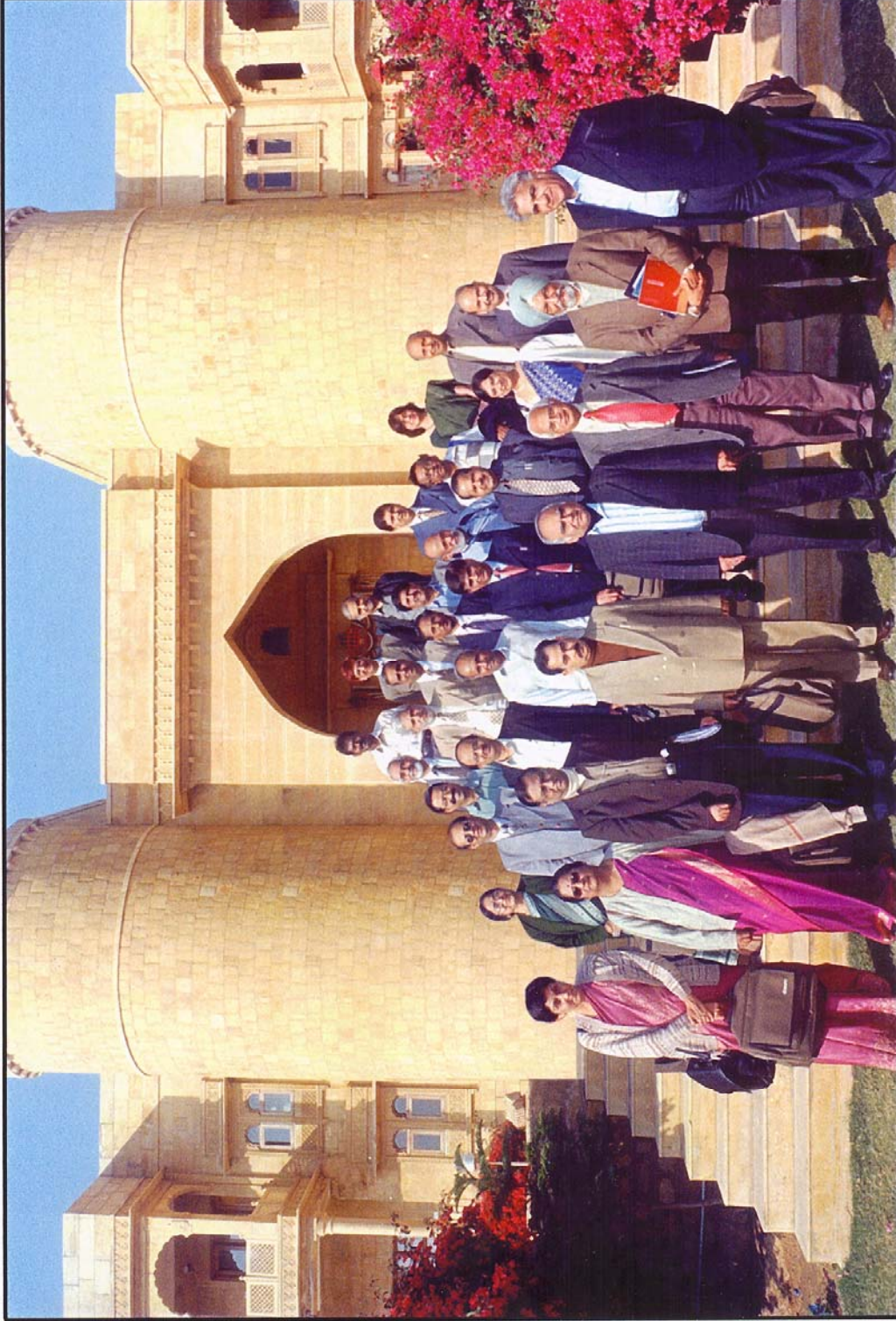




ICMR DIRECTORS CONFERENCE

December 23-24, 2006

Desert Medicine Research Centre, Jodhpur



Dr. Neeru Singh, Dr. V Muthuswamy, Dr. S K Bhattacharya, Dr. A P Dash, Prof. N K Ganguly (DG-ICMR), Sh P D Seth, Sh. Mohinder Singh, Dr. C P Puri
Dr. Kiran Katoch, Dr. V M Katoch, Dr. R S Paranjape, Dr. S K Satyanarayana, Dr. A C Mishra, Dr. B K Tyagi, Dr. Sunita Saxena, Dr. J Mahanta
Dr. P R Narayanan, Dr. M D Gupte, Dr. B C Das, Dr. P K Das, Dr. S Chakrabarti, Dr. Arvind Pandey
Dr. J M Deshpande, Dr. R J Yadav, Dr. K K Singh, Dr. Pradeep K Das, Dr. Bela Shah
Dr. P Vijayachari, Dr. R C Sharma, Dr. Lalit Kant

29th Jan to 9th Feb, 2007



19th Feb to 2nd Mar, 2007



12th Mar to 23rd Mar, 2007



Hindi Workshop June, 2006



Hindi Week 14-21, September, 2006



First Prize in work done in Hindi during the year 2005-06, October 2006



DMRC Day Celebration, July 2006



Independence Day August, 2006



Vigilance week November, 2006





Scientific Lecture Series in Hindi,
29th Mar, 2007

Dr. Madhu B. Singh, with WHO &
Micronutrient Initiative Expert Group
On Anaemia during First World
Congress of Nutrition at Barcelona,
Spain in September, 2006



“Jodhpur Gaurav Alankaran”
Conferred to Dr. Vinod Joshi
On 30th Mar, 2007