COMMUNITY HEALTH CELL LIBRARY

ISSN 0377-4910

ICMR BULLETIN

Vol. 29, No. 1 January, 1999

PROSPECTS OF USING BACILLUS SPHAERICUS IN THE CONTROL OF CULEX MOSQUITOES IN RELATION TO RESISTANCE DEVELOPMENT

Vector resistance to insecticides, high cost and environmental pollution associated with insecticide use have imposed severe constraints on vector control programmes. Consequently, vector control strategies have to be redesigned to minimize the use of chemical pesticides. *Bacillus thuringiensis* var. *israelensis* and *B. sphaericus* are being projected as promising biocontrol agents against the vectors of malaria, filariasis and *B. thuringiensis* against onchocersiasis[1-6]. Several field trials with *B. sphaericus* carried out against *Culex* and *Anopheles* mosquitoes breeding in different habitats produced variable results[7-20]. *B. sphaericus* has a narrow host range[21], is non-toxic to black flies, less active against most *Aedes* species and tends to be more effective against *Culex* followed by anophelines[22-37]. The euphoria of control of vector mosquitoes with this agent was short lived in view of reports of resistance development from various laboratory and field studies[15,18,35,38-40], which showed that high dosage and continuous exposure to *B. sphaericus* would result in extremely high or moderate levels of resistance in *Culex quinquefasciatus*.

The potential of using *B. sphaericus* as a biocontrol agent to control the vectors of the filarial parasites will depend upon the risk of emergence of resistance. This information is essential in formulating suitable strategies for resistance management. The future utility of *B.sphaericus* in the integrated vector management (IVM) programmes will depend on how the emergence of resistance in the field population is managed. This review highlights the mode of action of *B. sphaericus*, the current status of the resistance in *Culex* mosquitoes to *B. sphaericus* and its future implications in mosquito control programmes.

Mode of Action of *Bacillus sphaericus*

B. sphaericus (Neide) is an aerobic and spore forming soil bacterium. Laboratory assays showed that >100 spores/ml are required to kill larvae of *Cx. quinquefasciatus*[41]. Among the 48 strains, the most studied were 1593, 2297 and 2362 [7,29,42,43]. Pathogenicity is caused by the production of two kinds of toxic proteins *viz*. crystal and Mtx (100 and 30.8 kDa). The crystal toxin is more mosquitocidal than Mtx and is produced during sporulation and concentrated in the parasporal inclusions. These inclusions have a crystalline ultrastructure and are released into the medium along with the spore after the completion of sporulation. The Mtx is reported to be produced only during the vegetative phase[44-46]. The crystal toxin is composed of two proteins, P51 and P42 (42 kDa) that act in synergy. The P51 part is responsible for the initial binding of the crystal toxin to the receptor on the brush border membrane of the epithelial cells in the gastric caecae

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and posterior midgut regions and the 42 kDa component confers toxicity[47-50] But the relationship between toxicity of *B. sphaericus* to larva and spore germination is not yet understood[51]. The toxic process involves (i) the ingestion of the crystal/spore/cell complex; (ii) dissolution of the toxin at the alkaline pH of the cell wall in the midgut; (iii) activation of the protoxin (42 kDa) into the toxic form by gut proteases; (iv) binding of the binary toxin to the specific receptors on the brush border membrane of epithelial cells in the gastric caecae and posterior midgut regions; (v) entry of spores containing binary toxin into the midgut cells by forming pores on the cell membrane;and (vi) the pathogenic effects then involve basal separation and sloughing of the midgut epithelial cells, swelling of the midgut, that leads to tremors, sluggishness and eventual death of the larvae[21,52].

Differential Susceptibility to *Bacillus sphaericus*

Susceptibility of mosquito species to *B. sphaericus* varies widely[53] and at least one or more of four factors are responsible for such wide variation. They are rate of ingestion of spore containing toxin, dissolution of the toxin in the mosquitoe's gut, activation of the toxic protein and binding to the specific receptor sites. In general *Culex* mosquitoes are more susceptible than anophelines and *Aedes*[29,54-60]. The lack of susceptibility of *Aedes* mosquitoes (especially *Ae. aegypti*) to *B. sphaericus* is not due to rates of toxin ingestion/concentration, dissolution of the toxin, specificity of the gut proteases and/or activation of the crystal toxin by midgut proteases[38,61-66] but is due to the failure of toxins binding to the midgut epithelial cells[59,67] since no functional receptors were found in this species[31]. For *Anopheles* species, *B. sphaericus* toxins bind to the larval midgut cells but with low specificity and affinity, thus resulting in moderate degree of susceptibility of these species to the toxins[47,59]. In *Culex* species the toxins bind with high specificity and high affinity, thus leading to high susceptibility in these species[47,59,68]. In a recent study it was found that there was no relationship between toxicity and irreversibility of toxin binding on brush-border membrane[69].

Genetics and Mechanism of Resistance

Mosquito pathogenic strains of *B. sphaericus* have been assigned to DNA homology group IIA[70]. Resistance to *B. sphaericus* is monofactorial involving a single sex linked recessive gene[44] which segregates independently from the overproduced esterases for organophosphorous resistance. These esterases are slightly more active in the *B. sphaericus* resistant than the susceptible strains and may not account for the high level of resistance[19]. Heterozygotes were only 3 times less susceptible than the parental field population. Resistance is found to be highly specific affecting only members of the strain used in the selection[38]. Several mutated genes, each coding for different mechanisms of resistance, may be present in natural populations at frequencies equal to the mutation rate.

From various laboratory and field studies it was found that the *B. sphaericus* exposed populations of *Cx. quinquefasciatus* were 4 to 100,000 times less susceptible than unselected populations (Table). This wide variation might be due to different factors influencing selection[71]. Factors such as selection pressure, the proportion of population exposed, the gene frequency of the resistance alleles in the population prior to selection, existence of untreated refugees, reduced ingestion rate, low biocides' persistence and/or availability because of settling of large proportion of spores at the bottom of the mosquito breeding habitat within 2 h of treatment, influx of resistant individuals, changes in the mosquito larval midgut receptor and physico-chemical factors of the breeding habitats are important factors that determine the development of resistance by mosquitoes[35,38,39,41,44,71-81]. On the contrary, the factor that could contribute for the slow process of resistance development is the phenomenon of back selection (or removal of selection) which results in lower survivorship of the resistant individuals because of their lower fitness[82]. However, continued exposure may improve the fitness of the target population through co-adaptation of resistant genomes by modifier genes which lead to a more stable resistance[83]. High and moderate levels of resistance in *Cx. quinquefasciatus* to *B. sphaericus* both in the laboratory and under field conditions were attributed to repeated intense selection pressure with low and slow mixing of non-selected populations[19,35,38].

Ingestion

Mosquito larvae are filter feeders and they normally exclude large particles[84]. They feed on particulate matters ranging from microscopic bacteria to clearly visible particles such as aquactic algae[85]. Species belonging to the genus *Anopheles* are surface feeders whereas *Culex* species feed below the water surface as well as at the bottom[84,86]. The presence of plenty of bacteria and other suspended matter in polluted breeding habitats is reported to influence the rate of ingestion of bacterial spores and digestability[87] which would account for the wide variation within the genus[86,88]. Earlier Sun *et al.*[89] showed no effect

Table. Selection of resistance in *Culex quinquefasciatus* to *Bacillus sphaericus*.

Strain	Mosquito strain	Selection (Lab./field)	Selection Pressure (%)*	Generation	Resistance level (fold)**	Source/ Country
2362	Lab.	Lab.	90	F_4	LC_{90} - 60	Georghiou *et al.*
	Field	Lab.	94-98	F_5	LC_{50} - 1000	(1992): USA[38]
			94-98	F_{12}	LC_{50} - 100.000	
2362	Lab.	Lab.	80	F_{24}	LC_{50} - 43	Rodcharoen &
	Lab.	Lab.	80	F_{55}	LC_{50} - 36	Mulla (1991)
	Field	Lab.	80	F_{35}	LC_{50} - 12	USA[40]
	Field	Lab.	80	F_{55}	LC_{50} - 24	
	Lab.	Lab.	80	F_{80}	LC_{50} - 37	Rodcharoen &
	Field	Lab.	80	F_{80}	LC_{50} - 10 - 30	Mulla (1994): USA[35]
	Lab.	Lab.	80	$\approx F_{100}$	LC_{50} - 37	Rodcharoen &
	Field	Lab.	80	$\approx F_{100}$	LC_{50} - 31	Mulla (1996): USA[39]
2362	Field	Field (8 years)	18 treatments		LC_{50} - 16.000	Sinegre *et al.* (1994) France[15]
2362	Field	Field	37 treatments (2 years)		LC_{50} - 10 †	Silva-Filha *et al.* (1995); Brazil[72]
1593 M	Resistant field strain	Lab.	75	F_{18}	LC_{50} - 6.223 LC_{90} - 31,325	Rao *et al.* (1995) India[18]
	Field	Field	35 treatements (2 years)		LC_{50} - 146 LC_{90} - 180	
1593	Lab.	Lab.	80	$\approx F_{100}$	LC_{50} - 22	Rodcharoen &
	Field	Lab.	80	$\approx F_{100}$	LC_{50} - 6	Mulla (1996); USA[39].
2297	Lab.	Lab.	80	$\approx F_{100}$	LC_{50} - 12	-do-
	Field	Lab.	80	$\approx F_{100}$	LC_{50} - 4	

* Per cent mortality at sublethal dosage.
** Difference in the susceptibility between the parental and selected population.
† Less susceptible than control field population
Superscript numbers refer to the sl. nos in the reference list.

of water depth between 1 and 5 cm on susceptibility. A recent study[90] is in aggreement with the observation of Nugud and White[91] that anophelines were less susceptible as they require higher concentraion of spores or their surface feeding habit does not allow them to ingest sufficient amount of bacterial spores. In a study it was found that larvae of the resistant strains (laboratory and field-collected) ingested *B. sphaericus* preparation at significantly lower rates than did larvae of the susceptible strains and yeast was ingested at a higher rate than the bacterial preparation when compared to susceptible strains. The reduced ingestion rates of *B. sphaericus* in resistant strains could possibly serve as a behaviouristic mechanism of resistance against this microbial larvicide[77].

Receptor site

Two different mechanisms conferring high levels of resistance to *B. sphaericus* toxin were reported in France and California. In the California study it was found that there was an alteration of the toxin binding functionality of the receptor[31]. Whereas, the mechanism of field-selected resistance in the French study is probably not caused by the impairment of the first step of toxin-receptor interactions at the cellular midgut level. It may involve

the midgut receptor, but not at the binding step level. However, the exact mechanism is not known[19]. Results of Brazil and Indian studies showed that *B. sphaericus* resistant field strain of *Cx. quinquefasciatus* was more susceptible than the normal population to *B.t.i.*[18,72], indicating that laboratory and field populations have different receptors. Such disparity is found in all species to any microbial insecticide. It remains to be seen whether the *B. sphaericus* resistance mechanism identified in California and France corresponds to different alleles of the same gene or to two different genes, and whether either of them is the gene conferring high resistance in India[18,19]. Binding experiments using isolated midgut brush border membrane and radio labelled binary toxin of *B. sphaericus* indicated that there was a direct correlation between resistance and stable changes in the mosquito larval midgut receptor[71]. But various other studies showed no such relationship between receptor binding and mechanism of resistance development[44,72,92].

Potentiality of Resistance Development

Laboratory and field collected *Cx. quinquefasciatus* exposed to *B. sphaericus* strain 2362 for 35 generations in the laboratory showed a level of resistance 43 and 12 fold that of parental generation respectively[40]. Further, exposure to concentrations that yielded 80% mortality resulted in 37 and 31 fold increase respectively in the resistance in the laboratory and field collected colonies by $\approx F_{100}$[39]. An extremely high level of resistance of 100,000 fold in the 12th generation was obtained in the field collected colony using 94-98% selection pressure[38]. However, a field collected population of *Cx. quinquefasciatus* larvae, which had been subjected to 37 treatments with *B. sphaericus* 2362 for 2 years was found to be ≈10 fold less susceptible than a control field population[72]. Laboratory selection of wild caught *Anopheles stephensi* with Spherix (a formulation of *B. sphaericus*) at a concentration 2.8 times that of LC_{50} for 3 generations resulted in 232 times increase in LC_{50} value than that of the parental population[93]. It was concluded that the intensive selection of *Cx. quinquefasciatus* with the binary toxin of *B. sphaericus* 2362 (51 and 42 kDa proteins) could produce high resistance and high degree of specificity affecting members of the same strain (Table).

Field resistance to *B. sphaericus* 1593M was observed in a population of *Cx. quinquefasciatus* in Kochi, India, exposed to 35 rounds of spraying over a 2 year period. Larvae from the sprayed area gave LC_{50} and LC_{90} values that were 146 and 180 times greater than the corresponding values for a susceptible strain from an unsprayed locality[18]. But susceptibility status of *Cx. quinquefasciatus* in the control and trial areas prior to treatment was not studied in this trial. Since it is possible that different populations differ in their susceptibility to the pathogen, it is difficult to conclude that there was emergence of a population resistant to *B. sphaericus*. When larvae from a sprayed area were subjected to selection pressure in the laboratory, there was 6,223 and 31,325 fold increase in the LC_{50} and LC_{90} values respectively in the 18th generation in comparison with the susceptible strain.

Cross Resistance

Cross resistance studies showed that resistance to *B. sphaericus* strain 2362 did not influence the susceptibility of the mosquitoes to several other strains as well as towards toxins of *B.t.i.*[38]. Laboratory and field collected strains of *Cx. quinquefasciatus* that were exposed to *B. sphaericus* strain 2362 for about 100 generations and attained 37 and 31 fold resistance respectively, did not show marked cross resistance to *B.t.i.*; only 3-4 fold tolerance was noted. However, they showed significant levels of cross resistance to strains 1593 (6-22 fold) and 2297 (4-12 fold) of *B. sphaericus*[39]. This showed that any change in receptor sites or binding affinities in one of the *B. sphaericus* strains would confer cross resistance to other *B. sphaericus* strains because these strains produce similar binary toxins which bind to a single class of specific receptors[31,94]. The degree of cross resistance to strain 2297, which produces a different type of toxin, may be less pronounced[39,94]. Further, the absence of cross resistance between the two microbial agents *B. sphaericus* and *B.t.i.*[18] was due to differences in the structure and biochemistry of the toxins and differences in specific receptors for these toxins in *Cx. quinquefasciatus*[71]. Therefore, *Cx. quinquefasciatus* larvae, which were highly resistant (>100,000 fold)[38] and no longer had functional receptors for *B. sphaericus*, were still susceptible to *B.t.i.*[71]. Similarly, *B. sphaericus* toxin failed to bind to the midgut brush border membranes (BBM) of *Ae. aegypti* which is refractory to *B. sphaericus* but highly susceptible to *B.t.i.*[31,59]. Further, selection with biocontrol agents had no apparent effect on the resistance to organo phosphorus (OP), carbamate and synthetic pyrethroids when compared with the untreated controls[38,95].

Management of Resistance Development

Resistance management strategy is an important and integral part of any IVM programme which focuses on minimizing the economic and environmental impacts

resulting from pesticide treatments[75]. It is encouraging to note that resistance to pathogens by insects takes a longer time than is with chemical insecticides. This may be due to genetic changes at one or more loci against insect pathogens[97] resulting in poor biological fitness and reversal of resistance [78,79,97]

Studies on the biological fitness of laboratory and field collected strains of *Cx. quinquefasciatus*, susceptible and resistant (37 and 31 fold upon selection) to *B. sphaericus* showed that the resistant strains had significantly lower fecundity and fertility, but significantly higher survival rates than the susceptible strains. The lower production of eggs (fecundity) and lower percentage of hatch (fertility) in resistant population was found to be due to a lower nutrient reserve in the adults that developed from larvae treated with sublethal concentrations of pesticides[98-101]. The preadult stages from females of resistant strains developed at slightly faster rates than those of susceptible strains, which could result in a shorter generation time. However, lower fecundity was likely to lead to overall lower population growth rates than in the susceptible strains. Studies provided evidence that the resistant strains exhibited fitness disadvantages in the absence of *B. sphaericus*[78].

Resistance and survival fitness are reported to be independent[102]. Studies on the reversal of resistance to bacterial toxins in insect populations have shown that low and moderate resistance levels are unstable after the interruption of the treatment. This is mainly because of the decline in resistance gene frequencies due to mixing of a large proportion of susceptible individuals with the resistant ones due to immigration under field condition[40,103,104]. However, highly resistant populations seem to reach a plateau of resistance and require a long time to recover susceptibility[38,79,97,105]. It was observed that a larval population of *Cx. quinquefasciatus* breeding in habitats treated with *B. sphaericus* once a month for 26 months was 10 times less susceptible to *B. sphaericus* and slightly more susceptible to *B.t.i.* compared with the untreated population. The low level of resistance to *B. sphaericus* was unstable in the absence of selection pressure but reversed to be susceptible to *B. sphaericus* when the treatment was interrupted ofter 16 months[105].

However, the eventual acquisition of acquired resistance to microbial control agents among field populations of mosquitoes cannot be ruled out[106] as evidenced by recent findings in France and India[15,18]. This necessitates the formulation of rapid, elaborate and practical resistance management strategies. This is a very important consideration because of the efficacy of *B. sphaericus* against culicines, safety for the environment and usefulness in mosquito control programmes[72].

Among the various resistance management strategies, the use of biocides in rotation might be an efficient management measure because it disrupts selection pressure from the control agent and enables the insect population to recover susceptibility to the agent previously used[75]. Since the progression of resistance to *B.t.i.* was reported to be slow because of its multi-toxin complex[107], this biocontrol agent could be considered a good candidate for use in rotation with *B. sphaericus*, larvicidal oils and insect growth regulators (IGRs)[105]. Binding experiments with *B. sphaericus* radio labeled toxin in the presence of an excess of unlabeled *B.t.i.* toxin as a competitor, indicated that the receptors for *B. sphaericus* and *B.t.i.* toxins are different[31]. Other reports also indicated that *Cx. quinquefasciatus* larvae resistant to *B. sphaericus* (150 fold) were 7 fold more susceptible to *B.t.i.* than the untreated population[18]. Similar results have been reported for a population of *Plodia interpunctella* resistant to *B.t.i.*[108]. This allows for the use of *B.t.i.* for a relatively longer period, allowing the reversal of *B. sphaericus* resistance and consequently its reintroduction[105]. The use of synergists with *B.t.i.* proteins resulted in high toxicity and expanded host range[109]. In this respect it should be noted that studies on the interactions between toxins of *B.t.i.* and *B. sphaericus* and target insects would provide a useful basis for the appropriate use of these entomopathogens in large-scale programmes. This would facilitate exploring the possibility of combining *B.t.i* mosquitocidal toxin genes with *B. sphaericus* genes to increase toxicity and for delaying the development of resistance[44].

Further, there is no report of *B. sphaericus* linked to esterase overproduction conferring resistance to organophosphorous insecticides and no cross resistance to certain other *B. sphaericus* strains producing crystal toxins. This facilitates using this biocontrol agent in rotation with OP compounds[30].

The initial overproduction of anophelines in paddy fields could not be regulated by natural predators and parasites because of the time lag in the build up of the population of pests and natural enemies. The maximum production of mosquitoes occurs during the initial stages of plant growth when farmers usually apply fertilizers. These mosquito breeding habitats are so extensive that the use of larvicides as a control method will not be cost effective. As biocontrol agents such as *B. sphaericus* and *B.t.i* are non toxic to non-target organisms[110,111], it has

been recommended that they be applied along with fertilizer for the control of anopheline vectors in agro-ecosystems[112]. This strategy would check the rapid increase of pest population and subsequent control could be achieved by naturally occurring parasites and predators. However, farmers should be educated to avoid contact with the bacillus preparation as recent finding indicated that *B.t.i.* might cause serious wound infection in animals and man[113].

The combined use of natural enemies such as *Gambusia affinis*[28], *Toxorhynchites rutilus rutilus*[114], *Mesocyclops longisetus*[115] and *Notonecta irrorata*[116] and *B.t.i.* has been proved successful in the effective control of mosquitoes in a cost effective and environmentally safe manner. The control of *Culex tarsalis* larvae was achieved by integrating both *B. sphaericus* and *G. affinis*[117]. This approach may slow down resistance development among the vector populations by inflicting a disproportionately high mortality on resistant individuals[118]. However, if the natural enemies kill a higher proportion of susceptible individuals, with the biocides acting equally on both the populations, there may be an increase in the population of resistant individuals[119].

Future Prospects

Use of control release formulations such as calcium alginate/Carrageenan microcapsule and encapsulating the biocontrol agent in a living organism (*Tetrahymena pyriformis*) would retain the toxin on the water surface and act as a phagostimulant[5,120]. Genes encoding specific toxicity could be incorporated with the larval food sources, such as algae and *Caulobacter spp.* and the response of mosquito immatures and other aquatic organisms on modified food sources should be determined[121]. New toxins identified from *Clostridium bifermentans* strain and several *B.t.i* strains appeared to be good candidates for use in combination with *B. sphaericus* toxin[122]. However, Federici[34] cautioned that the release of transgenic insecticidal organisms used as larval food into the breeding habitat might be counter productive. New genetic engineering approaches such as recombinant DNA techniques should be envisaged to overcome these limitations that will allow stable expression of broad host range, buoyancy and resistance to ultraviolet light. The recombinant strains are relatively inexpensive, easy to mass-produce and possess excellent shelf-life[67,109,123-125]. The development of CytA/Cry4/Cry11 and CytA/*B.s.* models from a study on the effect of the CytA protein on resistance to *B.t.i* proteins and *B.s.* preparations provided a potential molecular genetic strategy for engineering resistance management for Cry proteins directly into bacterial insecticides[126,127]. The production of a bacterium combining the best attributes of both *B.t.i* and *B. sphaericus* and studies on the specificity of toxicity was suggested[41]. Grigorova *et al.*[128] demonstrated transfer of plasmids from *Streptococcus agalactine* and *S. pyrogenes* to *B.s.* but did not investigate further transfer among *B.s.* strains. Orzech and Burke[129] reported that *B.s.* 1593 could accept conjugated plasmid pAMβ1 from *Streptococcus faecalis* JH2.2 and could transfer pAMβ1 to another strain of *B.s.* The plasmid phSP11 that contained genes coding the binary toxins of *B.s.* was introduced into the Colombian strain of *B.t.i.* to study its expression and stability. The presence of the plasmid was detected after 8 h of growth, coinciding with the movement that the expression of the plasmid-coded toxins started[130]. Rodcharoen and Mulla[77] suggested that the possibility of encoding toxic genes of *B. sphaericus* being transferred to blue green algae if genetically engineered pathogens are used in a control programme, be explored. However, a recent observation opined that it was unlikely that genetically engineered *B.s.* carrying a recombinant plasmid could transfer that plasmid to other bacteria[131]

Conclusions

Though high levels of resistance in *Cx. quinquefasciatus* were reported from laboratory and field studies, none of the studies have examined the gut content of the larvae for spores. It is possible that due to bad formulation, the spores might have clumped and could not have been ingested, or sunk to the bottom. Reversal of resistance was observed in the field when the treatments were withdrawn[44]. There are no reports of cross resistance to *B.t.i*[38,39,71], and organophosphorous insecticides[38,95]. Cross resistance between strains of *B. sphaericus* harbouring different sequences has not been found to occur[38]. There have been considerable studies on the positive interactions of biocontrol agents with other control agents such as chemicals[132]. Under normal dosages *B. sphaericus* has a minimum impact on non-target organisms[11,52,110,111,133], and is non-toxic to mammals[134,135]. Therefore, there is scope for using *B. sphaericus* along with other control methods in the integrated vector management of *Culex* mosquitoes.

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a 3 fold risk was observed for those who consumed alcohol. It was observed that the patients who consumed eggs had nearly 2 fold risk of urolithiasis than those who did not consume eggs. Similarly a 2 fold risk of urolithiasis was observed in patients who consumed flesh food twice or even less than twice per week compared to vegetarian. Consumption of *rajma* more than twice a week also resulted in 2 fold higher risk of urolithiasis.

It was thus concluded that urinary tract infections, smoking, alcohol intake and consumption of egg, non vegetarian food items as well as *rajma* were associated with increased risk of urolithiasis.

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Ergonomic evaluation of different modes of load carrying and maximum permissible load for Indian women engaged in manual material handling.

The study was carried out to document the existing modes of manual load carrying by Indian women and determine the physiological cost involved during the process.

Two basic approaches (field and laboratory studies) were employed for the study. Several parameters like personal and medical history, sites of pain in the body, *etc.* were ascertained through a questionnaire. Anthropometry, physiological cost of the work, work-rest cycle, walking speed, environmental heat load and energy consumption were determined by direct measurements on the subjects in the field.

Women involved in construction activities carried the load on the head but in case of water carrying four basic modes like head, shoulder, waist and hand mode were observed. Besides these, women had adopted several other modes for carrying water like head and hand, head and waist, waist and hand *etc.* Ninety three women engaged in construction activities and 76 women engaged in water carrying activities were selected at random for detailed study. Anthropometric details were recorded on 47 women workers from construction and 25 women workers from water carrying activities.

In the case of women engaged in construction activities the average body height, weight and age were 149.7 (±4.7) cm, 42.8 (±5.5) kg and 28.5 (±7.1) yr, respectively. The percentage of fat, lean body weight and body surface area was 21.2 (±3.7)%, 34.0 (±0.9) kg and 1.5 (±0.1) m^2, respectively. The average energy intake was 1355 kcal. The average walking speed in the field was 3.4 km/h and the maximum heart rate of 163 beats/min was observed for women engaged in block carrying. Heart rates during other activities like carrying concrete for column and beam, carrying concrete for block making and mud transferring were between 110-135 beats/min. The O_2 demand varied from 0.77 l/min (45.31% of VO_2 max) to 1.41 l/min(82.7% of VO_2 max); being lowest at ground level and maximum after climbing to the 4th floor. The maximum aerobic power obtained was 1.71 l/min. A regression equation between heart rate and O_2 consumption was derived.

In case of women carrying water the average body height, weight and age was 150.0 (±5.3) cm, 50.4 (±10.9) kg and 30.5 (±8.1) yr, respectively. The fat content, lean body weight and body surface area was 25.6 (±5.9)% of the whole body weight, 36.6 (±6.0) kg and 1.6 m^2, respectively. It was evident that women involved in water carrying had significantly more body weight, fat content and lean body weight as compared to those involved in construction activity. The women involved in water carrying had work experience ranging from a few months to 25 years and more. The water was being carried by *HANDA* and load varied from 10 to 50 kg, while in case of construction activity the weight varied from 9-34 kg. Women carrying water reported the back, elbow, upper arm, calf and knee joint as the major sites of pain. Similar sites of pain were also reported by the construction workers. About 6.5% women water carriers reported miscarriage while miscarriage was reported by 8.3 women involved in construction activity. The average walking speed for water carrying was 3.8 km/h which was slightly higher than that of construction group. The average working heart rate was 129 beats/min.

The average maximum heart rate and O_2 consumption for women carrying water during maximum aerobic power was 182.3 beats/min and 1.47 l/min respectively, being lower than that of construction workers. This indicates that the physical work capacity of the construction workers was superior to that of women engaged in water carrying activity.

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ICMR NEWS

The following meetings of various technical groups/ committees of the Council were held:

Meetings of the Scientific Advisory Committee (SAC)/ Project Review Committee (PRC)/Task Forces (TFs) held:

PRC on Oncology and Pathology	December 17-18, 1998 (at New Delhi)
TF on Drug Resistance in Malaria	December 31, 1998 (at New Delhi)
TF on Diagnostics in Malaria	January 11, 1999 (at New Delhi)
SAC of the Desert Medicine Research Centre, Jodhpur	January 12-13, 1999

Participation of ICMR Scientists in Scientific Events:

Dr. S.M. Mehendale, Dy. Director, National AIDS Research Institute, Pune and Dr. Nandini K. Kumar, Sr.Research Officer. ICMR Headquarters. New Delhi. participated in the workshop on International Research Ethics at Baltimore (January 5-6, 1999).

Dr. Neena Valecha, Asstt. Director, and Dr. C. Usha Devi, Sr. Research Officer, Malaria Research Centre, Delhi, participated in the workshop on Azithromycin in Malaria at Kathmandu (January 14, 1999).

Appointment:

Dr. Ashok Mukherjee took over as Director of the Institute of Pathology, New Delhi w.e.f. January 7, 1999.

Workshop:

An Indo-US Workshop on Behavioural Prevention Research on HIV/AIDS was organized in New Delhi during January 12-13, 1999 for identification of areas for collaboration through joint projects.

ICMR AIDED SYMPOSIA/SEMINARS/WORKSHOPS/COURSES/CONFERENCES

Symposium/Seminar/Workshop/ Course/Conference	Date & Place	Contact Address
LXXXVI Session of the Indian Science Congress Association	January 3-7. 1999: (at Chennai)	Prof. Kunthala Jayaraman. Dean of Technology & Professor of Biotechnology. Anna University. Chennai-600025.
V International Symposium on Biochemical Roles of Eukaryotic Cell Surface Macromolecules	January 4-8. 1999: (at Bangalore)	Dr. A. Surolia. Convener of the Symposium. Molecular Biophysics Unit. Indian Institute of Science. Bangalore-560012.
Workshop on Scientific Communication	January 9-10. 1999: (at Mumbai)	Dr. S.R. Naik. Professor & Head. Department of Gastroenterology. Sanjay Gandhi Postgraduate Institute of Medical Sciences. Lucknow-226014.
National Workshop on Helicobacter/Campylobacter	January 19-23. 1999: (at Lucknow)	Dr. A. Ayyagari. Organising Secretary of the Workshop. Department of Microbiology. Sanjay Gandhi Postgraduate Institute of Medical Sciences.. Lucknow-226014.
National Symposium on Biological Control of Insects in Agriculture. Forestry. Medicine and Veterinary Science	January 21-22. 1999: (at Coimbatore)	Dr. K. Murugan. Organising Secretary of the Symposium. Division of Entomology. Department of Zoology. Bharathiar University. Coimbatore-641046.
XVII National Conference of the Society for Reproductive Biology and Comparative Endocrinology	January 23-25. 1999: (at Surat)	Dr. Nandita J. Arbatti. Organising Secretary of the Conference. Department of Biochemistry. Government Medical College. Surat-395001.

ICMR NEWS

The following meetings of various technical groups/ committees of the Council were held:

Meetings of the Scientific Advisory Committee (SAC)/ Project Review Committee (PRC)/Task Forces (TFs) held:

PRC on Oncology and Pathology	December 17-18, 1998 (at New Delhi)
TF on Drug Resistance in Malaria	December 31, 1998 (at New Delhi)
TF on Diagnostics in Malaria	January 11, 1999 (at New Delhi)
SAC of the Desert Medicine Research Centre, Jodhpur	January 12-13, 1999

Participation of ICMR Scientists in Scientific Events:

Dr. S.M. Mehendale, Dy. Director, National AIDS Research Institute, Pune and Dr. Nandini K. Kumar, Sr.Research Officer. ICMR Headquarters. New Delhi. participated in the workshop on International Research Ethics at Baltimore (January 5-6, 1999).

Dr. Neena Valecha, Asstt. Director, and Dr. C. Usha Devi, Sr. Research Officer, Malaria Research Centre, Delhi, participated in the workshop on Azithromycin in Malaria at Kathmandu (January 14, 1999).

Appointment:

Dr. Ashok Mukherjee took over as Director of the Institute of Pathology, New Delhi w.e.f. January 7, 1999.

Workshop:

An Indo-US Workshop on Behavioural Prevention Research on HIV/AIDS was organized in New Delhi during January 12-13, 1999 for identification of areas for collaboration through joint projects.

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International Workshop on Hypospadias and the Child with Undetermined Sex	January 30-31, 1999: (at New Delhi)	Dr. D.K. Gupta, Convenor of the Workshop, Department of Paediatric Surgery, All India Institute of Medical Sciences, New Delhi-110029.
Seminar on Low Level Electromagnetic Field Phenomena in Biological System	February 3-4, 1999: (at New Delhi)	Dr. J. Behari, Professor of Environmental Physics, School of Environmental Sciences, Jawaharlal Nehru University, New Delhi-110067.
I National Conference on Hepatitis and Renal Medicine	February 13, 1999: (at New Delhi)	Dr. Sanjay K. Agarwal, Organising Secretary of the Conference, Department of Nephrology, All India Institute of Medical Sciences, New Delhi-110029.
VII Annual Meeting of Molecular Immunology Forum - 1999	February 13-15, 1999: (at Hyderabad)	Dr. Ashok Khar, Convener of the Meeting, Centre for Cellular and Molecular Biology, Hyderabad-500007.
IV International Symposium on Vectors and Vector-borne Diseases	February 18-20, 1999: (at Gwalior)	Dr. P.R. Malhotra, Convener of the Symposium, Defence Research and Development Establishment, Gwalior-474002.
XVIII Annual Convention of Indian Association for Cancer Research and National Symposium on Molecular Biology of Cancer	February 19-21, 1999: (at New Delhi)	Dr. Neeta Singh, Addl. Professor, Department of Biochemistry, All India Institute of Medical Sciences, New Delhi-110029.
AOCS-OTAI International Conference and Exhibition on Oil Seeds and Vegetable Oil Processing	February 19-21, 1999: (at New Delhi)	Sh. K.P. Sharma, Organising Secretary of the Conference, Oil Technologists Association of India, 1205, Nirmal Tower, Barakhambha Road, New Delhi-110001.
Technical Training Workshop on Industrial Hygiene Measurements and National Seminar on Occupational Safety and Health	March 1-6, 1999: (at Chennai)	Dr. Kalpana Balakrishnan, Organising Secretary of the Workshop, Sri Ramachandra Medical College and Research Institute, Chennai-600016.

COUNCIL'S TRAINING PROGRAMMES FOR 1999-2000

Reproductive Biology

At the Institute for Research in Reproduction, Mumbai:

- Training Course in Molecular Techniques in Ovarian Function (September 27-October 2, 1999).
- Training Course on Cytological Detection of Reproductive Tract Infections (December 13-17, 1999).

Endocrinology

At the National Institute of Nutrition,, Hyderabad:

- Annual Certificate Course on Endocrinological Techniques and their Application (August 1 - September 15, 1999).

Nutrition

At the National Institute of Nutrition, Hyderabad:

- M.Sc. in Applied Nutrition (June 1, 1999-February 28, 2000).
- Postgraduate Certificate Course in Nutrition (December 1, 1999-February 28, 2000).

Laboratory Animal Technology

At the National Centre for Laboratory Animal Science, National Institute of Nutrition, Hyderabad:

- Training Course for Laboratory Animal Technicians (June 14-July 30, 1999).

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- Training Course for Laboratory Animal Supervisors (September 1-November 30, 1999).

Medical Entomology:

At the Vector Control Research Centre, Pondicherry:

- Postgraduate Diploma Course in Medical Entomology (from July 1999).

Biomedical Statistics

At the Institute for Research in Medical Statistics, Chennai:

- Basic Course in Statistics for Medical Officers (July 1999).
- Training Course in Field Epidemiology (November, 1999).

ICMR PUBLICATIONS

	Price(Rs.)
Nutritive Value of Indian Foods (1985) by C. Gopalan, B.V. Rama Sastri and S.C. Balasubramanian, Revised and Updated (1989) by B.S. Narasinga Rao, Y.G. Deosthale and K.C. Pant (Reprinted 1996)	23.00
Growth and Physical Development of Indian Infants and Children (1972, Reprinted 1989)	10.00
Studies on Weaning and Supplementary Foods (1974, Reprinted 1996)	15.00
A Manual of Nutrition (Second Edition 1974, Reprinted 1995)	6.00
Low Cost Nutritious Supplements (Second Edition 1975, Reprinted 1996)	5.00
Menus for Low-Cost Balanced Diets and School-Lunch Programmes Suitable for North India (Second Edition 1977, Reprinted 1994)	4.50
Menus for Low-Cost Balanced Diets and School-Lunch Programmes Suitable for South India (Fourth Edition 1996)	6.00
Some Common Indian Recipes and their Nutritive Value (Fourth Edition 1977, Reprinted 1995) by Swaran Pasricha and L.M. Rebello	10.00
Nutrition for Mother and Child (Fourth Edition 1994, Reprinted 1996) by P.S. Venkatachalam and L.M. Rebello	11.00
Japanese Encephalitis in India (Revised Edition 1980)	5.00
Some Therapeutic Diets (Fifth Edition 1996) by Swaran Pasricha	7.00
Nutrient Requirements and Recommended Dietary Allowances for Indians (1990,Reprinted 1995)	16.00
Fruits (Second Edition 1996) by Indira Gopalan and M. Mohan Ram	20.00
Count What You Eat (1989, Reprinted 1997) by Swaran Pasricha	15.00
*The Anophelines of India (Revised Edition 1984) by T.Ramachandra Rao	150.00