6.0 DISCUSSION
Fig. 25: Initial Predation (IP) and Final Predation (FP) of Guppy and Panchax individually on *Anopheles stephensi* larvae
6.0 DISCUSSION

A. Density of wild caught mosquitoes in different biotopes and its relation to malaria cases.

The literature available on control of malaria in Calcutta itself revealed involvement of much time and money for undertaking general measures exclusively, even without making any preliminary study of the nature of the local problem. Every malaria problem, especially the vector problem, should first be subjected to field studies to determine its specific nature and define its limits (Samuel, 1926).

*Anopheles stephensi* is the recognised and incriminated potential vector of malaria in the city of Calcutta (Siddons, 1946, Mukhopadhyay 1980, Hati et al., 1987). The density of the vector is one of the major factors affecting the epidemiology of malaria. It determines the degree of contact between man and the vector and for that matter the intensity of malaria transmission. But the information regarding density factor of *Anopheles stephensi* in this metropolis is far from adequate, practically due to non-availability of species from the conventional resting places. Hence, the density factor of *Anopheles stephensi*, the age-old problem in city, remains yet to be settled.

The objectives were to get an idea what were the species of mosquitoes available in a malaria endemic area, whether there would be any seasonal fluctuation of studied vector species, with special emphasis on *Anopheles stephensi* in two consecutive years and whether association could be established between the density of vector mosquitoes and the occurrence of clinical infections in the human population of the study area. Such type of longitudinal study extending over two consecutive years had not been conducted earlier.

Altogether 9 species of mosquitoes were obtained from the six study sites during the study period belonging to the genera *Culex, Anopheles, Aedes* and *Armigeres* which came to take rest in human habitations. Besides, brick-built rooms, slum rooms and shanties were also searched for the presence of *Anopheles stephensi*, throughout the study period. The mosquito fauna was represented by the only species of *Culex*, namely, *Culex quinquefasciatus*.

The total number of *Culex quinquefasciatus* obtained in two years was 38,915. In the genus *Anopheles*, there were six species. The names of the species and the total numbers in two years were, *Anopheles subpictus* (876), *Anopheles vagus* (427), *Anopheles stephensi* (171),
Anopheles barbirosiris (45), Anopheles annularis (39) and Anopheles hyrcanus (4). 150 Aedes aegypti and 558 Armigeres subalbatus mosquitoes were also obtained.

Besides six study sites, 40 brick-built rooms, 30 shanties and 25 slum rooms were also searched, from where 76 Anopheles stephensi were obtained. Among the above three biotopes, it was found that the number of collection of Anopheles stephensi was significantly higher in shanties (39) in comparison to slum room (22) and brick-built rooms (15).

So far as the collections of mosquitoes were concerned, in the human habitations in the two years of consecutive study period i.e. July 2000-June 2001 and July 2001-June 2002, the first position was taken by Culex quiquefasciatus in both the years.

Among the Anopheles mosquitoes, Anopheles subpictus, Anopheles vagus, Anopheles stephensi, Anopheles barbirosiris, Anopheles annularis and Anopheles hyrcanus occupied the first, second, third, fourth, fifth and sixth position respectively in both the years.

Altogether 9 species of mosquitoes were obtained from the six study sites in both the years. In Monoharpukur, Anopheles stephensi were not found in the month of October 2000 to February 2001, May 2001, July-August 2001, October-November 2001, February 2002 and April-May 2002 respectively. Anopheles subpictus were not found in the month of October-December 2000, February 2001, September-November 2001, January 2002 and April 2002 respectively.

In Taratala, only 19 Anopheles stephensi were found in the month of July 2000, September 2000, November 2000, June 2001, August 2001, January 2002 and April-May 2002 respectively. Similarly, Anopheles subpictus were found in the month of July-August 2000, February-April 2001, June 2001, August 2001, October 2001, December 2001, March-April 2002 and June 2002 respectively. Though Anopheles annularis were not found in Taratala. But few species of Aedes aegypti (19) were found in Taratala.

In Gajtala, Anopheles stephensi were found in the month of July-August 2000, March 2001, May-July 2001, September-October 2001 and March 2002 respectively. Similarly, Anopheles subpictus were found in the months of July-October 2000, January 2001, March-May 2001, July 2001, September 2001, December 2001, Feb-March 2002 and May-June 2002 respectively. Though Aedes aegypti were not found in Gajtala, Anopheles annularis were found in small number.

In Kankurgachi, *Anopheles stephensi* were found in the month of July-September 2000, November 2000, March-April, August 2001, December 2001 and May-June 2002 respectively. *Anopheles subpictus* were found in the month of July 2000, September 2000, December 2000, February 2001, April-July 2001, October 2001, March 2002 and May 2002 respectively. *Anopheles annularis* were not found but *Aedes aegypti* were found few in number.

In Kasba Golpark, *Anopheles stephensi* were found in the months of Aug-Oct 2000, Feb 2001, April-May 2001, Aug. 2001 and May-June 2002 respectively. *Anopheles subpictus* were found in the months of July-Dec 2000, March 2001, May 2001, July 2001, December 2001 and March-April 2002 respectively. *Anopheles annularis* and *Aedes aegypti* both were found, though they are few in number.

Amongst the Anopheline species, available both the years from the six study sites, *Anopheles stephensi* were caught in maximum number from Monoharpukur (57) and least from Kasba Golpark (12). Only 20 *Anopheles stephensi* mosquitoes from Gajtala, 19 from Taratala, 38 from Bowbazar and 25 from Kankurgachi were found from the rest four study sites. *Anopheles subpictus* were found maximum in Bowbazar (191) and least in Taratala (85). 161 *Anopheles subpictus* mosquitoes from Monoharpukur, 169 from Gajtala, 149 from Kankurgachi and 121 from Kasba Golpark were found from the rest four study sites. *Anopheles annularis* were found from the peri-urban area of Calcutta, i.e. only *Anopheles annularis* were found from Gajtala (11) and Kasba Golpark (28) Seasonal fluctuations in population density of the Anopheline species were also noticed amongst the study sites.

Taken as a whole, amongst the six study sites, during the first year in Monoharpukur *Anopheles stephensi* collection was highest in Summer (18) than that of the Rainy season (14). During the second year, collection of *Anopheles stephensi* was (8), both in Summer and Rainy season. In Winter, maximum (9) *Anopheles stephensi* were collected.
During the first year, in Gajtala, maximum collection of *Anopheles stephensi* was in Summer (7) than that of Rainy (6) season. During the second year, maximum collection (6) was in Rainy season than that of Summer (1). In Taratala, during the first year, collection of *Anopheles stephensi* was (5), each in Summer and Rainy season. In Winter, only 1 (one) *Anopheles stephensi* was collected. During the second year, collection of *Anopheles stephensi* was (2), each in Summer and Winter season. Maximum *Anopheles stephensi* collection (4) was observed in Rainy season.

In Bowbazar, maximum *Anopheles stephensi* collection (11) was in Rainy season than that of Summer season (8), during the first year. During the second year, maximum *Anopheles stephensi* collection was encountered in Rainy season (10) than that of Summer (7) and Winter (2) season.

In Kankurgachi, during the first year, maximum *Anopheles stephensi* collection was in Rainy (9) than that of Summer (3) and Winter (2) season. During the second year, maximum collection of *Anopheles stephensi* was found in Summer (6) than that of Rainy (2) and Winter (3) season.

In Kasba Golpark, during the first year, maximum collection of *Anopheles stephensi* was found during Rainy (5) than that of Summer (3) and Winter (1). During second year, maximum collection of *Anopheles stephensi* was Summer (2) than that of Rainy (1) season.

So far as *Anopheles stephensi* was concerned, there was some variation as regards the density in different season in both the years. This points out that *Anopheles stephensi* can effectively utilise different seasons to build up its population in comparison to *Anopheles subpictus* and *Anopheles annularis* which is unique.

The study reveals that so far as vector mosquitoes belonging to the genera *Culex, Anopheles, Aedes* are concerned, the situation in Calcutta is very bleak. Some Anopheline species of mosquitoes were available in maximum number, high population density of some species may be achieved suddenly, some species were highly sensitive to seasonal changes, and their population to seasonal changes differs significantly from year to year. In the present study, it was seen that maximum number of *Anopheles stephensi* were found in Rainy season than that of Summer and Winter season from all the study sites. Hati, et al. 1987 also made the similar observation. During their study, maximum collection of *Anopheles stephensi* mosquitoes (90%) was observed during the monsoon and post-monsoon months. It is interesting to note that 29.1% *Anopheles stephensi* adults were collected in July. From November 1985 to May 1986, the density was very low and actually no *Anopheles stephensi* was obtained in the month of January and March.
1986 Chatterjee et al (1993) also found similar observations. They found maximum number of *Anopheles stephensi* during the rainy season. The observation of Hati et al (1987) and Chatterjee et al (1993) corroborates with the present study, which suggests that in Calcutta, rainy season is more conducive to the building up of adult population of *Anopheles stephensi* than Winter and Summer seasons. The seasonal fluctuation of *Anopheles subpictus* was also noticed in the present study. Except Taratala and Kankurgachi, during the first year, *Anopheles subpictus* were found to be maximum in the Rainy season than that of Summer and Winter season in Monoharpukur, Gajtala, Bowbazar and Kasba Golpark. During the second year, the scenario was not the same. Bowbazar and Kasba Golpark encounters least number of *Anopheles subpictus* in the Rainy season than that of Summer and Winter season. But during the first and the second year, *Anopheles annularis* collection was more in the Rainy season than that of Summer and Winter.

The per man hour (PMH) collection of *Anopheles subpictus* was found to be higher amongst the six Anopheline species caught during the study period. Amongst the six study sites, per man hour collection of *Anopheles subpictus* was highest in Bowbazar (191, 0.49) and lowest in Taratala (85, 0.22). Similarly, per man hour collection of *Anopheles stephensi* was highest in Monoharpukur (57, 0.14) and lowest in Kasba Golpark (12, 0.031). Maximum number of *Anopheles annularis* was found in Kasba Golpark (28, 0.07) and minimum in Gajtala (11, 0.02). Per man hour collection of *Anopheles vagus* was found to be highest in Bowbazar (126, 0.32) and lowest in Taratala (15, 0.039) and that of *Anopheles barbirostis* was highest in Monoharpukur (29, 0.075) and lowest in Gajtala (16, 0.041).

To determine biotope-wise prevalence of *Anopheles stephensi*, different biotopes, namely brick-built rooms, slum rooms and shanties were searched. According to the observations made by the early investigators (De, 1923, Senior White, 1934, Knowles and Basu, 1934, Ganguly, 1935, Roy et al., 1938, Senior White, 1940, Siddons, 1943, Mukhopadhyay, 1980), *Anopheles stephensi* was of secretive habit and its presence was not easily detected. But from the present study, it would be clear that *Anopheles stephensi* lost its secretive habits being found in considerable number from different biotopes of Calcutta.

The determination of “per man hour collection” has been used most extensively, not only to study fluctuations in the abundance but also to assess the success or failure of control measures (Service, 1976). In this study, the maximum per man hour collection of *Anopheles stephensi*...
was in the month of June '01 (0.37) in brick built rooms. The corresponding data in slum rooms and shanties were 0.60 (November, 2001) and 0.42 (July, 2000 and June, 2001) respectively. The collection made from the shanties, where per man hour collection (0.42) was found to be higher than all the habitations searched, should be regarded as an exception. Chatterjee & Hati (1986) also made similar observation.

The previous investigators (Stickland et al., 1936; Roy et al., 1938, Senior White, 1940, Mukhopadhyay, 1980) found that *Anopheles stephensi* were available in brick-built rooms of Calcutta. But it would be clear from the present study that *Anopheles stephensi* was found maximum in shanties and slum rooms. This clearly indicate that they have changed their preference of resting habits abruptly.

Nursing et al. (1938) reported that in Mysore (Karnataka), *Anopheles stephensi* showed a decided preference in human habitations for day time resting places. It preferred to rest in poor habitations and cattlesheds indoors in Kutch of Gujrat (Afridi et al., 1938) *Anopheles stephensi* were available in human dwellings in Poona (Barber and Rice, 1938) and Vizagapatnam (Senior White and Rao, 1941). Maximum preference was found in mixed (human and cattlesheds) dwellings in Madras (Bhaskar Rao et al., 1946). It was obtained from “pucca” houses, cattlesheds and godown indoors and walls of the wells in Salem, Tamilnadu (Batra et al., 1979).

The indoor resting of *Anopheles stephensi* was of very high density as reported from different parts of the country and abroad (Subbarao and Apparao, 1945, Bhaskar Rao et al., 1946, Subbarao et al., 1984, Singh et al., 1985, Sharma et al., 1985). In Iran, maximum density of *Anopheles stephensi* was found to be 200 per room (Zahar, 1974), though such a high density of *Anopheles stephensi* was not recorded during the present study.

*Anopheles stephensi, Anopheles subpictus* and *Anopheles annularis* are the recognized malaria vectors in India (Rao, 1984, Panicker et al., 1981, Kulkarni, 1983). For the last few decades, investigators, however few, have been referring *Anopheles stephensi* as the only vector in Calcutta. Albeit no attempt has so far been made to find out the role of other Anopheline species, namely, *Anopheles subpictus, Anopheles annularis* in the transmission of malaria in Calcutta. Bhattacharya and Mukherjee (1996) stated that there is prenual transmission of malaria in Calcutta and there exist seasonal variations in the incidence of malaria.
B. Resting habitat of *Anopheles stephensi*

The determination of indoor resting sites of any anthropod vector is of paramount importance for the estimation of the population indices before and after control operations for an appropriate evaluation, to adopt proper chemical control measure, as well as for the determination of infection rates.

The adult *Anopheles stephensi* preferred to rest upon coloured funnels, cotton clothes, fans, etc and were easily available in wardrobes which were left open during night and filled with clothes, beneath tables or a desk left in a corner of a room or behind the dark curtain. Interestingly in the present study, *Anopheles stephensi* were captured from folded umbrellas, gunny bags, inside empty tin drums, furniture ceiling, iron pillars, etc in different biotopes i.e. brick-built rooms, slum rooms and shanties. List of the resting sites of *Anopheles stephensi* as described by Roy et al (1938) also did not corroborate the findings of the present study. Rafi (1955) witnessed in Punjab (Pakistan) resting of *Anopheles stephensi* throughout the height of the wall inside rooms. The observation made in this study does not reinforce this view as in the present cases, *Anopheles stephensi* was found in lower number in brick built rooms in Calcutta.

C. Manbait experiment

The direct collection of mosquitoes coming to bite man gives important epidemiological information and this can be measured with a reasonable degree of accuracy, if properly planned and carried out (Pant et al, 1980).

In the study area, monthly captures during investigations over two years has yielded altogether 8 species of mosquitoes from Manoharpukur and 9 species of mosquitoes from Gajtala on man baits at night hours from 6pm to 6am. From the data, per man per night collection of mosquitoes can be calculated in an area, where malaria is endemic.

Culicine mosquitoes were attracted to human beings in much greater number in comparison to Anopheline mosquitoes (7483 *Culex* mosquitoes against 436 *Anopheles* mosquitoes). The overall per man per night collection of *Culex quinquefasciatus, Anopheles subpictus, Anopheles vagus, Anopheles stephensi* and *Anopheles barbirostris* was 65.47, 3.34, 0.79, 0.95 and 0.12 respectively in Manoharpukur. Similarly, the overall per man per night collection of *Culex quinquefasciatus, Anopheles subpictus, Anopheles vagus, Anopheles stephensi, Anopheles barbirostris, Anopheles hyrcanus* and *Anopheles annularis* was 90.41, 1.56, 1.12, 0.42, 0.03.
Anopheles subpictus constituted 4.57% of the total catch, followed by Anopheles stephensi (1.31%) in Monoharpukur. Similarly Anopheles subpictus constituted 1.53% of the total catch, followed by Anopheles stephensi (0.41%) and Anopheles annularis (0.29%) respectively in Gajtala.

The results revealed that at night, a total of 66 Anopheles stephensi mosquitoes landed on human bait in two years. The number is less in comparison to the finding of Chatterjee & Hati (1986). Anopheles annularis is considered a rural vector species (Ghosh et al., 1985) but its presence in certain pockets in Calcutta has been reported by Sen (1938), Pramanik et al., (1992) and Bhattacharya et al., (1996). In fact, ecological condition of some areas under Calcutta Municipal Corporation is almost similar to rural settings.

Anopheles stephensi has been collected more in outdoors than indoors in all the months except in July 2000 and July 2001 at Monoharpukur. Man-vector (Anopheles stephensi) contact was absent both indoors and outdoors in the month of September 2000, November 2000, December 2000, January 2001 to May 2001, September 2001, December 2001 and January to June 2002 at Monoharpukur. Similarly, man-vector (Anopheles stephensi) contact was absent both indoors and outdoors in the month of January to May 2001, November 2001, December 2001 and January to May 2002 at Gajtala.

Samini (1966) in Southern Iran reveals that Anopheles stephensi landed on man bait both indoors and outdoors but proportion of landing outdoor was greater than indoor. But the study of Chatterjee and Hati (1986) showed that landing indoor (42) was greater than outdoor (12) which was significant at 5% level. The present study corroborates with the findings of Samini in Southern Iran but differs with the study of Chatterjee and Hati in Calcutta.

In the study period spanning over two years, altogether 8 species of mosquitoes, namely, Culex quinquefasciatus (PMH 5.45), Anopheles subpictus (PMH 0.28), Anopheles vagus (PMH 0.06), Anopheles stephensi (PMH 0.07), Anopheles barbirostris (PMH 0.01), Aedes aegypti (PMH 0.13), Armigeres subalbatus (PMH 0.04), and Mansonia annulifera (PMH 0.001) landed on human bait both indoors and outdoors at Monoharpukur. Similarly, 9 species of mosquitoes, namely Culex quinquefasciatus (PMH 7.53), Anopheles subpictus (PMH 0.13), Anopheles vagus (PMH 0.09), Anopheles stephensi (PMH 0.03), Anopheles barbirostris (PMH 0.027), Anopheles hyrcanus (PMH 0.01), Anopheles annularis (PMH 0.024), Armigeres subalbatus (PMH 0.62) and Mansonia annulifera (PMH 0.005) landed on human bait both indoors and outdoors at Gajtala.

Per man per night contact of Anopheles stephensi at Monoharpukur was
0.95 and at Gajtala was 0.42. Per man hour collection at night was 0.023 in 1980 as cited by Mukhopadhyay, 1980. Chatterjee and Hati (1986) showed that per man per night contact of *Anopheles stephensi* was found to be 1.12. The above data reveals that during eighties per man per night contact was significantly low (0.023) but from mid-eighties the figure rose as high as 1.12 till nineties. During the present study, the figure again falls. It was 0.95 at Mohaharpukur and 0.42 at Gajtala. So there is a variation of PMPN density of *Anopheles stephensi* landed on human bait in different areas and years. Per man per night contact of *Anopheles subpictus* was 3.34 at Monoharpukur and 1.56 at Gajtala, which was significantly higher than that of *Anopheles stephensi*.

The density of *Anopheles subpictus* mosquitoes increased in the IV quadrant of night (74, 46.25%) at Monoharpukur and in the III quadrant of night (45, 60.00%) at Gajtala. No *Anopheles annularis* was found at Monoharpukur during man-bait. *Anopheles annularis* came to bite at 12-01 am, 01-02 am, 02-03 am, 03-04 am, 04-05 am and 05-06 am during first year (July 2000 to June 2001) and at 12-01 am, 01-02 am, 02-03 am, 03-04 am, 04-05 am and 05-06 am during second year (July 2001 to June 2002) at Gajtala. The peak biting activity of *Anopheles annularis* was observed between 04-05 am. The density of *Anopheles annularis* increased in the IVth quadrant of night (9, 64.28%)

Monthwise variation was also noticed in respect of prevalence of *Anopheles stephensi* on man bait. The collection reached the peak in the months of July 2001 (4, 26.67%) and August 2000 (3, 20.00%) in indoor and in the months of September 2001 (5, 16.12%) and October 2000 (4, 12.90%) at outdoor in Monoharpukur. Similarly, the highest collection of *Anopheles stephensi* was observed in the months of July 2001 (3, 15.00%) at outdoor in Gajtala. No *Anopheles stephensi* collection was observed during Summer months (March-June). As *Anopheles stephensi* is a fresh water breeder, it breeds more in the rainy season, thereby increasing the number of manlanding mosquitoes in that season in Calcutta.

**D. Infection and Infectivity rate of Malaria Vectors**

Iyenger, 1933, Strickland et al 1936, Knowles and Basu, 1944 stated that in Calcutta *Anopheles stephensi* was proved to be a good reservoir of both *Plasmodium vivax* and *Plasmodium falciparum* infection in the laboratory conditions. Siddons, 1946 first incriminated *Anopheles stephensi* as a vector of malaria in Calcutta. The natural infection rate was found to be 0.85 (oocyst 0.58 and sporozoite 0.56) in the year 1946 by Siddons, which rose to 1.56 in 1980, as
detected by Mukhopadhyay Chatterjee (1991) reported that the natural infection rate was 1.78 (2 sporozoite infection, out of 112 specimens dissected). It was also stated by Neogy and Sen (1962), that the infection rate in Calcutta was higher (0.51) than West Bengal. In the present study the natural infection rate was 0.26 (1 sporozoite infection in *Anopheles stephensi* was found in September, 2001 out of 386 wild caught *Anopheles stephensi* mosquitoes collected from Monoharpukur, dissected for infection, if any) This might led to an inference that *Anopheles stephensi* is still a potential vector of malaria in Calcutta.

The sporozoite and oocyst rate of infection were 2.40 and 7.40 respectively in Bombay amongst *Anopheles stephensi* showing thereby that in Bombay the species was more active so far as transmission of malaria was concerned than that of Calcutta Banerjea (1930), reported the infection rate of *Anopheles stephensi* was 9.30 in Lucknow, which was significantly higher than Calcutta and Bombay The infection rates were lower in Andhra Pradesh (King and Iyer, 1929); Delhi (Hodgson, 1914), Madras (Ramsay and Macdonald, 1936); Ahmedabad (Singh and Jacob, 1939) than Calcutta Outside India, (Afridi and Majid, 1938 and Monouchchri, 1976) the infection rates were found to be lower than Calcutta. Besides, *Anopheles stephensi*, one sporozoite infection was also found in *Anopheles subpictus* from wild caught collection (Kasba Golpark) in the present study The sporozoite infection rate was 0.27 (1 out of 368 wild caught mosquitoes) Chatterjee et al (2003) also found the natural sporozoite infection in *Anopheles subpictus* in Hooghly, 60 kms away from Calcutta. Average sporozoite rate was 0.32%. These findings are very important and significant since there was no report available of any malarial parasite infection in *Anopheles subpictus* in West Bengal, specially in Calcutta. *Anopheles annularis* have been obtained in certain pockets of Calcutta, specially in the peri-urban area The density of this species is found to be extremely low both in indoor and on man-bait. No malarial parasite infection was detected in this mosquito during the study period. Naturally its presence possibly has got no epidemiological significance in the present ecological scenario in Calcutta.

### E. Vector density and man/vector contact in relation to malaria cases

2002. But malaria cases were observed throughout the study period, encompassing all the months with maximum in the month of September 2000 and minimum in the month of April 2001.

Several human-bait experiments with mosquitoes conducted in Calcutta also indicate no *Anopheles stephensi* /man contact was evident in certain months of the year (Mukhopadhyay, 1980, Chatterjee & Hati, 1995, Bhattacharya et al., 2002). Although clinical manifestations of the infections were detected in every month (Hati, 1991, Mandal et al., 1998) Man-landing catches conducted in Calcutta also revealed that the density of *Anopheles stephensi* was very low and most of the captures were made in the rainy season (Hati et al., 1998). Data of the present study also show that in Calcutta malaria transmission has been going on perennially.

Considering the above findings and owing to the existence of some recognised malaria vectors (Tandon & Tandon, 1994) namely, *Anopheles annularis* and *Anopheles subpictus* in Calcutta (Sen, 1938; Pramanik et al., 1992, Bhattacharya et al., 1996), apprehension has been raised in certain quarters that these species might have some complementary role (Bhattacharya et al., 1999) during the absence or at the time of low density of the vector, *Anopheles stephensi*. Recent report of sporozoite infection in *Anopheles subpictus* in Hooghly by Chatterjee et al. (2003) and the detection of the same in *Anopheles subpictus* in the present study have added a new dimension in malaria epidemiology in West Bengal, specially in Calcutta and also reinforces the apprehension of multivectorial involvement (Bhattacharya et al., 1993, Bhattacharya et al., 1999).

Man-*Anopheles subpictus* contact was observed almost throughout the study period except in few months. In indoor, man/*Anopheles subpictus* contact was absent in the month of February 2001, October 2001, April 2002 and June 2002, whereas in outdoor, man/*Anopheles subpictus* contact was absent in the month of January 2001, January – February 2002 and May 2002. But malaria cases were found throughout the year, encompassing all the months.

It was also noted that wild caught collection of *Anopheles stephensi* was absent in the months of October – December 2000, January – February 2001, May 2001, July – August 2002 and May 2002 in Ward no. 84 under CMC, but malaria cases were there throughout the year.

Similarly wild caught collection of *Anopheles subpictus* was absent in the months of October – December 2000, February 2001, October – December 2001, January 2002 and April 2002; but malaria was prevalent throughout the year.

In spite of perennial transmission of the disease, no man/*Anopheles stephensi* contact was evident in several months in Monoharpukur during the study period. Alteration of host-parasite
relationship, addition and alteration of mosquito vectors, and vectorial capacity may have some role in the present malaria situation in West Bengal (Bhattacharya et al. 1993). In a given ecological stratum the stability and complexity of mosquito-borne disease transmission increases with the increasing diversity of the vectorial system.

The apprehension of multivectorial involvement in malaria epidemiology in Calcutta is further reinforced by the study of Chakraborty and Tandon (1995) who opined, that, *Anopheles stephensi*, the principal vector of malaria in the metropolis (Calcutta) is zoophilic and prefers to feed on bovine blood irrespective of the biotope and availability of the host.

F. Studies on Epidemiology of Malaria

Epidemiological studies on malaria in Calcutta are of considerable importance in view of the magnitude of the malaria problems. Malaria transmission is perennial in Calcutta. The problem of malaria epidemiology in Calcutta presents not only a difficult proposition but also a colossal one, considering its variegated topography, water bodies, its tropical monsoon climate with summer rains, water storing habits of the community, all contributing to the growth of the vector population. Malaria being a vector borne disease, where vector biology and vector ecology, with adaptive modifications under different sets of conditions exert a profound influence on the epidemiological picture. It is, therefore, obvious that any effective measure aiming to interrupt transmission of the disease must be based on precise and relevant epidemiological information.

Total malaria cases reported in India during 1980-1990 were 28,96,000, 26,66,244, 21,82,303, 18,95,297, 21,84,446, 18,64,380; 17,92,168, 16,47,485; 18,50,060, 20,20,000 and 20,20,000 respectively. There was an increasing trend of malaria during the period 1991-1996 in India. In 1997, 1998 and 1999 there were 26,60,557, 20,98,356; and 18,65,874 malaria cases respectively. Number of *Plasmodium falciparum* cases shows an increasing trend since 1980 till 1999 with little fluctuation during 1982 (55,10,57 cases), 1985 (54,50,05 cases), 1987 (61,15,88 cases); 1991 (69,00,00 cases); 1993 (85,00,00 cases), 1997 (1,00,73,66 cases), 1998 (91,45,84 cases), and 1999 (88,24,12 cases) respectively. Maximum number of deaths due to malaria was reported during 1994 (1422) as cited by Hati (2001).

During 1980-1990, in West Bengal, the total malaria cases reported were 22219, 30717, 34237, 41861, 46340, 46510, 53620, 46029, 36318, 35624 and 27531 respectively. During 1990-1999 maximum number of *Plasmodium falcitarum* malaria cases were reported in the year 1999 (57245), along with 155 death cases (Hati, 2001).
Calcutta


The data on the incidence of malaria were collected for three consecutive years (Jan'02-Dec'02) from Calcutta Municipal Corporation and they were analysed critically. Seasonal variation of malaria incidence were noticed. It has been seen that maximum number of malaria cases were reported in the Rainy season than that of Summer and Winter season. In the Rainy season, maximum number of malaria cases (35942) were reported during the second year, than that of first year (29768) and third year (6692). In Summer, maximum number of malaria cases were found during the first year (18383) than that of second year (7226) and third year (2188) respectively. Similarly, in Winter, maximum number of malaria cases were found during the second year (22772) than that of first year (19031) and third year (3912) respectively.

So, it has been seen that there was not only the seasonal variation of the malaria cases in three consecutive years but also there was fluctuation of the malaria incidence amongst different years of the study period. Seasonal variation of malaria incidence is not same in different parts of India. In Delhi, maximum prevalence was found in Summer (1577) followed by Rainy (264) and Winter (113) seasons (Bhatia et al, 1958). In Madras, maximum abundance was obtained in the Winter season (976) followed by Rainy (678) and Summer (363), whereas in Mandora, Haryana, highest number was reported in the month of February (Subbarao et al, 1984). In the present study, clinical manifestations of the infections were observed in all the years encompassing all the meterologically distinguishable three seasons with a pronounced peak in the Rainy season. These observations suggest that there is perennial transmission of the disease with seasonal fluctuations. The present observation corroborates with the findings of Chatterjee & Hati 1986, Bhattacharya and Mukherjee, 1995 and Bhattacharya et. al, 1996.

During the study period, it was seen that the percentage of mixed cases were more in second year (12.35%) than that of first year (0.18%) and third year (0.15%) respectively. Percentage of *Plasmodium vivax* infection was more or less same in all the years of study, but the percentage of *Plasmodium falciparum* was less in the third year (8.25%) than that of first year (17.56%)
and second year (18.2%). Interestingly, it was found that *Plasmodium vivax* cases were more in the monsoon and post-monsoon months and less during late Winter months, whereas *Plasmodium falciparum* cases were maximum in early Winter months and minimum in Summer months. This observation would be helpful in formulating species-wise and season-wise strategy for the containment of the disease. Death cases were reported only in the third year of study period and all of them were due to *Plasmodium falciparum*, which suggests that it is still more virulent than *Plasmodium vivax*.

It can be concluded that there was perennial transmission of the disease in all the three years of the study period. Mixed infection were present in all the three seasons i.e. Summer, Rainy and Winter. *Plasmodium vivax* mainly dominates the Rainy season, whereas *Plasmodium falciparum* dominates the Winter season.

**G. Incidence of malaria cases of Ward no 84 under CMC**

The incidence of malaria cases were studied in Ward no 84 under CMC for three consecutive years (January 2000 to December 2002). During the first year, maximum number of malaria cases were found in the month of September and least in the month of December. During the second year, maximum and minimum malaria cases were found in the month of August and April respectively. Similarly, during the third year, maximum and minimum cases were in the month of September and April respectively. Comprising three years of study period, maximum and minimum cases were found in the month of September 2000 and April 2001 respectively.

During the study period, it was seen that maximum number of malaria cases were reported in the Rainy season than that of Summer and Winter season. Altogether 2049 malaria cases were reported during the Rainy season. The corresponding figures of Summer and Winter season were 683 and 991 respectively. It was seen that *Plasmodium vivax* cases were more in the Rainy season whereas *Plasmodium falciparum* cases were more in Winter season. The trend of species-wise case incidence in Ward No. 84 was found to be similar to the overall scenario of Calcutta.

The maximum number of malaria cases was encountered in the monsoon and post–monsoon months, which suggests that in Calcutta, Rainy season in more conducive to the building up of adult population of *Anopheles stephensi* than Winter and Summer seasons. The present study corroborates with the findings of Chatterjee and Hati, (1986).
During Jan 2000 – December 2002, 526,144 and 422 *Plasmodium vivax* cases were reported during Summer, Rainy and Winter season respectively. Whereas 102, 547 and 553 *Plasmodium falciparum* cases were reported in Summer, Rainy and Winter season respectively. Interestingly it was found that *Plasmodium vivax* were more in Rainy season, whereas, *Plasmodium falciparum* were more in Winter season.

Both *Plasmodium vivax* and *Plasmodium falciparum* cases were found to be maximum amongst the people who are above 15 years of age in all the three years (Jan 2000 to December 2002) During the first year (Jan 2000 – Dec 2000) maximum number of *Plasmodium vivax* cases were reported during September and minimum during December Similarly, maximum number of *Plasmodium falciparum* cases were reported during September and minimum during April. Interestingly, no *Plasmodium falciparum* cases were found among the age group of 0 – 5 years during January, May, August and November

During the second year (January 2001 – December 2001), maximum number of *Plasmodium vivax* cases was reported during September and minimum cases during January. Similarly, *Plasmodium falciparum* cases were maximum during August and minimum during March, May and June. In both the cases, people above 15 years of age were mostly affected. No *Plasmodium vivax* cases were found amongst the age group of 0–5 years during January. Similarly *Plasmodium falciparum* cases were not found amongst the age group of 0–5 years during April–September and December and amongst the age group of 6–15 years during February –July

During the third year (January 2002–December 2002), maximum number of *Plasmodium vivax* cases was reported during September and minimum cases during February. Similarly, *Plasmodium falciparum* cases were maximum during November and minimum during August. In both the cases people above 15 years of age group were mostly affected. No *Plasmodium vivax* cases were found among the age group of 0–5 years during April. *Plasmodium falciparum* cases were not found amongst the age group of 0–5 years during January, March–September, and November, and amongst the age group of 6–15 years during February–August and November.

Ward no. 84 comprises of mainly middle class people. It was seen from the random survey, that the 65% people below 15 years of age use mosquito nets and mosquito repellents, whereas 82% people above 15 years of age did not use mosquito nets. Moreover, during monsoon, due to hot and humid climate, they open the windows during night, so the chances of man-mosquito contact
and the incidence of malaria cases were studied for two years i.e. July 2000 – June 2002 both in Calcutta and also in Ward No. 84 under CMC.

**Calcutta**

During July 2000 to June 2002, mean maximum temperature (m.m.t.) varies between 26.01°C – 36.72°C, encompassing 24 months. Altogether, 1,04,701 malaria cases were registered. 77.81% cases were found to occur between the m.m.t 29° – 32°C. No rainfall was reported in December 2000, 2001; March 2001 and February 2002. Although malaria cases were found in both the years, encompassing all the months with a pronounced peak in the monsoon (58.90%). This findings corroborates with the findings of Hati (2001), who stated that malaria is prevalent from July to November, with the peak incidence in September, in Central India and Indo-Gangetic Plain. In Assam, malaria season extends from March to June. This observation suggests that despite the absence of rainfall in some months, there is a perennial transmission of the disease with seasonal fluctuations in its maintenance cycle in Calcutta. Average monthly rainfall varies from 0 to 13.06mm during the study period. 60.46% cases were reported when the monthly rainfall was between 0.22 to 5.37 mm. Naturally, to draw conclusion regarding a causative association between maximum rainfall and higher case incidence is difficult. When temperature varies from 26 01°C – 33.81°C coupled with an average rainfall ranges between 0 mm – 8.56mm, 91.03% malaria cases have been encountered.

Temperature and humidity not only affect the growth of parasites inside the vector, but also determines the longevity and biting capacity of the mosquitoes (Hati, 2001). Studies in this direction are necessary in Calcutta in order to substantiate the above contention. There is no scope of studying the above factors in the present context.

**Ward No. 84 under CMC**

Altogether 3723 malaria cases were registered from Monoharpukur. 74.66% cases were found between the mean maximum temperature (m.m.t) 29° – 32° C. 61.75% cases were reported when the monthly average rainfall varies between 0 to 7.54 mm. 94.91% cases were found when the mean maximum relative humidity was between 92 – 97%. Albeit few studies have been done in order to establish a relationship between climatic variables and the rate of malaria incidence in India, many investigations have addressed the facts, that other factors have affected the rates. Since these factors often compound the effect of meteorological variables on malaria, it is important to delineate and understand them.
I. Collection of larvae of *Anopheles stephensi* from different water bodies

The different breeding spots of *Anopheles stephensi* have been reported by different workers from the various parts of the world *Anopheles stephensi* used to breed in Garden tubs (Iyenger, 1920, Covell 1944), cisterns (Iyenger 1920, Covell 1928 and 1944, Mukhopadhyay 1980), shallow pits (Iyenger 1920), filtered and unfiltered water (Basu 1930), open earth drains (Roy 1931) ponds (Roy 1931), earthen handis (Knowles and Basu, 1934), earthen tubs (Knowles and Basu 1934) jars (Knowles and Basu 1934), Kerosene tins (Knowles and Basu 1934), iron tubs (Knowles and Basu 1934), sewered areas of Calcutta (Senior White 1934), cemented floors (Covell 1944, Mukhopadhyay 1980), key holes of Corporation (Mukhopadhyay 1980), fountain (Covell 1944, Mukhopadhyay 1980), burrow pits (Sinton 1917, Mukhopadhyay 1980), marble pots (Mukhopadhyay 1980) flower tins (Mukhopadhyay 1980), broken porcelain pots (Mukhopadhyay 1980), wells (Hodgson 1914, Afridi 1938, Russel and Rao 1941, Abraham and Samuel 1944, Senior White and Rao 1944, Covell 1944, Monouchchri 1976, Batra and Reuben 1979, Kaur and Reuben 1981), pools (Hodgson 1914, Sinton 1917), shallow stream (Hodgson 1914, Covell 1928 and 1944), rain water puddles (Sinton 1917), grass grown ditches (Sinton 1917), dark places (Covell 1928), roof gutters (Covell 1928 and 1944, Mukhopadhyay 1980), terraces (Covell 1928), rivers (Abraham and Samuels 1944), troughs siphons and reservoirs (Afridi 1938, Covell 1944), salt water (Bana 1943, Zuluesta 1968), irrigation channels (Covell 1944), Gulf of Suez (Gad 1967), hoof prints of animals (Monouchchri 1976), rice fields (Monouchchri 1976), goat skin bags (Monouchchri 1976), overhead tanks, cement, earthen, stone and wooden pots (Tandon and Chakroborty, 1998), old tyres, cement cisterns for soaking bricks (Hati et al, 1987) and earthen, stone, cement and wooden pots (Chakroborty et al, 1998)

Calcutta Municipal Corporation of Calcutta has two types of water supply system. The filtered water for cooking, drinking and bathing purposes which usually stored in masonry vats under the ground level. Affluent dwellers lifted the stored water to the overhead tanks by an electric pump. The slum dwellers and the people lived in shanties usually stored water in different kind of pots, which sometimes do not have lead on them [Plate 11 & 12]. Sometimes it also appears, that the overhead tank is left open without any lead. *Anopheles stephensi* finds a suitable breeding ground in these places.

In the present study, different perennial water bodies have also been searched for the presence of *Anopheles stephensi* larvae. But for the last couple of decades, investigators, however few,
Plate 11 & 12: Water storing habits of the slum dwellers in Calcutta
were attempted to find out the larvae of *Anopheles stephensi* only from the artificial containers, construction sites, overhead tanks and other man-made residential situation. But no attempt had been made to find out the larvae of *Anopheles stephensi*, if any, in the perennial water bodies such as ponds, ditches and lakes which are abundant in the CMC area in Calcutta. In the present study, however, such an attempt had been made. The density of *Anopheles stephensi* larvae in the said perennial water bodies was found to be significantly low in comparison to the artificial containers mentioned above. It was seen that both the years of study, maximum number of larvae were collected from ponds than that of ditch and lakes. The collection of *Anopheles stephensi* larvae were maximum (176) in the Rainy season from Ponds, Ditch and Lakes than that of Summer (60) and Winter (28) season. For the first time such an extensive study was made and surprisingly it was noted that *Anopheles stephensi* larvae breeds in these biotopes (Ponds, Ditch and Lakes). Besides the perennial water bodies, artificial water bodies (Cement tanks, Earthen tubs, Tin drum and Plastic bucket) were also searched and found that *Anopheles stephensi* breed mostly in Cement tanks and least in Plastic buckets.

In artificial containers, *Anopheles stephensi* was found to breed in close association with *Aedes aegypti*. Larvae of *Culex quinquefasciatus* and *Armigers subalbatus* were frequently found to breed with *Anopheles stephensi*. Seasonwise prevalence of water holding containers with larval infestation showed quite higher larval breeding during Rainy season as it was pointed out by Paiva way back in 1912 in Calcutta. When seasonwise larva breeding of individual mosquito species was considered, *Anopheles stephensi* and *Aedes aegypti* larvae breeding spots were found to be significantly higher during the Rainy season than those of other seasons which might be due to influence of a number of climatological factors, especially the higher rainfall and suitable relative humidity, which prevailed during the Rainy seson (Wattal 1964, Rao 1967, Basu 1930). As *Anopheles stephensi* and *Aedes aegypti* are absolutely fresh water breeders (Basu 1930, Paiva 1912), their breeding capacity was found to be significantly lower during the Winter and Summer seasons, only due to paucity of fresh water. The findings of the present study corroborates with the data of Chatterjee & Hati (1986) and Chakraborty et.al. (1998). *Anopheles subpictus* was also found to breed in cement tanks and earthen tubs.

When the relative abundance of *Anophleses stephensi* larvae during various seasons was shown, it had been found that there was an increasing trend from pre-monsoon to post-monsoon periods and this may be due to the accumulation of water in the natural and artificial containers, indicating
the influence of monsoon rain, which enhanced the number of breeding sites and subsequently increased the density of the mosquitoes. Roy (2001) was also made the similar observation.

J. Fish as a bio-control agent

In view of the growing concern over insecticidal contamination of the environment and its subsequent detrimental effect to biotic community along with the continuing appearance of multiple resistance of insect pests to most chemical insecticides with their resing costs, search for alternative vector control strategies, has thus assumed a priority demand. The use of environment friendly materials for vector control initiated a vigorous search of biological agents either as an alternative and/or complementary to chemical treatment. Potential biological control agents have so far been selected from organisms naturally found in the field. Various species of fish have been found to posses enormous potentiality of being a biocontrol agent against mosquitoes. (Bhattacharya ,1992).

In the present investigation with mosquito larvae, whether individually or in crowded condition Poecilia reticulata (Guppy) feeds more than Aplocheilus panchax (Panchax). So in the same weight Poecilia reticulata is found to be more efficient than Aplocheilus panchax. From the present observation, it can be concluded that the satiation quantity of Guppy is more than Panchax when kept seperately. The amount of larvae, and the weight of the fishes were same in all the cases. But when they were kept in crowded condition (i.e. both Guppy and Panchax in the same biotope) their satiation quantity decreases.

K. Resurgence of Malaria in India

Malaria has returned as a major scourge causing enormous loss to the economy of the people of India. While malaria was resurging, it was ignored and given low priority. This provided an oppurnunity for malaria to return with venegence and occupy new paradigms. By the 1970s malaria was noticed occupying areas at one time free by the National Malaria Eradication Programme (NMEP). In 1976 epidemic situations returned with 6.5 million cases. In 1977 the eradication strategy was replaced by control under the modified plan of operation, prioritizing control objectives. In high risk areas spraying was restrated resulting in initial decline and incidence stabilizing at about 2 million cases. Forty percent of these constitute Plamsodium falciparum, a dangerous parasite responsible for almost all deaths due to malaria. Seen in the background of changing malaria profile, return of malaria has its own charesteristic features involving vector, parasite, host and the environment establishing new malaria paradigms. This
is facilitated by control failures due to blunt control tools aided and abetted by poor understanding of the malaria transmission dynamics. Resurgence of malaria is due to various factors some of which may be elucidate as follows :-

1. Technical, such as insecticide - resistance ability of mosquitoes to escape contact with the insecticides, minimal effective dose of DDT and resistance of Plasmodia to antimalarial drugs;

2. Operational, such as movement of population, lack of sufficient transport, excessive reliance on active case detection, refusal of spraying by the people on various grounds, habits of plastering walls with mud;

3. Financial problem;

4. Administrative, such as shortage of personnel and insecticide, inadequacy of geographical reconnaissance and imperfect coverage.