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ETHOLOGICAL CHANGES IN ANOPHELES PSEUDOPUNCTIPENNIS IN MEXICO
AFTER PROLONGED USE OF DDT

by

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The nation-wide malaria eradication campaign of Mexico, launched in 1955, has met with success in most of the malarious areas of the country. The campaign has been based on the application of DDT to indoor surfaces of human dwellings at the dosage of 2 g/m^2 twice a year, a method which had already proved its value in pilot operations in several parts of the country. Dieldrin had been used in the campaign in areas of difficult access where it was thought that an annual application of this insecticide would be sufficient to interrupt malaria transmission. The rapid development of dieldrin resistance in 1959 in the two main vectors of the country - Anopheles albimanus and Anopheles pseudopunctipennis - made it necessary to discontinue the use of dieldrin in those areas and to replace it by DDT.

Four years' spraying of residual insecticides brought the interruption of malaria transmission in 77% of the malarious areas of the country but in the rest of the malarious territory it was found that, apart from operational problems inherent to areas of difficult topography and poor communications, the disease persisted despite adequate spraying coverage and in the absence of physiological resistance to DDT. Investigations carried out in these problem areas, particularly in the State of Oaxaca by de Zulueta & Garrett-Jones (1963), showed that the main cause accounting for the persistence of transmission was the behaviour in relation to DDT of the two vectors - A. albimanus and A. pseudopunctipennis. The females of both species were able to penetrate the DDT-sprayed houses, bite the inhabitants inside them and escape without having picked up a lethal dose of insecticide.

When it became clear that mosquito behaviour was an important factor in the persistence of transmission, the question arose as to whether the behaviour observed had existed before the introduction of DDT or was a development brought about by the prolonged use of the insecticide. In a country-wide campaign, like the one carried out in Mexico, all human dwellings in the malarious territory are regularly sprayed. This means in practice that the finding of an area untouched by insecticides where the original behaviour of vectors can be studied is virtually impossible. What can be done, and in fact was done in the present case, was to compare existing behaviour patterns with previous observations on the subject.

Downs & Bordas (1951) studied the behaviour of A. pseudopunctipennis in sprayed and unsprayed huts in Acatlipa, a village of the State of Morelos considered as representative of conditions in the Mexican plateau. Acatlipa is 1400 m above sea level and is situated in a rice-growing area with abundant breeding of A. pseudopunctipennis. Despite the annual application of DDT in houses during the previous five years, Bordas & Downs (op. cit.) found that the density of adult mosquitos in the occasional untreated houses in the periphery of the village was high and they decided to build there a series of experimental huts. These were 2 x 3 m structures of mud walls and thatch roofs, similar to those described by Muirhead-Thomson (1950) in Africa.

Preliminary work had indicated that the use of baffles or louvres easily deterred the approaching A. pseudopunctipennis from entering the huts but that an L-shaped tunnel permitted their entry and at the same time prevented their escape. The L-shaped tunnel proved very useful for studying the entry pattern of A. pseudopunctipennis and the egress was investigated by means of an exit trap fixed to the door of each hut. Observations on behaviour in untreated huts were made in 1949 (Bordas & Downs, 1950), and on the effect of DDT and BHC in 1950 (Downs & Bordas, 1951). Eleven years later, huts Nos. 1 (DDT) and 2 (control) of Downs & Bordas were still standing and it was decided to repeat the original observations to determine whether the prolonged use of DDT (Acatlipa had been sprayed since 1944) had produced any ethological changes in the local A. pseudopunctipennis.

The two huts had their mud walls replastered and their thatch roofs renewed and were fitted with L-shaped entry tunnels and exit traps identical to those used in the studies of 1949-1950. Throughout the present investigations the huts were operated four nights every week from dawn to dusk - weeks of entry captures alternating with weeks of exit collections. In the former the mosquitos were caught at the time they reached the entry tunnel (as had been done in the early studies) and in the latter they were removed every hour from the exit trap. In both types of captures counts were made of mosquitos found dead on the floor.

Observations were made for a period of six weeks before any of the huts were treated. On 11 August 1962 hut No. 1 was sprayed indoors with DDT w.d.p. at the dosage of 2 g/m². This had been the hut treated and the amount of insecticide used in the 1949-1950 investigations. Hut No. 2, which had served then as control, was again left unsprayed and served once more as comparison hut. After the spraying of hut No. 1, ingress and egress captures were made in alternate weeks for another period of six weeks, after which hut No. 1 was replastered and its thatch roof was changed. On 10 October 1962 this hut was sprayed again, using the same DDT formulation and dosage as in the first spraying. Eight months later a new series of captures of incoming and outgoing mosquitos was carried out for another period of six weeks. Three different sets of observations had therefore been made in these investigations: (a) with the two huts unsprayed; (b) with one hut recently sprayed; and (c) with the same hut sprayed eight to 10 months before. Conditions in (c) reproduced exactly those of Downs & Bordas' investigations regarding season and time of spraying. According to their published records (Downs & Bordas, 1951), only exit captures were made after spraying and for this reason in the comparison of Downs & Bordas and our own results made in Table 1 only results from exit captures are given.

As can be seen from these results, the number of mosquitos caught in huts Nos. 1 and 2 before DDT spraying was very similar and this, it should be added, is in agreement with what had been observed by Bordas & Downs (1950). After the introduction of DDT, however, an important difference is noticeable between the early investigations and the present ones. In Downs & Bordas' (1951) observations the number of males and females of A. pseudopunctipennis caught after spraying in hut No. 1 was considerably

lower than in hut No. 2. The treated hut caught only 65.7% of the males and 56.2% of the females caught in the control hut. In the present observations there was no reduction in the number of A. pseudopunctipennis caught in the DDT-treated hut. As Table 1 shows, there was in fact an increase in the number of males and females caught in the treated hut immediately after spraying. A slight reduction, it is true, was observed eight to 10 months after treatment but this is a trend without significance. If the numbers caught in each hut are added up, it will be seen that the treated hut caught slightly more mosquitos than the unsprayed hut. This, as pointed out before, is in strong contrast with the observations of Downs & Bordas (1951).

Although ingress captures do not appear to have been made by these investigators after spraying of the huts, they were made in the present investigations both before and after spraying. Their results confirm the lack of a reduced entry in hut No. 1 after spraying. Ingress captures in hut No. 1 yielded 258 males and 276 females immediately after spraying and 212 and 256 males and females respectively, eight to 10 months after. Corresponding figures for hut No. 2 (control) were 317 and 307; 168 and 201. There seems to be a slight reduction in numbers in hut No. 1 immediately after spraying but this is compensated by a small increase eight to 10 months after spraying. Altogether more mosquitos were caught in the treated hut than in the control, as was the case with the exit trap captures. And here it should be pointed out that in the ingress captures mosquitos are caught (by the men in charge of the captures) at the very moment they reach the ingress tunnel so that an unrecorded escape from the huts (a possibility in the exit captures) is ruled out here.

The complete lack of deterrency, a term proposed by Zulueta & Cullen (1963) to embrace all possible causes that prevent mosquitos from entering sprayed structures, so clear in the present observations, is remarkable in more than one respect. In the first place this is not only different from what had been observed before in the area but it is also in contrast with observations made in other parts of the world (e.g. Muirhead-Thomson, 1947; Zulueta et al., 1961; Kuhlman, 1962; and Slooff, 1964) which show that deterrency is in fact a normal effect of DDT. Its absence from an area like Acatlipa, where it had been previously observed, must therefore be considered as a very significant finding.

There is another aspect which is also interesting in the present observations and that is that the lack of deterrency is observed among mosquitos which for the most part are seeking shelter and not a blood meal (e.g. males and engorged females). A behaviour like this suggests immediately a development of physiological resistance but periodical checks on the susceptibility of the local A. pseudopunctipennis gave results which indicated normal susceptibility of the species to DDT. If the susceptibility under test conditions seemed to be unchanged, the same cannot be said of the lethal effect produced by DDT in the treated hut. Immediate mortality in hut No. 1, based on numbers of mosquitos dead at the time of collection (window-trap and floor collections) was in Downs & Bordas' (1951) observations 49% in males and 85.9% in females. Corresponding figures in the present observations were 45.6% and 59.6% soon after spraying and 34.3% and 44.1%, eight to nine months after spraying. These results seem to indicate that, in spite of unchanged susceptibility, A. pseudopunctipennis avoided the lethal effect of DDT better in 1962 and 1963 than it did in 1950. Like in the case of deterrency, a change of behaviour seems to have taken place here after 11 years' use of DDT.

The operational significance of the changes observed in Acatlipa must be very considerable. For if the results obtained in the small-scale experiments described here reflect the situation in other parts of Mexico, this means that A. pseudopunctipennis must now be entering treated houses in greater numbers and avoiding the lethal effect of DDT better than it did before. That this is probably the case is borne out by the fact that in areas where transmission persists, A. pseudopunctipennis has been observed visiting houses in numbers which do not seem to be affected by the presence of DDT (de Zulueta & Garrett-Jones, 1963). There, however, the lack of early quantitative data does not permit a comparison to be made like in the case of Acatlipa.

From what has been said in this note the importance of mosquito behaviour in anti-malaria work becomes evident. The case of Mexico is probably not an isolated one. Other countries exist where in spite of normal susceptibility of the vectors the effectiveness of DDT seems to have been blunted after prolonged use of the insecticide. There changes of behaviour may provide the clue to difficulties and failures.

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RESUME

Les progrès de la campagne d'éradication du paludisme au Mexique ont montré que, pendant les dernières années de l'existence des zones difficiles et malgré la couverture totale aux insecticides, la transmission du paludisme persiste toujours. En 1962, on a découvert que la persistance de la maladie dans certains de ces zones était due au comportement des vecteurs en rapport avec le DDT et c'est pourquoi l'on s'est demandé si ce comportement a toujours existé ou si un changement a eu lieu après l'emploi prolongé du DDT.

Un essai a eu lieu dans la localité d'Acatlipa (Etat de Morelos), où des études avaient été faites en 1950 sur le comportement du vecteur local, A. pseudopunctipennis, dans des huttes expérimentales traitées ou non traitées aux insecticides à action rémanente. Onze ans après, durant lesquels le DDT a toujours été utilisé, l'on a répété cet essai ce qui a permis de déceler deux changements de comportement importants:

- (1) la présence du DDT ne diminue pas les entrées d'A. pseudopunctipennis dans la hutte traitée, comme on l'avait constaté lors des observations de 1950;
- (2) la proportion des moustiques trouvés morts sur le sol et dans le piège de sortie de la hutte traitée est plus basse que dans les observations précédentes. L'importance que ces changements peut avoir dans les opérations d'éradication du paludisme au Mexique est soulignée dans l'article.

TABLE 1. RESULTS OF CAPTURES OF A. PSEUDOPUNCTIPENNIS
 IN EXPERIMENTAL HUTS AT ACATLIPA (MORELOS)
 (EXIT TRAP AND FLOOR COLLECTIONS)

Time after spraying	No. of captures	Hut No. 1 (DDI')		Hut No. 2 (control)	
		No. of males	No. of females	No. of males	No. of females
Original experiments of Downs & Bordas, April - July 1950					
8-10 months	21	670	510	1 185	907
Present observations, July 1962 - August 1963					
Before spraying	12	444	438	367	412
2-35 days	12	425	374	315	256
8-10 months	12	353	245	338	252

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