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Genetics of resistance to permethrin in *Anopheles stephensi*

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Pyrethroid resistance has been viewed as a very serious threat to the future of malaria vector control[1]. However, recently two contrasting outcomes have been reported where resistance has been reported in *Anopheles* malaria vectors. In West Africa a high frequency of the *kdr* gene in *Anopheles gambiae* has not prevented good results with pyrethroid treated bednets in laboratory simulations[2], in experimental huts[3,4] and in malaria control trials in villages[5]. However, in South Africa, pyrethroid resistance in *An. funestus* due to a metabolic mechanism[6,7] had a serious impact on the indoor residual spraying programme using a pyrethroid and a steep rise in malaria cases was only reversed by switching back to spraying DDT[8] to which the metabolic mechanism does not confer resistance.

Thus there seems to be a crucial difference between the impact on control operations of resistance due to *kdr* and to at least one type of metabolic resistance. It is of interest, therefore, to assess when *kdr* and a metabolic resistance gene occur together in the same insect whether one or the other has a predominant effect in causing resistance, or whether they act like polygenes, each contributing a limited amount to the overall resistance.

We have such a multi-resistant strain in *An. stephensi* from Dubai origin which carries both *kdr*[9] and a gene for an elevated P-450 oxidase mechanism[10] and possibly other metabolic mechanisms.

We observed the relationships of mortality to time of exposure to 0.25% permethrin papers of the laboratory selected Dubai resistant stock (Dub234), a susceptible *An. stephensi* stock of Indian origin (Beech) and the F_1 hybrids from crossing female Dub234 to male Beech. The data on probit mortality and log time scales are shown in Fig. l with 95% confidence limits based on the variation between replicates. Resistance shows intermediate dominance. A backcross was made of the F_1 hybrid males to the females of the Dub234 stock and the observed data for the backcross at a series of exposure times are shown in Fig. 2. There are definite signs of a "kink" at about 50% in the dose response relationship, suggesting the action of a major resistance gene.

On Fig. 2 have been marked the expected relationship if there was only a single gene operating so that the backcross progeny would consist of 50% like the F_1 and 50% like the Dub234 stock. Also shown is the straight line relationship expected if several polygenes of small effect were contributing to resistance and segregating freely in the backcross progeny. The 95% confidence limits, calculated as above, on the observed data in several cases overlap with both of the hypothetical lines. However, in our judgement there is

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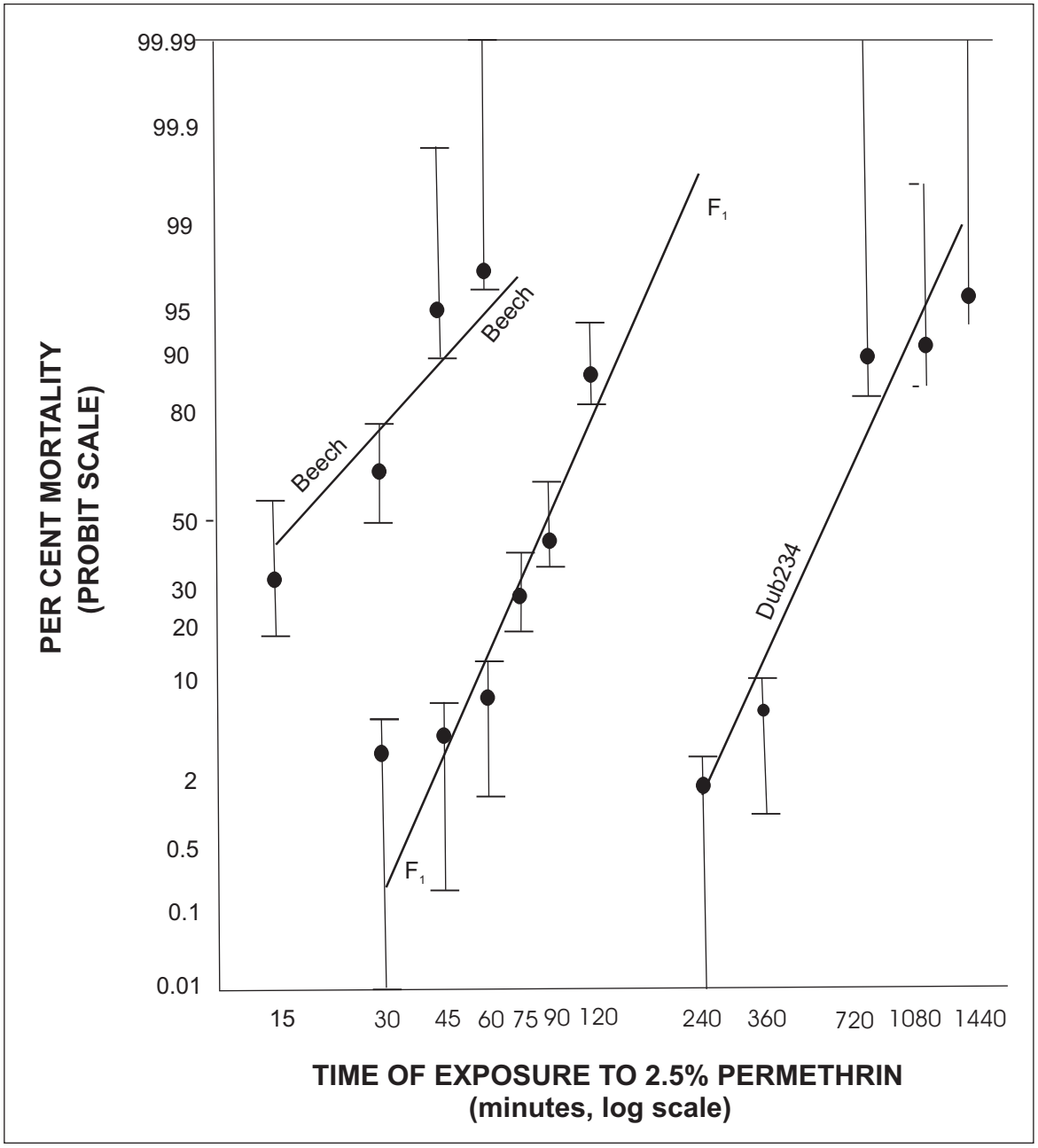

Fig. 1: Time-mortality (computed linear regression lines) for the susceptible (Beech) and the resistant (Dub234) strains of *Anopheles stephensi* and the F_1 from crossing them. 95% confidence limits are attached to the mean mortality at each exposure time based on the arcsine transformed mortalities in each of a series of replicates

a better fit with the hypothesis of control by a single major gene. We tentatively conclude that either the *kdr* or the P-450 gene is having a predominant effect on the resistance. However, these data do not exclude an appreciable contribution from the other mechanism.

The question of whether it is the *kdr* or the P-450 gene which is predominant could be approached by testing the *kdr* genotype in the backcross progeny which survive long exposures[11]. Conversely the effect on the shape of the dose response curve of a synergist which blocked P-450 would be informative.

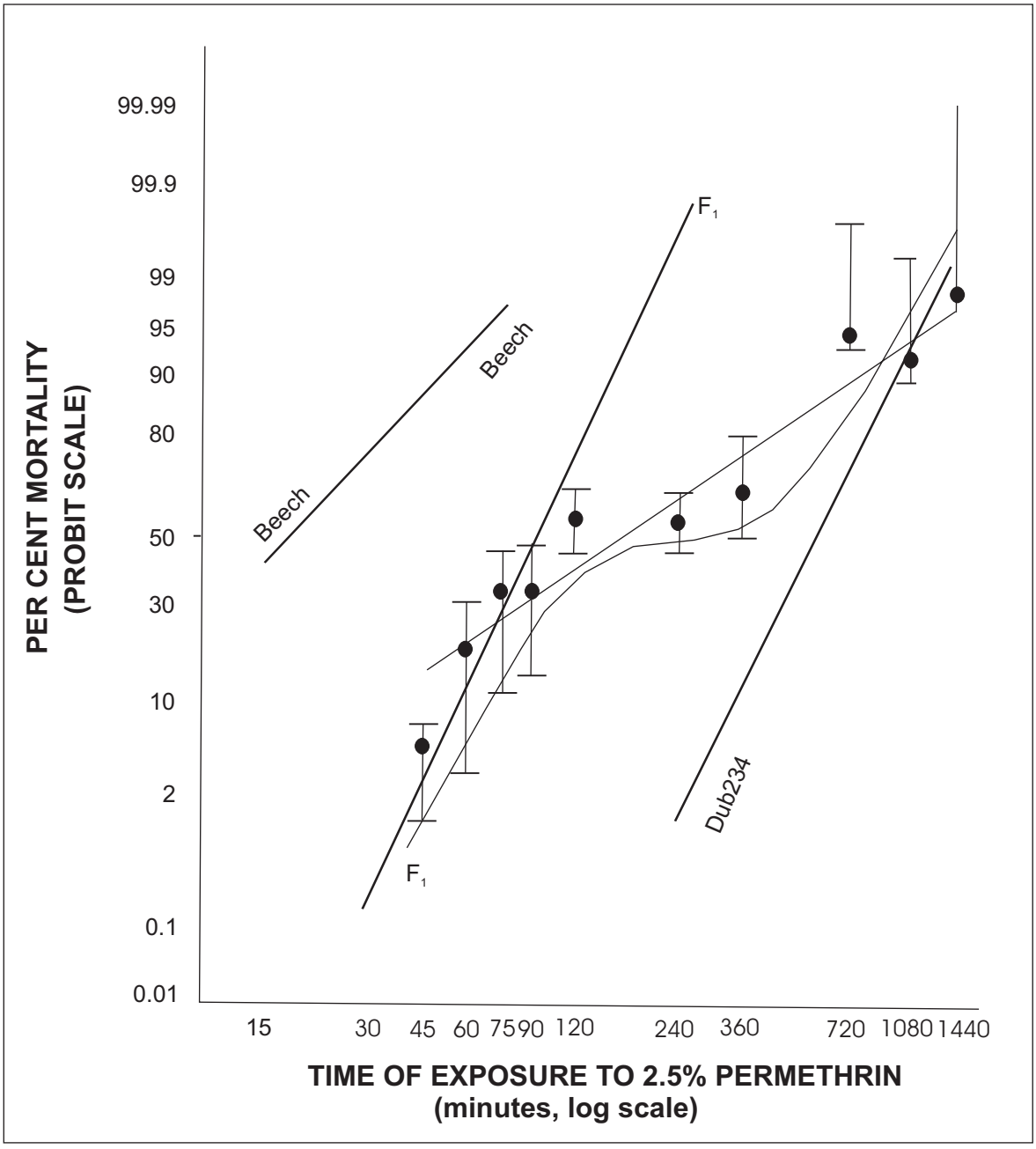

Fig. 2: Time mortality regression lines for the Beech, F_1 and Dub234 mosquitoes repeated from Fig. 1 and the observed data, with 95% confidence limits, for the backcross of the F_1 to the Dub234 stock. Also shown are the expectations for the backcross on the hypotheses (kinked line) of single major resistance gene giving a 1:1 ratio like the F_1 and like the Dub234 stock or (thin straight line) for polygenic control of resistance

We also bred a F_2 by intercrossing the F_1 males and females. The dose response data (not shown) and the hypothetical relationships for a single major gene—a 1:2:1 ratio of SS:RS:RR or polygenes are all so close to a straight line that these data do not help to decide the question about whether a single resistance gene is predominant.

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