

Insecticide Resistance in *Anopheles gambiae* Giles: a Case of Simple Mendelian Inheritance

A STRAIN of *Anopheles gambiae* from Northern Nigeria, highly resistant to dieldrin and cross-resistant to the related chlorinated hydrocarbon insecticides (chlordane, aldrin, isodrin, endrin and gamma-BHC), but still susceptible to DDT, has recently been colonized in this Institute and the degrees of resistance to these insecticides reported¹. A colony of the same species from Lagos and susceptible to all these insecticides has also been maintained in the same Institute for some four years.

Crossings between the two strains have now thrown light on the mechanism of the inheritance of this particular type of resistance in *A. gambiae*.

Crossings have been possible both ways (male × female and female × male), and result in hybrids which are identical and homogeneous so far as their susceptibility to insecticides is concerned (so far only gamma-BHC and dieldrin have been tried). Homogeneity is indicated by the narrow range of dosages of these insecticides necessary to kill all these hybrids, the dosages being between those necessary to kill the susceptible strain and those necessary to kill the resistant strain. Table 1 shows the LD50 for the two strains and the two hybrids when exposed for 1 hr. to solutions of dieldrin and gamma-BHC in Risella oil². The LD50 of dieldrin was impossible to determine for the resistant strain when exposure for 1 hr. only was made, as a saturated solution of this insecticide (4 per cent) gave no kill. By increasing the exposure time, however, it has been shown to be about 800 times as resistant to dieldrin when compared with the Lagos

strain, and is also some twenty-six times as resistant to gamma-BHC¹. The hybrids proved to be some thirty times as resistant to dieldrin and seven times to gamma-BHC. The males of both strains were slightly more susceptible than the females.

Thus it has been possible to choose two separating dosages of both insecticides, one killing all susceptibles but not hybrids or resistants, and the other killing all hybrids and susceptibles but not resistants. These dosages for an exposure of 1 hr. are 0.33 and 4.0 per cent dieldrin and 0.025 and 0.1 per cent gamma-BHC.

The usual method of determining the number of genes involved in a particular character by ascertaining the sort-out in the F_2 generation was not possible with these two strains of *A. gambiae*, because the F_1 hybrids were almost sterile. Though eggs were readily laid by the hybrid females, only a very occasional one proved to be viable. The sterility was eventually shown to be in the male hybrid.

It has been possible, however, to back-cross the two hybrid females with the parent susceptible and resistant males. The offspring of the four possible back-crosses have been exposed to the established separating dosages and the resulting proportions of susceptibles, hybrids and resistants obtained are given in Table 2. These results show quite conclusively that the inheritance of resistance to the dieldrin-chlordane-gamma-BHC group of insecticides is monofactorial with incomplete dominance.

A complete proof of this 50 : 50 separation in the offspring of the back-cross was given when the offspring of the back-cross of the hybrid female ($R♀ \times S♂$) with the susceptible male parent were exposed to the dosage eliminating susceptibles and the survivors were crossed with the resistant male parent. Subsequent offspring were exposed to the dosage killing hybrids and the following proportions obtained :

	Hybrids	Resistants
Males	53 per cent (86)	47 per cent (75)
Females	41 per cent (119)	59 per cent (170)

The surviving males and females from this latter exposure, on being crossed with the parent resistant strain, yielded pure resistant strains again.

In addition to the selection of this resistant strain from the back-cross of the hybrid with the susceptible parent, the offspring of this back-cross were shown to be composed of hybrids and susceptibles only, by exposing them to the dosage which kills hybrids. Such exposure resulted in 100 per cent kill. Similarly, exposure of the offspring of the back-cross of the hybrid with the resistant parent to a dosage which kills susceptibles only resulted in no kill at all.

Table 1. MEDIAN LETHAL CONCENTRATIONS (PERCENTAGES IN RISSELLA OIL) OF DIELDRIN AND GAMMA-BHC WHEN SUSCEPTIBLE, RESISTANT AND HYBRID STRAINS OF *Anopheles gambiae* ARE EXPOSED FOR 1 HR.

	Susceptible strain (S)		Resistant strain (R)		Hybrid $R♀ \times S♂$		Hybrid $R♂ \times S♀$	
	♀	♂	♀	♂	♀	♂	♀	♂
Dieldrin	0.08	0.05	>4.0	>4.0	2.6	1.5	2.3	1.6
Gamma-BHC	0.007	0.006	0.21	0.18	0.053	0.038	0.062	0.038

Table 2. PERCENTAGES OF SUSCEPTIBLES, HYBRIDS AND RESISTANTS RESULTING FROM THE FOUR BACK-CROSSES OF THE TWO HYBRID (RS) FEMALES WITH THE PARENT RESISTANT (R) AND SUSCEPTIBLE (S) MALES
The actual numbers of mosquitoes involved are given in brackets

Parents	$R♀ \times S♂$				$R♂ \times S♀$			
	$RS♀ \times S♂$		$RS♀ \times R♂$		$RS♀ \times S♂$		$RS♀ \times R♂$	
Back-cross	Susceptible	Hybrid	Resistant	Hybrid	Susceptible	Hybrid	Resistant	Hybrid
Back-cross offspring								
Males	57 (334)	43 (295)	48 (167)	52 (181)	49 (195)	51 (202)	45 (133)	55 (161)
Females	49 (364)	51 (380)	51 (241)	49 (235)	56 (194)	44 (150)	49 (195)	51 (207)

This finding of a monofactorial inheritance of resistance indicates the necessity for a change in the method of detecting resistance in the field. The present method of comparing the LD_{50} of a particular insecticide in an insect population where resistance is suspected with that in a population where the insecticide has never been used may show only a small difference, in some cases not even statistically significant, depending on the proportions of the three groups (susceptibles, hybrids and resistants) in the population examined. Thus, in Western Sokoto, from where these dieldrin-resistant *A. gambiae* originated, the degree of resistance was recorded³ as only eight times in 1955. The use of separating or discriminating dosages would have enabled a selection of the pure resistant strain, which is in fact some 800 times as resistant as the susceptible. It is suggested, therefore, that instead of a comparison of LD_{50} in the two populations, the minimum LD_{100} be established in the susceptible population, and the suspected population exposed to this dosage. Survivors, if any, can then be reared and their offspring exposed to higher dosages to select the resistant strain.

Alternatively, an accurate dosage/mortality regression line can be made in the suspected area, using a large number of dosages of the insecticide. If resistance is present, it will be shown by two steps in the line where increase in dosage produces no change in kill. These are the discriminating dosages between susceptibles and hybrids and between hybrids and resistants.

G. DAVIDSON

Ross Institute of Tropical Hygiene,
Keppel Street,
London, W.C.1.
Aug. 27.

¹ Davidson, G., *Nature* [178, 705 (1956)].

² World Health Org., Tech. Series Report No. 80 (1954).

³ Elliott, R., and Ramakrishna, V., *Nature*, 177, 532 (1956).