DDT RESISTANCE IN *ANOPHELES KOLIENSIS* (DIPTERA: CULICIDAE) FROM NORTHEASTERN IRIAN JAYA, INDONESIA

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ABSTRAK

Nyamuk Anopheles koliensis, *adalah perantara (vektor) penyakit malaria yang penting di daerah pedalaman Irian Jaya, Indonesia, yang telah dievaluasi kerentanannya terhadap DDT dengan menggunakan test kit diagnostik dan kertas yang telah diresapi sesuai dengan standar WHO. Serangkaian tes telah dilakukan di ARSO PIR I, yang merupakan tempat pemukiman para transmigran yang terletak 60 km sebelah selatan Jayapura. Pemeriksaan tersebut dilakukan mulai bulan Januari 1988 sampai dengan Mei 1989. DDT telah diuji pada dosis diagnostik yang telah direkomendasikan untuk jangka waktu tertentu pula. Daya tahan terhadap DDT diamati baik pada populasi nyamuk* An koliensis *maupun nyamuk* Culex quinquefasciatus. *Kira-kira 30% dari populasi nyamuk* An. koliensis *(sejumlah 468) ternyata tahan terhadap 4% DDT dalam kurun waktu 1 dan 2 jam paparan. Penemuan ini menunjukkan bahwa penggunaan DDT secara rutin di ARSO PIR I untuk penyemprotan di dalam rumah diperkirakan efektivitasnya terbatas, antara lain disebabkan oleh daya tahan fisiologik. Walaupun demikian, penggunaan insektisida alternatif ini akan lebih mahal dan mungkin terbukti sama tidak efektifnya karena sifat eksofilik dari jenis nyamuk tersebut. Makalah ini merupakan laporan pertama yang diperkuat dengan pengamatan yang dilakukan secara berulang- ulang mengenai kerentanan nyamuk* Anopheles koliensis *asal Indonesia terhadap DDT.*

INTRODUCTION

Beginning in the mid-1950's and until recently, the use of DDT had long been the insecticide of choice for malaria control in Irian Jaya, Indonesia.[1] However, its effectiveness in holo/hyperendemic areas was called into question by early workers in Irian Jaya.[2-4] The concern was not the development of physiologic resistance but the vectors exophilic resting behavior allowing it to avoid contact with interior residual spray deposits. Slooff[3] observed that anophelines entering houses usually returned to outdoor shelters soon after feeding further limiting the effects of insecticide usage.

Others during the same period were more optimistic about the effect of DDT on vector species and malaria control in the region.[5]

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Reassessment by Metselaar[1] later reported on various technical and logistic problems that had produced disappointing results in some localities, including the Arso area.

Anopheles koliensis Owen is one member of the *Anopheles punctulatus* complex which together form the major group of malaria vectors in Irian Jaya, Indonesia.[6] Various studies have either given favorable or less than salutary reviews of the real or potential effectiveness of DDT to control malaria in Irian Jaya.[7] The principal concern has been the facultative exo/endophagic and exophilic resting behavior of *Anopheles koliensis* and not development of physiologic resistance. Peters and Standfast[8] in New Guinea found *An. koliensis* the least susceptible to DDT of the species tested, yet DDT was still considered within the range for effective control. After several years of close observation, Slooff[2] could not demonstrate resistance to DDT in the Arso area. Spencer et al.[9] reported that *An. koliensis* remained susceptible to DDT in neighboring Papua New Guinea. *Anopheles koliensis* was not included for insecticide resistance status in the fifth report of the Expert Committee on Vector Biology and Control.[10] Several reviews reported preliminary data suggesting DDT resistance in this species in tests conducted in 1980, and acknowledged confirmatory tests were required.[11,12]

There were several incomplete and unpublished Indonesian Government reports of DDT resistance in *An. koliensis* during 1980 and 1989.[13,14] Small sample size resulted in equivocal findings from the Nimboran and Jayapura districts. The 24-hour mortalities ranged from 25 to 52% indicating some resistance with 4% DDT at one hour exposure. Brown[15] and WHO[16] has recently listed *An. koliensis* in Indonesia as resistant to DDT; however, the specific source was not referenced.

Dieldrin was used in two areas during early pilot projects and was found to adequately control house entering anophelines. However, because the residual action of dieldrin was found inferior to DDT, routine house spraying was conducted with the latter and dieldrin was held in reserve.[1] Before this investigation no other insecticides had been used to control malaria in this region. To our knowledge susceptibility tests with other insecticides have not been conducted in Irian Jaya.

Arso PIR I is hyperendemic for malaria and has recently been well described.[17] *Anopheles koliensis* has been documented as the principal malaria vector in the Arso region.[3] During investigations conducted concurrently with this study, *An koliensis* represented over 90% of the anophelines collected and 95% of the sporozoite infected vectors.

Because *Culex quinquefasciatus* has shown multi-resistance to many classes of insecticides worldwide[15,16] we had the opportunity to screen the abundant Arso PIR I population for sensitivity.

The purpose of this study was to determine if DDT resistant *An koliensis* individuals were present in the Arso PIR I population at the recommended diagnostic dosages and to verify previous reports suggesting DDT resistance in northeast Irian Jaya.

METHODS AND MATERIALS

The study site, Arso PIR I is 60 kilometres south of the Pacific Coast of Irian Jaya and 24 kilometres west of the international border with Papua New Guinea (140° 47' E, 2° 56'S elevation < 50 meters). It is near the southern extreme of a narrow coastal plain that remains primarily secondary jungle. The area has been extensively cleared for commercial oil palm estate's and settlements. Arso PIR I is one of several settlement sites in the Arso area that is principally populated by transmigrants from the island of Java. This area has proved fertile ground for intense malaria transmission.[17]

A WHO standard diagnostic test kit with unexpired pretreated insecticide impregnated and control papers were used following the protocol set forth in "Instructions for determining the susceptibility or resistance of adult mosquitoes to organochlorine, organophosphate and carbamate insecticides - diagnostic test".[18]

All female mosquitoes tested were captured from nightly human-landing or animal-shed resting collections. These collections were made from sprayed and unsprayed homes and from indoor and outdoor locations. In most cases, collected specimens were fully blood-fed or were allowed to feed on chickens before test exposure. To avoid against damaged specimens, a pre-test holding period of one hour was conducted. Those refusing to fly or obviously disabled were discarded. Depending upon availability, each test and control cylinder contained between 15 and 30 mosquitoes. Mosquitoes were exposed to the diagnostic dosages of 4% DDT for the recommended time period, (1 and 2 hours) then carefully removed and placed in clean holding tubes. Mosquitoes found dead after one hour exposure were immediately removed and recorded. Holding tubes were held in a secluded, unsprayed indoor shaded area for 24 hours, after which mortality was recorded. Holding and test cylinders were kept separate to avoid cross contamination and were not interchanged with other insecticides at any point during the study.

All tests were conducted under as similar conditions as possible. Ambient temperature and relative humidity was recorded during actual exposure period and maximum-minimum temperature was recorded over the 24-hour holding period. Age-grading dissection was done each test period on a 10% random sample of the nightly catch using ovarian tracheole condi‾ .... .. uifferentiate between parous and null.parous females.[19]

Controls were conducted on nearly all replicates for observing natural mortality rates. When applicable, test mortalities were corrected by Abbott's formula. The probable insecticide resistance status was based on WHO approved criteria for diagnostic dosages.[20]

RESULTS

DDT toxicity results are summarized in Table 1. Ambient temperatures and relative humidity during the exposure and 24-hour holding periods were nearly identical among all tests and controls, respectively. Exposure period temperatures ranged from 25-27°C while holding temperature ranged from 23-32°C under 60-95% relative humidity.

Tabel 1. **DDT resistance in** *Anopheles koliensis* **in Arso PIR, Irian Jaya, Indonesia.**

Date : 29 Jan - 04 Feb 1988

Treatment 1 hour exposure	% mortality TEST #			Mean* (A)
	1	2	3	
DDT* (n = 240)	88.8 (86.7)	70.0 (68.4)	61.3 (57.0)	73.4 (70.1)
Control	15.8	5.0	10.0	10.2

Date: 10-17 Jan 1989

Treatment 1 hour exposure	TEST #					Mean
	1	2	3	4	5	
DDT* (n = 164)	(91.5)	60.0	73.1	58.8	52.4	71.6
Control	20	39	34.5	--	--	--

Treatment 1 hour exposure	TEST #			Mean
	1	2	3	
DDT* (n = 64)	(73.1)	75.0	50.0	67.2
Control	7.0	--	--	--

* % mortality after 24 hours to 4% DDT.

( ) Abbott's formula

$$\frac{\%\ \text{test mortality - \% control mortality}}{100 - \%\ \text{control mortality}} \times 100$$

In January 1988, three test series involving 240 mosquitoes resulted in a mortality of 70.1% after 1-hour exposure to 4% DDT. Using normal approximation, the 95% confidence interval (C.I.) is 64.3% to 75.9%. No mortality was seen during the 1-hour contact period. The average parity rate was 34%. One year later, a 5 test series (N = 164 produced a similar mortality of 71.6% (95% C.I. 64.7% - 78.5%). Fewer mosquitoes were tested against DDT in 1989 because of low collection numbers. Moreover, control mortalities were much greater during this period, possibly the consequence of an older population sampled (63.8 - 72.8% parous). One-hour mortality also varied with each test from 0-28%. In 1989, three tests with 2-hour DDT exposure produced 67.2% mortality after 24 hours. Between 28.4% and 32.4% of the population in Arso PIR I showed some degree of resistance to 4% DDT at one and two hour exposures. Because *An koliensis*, on average, had test mortalities below 80%, this species is considered resistant based on WHO criteria.

Culex quinquefasciatus, a significant pest species in the Arso area, when exposed to 4% DDT for 4 hours produced an average mortality of only 69% (N = 143). Mortality was not seen in the control. Although present, to few *Anopheles punctulatus* and *An. farauti* were available to conduct meaningful tests.

DISCUSSION

Irian Jaya is the most remote and least developed province in the Indonesian archipelago. Many areas of Irian Jaya have meso-to-holoendemic malaria, creating a serious public health problem. [11] Dutch and American workers from the 1940's to early 1960's published many reports concerning vector incrimination, bionomics and repeated attempts at malaria control. [7] Up to the present, malaria control efforts in Irian Jaya have given poor or only passing relief.

DDT use in the Arso region of northeastern Irian Jaya began around 1960 and has continued nearly uninterrupted up to the present study. DDT has had little impact on malaria transmission in the Arso region presumably because of vector behavior. [1] Slooff [2] believed the exophilic behavior of *An. koliensis* would limit the effectiveness of residual spraying and thereby impede the development of resistance to residual insecticides like DDT. With regard to efficacy, Slooff believed that the shorten degree of contact with insecticide was more important than susceptibility. After intensive investigation, Slooff [3] concluded it would be nearly impossible to interrupt transmission in Arso through use of DDT alone. Garret-Jones and Grab [21] reviewed Slooff's data on DDT spraying and concluded that the overall impact on the vectorial capacity was minimal. In effect, DDT would not be expected to interrupt transmission in areas of high perennial incidence. The analysis of these workers agreed with Ford's [22] belief that systematic interior residual spraying would have limited effectiveness owing to the outdoor resting habits of this species complex. Regarding the actual effect of an insecticide on disease transmission, the interpretation of bioassays as conducted in this study can be misleading as they fail to take into account vector behavior when near or in contact with sprayed surfaces. This is especially true with the excito-repellent properties of DDT. [23]

In 1956, Smith[24] stressed the need for supplementary or alternative methods of control in Indonesia. As he put it, "There would appear to be ample evidence to cast serious doubt on the ability of the one method of residual spraying to control malaria adequately in Indonesia under existing conditions and in a reasonable period of time." Today, residual insecticide usage in Irian Jaya is being scrutinized to establish its efficacy and sustainability. Alternative approaches to vector control are also being planned and evaluated (e.g. impregnated bednets, source reduction, biological control).

In 1991, all uses of DDT, including public health applications was banned. However, remaining stock has been reserved for vector control in areas of economic and social development. Bendiocarb is now being evaluated as a country-wide replacement for DDT. Nonetheless, reliance on an insecticide alone to control malaria is usually too expensive and impractical. Community- based integrated approaches involving timely diagnosis and treatment, source reduction, personal protection, health education, and chemical control, concurrent with socio-economic development, may be the best strategy.

Although this is the first report, based upon repeated measurements of resistance to DDT in *An. koliensis* from Irian Jaya, resistance has probably been present since 1980 or even longer. The results of the work in Arso emphasizes the need for reassessment of insecticide use in areas like Irian Jaya for long-term control. However, alternative chemicals should continue to be evaluated for both physiological response and behavioral effects on the vector.[25] Fortunately, resistance to DDT does not promote resistance to organophosphate compounds.[15] However, the lethal effect may not be the only desirable characteristic of an insecticide. Instead, insecticides that alter vector feeding behavior and reduce human- vector contact might offer the best means of sustainable control when integrated with other measures.

Infrastructure, education and communications remain primitive in most of Irian Jaya. The many different languages and cultures create imposing obstacles to development. Nonetheless, progress is being made to overcome these obstacles that hamper abatement of malaria. Increased malaria control will come as a natural consequence of accelerated socio-economic development may be the best overall long-range approach to malaria control.

The findings in this report suggest the presence of significant resistance to DDT in the principle vector of malaria in the Arso region. Replacement of DDT with other insecticides such as malathion, bendiocarb or fenitrothion may not prove efficacious because of vector exophilic behavior. Cross-resistance to pyrethroids may also be present, thereby excluding the use of some of these chemicals.[26] If other compounds are found to be ineffective or too expensive, the continued use of DDT *in combination* with other control methods may still be appropriate. However, consideration and development of alternative strategies for long-term vector control in Arso PIR is sorely needed.

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