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ACTION OF BENDIOCARB AGAINST TRIATOMINE (HEMIPTERA: REDUVIIDAE) VECTORS OF CHAGAS' DISEASE[1]

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Abstract. Laboratory and field trials were conducted to determine the action of bendiocarb against triatomine vectors of *Trypanosoma cruzi.* Bendiocarb concentrations of 100 mg active ingredients/m² killed all exposed triatomines; at lower concentrations *Triatoma infestans* was the most susceptible, followed by *Rhodnius prolixus* and *Panstrongylus megistus.* The ovicidal effect of bendiocarb was slight. Residual activity of bendiocarb applied to earthen walls ("barro") was depleted by ⅔ within 2 months postspray and virtually nil 4 months after application. Field trials in an endemic area of Brazil where *P. megistus* is the sole domestic triatomine demonstrated that bendiocarb is effective in reducing triatomine densities. Household infestation rates were reduced from 18% prespray to 7% postspray. *Panstrongylus megistus* density was reduced from 7 per man-h prespray to 1.5 per man-h postspray. The practical use of bendiocarb in large-scale campaigns for the control of Chagas' disease will depend on cost.

Trypanosoma cruzi presently infects about 10 million persons in Brazil (Barretto 1979). Improvement of socioeconomic conditions and subsequent housing improvement in endemic areas should be the definitive vector-control measure. However, such improvement is a slow process. Thus, insecticides are used as an intermediate step to control transmission of *T. cruzi,* the etiologic agent of Chagas' disease (Sherlock 1976).

Well-conducted programs relying on benzene hexachloride (BHC) application have controlled *Panstrongylus megistus* (Burmeister), the principal vector of Chagas' disease in an extensive area of Brazil (Sherlock & Serafim 1972, 1974, Sherlock 1979a). Such application can reduce triatomine populations, thereby greatly reducing transmission of *T. cruzi* to man (Dias 1959, Sherlock & Muniz 1974/75, Silva 1979). However, BHC has proven toxic to man and domestic animals (Dias 1959, Leal et al. 1965, Sherlock & Muniz 1974/75, Sherlock 1976, 1979b, Silva 1979). In addition, resistance to chlorinated hydrocarbons has been detected in populations of Venezuelan *Rhodnius prolixus* Stål (Valdivieso et al. 1971, Cockburn 1972, Nocerino 1976, Silva 1979). Consequently, the testing of more efficient and safer insecticides, as well as alternative triatomine control measures, are urgently required.

We have already tested the actions of several insecticides against *P. megistus* in the field (Sherlock & Muniz 1974/75, 1975, 1977, Sherlock et al. 1976, Sherlock 1979b). The new carbamate insecticide bendiocarb provided excellent results when used in Britain for the control of a variety of domestic insects (Story 1972, Lemon & Bromilow 1977, Traub & Starke 1980), and in Iran for the control of *Anopheles stephensi* (Eshghy et al. 1980). Moreover, initial laboratory tests against *R. prolixus* proved promising (Nelson & Colmenares 1979). Accordingly, we determined in the laboratory the effect of bendiocarb against the principal vectors of Chagas' disease. Subsequently, we evaluated the efficacy of bendiocarb for the control of *P. megistus* in a Chagas' disease endemic area.

MATERIALS AND METHODS

Insecticide. Bendiocarb, 2,2-dimethyl-1,3-benzodioxol-4-yl methylcarbamate, was discovered in 1967 and designated WHO 1394 (bendiocarb) (WHO 1970, Story 1972, Traub & Starke 1980). We received bendiocarb in the form of Ficam W®, which was kindly donated by the manufacturer, FBC Limited Company of England. The crude product is a wettable powder containing 80% (nominal) carbamate, the active ingredient of bendiocarb. Bendiocarb is highly active as a contact and ingested poison for various domestic insects. The acute oral LD_{50} of rat-formulated product is 179 mg/kg, active ingredient (lab. solvents) 50 mg/kg; acute dermal LD_{50} of rat-formulated product is 1000 mg/kg (WHO 1970, Story 1972, Traub & Starke 1980). Since bendiocarb is a cholinesterase inhibitor, the manufacturer recommended specific

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TABLE 1. Mortality of 4th- and 5th-instar triatomine nymphs exposed to bendiocarb-sprayed filter paper (500 mg a.i./m²).

	Treatment	No. nymphs observed	Cum. % mortal. after exposure (days)			
			1	2	3	7
P. megistus	bendiocarb	20	97	97	97	100
	control-moist	20	0	0	0	0
	control-dry	20	0	0	0	0
T. infestans	bendiocarb	20	100	—	—	—
	control-dry	20	0	0	0	0

safety precautions for its experimental use (Bonsall et al. 1981, Motabar et al. 1981).

Laboratory studies. Standard insecticide test methods were used in the laboratory (Busvine 1961, WHO 1970). We prepared filter papers containing measured concentrations of bendiocarb. Acetone solutions containing sufficient bendiocarb to give final concentrations of 100, 50, or 10 mg active ingredient (a.i.)/m² were placed in petri dishes containing filter paper 10 cm in diam. Treated filter papers were allowed to dry for 1 h before triatomines were exposed. Filter papers impregnated with aqueous solutions of bendiocarb applied in the field contained 500 mg a.i./m² (see below). Triatomines were exposed to bendiocarb-treated or nontreated (control) filter papers inside petri dishes or plastic cups covered with cotton mesh. Samples of earthen wall surfaces ("barro") were removed from houses immediately after bendiocarb (500 mg a.i./m²) spraying. Triatomines were placed into glass jars containing the bendiocarb-treated barro on 1, 50, and 120 days postspray. The ovicidal effect of bendiocarb was tested by observing daily, for 30 days, the proportion of eggs eclosed and the mortality of eclosed nymphs (age 0–24 h) in contact with bendiocarb-treated (500 mg a.i./m²) filter papers. All tests were conducted in the Centro de Pesquisas Goncalo Moniz (CPqGM, Salvador, Bahia) at room temperature (25–30 °C, 70–90% RH).

Triatomine bugs used in these studies were obtained from colonies maintained at CPqGM for up to 10 years. Colonies were maintained as previously described (Szumlewicz 1976, Sherlock 1979a). The origins of colonies used in this study were: *P. megistus,* state of Bahia, Brazil; *Triatoma infestans* (Klug), states of Bahia and São Paulo, Brazil; *R. prolixus,* Venezuela; and *Triatoma dimidiata* (Latreille), Belize.

Study area. The study area, in the municipio of Castro Alves, state of Bahia, Brazil, has been previously described (Sherlock & Muniz 1975, Mott et al. 1976). Chagas' disease is highly endemic in the area; a serological survey of residents prior to the evaluation of bendiocarb demonstrated a rate of 44% seroreactivity to *T. cruzi* (Mott et al. 1976). *Panstrongylus megistus* was the only triatomine commonly infesting houses within the area, and its infection rate with *T. cruzi* was 38% (Sherlock & Serafim 1972, 1974, Sherlock & Muniz 1975, Mott et al. 1978). Other insecticides have been evaluated in the Castro Alves study area (Sherlock & Muniz 1975, Sherlock et al. 1976).

Ten geographic subunits, defined as fazendas by local residents, were included in this study. The closest is ca. 12 km from the city of Castro Alves by road. Some fazendas are quite distant and are located in rough terrain impossible to reach by motor vehicle. A census of the 10 contiguous fazendas prior to our study identified 232 houses containing 1055 persons.

Field trial. To determine the density of *P. megistus* household infestations, we carried out timed searches following application of the excitatory repellent pyrethrum. Pyrethrum was applied, bug densities were determined, and bugs examined for infection with *T. cruzi* as previously described (Paulini et al. 1961, Sherlock 1976, 1979b, Mott et al. 1978). All houses in the 10-fazenda study area were searched for bugs during December 1979, before application of bendiocarb, and in December 1981, 6 months after the final bendiocarb application. The preliminary and final searches were performed by the same individual.

Bendiocarb was applied to the inside and outside walls of every house in the area. The initial application was performed immediately following the preliminary timed search for triatomines in December 1979. Subsequently, bendiocarb was applied to all houses at 6-month intervals during 2 years. Bendiocarb, 80% a.i., was provided in packets of 125 g. Each packet was mixed with 10 litres of water inside Blemco hand sprayers. Sprayers were fitted with HSS-8002-NOZZI nozzles cali-

TABLE 2. Mortality of 4th- and 5th-instar triatomine nymphs exposed for 24 h to filter papers containing varying concentrations of bendiocarb.

	Insecticide conc. (mg a.i./m²)	No. nymphs exposed	Mortality (%)
P. megistus	100	20	100
	50	20	0
	10	20	0
	*	20	0
R. prolixus	100	20	100
	50	20	60
	10	20	0
	*	20	0
T. infestans	100	20	100
	50	20	100
	10	20	0
	*	20	0

* Acetone (control).

brated to deliver about 760 ml of suspension/min. The contents of each sprayer covered 1 house, delivering a final bendiocarb concentration of 400–500 mg a.i./m^2.

RESULTS

Laboratory studies. As a preliminary experiment, we determined whether the aqueous solution of bendiocarb applied in the field at a concentration of 500 mg a.i./m^2 was effective in killing triatomines. Filter papers were placed on wall surfaces during the spray operation. Fourth- and 5th-instar *P. megistus* and *T. infestans* were placed in contact with exposed filter papers 24 h postspray. Virtually all triatomines exposed to treated filter papers died within 24 h (Table 1). One *P. megistus* survived until the 7th day of exposure. Thus, bendiocarb sprayed at a concentration of 500 mg a.i./m^2 was extremely effective in killing triatomines.

The concentration of bendiocarb required to kill triatomines was determined. Bendiocarb concentrations of 100 mg a.i./m^2 killed all exposed triatomines during the 24-h test period; there was no mortality in the group exposed to 10 mg a.i./m^2 (Table 2). Concentrations of 50 mg a.i./m^2 killed all exposed *T. infestans,* the majority of exposed *R. prolixus,* and none of the exposed *P. megistus.* Thus, *P. megistus* was the least susceptible to bendiocarb of the 3 triatomine species examined.

We studied the residual effect of bendiocarb when sprayed onto the earthen wall surface (barro) typical of mud-stick houses in the endemic area. The majority of triatomines exposed to barro 1 day postspray died within 24 h and virtually all such triatomines died within 1 week of exposure (Table 3). However, less than ⅓ of triatomines exposed to barro 50 days postspray died. Bendiocarb-treated barro had virtually no effect on triatomines exposed 4 months postspray. The residual activity of bendiocarb applied to earthen surfaces was short-lived, lasting less than 2 months.

The ovicidal effect of bendiocarb was determined. The proportion of eclosion among bendiocarb-exposed eggs was less than control (nonexposed) eggs for all 4 species (Table 4). However, the proportion of eggs eclosing was reduced to less than ½ that of control eggs in *R. prolixus* but not in *P. megistus, T. dimidiata,* or *T. infestans.* Overall, the ovicidal effect of bendiocarb was slight. In contrast, 100% of 1st-instar nymphs died within 24 h

TABLE 3. Mortality of 4th- and 5th-instar triatomine nymphs exposed to bendiocarb-sprayed barro (500 mg a.i./m²).

	Time between insecticide appl. and exposure (days)	Treatment	No. nymphs observed	Cum. % mortal. after exposure (days) ⅓	1	2	3	7
P. megistus	1	bendiocarb	30	7	60	77	90	93
		control	20	0	0	0	0	0
	50	bendiocarb	30	0	10	13	17	30
		control	20	0	0	5	5	5
	120	bendiocarb	30	0	0	0	3	7
		control	20	0	0	0	0	5
T. infestans	1	bendiocarb	20	10	60	85	100	100
		control	20	0	0	0	0	0
	50	bendiocarb	20	5	10	15	15	25
		control	20	0	0	0	0	0
	120	bendiocarb	20	0	0	0	5	5
		control	20	0	0	0	0	0

TABLE 4. Development of triatomine eggs exposed to bendiocarb-sprayed filter paper (500 mg a.i./m²).

	TREATMENT	NO. EGGS OB-SERVED	% EGGS ECLOSED	% MORTAL. OF ECLOSED NYMPHS
P. megistus	bendiocarb	92	29	100
	control	48	54	0
T. dimidiata	bendiocarb	71	35	100
	control	40	53	0
T. infestans	bendiocarb	80	46	100
	control	62	87	6
R. prolixus	bendiocarb	100	15	100
	control	40	85	0

of eclosion. A similar effect was noted when eggs of *P. megistus* were placed in contact with bendiocarb-treated barro (Table 5). Eclosion was slightly reduced and 9/10 of the 1st-instar nymphs died within 24 h of eclosion.

Field trial. We evaluated the efficacy of bendiocarb for the control of *P. megistus* by comparing the prespray (Dec. 1979) and postspray (Dec. 1981) prevalence and density of household *P. megistus* in the 10-fazenda study area. Although 232 houses were examined in the preliminary timed search, many of these houses were demolished or abandoned during the 2-year study period. Accordingly, only those houses (156) that remained intact and inhabited during the study were included in the final analysis. The proportion of houses found to be infested by *P. megistus* dropped from 18% prespray to 7% postspray (Table 6). In addition, while 13% of houses contained *T. cruzi*-infected *P. megistus* before spraying, none of the houses contained infected *P. megistus* after spraying.

The density of *P. megistus* was reduced from a prespray level of 7 per man-h to a postspray level of 1.5 per man-h (Table 7). Interestingly, most triatomines collected in the prespray search were nymphs, whereas about 90% (33/37) of the triatomines collected postspray were adults. Since winged adults are able to disperse whereas nymphs are not, the predominance of adults in houses post-

TABLE 5. Development of triatomine eggs exposed to bendiocarb-sprayed barro (500 mg a.i./m²).

	TREATMENT	NO. EGGS OB-SERVED	% EGGS ECLOSED	% MORTAL. OF ECLOSED NYMPHS
P. megistus	bendiocarb	40	25	90
	control	20	75	0

TABLE 6. Control of domestic *Panstrongylus megistus* by semiannual application of bendiocarb in Castro Alves, Bahia. Proportion of houses infested.

	NO. HOUSES EXAMINED	NO. HOUSES (%) INFESTED	NO. HOUSES (%) WITH INFECTED BUGS
Dec. 1979 (prespray)	156	28 (18)	18 (13)
Dec. 1981 (postspray)	156	11 (7)	0

spraying may have been the result of reinfestation by adults from outside. In addition, the decrease in the proportion of *P. megistus* infected by *T. cruzi*, dropping from ¼ in 1979 to none in 1981, suggested that the adult *P. megistus* collected postspray came from sylvatic ecotopes where *T. cruzi* was rare or absent.

To confirm whether adults were indeed numerically predominant over nymphs and whether *T. cruzi*-infected *P. megistus* were absent in the postspray domestic *P. megistus* population, we reexamined 22 houses judged to be infested in 1979 but noninfested in 1981. This reexamination lasted 30 min, as opposed to the standard 10 min used in the original timed searches. The reexamination of 22 houses took place in February 1982, 2 months after the postspray timed search in December 1981. Bendiocarb had been applied to all houses in the study area immediately following the December 1981 collection. Of these 22 houses, 9 (41%) were infested by *P. megistus*. Of the 28 specimens collected (3.8 bugs/man-h), 12 were adults and 16 were nymphs. Twenty-three of these *P. megistus* were examined, 12 (53%) of which were infected by *T. cruzi*. Thus, comparison of prespray and postspray timed searches demonstrated a marked relative reduction in the *P. megistus* population. However, the more thorough reexamination demonstrated that *T. cruzi*-infected *P. megistus* continued to infest houses after 2 years of bendiocarb treatment.

DISCUSSION

Many insecticides are highly active against triatomines in the laboratory but lose their effectiveness when applied under field conditions (Sherlock 1976). Insecticide residual activity is often reduced when applied to natural substances such as earthen walls (barro). Barro markedly depleted the residual activity of bendiocarb within 2 months after application. Prolonged residual activity is especially important in insecticides used to combat triatomines, since triatomines can reinfest houses once insecticides have lost their activity (Sherlock 1976).

Reinfestation is probably the result of adult triat-

TABLE 7. Control of domestic *Panstrongylus megistus* by semiannual application of bendiocarb in Castro Alves, Bahia. Density of triatomine populations.

	No. houses examined	No. *P. megistus* coll.			No. coll. per man-h	*P. megistus* infected with *T. cruzi*	
		Adults	Nymphs	Total		No. examined	No. infected (%)
Dec. 1979 (prespray)	156	62	102	164	7	134	33 (25)
Dec. 1981 (postspray)	156	33	4	37	1.5	28	0

omines flying into houses from either sylvatic habitats or neighboring infested dwellings. Freitas (1963) emphasized the dispersal tendency of newly emerged adult *P. megistus*. The lack of nymphs in the postspray timed search suggested that reproduction of *P. megistus* had been interrupted in sprayed houses and that the adult *P. megistus* collected had flown in from outside. However, the more detailed reexamination detected additional nymphs, indicating that triatomines had survived in sprayed houses; triatomine reproduction had been diminished but not interrupted. Triatomines can avoid contact with insecticide-treated surfaces in mud-stick houses by hiding in places not reached by the chemical or where its effects are minimal (Sherlock 1976). Presently, we cannot establish whether triatomines detected in houses after insecticide treatment reinfested these dwellings or survived insecticide application.

One of the principal advantages of bendiocarb was the ease of application. The convenient packets facilitated suspension preparation. Since bendiocarb is water soluble, mixing was easy in the field. Moreover, the problem of plugging of spray nozzles, often observed with other insecticides (Sherlock & Muniz 1974/75, Sherlock 1979b, Silva 1979), was absent with bendiocarb. Finally, bendiocarb was nearly odorless, with a smell similar to rain-moistened fields. The lack of the unpleasant odor often associated with insecticides enabled us to obtain the complete cooperation of residents during the spray operation.

No signs of immediate toxicity were evident in spraymen and residents during bendiocarb application. Spray personnel were instructed, as per manufacturer's safety precautions, not to expose persons and domestic animals to insecticides. Nevertheless, domestic animals were commonly, and residents occasionally, exposed. A kitten was sacrificed by its owners when it became agitated 1 h after contact with bendiocarb. A dog and a cat were sprayed directly, upon owner's request, to combat flea infestation. Both animals became severely ill but convalesced. Overall, we observed less toxicity to residents, spray personnel, and domestic animals than had been experienced in the same area with other insecticides (Dias 1959, Freitas 1963, Sherlock 1979b, Silva 1979).

The field-trial study plan, while not ideal, is one that the principal author has adapted after many years of experience with insecticide trials (Sherlock & Muniz 1974/75, 1975, 1977, Sherlock et al. 1976, Sherlock 1979b). First, insecticide was applied at 6-month intervals. Whereas more frequent application would surely increase the efficacy of the test insecticide, economic and bureaucratic considerations make it impossible to apply insecticide more frequently than semiannually in Brazil (Sherlock & Muniz 1975, 1977, Sherlock 1976). Second, comparison of prespray and postspray triatomine levels is less meaningful than a comparison of sprayed to nonsprayed (control) areas. However, ethical and humanitarian considerations prevent the designation of control houses in the endemic area. Third, the 10-min timed search gives only a relative estimate of triatomine density. Triatomines may be present in houses that are negative in the 10-min timed search, as shown by our discovery of 9 infested houses during our reexamination. However, absolute triatomine densities can only be determined by house demolition, a technique that is obviously impractical (Sherlock & Muniz 1975, Sherlock 1976). Therefore, 10-min timed searches following pyrethrum application are the most practical means of evaluating triatomine densities during insecticide trials. Indeed, this technique has been adopted by official control campaigns in Brazil (Paulini et al. 1961, Sherlock 1979b). Finally, housing improvements may lead to an overestimate of an insecticide's effect. Owing to informal sanitary education, residents are often motivated to improve dwellings to combat vectors of Chagas' disease (Sherlock 1979b). Thus, some houses were plastered and painted during the 2-year study period. However, marked reductions in triatomine populations took place in many mud-stick houses that were unchanged except for bendiocarb application.

Bendiocarb is a promising insecticide for *P. megistus* control. Among the important vectors of Chagas' disease, *P. megistus* is the least susceptible to insecticides (Sherlock 1979b). In our laboratory tests, *T. infestans* appeared to be the most susceptible to bendiocarb, followed by *R. prolixus* and *P. megistus.* A recent trial involving 16 infested houses in La Guaca, Venezuela, showed bendiocarb to be efficient in the control of *R. prolixus* (Dr J. Goose, pers. commun.). We found bendiocarb to be as effective in the control of *P. megistus* as were BHC (Sherlock & Muniz 1974/75), malathion (Sherlock & Muniz 1975, Sherlock et al. 1976), and Baygon® (Sherlock & Muniz 1977). Accordingly, bendiocarb may prove useful in the control of Chagas' disease. Its practical use in large-scale campaigns for the control of Chagas' disease will depend on cost. Meanwhile, the search for other insecticides and alternative means of controlling triatomines must continue.

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