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INVESTIGATIONS ON THE USE OF INSECTICIDE-BASED PAINTS FOR THE CONTROL OF TRIATOMINE BUGS

by

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and
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EXECUTIVE SUMMARY

A recent development in the control of triatomine vectors of Chagas' disease is the incorporation of insecticides into paint and other matrices to provide long lasting insecticidal activity. Initial evaluation of a commercially available insecticide-based paint, conducted at the Centro Internacional de Entrenamiento e Investigaciones Médicas (CIDEIM) in Cali, Colombia and at the Division de Control de los Vectores (DCV) in Tegucigalpa, Honduras provided promising indication of the potential for residual control of triatomine bugs.

The paint, a chlorpyriphos-based acrylic (Super IQ, A.I. 0.9%) with prolonged demonstrated residual activity against cockroaches was evaluated under laboratory conditions in Cali, Colombia against fifth stage nymphs of Rhodnius prolixus on fired and unfired bricks and wood substrates. The material was most active on wood substrates producing 5, 17, 20 and 100% mortality after 2, 4, 8, and 16 hours of exposure, respectively. Sixteen hours of exposure on fired and unfired brick substrates resulted in 53 and 30% mortality, respectively. Considerably less exposure of first stage nymphs of R. prolixus was required to produce significant mortality. After 0.5, 1, and 4 hours exposure to painted wood substrates, the nymphs responded with 30, 81, and 95% mortality, respectively. Nymphs of four other species were also used for bioassays of Super IQ. After 8 hours of exposure to painted wood substrates, 100% mortality was observed in Dipetalogaster maximus and Triatoma infestans. Nymphs of Triatoma pallidipennis and Triatoma leticularia responded with 33 and 95% mortality, respectively. Studies on the residual activity of Super IQ are currently in progress at CIDEIM in Cali.

Contact between individuals concerned with control of Chagas' disease in Honduras and Dr. D'Alessandro in May 1987, indicated an interest in the use of insecticide-based paints. The current visit was intended to initiate studies on the residual activity of the chlorpyriphos-based paint under field and simulated field conditions, and to draw up a work outline for a pilot study on the use of the paint to interrupt transmission.

Application of the paints onto materials commonly used for construction of dwellings was done in Tegucigalpa with the help of Dr. Luis Rivera and the staff from Division de Control de los Vectores (DCV). Initial results of bioassays on the painted surfaces with unfed field collected R. prolixus indicate activity comparable to that observed in the Colombia tests.

Field trips to small towns near Tegucigalpa (Santa Lucia and Valle de los Angeles) revealed low levels of infestations with Triatoma dimidiata. A visit to a small village outside of San Marcos de Colon (Dept. Choloteca) yielded incredibly high infestations of R. prolixus in certain thatched roofed houses.

The pilot study evaluation of Super IQ as a control intervention is proposed in three phases. The preliminary phase would include the evaluation of Super IQ on various types of walls (constructed of brick, mud, cement, and wood). Initial and residual insecticidal activity will be assessed using field collected and/or colony reared triatomine bugs.

Phase II will involve the application of Super IQ to the walls and ceiling of rooms used for sleeping in at least five houses in a Chagas' endemic zone that is not currently under a spray program. Evaluation will be chiefly through entomological surveys.

Phase III will involve application of Super IQ to the walls and ceilings (when appropriate) of rooms used for sleeping in all houses of one or more selected communities in which Chagas' disease is endemic.

Based on the benefits of home improvement and prolonged control of vector and pest insects, commercially available insecticide-based paints offer good potential for use in programs that will utilize community participation for application of the paint. Community health education will play a key role in maximizing the potential of such interventions.

I. INTRODUCTION AND BACKGROUND

Chagas' disease is found in virtually every country in South and Central America (including Mexico). In Honduras the two principal vectors of Chagas' disease are R. prolixus and T. dimidiata. The distribution and incidence of Chagas' disease in the country have been recently elucidated by Ponce and Ponce (unpublished report to TDR/WHO). Over 15% seropositivity for Chagas' is observed in much of the South, Central and Western portions of the country. Seropositivity of 4.4% is reported for the Northern areas. Many of the seropositive foci are not currently covered by regular insecticide applications.

When control efforts have been attempted, in Honduras and elsewhere, they have been conducted in the same manner as for the control of malaria vectors (i.e., through the application of residual insecticides to the walls of houses for control of the triatomine vectors). Residual activity of many of the insecticides used for control is often of insufficient duration to bring about control without repeated application throughout the year. For example, a case of extremely short lived control (17 days) of T. infestans is reported by Martinez et al. (1975) after an application of malathion. Other reports indicate variable duration of insecticidal activity of conventional insecticides when applied by conventional spraying. Residual activity is shortest on porous surfaces, especially those made of mud (Schofield 1985).

One possible solution to the short effective life of a single application is the incorporation of active material into paint and other slow release materials. Studies conducted by Oliveira Filho (1984) and Pinchin et al. (1978a) indicate that extremely prolonged residual insecticidal activity under laboratory conditions is possible using experimental slow release formulations of malathion and chlorpyriphos. The purpose of this assignment was to evaluate the efficacy of a commercially available chlorpyriphos-based paint for control of R. prolixus, and other triatomine bugs and the potential for its use in a pilot program in Honduras. The use of slow release insecticides with prolonged insecticidal activity applied by communities would enhance the feasibility of controlling Chagas' disease in Honduras and other developing countries in the forseeable future.

Based on interest among Honduran Ministry of Health professional staff for potential use of insecticide-based paints for control of triatomine vectors of Chagas' disease (see Annex 1), Drs. Antonio D'Alessandro and Lawrence Lacey visited Honduras to assess the possibility for developing a collaborative pilot

study on the efficacy and residual control of Chagas' vectors using a commercially available paint (Annex 2).

Laboratory bioassays on a chlorpyriphos-based paint (Super IQ) conducted in Cali, Colombia indicate that the paint has very good potential for control of a range of triatomine species including the two most important vectors in Central and South America, R. prolixus and T. infestans (see Annex 3).

Two of the key personnel proposed for collaborative studies, Dr. Carlos Ponce and Dr. Elisa de Ponce, were away on emergency TDY in Nicaragua. Discussions with Dr. Pereira, Laboratorio Central, Ministerio de Salud, and Dr. Rubio, Dr. Rivera and Mr. Gomez of the Division de Control de los Vectores of the same ministry, and correspondence with Dr. Rina Kaminsky, Project Hope, indicated a continued interest in innovative interventions for use in triatomid control.

The pilot study evaluation of Super IQ as a control intervention is proposed in three phases. The preliminary phase would include the evaluation of Super IQ on various types of walls (constructed of brick, mud, cement, and wood). Initial and residual insecticidal activity will be assessed using field collected and/or colony triatomine.

Phase II will involve the application of Super IQ to the walls and ceiling of rooms used for sleeping in at least five houses in a Chagas' endemic zone that is not currently under a spray program. Evaluation will be chiefly through entomological surveys.

Phase III will involve application of Super IQ to the walls and ceilings of rooms used for sleeping in all houses of one or more selected communities in which Chagas' disease is endemic. Evaluation of efficacy and residual activity will be through entomological and parasitological surveys. Detailed descriptions of each phase follow:

A. Phase I

Residual activity of Super IQ on various wall types.

1. Objective: To determine the activity of initial and residual insecticidal activity of Super IQ white paint on various wall types using field collected or colony reared R. prolixus and T. dimidiata. Ideally, assays would be conducted at monthly intervals until insecticidal activity subsides.

2. Procedures: Select walls to be treated in domestic or simulated domestic settings (for example, wall and other surfaces at the Division of Vector Control Offices). Surfaces should not be in an air conditioned environment, subject to being wetted by normal rainfall or exposed to direct sunlight.

Surfaces should be swept of loose dirt, cob webs, etc., and when possible, the manufacturer's instructions, on preparation of surfaces to be painted should be followed as closely as possible (Annex 4). Mud walls should be under-coated with white wash or other materials that would be normally used by local inhabitants. When surfaces are absolutely dry, apply one coat of the Super IQ white evenly over the surface. After at least 24 hours, commence initial bioassays using fifth stage nymphs of R. prolixus and, if possible, T. dimidiata. The bioassay procedure is as follows:

Securely tape WHO bioassay cones onto the substrates. Into each cone, place 10 fifth stage nymphs. Ensure that cones are taped securely in place. If the surface of the substrate is uneven, cotton can be stuffed into depressions near the edge of the cone before applying tape. The nymphs should not come into contact with sticky surfaces of the tape. If less than 10 nymphs are available per substrate and control, write the number used on tape and place the tape on the cone.

After 24 hours of exposure, remove the nymphs from the cones, recording the number dead and place the nymphs in holding cups, provided with paper-lined bottoms. Use one holding cup per cone and attach a label with the following information:

Date, number of nymphs used for test if other than 10, stage of nymphs if younger or older than fifth stage nymphs (5th stage nymphs can be recognized by wing pad development, the fifth stage of R. prolixus is considerably smaller than the fifth stage of T. dimidiata), and substrate type (wood, furniture, brick, mud, etc.).

Also, record this information on the register form that has been provided (Annex 5). Each day for five days, record the number dead that are found in the cups. After counting dead nymphs, remove them from the cups. Percent total mortality is calculated five days after the exposure period ends.

Ideally, the tests should be conducted with fifth stage nymphs of both R. prolixus and T. dimidiata. Protocols for Phase I in Spanish are provided in Annex 5.

B. Phase II

Approximately six months after the initiation of Phase I, five or more houses of typical construction should be selected for painting in an area endemic for Chagas' disease and in which moderate to high populations of R. prolixus or T. dimidiata can be found. Optimally, one or more areas, each containing one or both of the species should be selected.

Painting of walls should be done in all rooms that are used for sleeping. Application of paint should be with brushes or rollers to simulate conditions under which a community-based application program would operate. Preparation of surfaces to be painted would be as in Phase I.

Optional comparative studies could be conducted in two or more houses in which paint is applied using a paint compressor/sprayer. This application technique would be useful for painting ceilings especially those made of thatch.

Evaluation of this intervention would be provided chiefly through entomological survey. The standard 1-man hour/house search technique for presence of bugs (reviewed Schofield et al. 1987) would be used prior to and at monthly intervals after the application of paint. Two unpainted houses in each area will also be searched for presence of bugs as controls. This procedure should be followed for at least one year or until populations of triatomines rebound.

Supportive studies should include (1) determination of baseline susceptibilities of resident bugs to technical deltamethrin (or another pyrithroid), fenitrothion and chlorpyriphos using standardized WHO techniques (see Nocerino and Hernandez 1986), (2) mark-release recapture studies on bugs from habitats surrounding painted houses to determine extent of reinvasion, and (3) determine other less labor intensive survey methods.

C. Phase III

Pilot program to determine efficacy and residual activity of insecticide-based paint in two small communities.

Application of paint and entomological evaluation will be as in Phase II. Parasitological studies are outlined below.

Work outline for parasitological studies.

1. Obtain the information gathered during the national survey for the houses to be included in the assessment of the insecticide-paint assay.

2. Survey prior to the application of the insecticide-paint as follows:

   a. Search for triatomine bugs. R. prolixus in the roof and wall crevices of sleeping areas of the house, expressed in man/hour search.

   b. Search for triatomine bugs. T. dimidiata, on the floor of sleeping areas but also elsewhere in human dwellings, chicken houses, roof tiles and wood piles (peridomestic). Also, expressed in man/hour search.

   c. Parasitological survey of captured triatomine, both for T. cruzi and T. rangeli. Feces, hemolymph and salivary glands of both species should be examined using the procedures described by D'Alessandro (1972). According with the number of specimens collected (few or many) individual or a 30% random sample by instar should be examined completely.

   d. Serological survey of the human population in houses examined in this phase (less than one year of age) and new inhabitant or people missed during national survey are particularly important to assess interruption of transmission. If possible, it would be advantageous to use the 90k and 72k glyco proteins of T. cruzi in the ELISA test and monoclonal antibody systems respectively (in addition to and in comparison with the IFT test carried out during the national survey). According to recent reports, these tests would prevent cross reactivity with Leishmania spp. and Trypanosoma rangeli which are both present in Honduras (Schechtner et al. 1983, Demsre et al. 1983).

3. Follow up survey, on a monthly basis after the application of the paint:

   a. Search for triatomine bugs. R. prolixus in the walls and ceilings of sleeping areas of the house, expressed in number per man-hour search.

   b. Search for triatomine bugs. T. dimidiata, on the floor, of sleeping areas but also elsewhere in the human dwelling, adjacent chicken houses, and (peridomestic) wood and tile piles. Also, expressed in number per man-hour search.

   c. Parasitological survey of captured triatomine, both for T. cruzi and T. rangeli. Feces, hemolymph and salivary glands of both species should be examined. According with the number of specimens collected (few or many) individual or a 30% random sample by nymphal instar should be examined completely.

   d. Serological surveys it should be carried out every two months, especially in children under one year of age both after the application of insecticide paint as well as in newcomers and people missed in the national survey.

4. The follow-up will be continued monthly for at least two years at the same time as the evaluation of the activity of the paint.

D. References Cited

D'Alessandro, A. 1972. New experimental vectors of Colombian Trypanosoma rangeli. J. Med. Entomol. 9: 187-195, 1972.

Demesre, J.L. et al. 1986. Specific and sensitive immunological diagnosis of Chagas' disease by competitive antibody enzyme immunoassay using a Trypanosoma cruzi specific monoclonal antibody. Am. J. Trop. Med. Hyg. 35: 86-93.

Lent, H. and P. Wygodzinski. 1979. Revision of the Triatominae (Hemiptera: Reduviidae) and their significance as vectors of Chagas' disease. Bull. Am. Mus. Nat. Hist. 163: 520 pp.

Martinez, A., J.A. Cichero, I.R. Alania and F.F. Gonzalez. 1975. Control of T. infestans (Klug) with malathion concentrate. J. Med. Entomol. 11: 653-657.

Nacerino, F. and A. Hernandez. 1976. Establishment of baseline data on the insecticide susceptibilities of the Chagas' disease vector Rhodnius prolixus in Venezuela. PAHO Bull. 20: 366-371.

Nelson, M.J., J.O. Williams, F. Nocerino, A.A. Arata and R.J. Tonn. 1983. Laboratory and field testing of insecticides against Rhodnius prolixus (Reduviidae, Triatominae), the vector of Chagas' disease in Venezuela. Wld. Hlth. Org. mimeo. doc. WHO/VBC/83.886. 18 pp.

Oliveira Filho, A.M. 1984. New alternatives for Chagas' disease control. Mem. Inst. Oswaldo Cruz. 79 (Suppl.): 117-123.

Pinchin, R., A.M. Oliveira Filho, C.A. Muller, M.J. Figueiredo, A. Prlowagora-Szumlewicz, and B. Gilbert. 1978a. Slow-release insecticides for Triatomine control: activity and persistence. Rev. Bras. Mal. Doen. Trop. 30: 45-55.

Pinchin, R., A.M. Oliveira Filho, C.A.C. Ayala and B. Gilbert. 1978b. Slow-release insecticides for Triatomine control: preliminary field trials. Rev. Bras. Mal. Doen. Trop. 30: 57-63.

Pinto Dias, J.C. and R. Borges Dias. 1985. Participaçao da comunidade no contrôle da doença de Chagas. Ann. Soc. Belge. Med. Trop. 65 (Suppl. 1): 127-135.

Schofield, C.J. 1985. Control of Chagas' disease vectors. Brit. Med. Bull. 41: 187-194.

Schofield, C.J., D.M. Minter and R.J. Tonn. 1987. The triatomine bugs - biology and control. Wld. Hlth. Org. mimeo. doc. WHO/VBC/87.941. 41 pp.

ANNEX 1

REPORT OF A VISIT TO HONDURAS BY DR. ANTONIO D'ALESSANDRO
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Late in August 1987, Dr. D'Alessandro visited Honduras, invited by Drs. Donald and Rina Kaminsky, Project Hope to give lectures at the University Hospital in Tegucigalpa. Due to discussions during VBC's meetings in Washington and information on Honduras' requests for action in the field of Chagas' disease, it was agreed that during his visit Dr. D' Alessandro would gather information regarding the status of Chagas' disease in the country and the interest of the Ministry of Health authorities in its control. Dr. Rina Kaminsky kindly made all necessary arrangements to visit with the key persons involved in this problem, and agreed to be the local coordinator of the control program if one could be developed.

The following is a brief summary of the visits Rina Kaminsky and I paid to Honduran scientists and of follow up activities.

1. Dr. Jose Ramon Pereira, Chief, Division of Laboratories, Ministry of Health. Dr. Pereira was very much interested in the possibility put forward by Dr. D'Alessandro of moving to a control effort using a paint-based insecticide as a follow up of the National Chagas' Disease Survey. This survey was carried out in Honduras by the Ministry of Health as a combined effort with the Division of Laboratories and of the Control of Vectors under TDR/WHO sponsorship. Mention was made of the possibility of interesting VBC/AID/Tulane in some of the initial evaluation phases of the control program. Dr. Pereira agreed that the Ponces should be the parasitologists participating in this program because of their heavy involvement in the national survey.

2. Drs. Carlos Ponce and Lisa Ponce, a husband-wife team, was responsible for the parasitological studies of the national survey. As a matter of fact, because I was aware of the importance of this study, as baseline information to establish a control program, I was interested in visiting Honduras "unofficially" on behalf of VBC.

ANNEX 1 cont.

The Ponces were very interested in the possibility of beginning a vector control program, and they agreed to participate as per letter of July 7, 1987 and provided copies of the final report to TDR of the National Survey and other publications. (A copy of this report was sent to VBC in September 1987 by Dr. D'Alessandro.)

3. Dr. Pineda, Chief of DCV, could not be visited because he was out of the country.

4. Dr. Barry Smith, A.I.D. in Tegucigalpa, also could not be visited because he was out of the country.

5. Dr. DeLeon, DCV, was very informative regarding vector-borne diseases and while with him Jeff Stivers, USAID personal services contractor, joined the group and both agreed that Chagas' disease was second (after malaria) in public health importance among vector-borne diseases in Honduras. Both welcomed the possibility of control measures.

6. Dr. Cosenza, Professor, Department of Microbiology. Dr. Cosenza, who considers himself an immunologist and biochemist, showed us the facilities which included an insectary. However, he stated that the Ponces were responsible for the work with the triatomines as well as for the field activities.

This information was presented orally to VBC and it was a subject of conversation at every meeting I have attended since.

On February 25, 1988, I wrote a letter to Mr. Robert Haladay, A.I.D. Health Officer in Tegucigalpa. He requested me to write to Rina Kaminsky who kindly visited him to assess his interest in the A.I.D. Mission of our proposed Chagas' control work. Following my letter, Mr. Haladay agreed with our visit to Honduras.

Note: During my membership of WHO/TDR/STAC and RSG (Scientific and Technical Advisory Committee and Research Strengthening Group), I became aware of the Chagas' disease control activities in Argentina and Brazil using two very practical and effective types of control measures developed in part under TDR sponsorship, and of the great interest and enthusiasm generated among the administrators of the program.

ANNEX 1 cont.

After my visit to Honduras in 1987, I attended the II Latin American Congress of Tropical Medicine in Bogotá and an opportunity to discuss this matter with Dr. Joao Pinto Dias, who knows the characteristics of Chagas' disease in Honduras and agreed that the insecticidal paint was better than the fumigant canister used in Argentina. He suggested using a French product; he sent me information regarding the French company, Artilin. Unfortunately, Artilin did not provide me with technical information nor samples of their product despite long distance phone calls and multiple efforts.

Dr. Lacey, also informed me of his unsuccessful attempts in obtaining Artilin materials and information. He then got in contact with Mr. Johnson of Envirochem who provided samples of Super IQ. Because of our interest in comparing Super IQ with the product used in Brazil, I contacted (through the PAHO office in Washington, Dr. Fernando Beltran) TDR in Geneva. Dr. Alvaro Moncayo, Secretary of the Steering Committee on Chagas' disease, explained to me that the product used in Brazil by Oliveira was not the French one, but a local formulation using malathion. Following a request by Dr. Moncayo, Dr. Oliveira very kindly sent his formulation to be assessed against Super IQ.

ANNEX 2

ITINERARY

June 8	Depart Washington, arrive in Cali (Dr. D'Alessandro arrived earlier on other business, and began preparation of substrates four days prior to arrival of Dr. Lacey).
June 9-14	Bioassays of insecticide-based paint at the International Center of Training and Medical Investigations (CIDEIM) in Cali
June 14	Depart Cali, arrive Bogotá
June 15	Depart Bogotá, arrive Tegucigalpa
June 16	Meeting with Dr. Pereira, Labortorio Nacional, Ministry of Health Drs. Carlos Ponce and Elisa de Ponce were unable to meet with us due to emergency TDY in Nicaragua.
June 17	Meeting with Dr. Jorge A. Rubio, Chief, Division of Vector Control (DCV), Dr. Luis A. Rivera, Chief of Entomology, DCV, and Mr. José R. Gomez, Chief of DCV Field Operations. Field visits to sites positive for Chagas' disease and T. dimidiata (Santa Lucia and Valle de los Angeles, within 20 km of Tegucigalpa).
June 18	Depart for Choloteca and to visit sites positive for Chagas' disease and R. prolixus in Las Tapias near San Marco de Colon.
June 19	Return to Tegucigalpa. Preparation and painting of walls used for long-term residual studies.
June 20	Preparation of report. Set up of bioassays on painted walls using field collected triatomines.
June 21	Continued preparation of report. Termination of bioassay. Briefings with Mr. Bob Haladay (Chief, HPN) Dr. Ernesto A. Pinto (Health Sector I), and Mr. Peter Kranstover (Project Planning Officer) of USAID.

ANNEX 2 cont.

June 22 Depart Tegucigalpa.

ANNEX 3

LABORATORY EVALUATION OF SUPER IQ INSECTICIDE-BASED PAINT

A commercially available formulation (Super IQ) of chlorpyriphos [0,0-diethyl 0-(3,5,6-trichloro-2-pyridyl) phosphorothioate] based paint was obtained for testing from Envirochem, Scottsdale, AZ. The paint composition is: pigment (clay) 0.5%; vehicle (vinyl acrylic emulsion, acrylic emulsion, water, glycols, and additives) 98.6%; and chlorpyriphos 0.9% with a pH of no more than 9.0 and a specific gravity of no more than 1.15 g/ml. The active ingredient, chlorpyrifos, is an organophosphate classified by WHO as moderately hazardous to mammals (LD_{50} (Rat) = 135 mg/kg). For comparative purposes, a sample of a slow release emulsifiable suspension (ES) (SRES-NPPN-1) containing 8% malathion was obtained from Dr. A.M. Oliveira Filho, Universidade Federal do Rio de Janeiro.

Five species of colonized triatomines were used for laboratory bioassays. Most of the tests were conducted with R. prolixus. Comparative assays were run with Dipetalogaster maximus (Uhler), T. infestans (Klug), T. leticularia (Stal), and T. pallidipennis (Stal). Rhodnius prolixus originating from a human dwelling in Choachi, Cundinamarca, Colombia were obtained from a colony maintained at least five years at the Universidad de Los Andes, Bogotá. The remaining four species were obtained from long maintained colonies at the Centro Internacional de Investigaciones Médicas (CIDEIM).

Substrates that were used for the evaluation of the chlorpyriphos-based paint (Super IQ white) were fired and unfired (adobe) bricks (10 cm x 20 cm x 6-7 cm) and squares of wood (20 cm x 20 cm x 2 cm). Only the upper surface of each substrate (broadest dimensions) was used for application of undercoatings and insecticide-based paint. Two days prior to application of the paint, two thirds of the bricks were painted with either a clay base wash or latex paint. The remainder were not given an undercoating. One half of the wood was undercoated with latex paint and the remainder was left natural. Two days after applications of undercoatings, the substrates were painted with a single coat of Super IQ and left for a minimum of 2 days prior to using for testing. Because of the variable porosity and roughness of the bricks, the amount of Super IQ required to evenly coat the surface ranged between 4.5 and 9 ml of paint (0.041-0.081 g A.I./brick or 2.03 - 4.1 g A.I./m^2). Smooth bricks and those previously receiving an undercoating required the least amount of Super IQ for thorough coverage (ca 5 ml/brick). The amount required for coverage of the wood was considerably less

ANNEX 3 cont.

variable, ranging from 4-5 ml/substrate (0.036 - 0.045 g A.I./ square of wood or 0.9 - 1.13 g A.I./m^2). The manufacture recommends application of primer before applying 1-2 coats of Super IQ. The product's spread rate to smooth prepared surfaces is 1000-1200 ft^2/gal (ca 25-30 m^2/liter). This would result in the application of ca 0.4-0.5 g A.I./m^2.

The malathion-based slow release ES was applied to the surface of fired bricks (no undercoating) at the rate of 2.5 g A.I./m^2 by using 0.64 ml of the ES/brick surface (200 cm^2). Due to the roughness and porosity of the bricks, each 0.64 ml of the ES required dilution with 15 ml of water in order to cover the upper surface of the bricks thoroughly and evenly.

The procedures of Nelson et al. (1983), modified in part, were used for evaluation of Super IQ. In preliminary bioassays, since no difference in insecticidal activity was observed between substrates that had been undercoated and those that had not, all bioassays except comparative tests with the malathion ES were conducted on undercoated substrates. Duration of adequate insecticidal exposure to Super IQ was determined for all three substrates using recently blood-fed (on chickens) fifth stage nymphs of R. prolixus. Plastic bioassay cones (9 cm diam.) obtained from the World Health Organization were fastened to the treated surface of the substrates with tape. Ten nymphs were gently placed onto each substrate through a hole in the top of the cone. Since the nymphs were unable to climb the sloping surface of the cone, the opening was left unplugged. Two replicates were used for each substrate type for each exposure period. One replicate of control nymphs was held on either untreated fired or unfired bricks or wood during each exposure period. After exposures of 2, 4, 8, 16 or 24 hours, the nymphs were removed from the cones using forceps and placed in filter paper bottom-lined plastic containers (230 ml, 6.5 cm x 6.8 cm diam.). One container was used for each replicate and control. The top of the containers were covered with organdy held securely in place with rubber bands. The nymphs were observed daily for five days following the termination of exposure. Mortality was noted and dead nymphs were removed each day. All tests and subsequent holding were at 27°C.

The effect of nymphal stage on insecticidal activity of Super IQ was determined using blood-fed first, fourth, and fifth stage nymphs. Exposures were made as in the previously described test, but on wood substrates only. First stage nymphs were exposed for 30 min., 1 and 4 hours. The older nymphs were only exposed for 4 hours. Procedures for post-treatment holding and

ANNEX 3 cont.

mortality determination were identical to those described above with the exception that first instars were held in Petri plates (100 mm diam. x 20 mm) on filter paper.

Susceptibilities of the aforementioned five species to an 8 hour exposure to the Super IQ was studied as in the exposure duration tests except that exposures were made on wood only and third instar D. maximus were used instead of fourth and/or fifth stage nymphs as with the other species. Third stage nymphs of D. maximus are comparable in size to fifth stage nymphs R. prolixus.

Comparative tests of the Super IQ and malathion slow release ES were made using fifth instar nymphs of R. prolixus exposed to treated fired bricks for 24 hours. As in the other tests, all treatments were replicated twice and mortality determined for 5 days post-treatment.

Results

The insecticidal activity of Super IQ on R. prolixus on the three substrates for variable exposure periods is presented in Table 1. A direct relationship between exposure period and mortality is apparent; no mortality was observed for the shortest exposure (2 hours) and near complete mortality was seen with the longest exposure (24 hour). The effect of substrate type on activity is most apparent in the 16 hour exposure. Although the highest insecticidal activity of Super IQ was seen on wood after 16 hours exposure, near complete mortality was also produced on fired bricks after 24 hours of exposure. The lowest insecticidal activity was see on treated adobe (unfired) bricks.

The effect of nymphal stage on susceptibility to Super IQ is presented in Table 2. A marked difference in susceptibility between first stage and older nymphs is apparent. It is also apparent from the data that a shorter period of exposure is required to produce mortality in the younger stages.

Data on the differential susceptibility of the five species tested is presented in Table 3. D. maximus, T. infestans, and T. leticularia were significantly more susceptible than T. pallidipennis and R. prolixus.

Initial mortality of fifth instar nymphs after 24 hours exposure to fired bricks treated with either Super IQ or malathion ES was 85% and 15%, respectively. Control mortality was nil. It should be noted that the dilution ratio used in our test was considerably higher than that recommended by Oliveira

ANNEX 3 cont.

(pers. comm.) and this could have some effect on efficacy. In this particular case, less dilution would have resulted in less even coverage of the brick surfaces.

Discussion

In most laboratory bioassays of candidate insecticides, the prolonged exposures that are utilized are unrealistic considering the duration of contact with treated surfaces under field conditions. Despite the fact that older nymphs of R. prolixus are not affected following fairly long exposures to Super IQ (4-8 hours), complete control of younger nymphs is possible after relatively brief exposure. Controlling only this segment of the vector population could result, over time, in a large reduction of the triatomine population in a dwelling and the interruption of disease transmission. Although flying adults can reinvade a cleared house, their numbers are somewhat limited and resulting offspring would be eliminated in the presnce of an efficacious and highly residual insecticide.

Rhodnius prolixus and T. infestans, because of their vector capacity and distribution, are two of the most important vectors of Chagas' disease in Central America/northern South America and the southern half of South America, respectively. Although older instars of R. prolixus were the least susceptible to an 8 hour exposure to Super IQ, mortality was complete in T. infestans before the exposure period had terminated. The other species tested are less important as vectors of human infection, but have all been found infected with T. cruzi Chagas in nature (Lent and Wygodzinski 1979).

Manufacturer literature and guarantees indicate that Super IQ is effective in controlling cockroaches and other household pests for at least 2 years when used according to manufacturer instructions. The high pH of the product and its various constituents apparently have not resulted in abbreviated insecticidal activity as have been observed with certain experimental formulations (Oliveira Filho 1984).

Undercoating increases ease of application of Super IQ and enables use of less of it on rough and porous surfaces. Undercoating, depending on material used, may also prolong residual activity by acting as a barrier between insecticide-based paints and mud walls. Periodic bioassays on the three substrates used in this study will be performed until insecticidal activity declines. Comparisons will also be made between adobe bricks with and without undercoating for possible influence on residual

ANNEX 3 cont.

activity. Data from our initial efficacy evaluations indicate that Super IQ warrants further evaluation under field conditions. Field trials of chlorpyriphos based slow release experimental formulations by Pinchin et al. (1978b) indicate prolonged activity that was greater than that of other insecticides, but the insecticidal activity was considerably reduced compared to laboratory bioassays of the same formulation.

Of the several interventions reviewed by Oliveira Filho (1984), Schofield (1985), and Schofield et al. (1987), only residual insecticides, housing improvements and public health education appear to offer the most promise for control of Chagas' disease in the forseeable future. Encouragement of community participation in the control of triatomid vectors will enhance the possibility of sustainable control using the various interventions and will reduce operational expenses (Pinto Dias and Borges Dias 1985). Considering the feeding sites of domestic and peridomestic triatomines, it may only be necessary to paint those rooms used for sleeping quarters. Partial painting of walls (only those portions the bugs cross over) may further reduce control costs. Based on the benefits of home improvement and prolonged control of vector and pest insects, commercially available insecticide-based paints offer good potential for use in programs that will utilize community participation for applying the paint. Health education at the village level will play a key role in maximizing the potential of such community based interventions.

Table 1. Effect of duration of exposure to Super IQ chlorpyriphos-based paint (0.9% A.I.) on mortality of fifth stage nymphs of *Rhodnius prolixus*

Substrate	Mean Percent Mortality[1] by Hours of Exposure				
	2	4	8	16	24
Fired brick	0	15	5	53	85
Unfired brick	5	-	0	30	-
Wood	5	17	20	100	-
Control	0	0	0	0	0

[1]Mortality 5 days after exposure.

Table 2. Effect of nymphal stage on susceptibility of *Rhodnius prolixus*, to Super IQ chlorpyrifos-based paint (0.9% A.I.) on wood substrate

Stage	Exposure	Mean % mortality
First	30 min.	40
	1 hr.	88
	4 hrs.	95
	Controls	20
Fourth	4 hrs.	0
Fifth	4 hrs.	17
	Controls	0

Table 3. Susceptibility of five species of triatomine bugs to Super IQ chlorpyrifos-based paint (0.9% A.I.) after 8 hours of exposure on wood (5 days post exposure)

Species	Stage	Mean % Mortality
Triatoma infestans	4-5	100
Controls		0
Triatoma pallidipennis	4	33
Controls		0
Triatoma leticularia	5	95
Controls		0
Dipetalogaster maximus	3	100
Controls		0
Rhodnius prolixus	5	20
Controls		0

ENVIROCHEM

SPECIFICATION

1. PRODUCT NAME

SUPER-IQ Insecticide All Purpose Transparent Coating (APT)
Semi-Gloss Finish
Interior/Exterior
EPA Reg. No. 49244-1

2. MANUFACTURER

Biochemico Dynamic Americas Corporation
P. O. Box 19307
Indianapolis, IN 46219
Phone: (317) 357-9680

3. PRODUCT DESCRIPTION

A. BASIC USES

Commercial, Industrial and Dwelling Premises: Exterior or interior structures of premises where protection against insect invasion is desired or required; such as exterior walls, concrete foundations, pilings, openings between walls and roofs, soffets, facias, doors, window frames, thresholds, concrete patios, refuse areas, garbage cans, trash dumpsters, dark and poorly accessible areas that are often infested with insects or contacted by insects.

Farms and Ranches: Exterior walls and supportive structures, barns, stables, stalls and outbuildings.

Mobile Homes: Exterior surfaces of buildings, particularly the bottom where there is often heavy insect invasions.

Gas and Electrical Industries: Equipment panels, meter boxes, housing, switchgear, etc.

B. SAFETY PRECAUTIONS

Keep container tightly and securely closed after each use. Do not take internally. Avoid skin and eye contact with product and inhalation of vapor. Keep out of reach of children. Do not reuse containers. Store in cool, dark place. Always mix product well before application to assure maximum homogeneity of the product.

C. LIMITATIONS

Do not use on metal surfaces without a suitable primer. Do not use on steps, walkways or other foot or vehicle traffic areas. Do not mix the product with ordinary paints, varnishes or other ordinary coating products. Do not dilute

SUPER IQ INSECTICIDE COATINGS

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with organic solvents. Efficacy of product may be severely damaged or destroyed by strong alkaline or acidic substances.

D. COMPOSITION AND MATERIALS

Pigment:		.50
Clay	100.00%	
Vehicle:		98.60
Vinyl Acrylic Emulsion	31.36	
Acrylic Emulsion	9.90	
Volatile Water	47.67	
Glycols	5.37	
Additives	5.70	
	100.00%	
Active Ingredient:		.90
Chlorpyrifos [0-0-Diethyl 0-(3,5,6-trichloro-2-pyridyl) phosphorothioate]	100.00%	
		100.00%

E. SIZES

One U.S. Quart (1.06 Litre)
One U.S. Gallon (3.785 Litre)
Five U.S. Gallon (18.925 Litre) containers

F. COLOR

SUPER-IQ APT is a white milky liquid suspension. It, however, produces a practically transparent coating after proper application and when dried. It is not to be tinted or colored with any colorants.

4. TECHNICAL DATA

Physical Status:	White milky homogenous liquid suspension
Odor:	Mild chemical
Viscosity:	No more than 90 K.U.
Specific Gravity:	No More than 1.15
PH:	No more than 9.0
Recommended Dry Film Thickness:	1 mil.
Flash Point:	Over 100 deg. C
Shelf Life:	Not less than 18 months

5. INSTALLATION

Surface Preparation: All surfaces must be clean, free from dirt, grease and oil, as well as loose scaly or crumbly old paint. Wipe off excessive dew and avoid painting when heavy dew or rain will strike before the paint has dried properly. Protected areas such as eaves should be scrubbed with a detergent to remove dirt and water soluable materials which accumulate and interfere with adhesion. Glossy surfaces must be sanded dull. Scrub mildrewed areas clean with a solution of household bleach to three quarts water followed with another detergent and three ounces of trisodium phosphate to one gallon of warm water. Wear eye protection and rubber gloves to avoid contact with the cleaning solutions. Thoroughly rinse treated areas and let dry before painting. In hot, windy weather dampen the exterior surface before painting.

Unpainted New Wood Surfaces: Counter sink nails and apply (oil base) white house paint primer on all woods. Caulk nail holes, joints and cracks. Follow with one or two coats of this finish. If the new wood contains knots or sap streaks remove

ANNEX 4 cont.

SUPER IQ T.M.

ART

INTERIOR-EXTERIOR
FLAT FINISH

FOR COMMERCIAL AND
INDUSTRIAL USE

ENVIROCHEM
13901 North 73rd Street
Suite 212
Scottsdale, AZ 85260
(602) 948-3842

Biochemico Dynamic Americas Corporation
Indianapolis, IN., U.S.A.

Product of U.S.A.

SUPER IQ T.M.

ART

INTERIOR-EXTERIOR
FLAT FINISH

NET CONTENTS — 1 GALLON

Keep Out of Reach of Children

CAUTION

See Back Panel for Additional Precautionary Statements

ANNEX 4 cont.

SUPER IQ T.M. APT

SUPER-IQ APT is an insecticide protective coating designed for use on walls, ceilings, woodwork, wallpaper, paneling, tiles and non-food contact surfaces in buildings such as offices, hotels, motels, theaters, sports arenas, factories, restaurants, domestic domiciles, and for use on farms, ranches and in dairy barns. It will control flies, mosquitoes, wasps, spiders, cockroaches, ants, moths, clover mites and gnats.

Active Ingredient:	
Chlorpyrilos [0-0-Diethyl 0-(3, 5, 6-trichloro-2-pyridyl) phosphorothioate]	0.90%
Inert Ingredients:	99.10%
TOTAL	100.00%

EPA Reg. No. 49244-1
EPA Est. No. 01990-MO-03

Biochemico Dynamic Americas Corporation
Indianapolis, Indiana

PRECAUTIONARY STATEMENTS
HAZARDS TO HUMANS AND DOMESTIC ANIMALS

CAUTION

Do not allow children or pets to contact treated surfaces until coating has dried. May be harmful if swallowed, inhaled or absorbed through skin. Avoid breathing vapors while paint is wet. Avoid contact with skin, eyes and clothing. Do not use in food areas of food handling establishments, restaurants or other locations where food is commercially prepared or processed. In non-commercial establishments do not apply to surfaces on which food will be prepared or served or in which animal feed will be served such as mangers and troughs.

NOTE TO PHYSICIANS: Chlorpyrilos is a cholinesterase inhibitor. Treat symptomatically. Atropine by injection is antidote of choice.

DIRECTIONS FOR USE

It is a violation of FEDERAL law to use this product in a manner inconsistent with its labeling.

Apply SUPER-IQ APT only on clean and dry surfaces.

MIX WELL BEFORE APPLICATION. Provide adequate ventilation of areas during application and while paint is drying.

Product may be applied with brush, roller or spray. It dries quickly and gives a beautiful finish. It is odorless when dried. Dried coat may be washed with water or water and mild non-akaline detergents.

Us in Domestic Domiciles: Apply to interior and exterior of window casings and thresholds. Coat a band of Super-IQ to baseboards and corners of walls. Band cabinets at base and corners where they join the walls. Coat underside and 3 sides of shelves and drawers. Do not coat surfaces where food may make contact. Each gallon covers approximately 450 to 500 sq. feet of surface. For best results, use without dilution and apply two coats.

STATEMENT OF PRACTICAL TREATMENT

If swallowed: Drink plenty of water. Call a physician or poison control center.
If inhaled: As with any freshly painted surface, get plenty of fresh air.
If in eyes: Flush with plenty of water. If irritation persists, get medical advice.
If on skin: Wash with plenty of water and soap.

ENVIRONMENTAL HAZARDS

This pesticide is toxic to fish, birds and other wildlife. Do not contaminate water by cleaning of equipment or disposal of wastes. Apply this product only as recommended on this label.

STORAGE AND DISPOSAL

Keep container tightly closed and store in a cool, dark place, preferably in a locked storage area.

Product Disposal: Partially filled can may be disposed of by securely wrapping original container in several layers of newspaper and discarding in trash.

Container Disposal: Do not reuse empty can. Wrap container and put in trash.

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with clear sanding sealer before priming. DO NOT APPLY THIS PRODUCT SELF-PRIMED.
Masonary: All new masonary surfaces should cure 60 days or be neutralized before before painting. Porous surfaces such as haydite or cinder blocks should be coated with latex block filler to give a sealed, smooth surface. Freshly poured concrete treated with form oils or curing compounds should be allowed to weather 2 to 3 years before painting. Finish with one or two coats of this product.
Metal: For iron or steel, prime with red metal primer or green equipment maintenance primer. Weathered galvanized metal with rust showing should be primed with gray metallic zinc. New galvanized surfaces should be weathered to a dull finish or treated with phosphoric acid solution and rinsed before painting. All peeling and scaly paint should be removed. Sand exposed area, clean and prime with proper metal primer. Following the appropriate treatment, all metal surfaces may be top coated with one or two coats of this product.
Previously painted Surfaces-Wood: For previously painted surfaces in good condition, clean the surface and apply one or two coats of this finish. Surfaces in poor condition need to be cleaned, wire brushed or scraped to a solid base. Spot prime bare wood with white house paint primer (oil base), then apply one or two coats of this finish.
Application: Apply SUPER-IQ APT with brush, roller, or spraying equipment. Make sure surfaces to be coated are clean and dry, primed or undercoated, if necessary, before applying the product. Keep premises well ventilated during application and until coating is thoroughly dried. Use suitable clothing and equipment to properly protect applicators from unnecessary and excessive skin contact and inhalation of vapor. Mix well before use.
Thinning: Use SUPER-IQ APT without any dilution or thinning.
Spread Rate: 2 coats 500-600 square feet per gallon.
Drying Time: To touch: 30 minutes; to recoat: 2-4 hours.
Equipment Clean Up: Use soap and water or detergent with water. Dry film may be dissolved and washed away with organic solvents such as acetone, xylene or mineral spirits.
Handling Precautions: Protect from freezing. Do not apply when surface, paint or air temperature is below 50. F (10 C). KEEP OUT OF THE REACH OF CHILDREN. Before using the product listed herein, carefully read each product label and follow directions for its use. Please read and observe all the warnings and precautionary information on the product labels. IMPORTANT: When applying coatings by spray equipment, observe all precautionary safety measures. Spray equipment must be handled with due care and in strict accordance with manufacturer's recommendations. If precautions are not taken, spraying of any material can be hazardous. When using or handling spray equipment, hoses and the like, observe all required safety practices. In addition, when spraying paint or coatings, wear respirator recommended for the product being handled. In all cases, wear protective eye equipment.

6. WARRANTY

SUPER-IQ Insecticide Coatings fully guarantees its products to meet its specifications, and will REPLACE THE MATERIALS OR REFUND THE FULL PURCHASE PRICE, where it is proven to be defective. With surfaces kept free of dirt and dust, and correct application, SUPER-IQ's carefully formulated insecticide feature will remain effective for A MINIMUM OF TWO YEARS. Biochemico Dynamic Americas Corporation will not be liable if the product is used other than as specified on the label, makes no warranties other than those set forth above, and DISCLAIMS ALL WARRANTIES OF MERCHANTIBILITY AND FITNESS FOR PARTICULAR PURPOSE.

BD/134D/85 Specification APT Semi-Gloss /3 of 3
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ANNEX 5

REGISTRO DE EXPERIMENTOS USANDO PINTURA-INSECTICIDA EN D.C.V., TEGUCIGALPA

SUBSTRATO	NUMERO DE CHINCHES MUERTAS DESPUES DE 24 HRS DE EXP. (EN HORAS)					
	Nº INICIAL DE MUERTAS	24 HS.	48 HS.	72 HS.	96 HS.	120 HS
FECHA DE EXPOSICION CON PINTURA-INSECTI.						
-CEMENTO						
-MADERA						
-MUEBLE						
-LADRILLO COCIDO						
-LADRILLO CRUDO						
SIN PINTURA-INSECTI. CONTROL						
FECHA DE EXPOSICION CON PINTURA-INSECTI.						
-CEMENTO						
-MADERA						
-MUEBLE						
-LADRILLO COCIDO						
-LADRILLO CRUDO						
SIN PINTURA-INSECTI. CONTROL						
FECHA DE EXPOSICION CON PINTURA-INSECTI.						
-CEMENTO						
-MADERA						
-MUEBLE						
-LADRILLO COCIDO						
-LADRILLO CRUDO						
SIN PINTURA-INSECTI. CONTROL						
FECHA DE EXPOSICION CON PINTURA-INSECTI.						
-CEMENTO						
-MADERA						
-MUEBLE						
-LADRILLO COCIDO						
-LADRILLO CRUDO						
SIN PINTURA-INSECTI. CONTROL						

ANNEX 6

PROTOCOL, IN SPANISH, FOR PHASE I

Fase I - Actividad residual del Super IQ en varios tipos de superficies.

Objetivos: Determinar la actividad insecticida inicial y residual del Super IQ blanco aplicado en varios tipos de superficies usando triatominos capturados en casas y vecindades o provenientes de colonias de R. prolixus y T. dimidiata. Dentro de lo posible, los ensayos se deberían conducir a intérvalos mensuales hasta que la actividad del insecticida deje de demostrarse.

Método: Seleccionar una superficie (pared, etc.) para ser pintada en un medio domestico o simulando un medio domestico (por ejemplo, paredes y estanteria en las oficinas de la División de Control de Vectores). Las superficies no deben estar en ambientes con aire acondicionado, o donde puedan mojarse o exponerse directamente a los rayos solares. Las superficies deben limpiarse para eliminar polvo, particulas, telas de araña, etc., y cada vez que sea posible se seguirán las instrucciones de los fabricantes del producto. Las paredes de barro deben prepararse con una o dos manos de cal o de otro material, de los que normalmente usan los habitantes del lugar. Despues que las superficies esten completamente secas, aplicar una mano uniforme de Super IQ blanco. De acuerdo con la superfici (más humedad inicial más tiempo para secar), comenzar con los bioensayos iniciales 24 o 48 horas después de aplicar el Super IQ usando ninfas de quinto estadio de R. prolixus, y si es posible, de T. dimidiata.

El bioensayo se realiza asi:

Asegurar los conos de WHO con cinta adhesiva sobre la superficie a ensayar. Colocar 10 ninfas de qunto estadio en cada cono. El cono debe quedar firmemente asegurado. Si la superficie es irregular, llene los espacios con algodón y luego aplique la cinta adhesiva. Las ninfas no deben ponerse en contacto con la superficie pegajosa de la cinta. Si hay menos de 10 ninfas por cada cono anote el número de ninfas usado en una cinta y adhierala sobre el cono.

Despues de 24 horas de exposición retire las ninfas de los conos anotando el numero de chinches muertes y coloque las ninfas vivas en un envase tipo copa y anote la información en el formulario. Debe haber un trozo de papel en el fondo del envase

ANNEX 6 cont.

para que absorba los liquidos y defecaciones de los triatominos. Cada envase debe marcarse con la siguiente información:

Fecha de la exposición de los triatominos, nombre de la especie usada, numero de insectos y estadio de los mismos, (el quinto estadio se conoce rapidamente por tener los muñones de las alas muy visibles), la superficie en la que se ensanyó la pintura-insecticida (pared de ladrillos cocidos, adobe, madera, etc.).

También anote esta información en la hoja de registro que está en poder del Dr. Rivera (Anexo _). Cada dia, por cinco dias, anotar en el formulario la mortalidad encontrada. Después de contar los insectos, los muertos se eliminan del envase de observación. Se calcula el porcentaje de triatominos muertos al final de los cinco dias de observación.

Si es posible, la prueba deberia conducirse con ninfas de quinto estadio, tanto de R. prolixus como de T. dimidiata.