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THE CAT FLEA, <u>CTENOCEPHALIDES</u> <u>FELIS</u>, AS A POTENTIAL VECTOR OF <u>TRYPANOSOMA LEWISI</u>

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A Thesis

Presented to the Department of Zoology and Entomology Brigham Young University

In Partial Fulfillment of the Requirements for the Degree Master of Science

> by Robert P. Belihar August, 1968



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This thesis by Robert P. Belihar is accepted in its present form by the Department of Zoology and Entomology of Brigham Young University as satisfying the thesis requirement for the degree of Master of Science.

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Typed by Audrey Wittwer

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INTRODUCTION AND LITERATURE REVIEW

The first hemoflagellate was discovered by Valentine in 1841; two years later Gruby established the genus <u>Trypanosoma</u> for a flagellate in the blood of frogs, and in 1879 Lewis in India described <u>Try-</u> panosoma lewisi Kent of the rat (Belding, 1965:141).

Considerable attention has been given to T. lewisi because of its close relationship to those species of trypanosomes pathogenic to man--T. gambiense, T. rhodesiense, and T. cruzi. Trypanosoma lewisi is frequently used in studies of trypanosomes because it is so easily maintained in the laboratory. Although it is a parasite primarily of rats, it has been found in guinea pigs (Nieschulz and Wawo-roentoe, 1919), mice (Bruynoghe and Vassiliadis, 1929), rabbits (Garnham and Lewis, 1959), and rarely in humans (Johnson, 1933). Trypanosoma lewisi is generally described as being non-pathogenic in rats, but it has been shown to cause death (Jurgens, 1902; MacNeal and Novy, 1903). Transmission takes place through the bites of fleas, and the following species have been shown to serve as experimental vectors (Steinhaus, 1967): Ceratophyllus lucifer, Ceratophyllus hirundinis, Ceratophyllus fasciatus, Pulex brasiliensis, Pulex irritans, Xenopsylla cheopis, Ctenocephalides canis, Leptopsylla segnis, and Ctenophthalmus agyrtes. The rat louse, Hematopinus spinulosus, may serve as a mechanical vector to rats, but apparently no developmental forms are seen in this insect as in the case of the flea. Trypanosoma lewisi occurs throughout the world, but the number of rats infected in each area varies greatly.

Lewis (1879) in his original research on <u>T. lewisi</u> in India found 29% of the rats infected. Some examples of the cosmopolitan distribution and frequency of occurrence of <u>T. lewisi</u> in rats are: London, 25% (Crookshank, 1886); Bombay, 12% (Carter, 1887); Berlin, 42% (Rabinowitsch and Kempner, 1899); Mexico, 3.6% (Beltran and Perez, 1947); United States, 13.2% (Eyles, 1952); Panama, 4.5% (Calero, 1952); and Puerto Rico, 5% (Fox and Thillet, 1962).

The first published experiments on the transmission of T. lewisi by fleas were performed by Rabinowitsch and Kempner (1899). They observed that three uninfected rats placed with others harboring trypanosomes in their blood subsequently became infected after 11-15 days. Fleas of an unidentified species were found on these rats. The fleas were removed from the bodies of the infected rats and inoculated into uninfected animals. Five of the nine uninfected rats became infected with T. lewisi. In another experiment Rabinowitsch and Kempner removed 20 fleas from an infected rat and placed them on an uninfected rat which became infected in two to three weeks. In four other experments, lice of an unknown species were removed from the rats harboring T. lewisi. These lice were inoculated into uninfected rats, but the results were negative. Wasielewski and Senn (1900) and Laveran and Mesnil (1901) substantiated the findings of Rabinowitsch and Kempner. MacNeal and Novy (1903) observed living T. lewisi in the stomachs of lice of an unidentified species. MacNeal (1904) reported that in one experiment several infected lice (with trypanosomes observed in their stomachs) were transferred to an uninfected rat with positive results---T. lewisi appeared in the peripheral blood after 14 days and persisted

for five weeks. He did not observe developmental forms, and concluded that the louse serves merely as a mechanical carrier of the trypanosomes from one rat to another. Prowazek (1905) described the development of T. lewisi in the rat louse, Hematopinus spinulosus, but failed to effect transmission from rat to rat by means of the lice. He believed that the lice could transmit despite his observations that they did not. He probably thought they were capable of transmitting on the basis of the developmental forms he had described. Nuttall (1909) experimented with the transmission of T. lewisi by fleas and lice. Four experiments with Ceratophyllus fasciatus as a vector were performed. In experiment 1, fleas from a wild rat infected with T. lewisi were placed on an uninfected white rat. The blood remained negative until the seventh day when T. lewisi was found. In experiment 2, the white rat of experiment 1 was killed four days after its blood was found positive. One fleat was recovered and placed on another laboratory rat, but no infection occurred. In experiment 3, ten infected fleas were placed on a laboratory rat. The peripheral blood remained negative until the eleventh day when trypanosomes were found. In experiment 4, three fleas from an infected wild rat were placed on a laboratory rat; the blood tested positive after ten days. Nuttall concluded that because of the ease with which infection occurred through the agency of fleas, they must be the chief vectors of the trypanosomes. In experiments dealing with lice, Nuttall demonstrated that lice were capable of transmitting T. lewisi from diseased to healthy rats. He examined large numbers of lice in order to observe development of the trypanosomes as described by Prowazek in 1905, but without success. He concluded that Prowazek

was probably deceived by extraneous flagellates. Strickland (1909) stated that Prowazek had most likely described a typical Crithidia species natural to the louse. Strickland could find no developmental forms of T. lewisi in either fresh or stained preparations of the gut contents from Hematopinus spinulosus fed on uninfected rats. Similar preparations made from lice fed on infected rats demonstrated T. lewisi in an unchanged morphological form in various parts of the alimentary tract. Examination of the gut of fleas belonging to the species Ctenophthalmus agyrtes revealed no trypanosomes despite their having fed on infected rats. Crithidia ctenophthalmi were observed in the gut of Ctenophthalmus agyrtes. Minchin and Thomson(1911) infected rats with T. lewisi by feeding them crushed fleas containing the trypanosome parasite, but conducted other experiments which led them to conclude that in nature, rats do not commonly become infected by eating fleas. The usual method is by the regurgitation of mature trypanosomes from the stomach of the flea into its proboscis, and then into the host's blood. Strickland(1911), however, performed experiments in which rats became infected largely by eating the fleas. The cycle of T. lewisi in the flea has been studied by many workers, but those of Minchin and Thomson in 1915 are probably the most detailed (Steinhaus, 1967:479).

The cat flea was described by Bouche in 1835, and 95 years later the genus <u>Ctenocephalides</u> was established by Stiles and Collins (Hubbard, 1947:60). The cat flea closely resembles the dog flea, <u>C</u>. <u>canis</u>, and although generally considered as a parasite of dogs and cats, it has been found on rats (Hicks, 1927), monkeys (Boero, 1945), mongooses (Haas, 1966), and numerous other mammals including man. The flea is

largely confined to its host inasmuch as 5-12 hours of feeding are needed before oviposition may occur; however, the fleas may at times leave their host (Belding, 1965:948). Ctenocephalides felis (Bouche) has a cosmopolitan distribution, although it prefers the temperate zones. Some representative areas of distribution and the frequency with which C. felis has been found on rats are: Shanghai, 1.2% (Hicks, 1927); Virginia, 0.6% (Hasseltine, 1929); Virgin Islands, 0.2% (Carnes, 1931); Puerto Rico, 1.7% (Fox, 1952); and Senegal, 3.6% (Boiron, 1952). The cat flea is capable of experimentally transmitting organisms such as the virus of Mexican typhus fever (Mooser and Castaneda, 1932), the nematode Dirofilaria immitis (Summers, 1940), the causative agent of murine typhus, Rickettsia mooseri (Irons, Bohls, Thurman and MacGregor, 1944), the causative agent of bubonic plague, Pasteurella pestis (Sapre, 1945), Brucella sp. (Tovar, 1947), and the tapeworms Hymenolepis diminuta and Dipylidium caninum (Belding 1965:799). No evidence that C. felis serves as a vector of trypanosomes was found in the literature. However, unknown species of Crithidia closely resembling trypanosomes have been observed in the alimentary tract of C. felis.

The principle objective of this study was to determine whether the cat flea, <u>Ctenocephalides felis</u>, is a potential vector of the blood flagellate, <u>Trypanosoma lewisi</u>, in rats. Included as supportive parts of this study were surveys of the rat population in San Juan, Puerto Rico, to determine the incidence of <u>C</u>. <u>felis</u> and <u>T</u>. <u>lewisi</u>. The reproductive potential of <u>C</u>. <u>felis</u> on laboratory rats was also investigated.

MATERIALS AND METHODS

Ectoparasite Survey

To provide specimens for determining the incidence of <u>Trypano-</u> <u>soma lewisi</u> and <u>Ctenocephalides felis</u> in the rat population, trapping techniques similar to those described by Fox and Garcia-Moll (1961) were used. Rats were captured in wooden box traps baited with bread. Traps were placed in and around homes in the areas of La Perla and Santurce near San Juan, Puerto Rico for periods of 24 hours, and then were taken to the laboratory.

In the laboratory each trap containing a rat was placed into a large, cloth bag which was put into a 50-gallon garbage can; 50 ml of commercial ether was added and the can was then tightly sealed. After anesthetization for two minutes, the rat was removed from the can and trap, placed into a smaller cloth bag, and put into a small metal canister with more ether. The trap and the cloth bag which contained it were shaken over a newspaper. The material was collected in a Petri dish and examined for ectoparasites under the microscope. After careful examination under a bright light to find ectoparasites, the rat was vigorously combed until much of its hair was removed. The hair was then placed into another Petri dish containing a detergent solution which caused many of the ectoparasites to detach from the hair. Those obtained using these methods were then identified. Small specimens were mounted on microslides for identification, and larger ones were identified under the dissecting scope.

Trypanosome Survey

To determine the presence of <u>Trypanosoma lewisi</u> in rats, both wet mounts and stained smears of peripheral blood were examined microscopically for parasites. Rats which had been trapped in the field were anesthetized for two minutes, as previously described, and blood samples were taken from the tail after snipping off the tip with a pair of scissors. Wet mounts were prepared by placing one or two drops of blood on a glass microslide, adding one or two drops of normal saline, and covering the mixture with a coverslip. The preparation was microscopically examined immediately for moving trypanosomes.

Permanent smears were prepared by spreading blood over a clean microslide in both thick and thin films for subsequent examinations. Thin films were treated as follows: (1) The preparation was fixed by immersing the slide in or applying absolute methyl alcohol for one minute, then allowing the slide to dry. (2) The slide was then immersed in or flooded with a dilute stain(0.5 to 1.0 cc stock Giemsa's and 10 cc distilled or buffered water, pH7) for 20 to 30 minutes. (3) The slide finally was washed in distilled or buffered water, and then allowed to dry at room temperature.

Thick smears were handled as follows: (1) The portion of the slide bearing the smear was immersed vertically in a solution of one part stock Giemsa's stain to 30 parts of distilled or buffered water for 25 minutes, or in a solution of one part stock Giemsa to 50 parts water for 45 minutes. (2) The slide was then removed, washed in distilled or buffered water for a sufficient time to clear the blood and differentiate the parasite colors, and allowed to dry. Washing time



varied from several dips to a five minute immersion, depending on the age and thickness of the smears. Older and thicker smears required longer washing. As a general rule, however, one minute was enough.

Trypanosome Cultures

An infected specimen of the species <u>Rattus norvegicus</u>, which had been trapped in the field, was the source for trypanosomes used in starting the cultures. The infected rat was anesthetized and its tail placed under a lamp for two to three minutes. The tip of the tail was then cut off, 20 drops of blood were placed into a Petri dish containing 1 cc of 2% sodium citrate solution, and 5 cc of physiological saline solution were added. The solutions were mixed by rotating the dish. Two to 3 cc of the resulting mixture was injected intraperitoneally into each of three white laboratory rats (<u>Rattus norvegicus</u>) which were used as a ready source of trypanosomes for subsequent transmission studies. This same procedure was repeated each week, except that an infected laboratory rat was used as the source of trypanosomes.

Flea Colony

In order to have an adequate, readily available supply of <u>Cteno-</u> <u>cephalides felis</u> for use in transmission studies, a colony was maintained within the laboratory. A revue of the literature did not reveal any acceptable, productive means of raising <u>C</u>. <u>felis</u> in the laboratory. Smith and Eddy (1954) described a method for raising <u>C</u>. <u>felis</u> using a dog as the host. Although relatively productive, their method required considerable space to house the dog in a small animal yard, and the



colony of fleas subsequently was difficult to control. It was necessary, therefore, to develop a technique whereby the fleas could be raised in a small area, and yet be produced in sufficient numbers. After experimenting with several arrangements the following method was used.

A small cat was placed into a wire cage which was 30 inches square, 18 inches high, and covered on all sides by one-quarter inch, galvanized wire mesh. A large, flat metal pan which could be removed for cleaning was placed beneath the cage. A smaller metal pan containing sand for the cat's excreta was put into the cage. Twenty specimens of C. felis (ten males and ten females) obtained from domestic cats in the San Juan area were used to start the colony. The eggs laid by the adult fleas fell onto the metal pan beneath the cage and were collected daily. They were placed into a container containing a mixture of 20 parts sterilized sand and one part food. The food, especially designed for flea larvae, was composed of 100 parts pulverized Gaines dog meal, 15 parts dried beef blood, and 10 parts dried powdered yeast. Pupation occurred in the sand-food mixture which was sifted daily to remove the pupae, which were subsequently placed into a 1000 cc Erlenmeyer flask where hatching occurred. The newly hatched fleas were used in the transmission studies with T. lewisi.

Transmission Experiments

Rats used in transmission studies were housed in special containers similar to those described by Fox, Bayona, and Rivera (1964). These consist of cylindrical, wire, test-tube baskets (five inches in diameter and six inches high) placed inside of cylindrical, glass,



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animal jars (eight inches in diameter and eight inches high) which contain a one-half inch layer of the sand-food mixture. The top of the glass jar was covered with wire mesh to prevent the rat's escape. A glass Petri dish lined with a paper towel was placed beneath the wire basket to catch the excreta. Experimental rats were fed a commercial preparation, and a water bottle was provided for one hour each day.

Four different types of transmission studies were conducted. (1) A rat which had been infected with <u>Trypanosoma lewisi</u> one week previously was placed into a container, and 50 newly-emerged <u>Cteno-</u> <u>cephalides felis</u> fleas were added. Infection of the host was confirmed by examination of blood taken from its tail. Ten days after the fleas had been placed on the rat they were killed, and smears were made of their gastrointestinal tracts. The smears were stained with Giemsa's stain and examined for the developmental forms of T. lewisi.

(2) An infected rat and a non-infected rat were placed together into a container, and 25 newly-emerged adult fleas added. After a tenday period, the peripheral blood of the non-infected rat was examined daily for parasites.

(3) A non-infected rat was placed into a container, and 25 newlyemerged infected fleas were added. Each day after the ten-day waiting period, the peripheral blood was examined for trypanosomes.

(4) Five infected fleas were homogenized in a 2 ml tissue homogenizer containing 1 ml of physiological saline. The solution was then transferred to a syringe and injected through a tube into the stomach of a non-infected rat. Wet mounts and stained smears of the peripheral blood were made each day following a ten-day waiting period,

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and examined for parasites.
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Reproductive Potential Experiment

The ability of the cat flea, Ctenocephalides felis, to successfully reproduce and colonize on a white laboratory rat, Rattus norvegicus, was determined in a series of experiments. The techniques used were similar to those in a study performed by Fox, Bayona, and Rivera (1964) in which they determined the reproductive potential of the oriental rat flea, Xenopsylla cheopis, on the white rat. In my experiments the container described previously was used to-house the rat. Five hundred milliliters of the sand-food mixture was placed into the bottom of the glass, animal jar. Four containers were used, each containing one rat. Two rats each received one male and one female flea, one received five males and five females, and the fourth received ten males and ten females. Each month for the next three months the larvae, pupae, and adults in each colony were counted. The rat was removed from the container, anesthetized, and examined thoroughly for adult fleas. The sand-food mixture was sifted through a wire strainer to find any pupae, and the mixture examined under a dissecting scope for larvae and eggs. The fleas used in these experiments were obtained from the regular colony maintained in the laboratory.



RESULTS

Ectoparasite Survey

A total of 225 rats, 206 <u>Rattus norvegicus</u> and 19 <u>Rattus rattus</u>, were examined during a seven-month period extending from June to December, 1964. One percent of the animals were infested with the cat flea, <u>Ctenocephalides felis</u>. Other ectoparasites encountered in the survey and the percentage of rats infested by each are: <u>Xenopsylla cheopis</u> (a flea) 14%; <u>Polyplax spinulosa</u> (a louse) 11%; <u>Listrophoroides expansus</u> (a mite) 6%; <u>Ornithonyssus bacoti</u> (a mite) 13%; <u>Rhipicephalus</u> sanguineus (a tick) 9%; and Ornithodoros puertoricensis (a tick) 7%.

Trypanosome Survey

Twenty-three (11.2%) <u>Rattus norvegicus</u> and two (10.5%) <u>Rattus</u> <u>rattus</u> were found infected with the blood flagellate, <u>Trypanosoma lewisi</u>. In the sub-standard housing area of La Perla, 26% of the rats were infected with <u>T. lewisi</u>, whereas in Santurce, a more modern portion of the city, only 2% carried the parasite.

Transmission Experiments

Transmission experiments that were conducted in four phases yielded the following results. (1) Thirty-seven of fifty non-infected fleas allowed to feed on an infected rat possessed developmental forms of <u>T</u>. <u>lewisi</u> in their gastrointestinal tract at the end of a 10-day period. (2) Seventy percent of these fleas transmitted <u>T</u>. <u>lewisi</u> from an infected rat to a non-infected rat. (3) Infection of non-infected rats using infected fleas occurred on eight of ten occasions. (4) Feeding non-infected rats homogenized infected fleas resulted in a 60% infestation efficiency.

Reproductive Potential Experiment

Over a three-month period, eggs, larvae, and pupae were not seen in any of the four colonies. At the end of the first month, adult fleas were found in only three colonies--one adult in the colony started with one pair, two in the colony started with five pairs, and three in the colony started with ten pairs. After two months, the only adult flea found was on the rat initially infested with ten pairs. No adults were found in any colony after three months.

DISCUSSION

Ectoparasite Survey

Data from this survey were compared with those of a similar study performed by Fox and Garcia-Moll (1961). The same localities were involved in both instances, and approximately the same numbers of rats were examined. The months during which the surveys were conducted also coincided. The study by Fox and Garcia-Moll covered a four-year period from 1957 through 1960, but only data obtained from June to December, 1957 were used for comparison.

The only ectoparasites that increased in number during the seven year period were fleas referrable to <u>Xenopsylla cheopis</u>--8% of the rats were infested in 1957, 14% in 1964. The rate of infestation for all other ectoparasites remained constant or decreased during this period. Those which decreased were the mite <u>Laelaps nuttalli</u> (87% in 1957, 84% in 1964), the mite <u>Listrophoroides expansus</u> (17% in 1957, 6% in 1964), the mite <u>Ornithonyssus bacoti</u> (25% in 1957, 13% in 1964), the louse <u>Polyplax spinulosa</u> (23% in 1957, 11% in 1964), and the tick <u>Rhipicephalus sanguineous</u> (36% in 1957, 9% in 1964). Those ectoparasites whose rates of infestation remained constant were the flea <u>Ctenocephalides</u> <u>felis</u> (1% in 1957 and 1964), and the tick <u>Ornithodoros puertoricensis</u> (7% in 1957 and 1964). An inverse relationship apparently exists between <u>X</u>. <u>cheopis</u> and other ectoparasites--when the former increases in abundance, the latter decreases. This same observation was made by Fox and Garcia-Moll (1961). They found that the inverse relationship between X. cheopis and L. nuttalli was particularly remarkable.

These changes in the ectoparasite population are difficult to explain on the basis of pest control measures. According to Fox and Garcia-Moll (1961), X. cheopis decreased from an infestation percentage of 54% in 1946-47 to 2.6% in 1959-60 without the influence of rat or flea control measures. The percentages of infestation of other ectoparasites increased during this 13 year period. Weather bureau records indicate that precipitation, humidity, and temperature varied only slightly from year to year, so these likely would not account for the rat flea reduction. Further studies on symbiosis and other ecological aspects of arthropods, particularly of rat fleas, are necessary to determine why the ectoparasite population varies from year to year.

Trypanosome Survey

The data of this study were compared with those obtained during 1957 and 1958 in a trypanosome survey performed by Fox and Thillet (1962). Rats of the same species were examined in 1957 and 1964 studies. From 1957 to 1964 there was a three-fold increase in the percentage of rats infected with <u>Trypanosoma lewisi</u> (3% in 1957, 11% in 1964). The infection percentage of 11% found in my survey is significantly above the 5% figure indicated by Fox and Thillet (1962). The infection percentage for Santurce, a modern area of the city, was the same (2%) for both surveys, whereas in La Perla, a slum area, a fourfold increase occurred in the percentage of rats infected (6% in 1957, 26% in 1964). This increase in number of rats infected with T. lewisi

may be attributed to the increase in the percentage of rats infested with <u>Xenopsylla cheopis</u>. In La Perla the percentage of rats infested with <u>X. cheopis</u> increased more than two-fold (13% in 1957, 30% in 1964). Slightly fewer rats of Santurce were infested with fleas in 1964 (5% in 1957, 4% in 1964). No other ectoparasite besides <u>X. cheopis</u> increased in frequency during the seven year interim period including <u>Ctenocephalides felis</u>. Fox and Thillet (1962) found that the percentage of rats infected with <u>T. lewisi</u> varied in direct proportion with the percentage of rats infested with <u>X. cheopis</u> in particular sections of the city.

Transmission Experiments

Experiments of this study have shown that <u>Ctenocephalides felis</u> can serve as a vector of <u>Trypanosoma lewisi</u>. The crithidial forms observed in the gastrointestinal tracts of infected fleas were probably those of <u>T</u>. <u>lewisi</u> and not extraneous flagellates, inasmuch as newlyemerged uninfected fleas did not possess these forms. Infection of non-infected rats which resulted from feeding them homogenized, infected fleas demonstrated that infection in nature may take place by ingestion of infected fleas by the rat. However, this does not preclude the possibility that infection occurs from the bite as well. Because of the evidence supporting <u>C</u>. <u>felis</u> as a capable experimental vector of <u>T</u>. <u>lewisi</u>, its ability to transmit trypanosome species pathogenic to man should be investigated.

Reproductive Potential Experiment

It is apparent that Ctenocephalides felis does not reproduce well on laboratory specimens of Rattus norvegicus. At the end of three months, C. felis could not be found where a colony had been started with ten pairs of adults. In a series of experiments dealing with the reproductive potential of Xenopsylla cheopis on laboratory rats. Fox, Bayona, and Rivera (1964) found that at the end of three months a colony begun with ten pairs of X. cheopis increased to 22,216 larvae, pupae, and adults. The reproductive potentials of these two fleas are strikingly different. Newly-emerged C. felis fleas actively fed on the shaved skin of white rats, but no signs of oviposition were ever observed in a colony started with cat fleas on a rat host. Several dead, adult fleas were found in the sand-food mixture when it was screened and examined at the end of each month. From these observations it is concluded that C. felis probably does not colonize on rats in nature, but is found only as an accidental parasite. There are several ways in which rats may become infested with C. felis in nature. (1) C. felis is known to leave its host on occasion (Belding, 1965:948) and may at this time find its way to a rat host. (2) Newly-emerged cat fleas may be exposed to the rat as their first available host. (3) Rats are often seen around dead cats and dogs and may at this time come in contact with the adult fleas. A review of the literature showed that C. felis is rarely found on more than 1-2% of the rats that have been examined in various localities throughout the world. I feel that this incidence is easily explained on the basis of the manner of infection of the rat by the three methods just described. Despite the fact that

<u>C. felis</u> has been shown to be a capable vector of <u>T. lewisi</u>, in view of the evidence available it is doubtful that it contributes significantly to its transmission in the field. There is little doubt that in Puerto Rico, <u>X. cheopis</u>, the only other proven vector of <u>T. lewisi</u>, is predominately responsible for the spread of <u>T. lewisi</u> in the native rat population. SUMMARY

The principle objective of this study was to determine whether the cat flea, <u>Ctenocephalides felis</u>, is a potential vector of the blood flagellate, <u>Trypanosoma lewisi</u>, in rats. Included as parts of this study were surveys of the native rat population in San Juan, Puerto Rico, to determine the incidence of <u>C</u>. <u>felis</u> and <u>T</u>. <u>lewisi</u>. Rats used in these surveys were captured in live-catch traps and taken to the laboratory where ectoparasites were removed and identified, and the rat's blood examined for trypanosomes. The reproductive potential of <u>C</u>. <u>felis</u> on laboratory rats was also investigated. Colonies of <u>C</u>. <u>felis</u> and <u>T</u>. <u>lewisi</u> were maintained in the laboratory to provide specimens for use in transmission experiments.

One percent of 225 rats examined were infested with <u>C</u>. <u>felis</u>. Other ectoparasites encountered were <u>Xenopsylla cheopis</u> (a flea) 14%; <u>Polyplax spinulosa</u> (a louse) 11%; <u>Listrophoroides expansus</u> (a mite) 6%; <u>Ornithonyssus bacoti</u> (a mite) 13%; <u>Rhipicephalus sanguineus</u> (a tick) 9%; and <u>Ornithodoros puertoricensis</u> (a tick) 7%. Comparison of these data with those of a similar study performed seven years earlier by Fox and Garcia-Moll (1961) revealed that only <u>X</u>. <u>cheopis</u> increased in frequency during the period between 1957 and 1964. The percentages for the other ectoparasites remained unchanged or decreased.

Eleven percent of the rats examined were infected with <u>T</u>. <u>lewisi</u>. In the substandard housing area of La Perla, 26% of the rats were infected with T. lewisi, whereas in Santurce, a more modern portion of the city, only 2% carried the trypanosome. The infection percentage of 11% found in my survey is significantly above the 5% figure indicated by Fox and Thillet (1962). The increase in the number of rats infected may be attributed to the increase in percentage of rats infested with Xenopsylla cheopis.

Experiments of this study show that <u>C</u>. <u>felis</u> can serve as a vector of <u>T</u>. <u>lewisi</u>. The crithidial forms of the parasite were seen in the gastrointestinal tracts of infected fleas, and successful transmission of the parasite from infected to non-infected rats using <u>C</u>. <u>felis</u> as a vector was accomplished on many occasions. Infection of non-infected rats which resulted from feeding them homogenized, infected fleas demonstrated that infection in nature may occur via ingestion of infected fleas. On the basis of these observations the ability of <u>C</u>. <u>felis</u> to transmit species of trypanosomes pathogenic to man should be investigated.

Reproductive potential experiments indicated that <u>C</u>. <u>felis</u> does not reproduce well on laboratory specimens of <u>Rattus norvegicus</u>. At the end of three months <u>C</u>. <u>felis</u> could not be found where a colony had been started with ten pairs of adults. Newly hatched fleas actively fed on the shaved skin of white rats, but no signs of oviposition were ever observed. From these observations it is concluded that <u>C</u>. <u>felis</u> does not do well on rats in nature, but is found only as an accidental parasite. Despite the fact that <u>C</u>. <u>felis</u> was shown to be a capable vector of <u>T</u>. <u>lewisi</u>, it is doubtful that it contributes significantly to its transmission in nature. In Puerto Rico, <u>Xenopsylla cheopis</u>, the only other proven vector of <u>T</u>. <u>lewisi</u>, is predominately responsible for the spread of this parasite in the rat population.

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ABSTRACT

The primary objective of this study was to determine whether the cat flea, <u>Ctenocephalides felis</u>, is a potential vector of the blood flagellate, <u>Trypanosoma lewisi</u>, in rats. Included as parts of this study are surveys of the rat population in Puerto Rico to determine the incidence of <u>C</u>. <u>felis</u> and <u>T</u>. <u>lewisi</u>. The reproductive potential of <u>C</u>. <u>felis</u> on laboratory rats was also investigated. Colonies of <u>C</u>. <u>felis</u> and <u>T</u>. <u>lewisi</u> were maintained in the laboratory to provide specimens for use in transmission experiments.

One percent of the 225 rats examined were infested with <u>C</u>. <u>felis</u>. The incidences of the other ectoparasites encountered were also determined. Comparison of the ectoparasite survey data with those of a similar study made seven years previously revealed that only the rat flea, <u>Xenopsylla cheopis</u>, had increased in frequency whereas all other ectoparasites remained unchanged or decreased in number.

Eleven percent of the rats examined harbored <u>T</u>. <u>lewisi</u>. This figure is significantly above the 5% incidence indicated in an earlier study, and is probably due to an increase in the percentage of rats infested with X. cheopis.

Transmission experiments indicated that <u>C</u>. <u>felis</u> can serve as a vector of <u>T</u>. <u>lewisi</u>. The crithidial forms of the parasite were observed in the gastrointestinal tracts of infected fleas. Successful transmission of <u>T</u>. <u>lewisi</u> using <u>C</u>. <u>felis</u> as a vector was achieved on numerous occasions. Infection of non-infected rats which resulted from feeding them homogenized, infected fleas demonstrated that infection in nature may occur via ingestion of infected fleas.

Reproductive potential experiments indicated that <u>C</u>. <u>felis</u> does not reproduce well on laboratory specimens of <u>Rattus norvegicus</u>. Newlyemerged fleas fed readily, but no signs of oviposition were ever observed. Despite the fact that <u>C</u>. <u>felis</u> was shown to be capable of transmitting <u>T</u>. <u>lewisi</u>, it probably contributes little to the spread of the parasite in the rat population.