THE STRUCTURE AND LIFE HISTORY OF *CRITHIDIA*
PULICIS, N. SP., PARASITIC IN THE ALIMENTARY
TRACT OF THE HUMAN FLEA, *PULEX IRRITANS*.

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With Plate X.

CONTENTS.

	PAGE
Introduction	237
Material and Methods	238
Distribution of <i>Crithidia pulicis</i> in <i>Pulex irritans</i> . . .	239
Movements	239
Morphology	240
The Pre-flagellate Stage	240
The Flagellate Stage	241
The Post-flagellate Stage	242
Division	243
Method of Infection	244
Some Remarks on Insect Flagellates, especially of the Genus <i>Crithidia</i>	244
Summary	251
References	252
Explanation of Plate	253

Introduction.

FOR some years now much of my attention has been given to the investigation of various Flagellates parasitic in Invertebrates. These parasites were various *Crithidia* and *Herpetomonads* and the hosts have been chiefly Hemiptera and Diptera. During the last three years, I have conducted research on the endoparasites of the fleas found on the

human body, mainly using *Pulex irritans*, the common human flea. Some of the *P. irritans* contained the parasite that I have named *Crithidia pulicis*, a typical *Crithidia*, with its life history complete within the flea and to this parasite the present paper refers.

Material and Methods.

The fleas used for the study of *C. pulicis* were specimens of *P. irritans* obtained from various parts of England. The work has involved much time and trouble, for I used only *bred* fleas for my research, and, at considerable personal inconvenience, bred and reared the fleas upon my person, keeping them in confined areas by means of celluloid and rubber "flea-cages" of my own designing. In order to avoid all possibility of contamination of the fleas by feeding on other animals, *e.g.* cats, dogs or rats, the fleas were bred to the third generation and these were the fleas used for dissection. No blood other than my own was used for feeding the fleas, a point on which I would lay stress. An interval of two days after the last feed was usually allowed to elapse before dissection as the preparations of *C. pulicis* were cleaner than if the fleas were dissected immediately after feeding.

The alimentary canals of about 200 adult and larval fleas were carefully dissected and preparations made of their contents. Much time was given to the examination of fresh preparations of the *Crithidia* contained therein. The percentage of infected fleas was low, and hence much waste of time occurred in the breeding of "unprofitable" hosts and in examination of the same. All other organs of the fleas were examined and faeces of the fleas were constantly searched.

With regard to stained preparations, both wet and dry methods of fixation were used. The chief stains used were iron-haematoxylin, Delafield's haematoxylin and modifications of the Romanowsky stain, especially Giemsa's stain. Methylene blue was useful for *intra vitam* staining. Fixation by osmic vapour followed by absolute alcohol proved quite satisfactory. Formalin vapour followed by absolute alcohol also was useful.

In connection with the occurrence of a *Crithidia* in the human flea bred in the manner described previously, I should like to state that the numerous detailed examinations made of my blood by smears, thick films and cultures have entirely failed to reveal the presence of any trypanosome. The bearing of this statement will be discussed later.

Distribution of Crithidia pulicis in Pulex irritans.

Crithidia pulicis shows three characteristic phases in its life history and is thus a typical *Crithidia*. It occurs throughout the alimentary tract of its host, both adult and larval fleas containing the parasite. The small oval or rounded pre-flagellates, having some resemblance to the Leishman-Donovan bodies, are most abundant in the entire alimentary tract of the larval fleas and in the crops of young adults. Fully developed flagellates, either growing or in process of division, are best observed in the midgut and intestine of the adult flea, while the post-flagellates are most numerous in the rectum of mature fleas and have often been recovered from the faeces of infected hosts.

Forms other than non-flagellates have not been found in the mouth or proboscidal regions of the flea, nor have fully developed flagellates been observed in the oesophagus of adult fleas.

Up to the present, there is no evidence of the occurrence of *C. pulicis* in organs other than those of the digestive tract. Hereditary infection, which has been fully demonstrated in *C. melophagia*, has not been shown so far in *C. pulicis*.

Movements.

C. pulicis is very active in its movements which are brought about chiefly by the undulating membrane, the flagellum being relatively short compared with that of such *Crithidia* as *C. gerridis*. *C. pulicis* moves with the anterior (flagellar) end forwardly directed and progression is aided by waves of contraction that pass from behind in a forward direction along the body. The wave-like motion is due to the myonemes in the body and membrane, which myonemes can be seen in life by the aid of the paraboloid condenser. The same phenomenon has been observed in living Spirochaetes and in *C. melophagia*. The myonemes in neither of these cases are artifacts, as some recent dogmatic writers would have us believe. Living organisms do not consistently display artifacts. The myonemes of the membrane are more marked than those of the body.

C. pulicis is capable of rapid reversal of its direction of motion. This is brought about by the organism swinging rapidly in a semi-circle, the posterior end acting as a centre of rotation.

Movements of flexion are more common in *C. pulicis* than in any other *Crithidia* that I have examined. The posterior end of the body

is very mobile and at times folds completely over so that the posterior end of the body lies parallel to the anterior end and the parasite is looped like a **U**. In ordinary quiet movement the body of the parasite may turn on itself, so that the posterior end seems folded just above the nucleus. (Pl. X, fig. 13.)

Entanglement movements were fairly common, but aggregation rosettes of *C. pulicis* were not nearly so numerous as in the case of *C. melophagia* or *C. gerridis*. One point about the aggregation rosettes of *C. pulicis* is the remarkable billowy effect due to the undulations of the membranes of the parasites.

Movements occurring during longitudinal division will be described in the section dealing with division.

MORPHOLOGY.

Crithidia pulicis, like other insect flagellates, exhibits the three typical phases of a *Crithidia* in its existence. While the general life history is quite typical, there are many points that apply to *C. pulicis* only, and hence I prefer to establish a new species for it, rather than amalgamate it with *C. ctenophthalmi*, the parasite of the rat flea, *Ctenophthalmus agyrtes*.

The Pre-flagellate Stage.

Pre-flagellates (Pl. X, figs. 1-8) of *C. pulicis* are most abundant in the whole length of the alimentary canal of the larva of *P. irritans* or in the crops of young adults. They are oval or rounded bodies in their simplest form, measuring $2.3\ \mu$ to $7.0\ \mu$ by $1.5\ \mu$ to $4.5\ \mu$. The general cytoplasm is faintly granular (figs. 1-8). The nucleus is well marked (figs. 1, 5, 6), and contains more chromatin than that of most other *Crithidia*. A small karyosome may be present (figs. 1, 6). The shape of the nucleus has a general resemblance to that of the entire pre-flagellate. The blepharoplast is large (figs. 4, 7, 8) and is either straight (figs. 2, 8), oval (fig. 7), or slightly curved (fig. 1). It is very obvious in life. In the older pre-flagellate (figs. 6-8), a clear area can often be distinguished in life, either to one side of the nucleus or below the blepharoplast, its position depending on the amount of elongation that the parasite has undergone. This is the area from which the flagellum develops. It takes the chromatin colouration in stained preparations. When the pre-flagellate elongates, the flagellar

end is the more rapid in growth, the endoplasm flowing forwards and pushing the ectoplasm before it. The contents of the chromatophilic area concentrate and gradually differentiate into a thread (fig. 4), which gains the surface and forces the ectoplasm outwards forming the wavy membrane. The final portion of the thread protrudes beyond the body as the free flagellum (figs. 6-8). The posterior end of the flagellate develops less rapidly than the anterior end. Division may occur while the pre-flagellate stage is merging into that of the flagellate, and repeated division produces rosettes (figs. 3, 5, 8). All the pre-flagellates do not develop at the same rate, consequently some of the members of a rosette are still oval while others have already developed their flagella and membranes to some extent (fig. 8).

The somewhat frail appearance of the pre-flagellates is certainly in contrast with the relatively gross facies of the mature flagellate, but the whole development has been watched repeatedly in life and is unmistakable.

The Flagellate Stage.

The flagellate form of *C. pulicis* (figs. 9-17) is an organism measuring from 26 μ to 65 μ long, the free flagellum being included. The body is elongate and the flagellar end prolonged and finely tapering. The aflagellar posterior end is relatively blunt, being somewhat rounded in the fully formed individual (figs. 16, 17). The general protoplasm is richly granular but the granules are fine. The nucleus is oval, rich in chromatin, the chromatin being in the form of refractile granules in life and staining well whatever stain be employed (figs. 10, 12, 17). Its position in the body varies somewhat. Frequently it is about a third of the way between the two ends of the organism (figs. 14, 16, 17), but occasionally after periods of great activity of the parasite, it is found nearer the flagellar end.

The blepharoplast is large, usually lying horizontally across the body of the organism (figs. 9, 11, 15), and as a rule extending almost across the transverse diameter. Commonly it presents a homogeneous structure except when in division. Its shape may be oval (figs. 10, 15-17), rod-like (figs. 11, 13), or curved slightly. All the forms have been seen in life and the stained specimens merely corroborate the observations *in vivo*. In a few cases, a vertical blepharoplast has been found. This is uncommon but it has been recorded in many other *Crithidia*. There are a few chromidia (fig. 16) present in many of the

flagellates, and there seems to be a tendency for their number to increase just prior to division. As at this time the organism is passing through a critical period, the migration outwards of chromidia may be an adaptation to secure a more perfect equilibrium between the general cytoplasm and the nuclear elements.

One of the most noticeable features about *C. pulicis* is the beauty of its membrane. The undulating membrane is large and well marked. Its chromatic border is highly refractile in life, and stained specimens show it as a well-marked chromatin containing edge to the membrane (figs. 15-17), continued outwards just beyond the body as the free flagellum. Contractile myonemes (figs. 13, 15, 16, 17) are present in the membrane and faint body myonemes (figs. 9, 16) are also seen occasionally. The free flagellum and the chromatic border of the membrane show a transverse striation in a few cases. There is much morphological variation among the full-grown flagellates, depending on the frequency of and variation in division.

The Post-flagellate Stage.

The post-flagellates of *C. pulicis* (figs. 23-36) again show differences from most other *Crithidia*. In the majority of these organisms division into four without subsequent growth seems to be a common procedure prior to the formation of post-flagellates. In *C. pulicis* division into two, followed by some slight amount of growth seems general. But as flagellates of different sizes may divide, the range of size of the cysts is fairly great. The fully formed post-flagellate is from 3μ to 6μ long and from 2.0μ to 4.6μ broad in those that I have examined. The processes leading up to the full post-flagellate stage may be herewith summarised. Retraction of protoplasm from both ends of the organism towards the nucleus occurs (figs. 23-25); part of the protoplasm is absorbed. The chromatin constituent of the membrane appears to be absorbed and the free flagellum shortens (figs. 23-28). General fusion of membrane and body occurs and the organism becomes ovoid (figs. 29-36). A thin, gelatinous secretion appears on the outside of the ovoid mass and hardens into a varnish-like cyst wall around the contents. The fully formed post-flagellate (figs. 29-36) consists then of a thin coat enclosing the nucleus and blepharoplast together with a fair amount of protoplasm. The nucleus is often round but it may be oval. As a rule, the nucleus and blepharoplast remain quite distinct and slightly separated one from

another (figs. 28, 33, 35, 36) but in a few cases the nucleus and blepharoplast were apposed (figs. 30, 31, 34). On rare occasions the contents were slightly contracted from the varnish-like wall (figs. 34-36).

DIVISION.

(a) *The Pre-flagellate.*

After the introduction of the parasite into a new host by way of the mouth, division occurs. As a result of repeated binary fission followed by growth, rosettes of pre-flagellates (figs. 3, 5, 8) are fairly common. The sequence of events in nuclear division resembles that of the flagellate stage described below. The chromatophile area of the pre-flagellate, however, divides, generally symmetrically, and hence each new pre-flagellate is a replica of its parent.

(b) *Division of the Flagellate.*

Longitudinal division of *C. pulicis* (figs. 18-22) is of two types:—(1) equal or symmetrical division; (2) unequal or asymmetric fission. The processes have been observed in life and corroborated by reference to stained specimens. Division is initiated by the concentration of the substance of the blepharoplast into two masses, one at either end (fig. 18). The blepharoplast becomes bowed and presents a dumb-bell appearance (fig. 19). The two heads of the dumb-bell finally become separated (figs. 20, 21). Soon after division of the blepharoplast has commenced, that of the nucleus begins, but the process is less rapid than is that of the blepharoplast. The flagellum and membrane next commence to split and as the flagellum violently lashes outwards, the division of the body follows. The two bodies thus formed gradually diverge (fig. 21) and a V-shaped organism then is seen. Ultimately the diverging arms of the V lie in a straight line (fig. 22) and the two daughter forms finally separate by constriction at the posterior end.

In asymmetric division two types are encountered:

(1) The blepharoplast and nucleus divide into unequal portions and the body fission follows that of the chromatic masses (fig. 22).

(2) On rare occasions the nucleus and blepharoplast may divide symmetrically while the general protoplasm divides asymmetrically.

Division is a rapid process and there is a great variety of forms resulting therefrom. I have not found division rosettes of the flagellate form of *C. pulicis* at all common. After the primary division, a second

may rapidly follow, but the original dividing organisms have separated from one another as a rule, so that rosettes are not produced.

Division of the flagellate form occurs just previous to the formation of the post-flagellates, but the number of divisions appears to be less than in the other *Crithidia* I have studied, and as a result, the ovoid post-flagellates of *C. pulicis* are relatively larger.

The form of the parasite is influenced to some extent by the diet of the host. In a starved flea the body cytoplasm of the flagellates is very finely granular. A short time after fresh blood reaches these parasites, they become larger, more richly granular and division commences. Flagellates occurring in the semi-digested "dark" blood in the hinder part of the alimentary canal of the flea are larger on the average than those in the fore part, the food medium apparently being more suited to their needs.

Method of Infection.

The method of infection of *P. irritans* by *C. pulicis* is purely a contaminative (casual) one, so far as I can ascertain. The faeces of already infected *P. irritans* contain numbers of post-flagellate forms of *C. pulicis* and sometimes also some active flagellates. Other fleas may obtain blood near the spot of skin fouled by their neighbours and thereby ingest some of the post-flagellates. In other cases, as the insects moved about, especially in the confined space of my "flea cages," fluid dejecta contaminated their bodies and legs. In the removal of the offending material, the post-flagellates are brought in contact with the mouth parts and thence find their way into the alimentary canal of the host.

I have seen no indications of hereditary infection. Careful dissection of both male and female genitalia failed to show any form of the parasite therein on examination. Breeding experiments confirmed the absence of *Crithidia* in the eggs. Clean fleas bred clean. Eggs of infected fleas hatched apart from the parents also yielded uninfected stocks. I have never found *C. pulicis* in any situation other than the alimentary tract and dejecta of *P. irritans*.

Some Remarks on Insect Flagellates, especially of the Genus Crithidia.

Earlier in this paper I referred to the fact that bred fleas were used during my investigations and that my blood was the medium on which they fed. This raises two possibilities. According to the

dogmatic hypotheses of certain writers, *C. pulicis* would be a stage of a trypanosome. No trypanosome has ever been observed in my blood, whether examined by smear, thick film or culture, and this examination has been frequently made by highly competent authorities. Further, the extended period over which observations have been made is sufficient to put this supposition entirely out of court.

Again, no sore of any kind has ever developed on my body, though bites of the usual character have been made by the fleas repeatedly. These observations exclude all possibility of *C. pulicis* being a stage or stages in the life-cycle of other flagellates, such as those described by C. Basile, and effectually dispose of the wild hypothesis laid down by a would-be authority, that flagellates of sanguivorous insects must be regarded as stages in the life-cycle of a vertebrate trypanosome.

Also, I have thought it well to investigate fleas from outside sources, and through friends, to whom my best thanks are due, obtained fleas from several places in each of the following counties:—Sussex, Surrey, Hampshire, Gloucestershire, Wiltshire, Somerset, London, Lancashire, Cambridge and Essex. Some of the fleas from each of the above-named counties contained *C. pulicis*. In every case, the parasites presented identically the same morphology and life history as those obtained from my bred *Pulex*. Recently, a criticism (*Bull. Sleeping Sickness Bureau*, May, 1911) was made of a paper by Swingle on the transmission of *T. lewisi* by rat-fleas, in which the critic "presumes" that the fleas used were "wild" fleas, and adds:—"in the present state of our knowledge, in the case of flagellates found in wild insects, it is almost guesswork to say that certain forms are natural flagellates and others stages of blood trypanosomes." Considering the evidence that is steadily accumulating of the occurrence of natural flagellates in blood-sucking insects, the above statement seems a rash and inexact one. Bruce and his colleagues have recently shown that natural flagellates, *Crithidia*, occur in the sanguivorous Tabanid flies suspected of transmitting *Trypanosoma pecorum*, and that the *Crithidia* are flagellates of the insects and not developmental forms of a trypanosome. Wild insects include sanguivorous and non-sanguivorous forms; while the case of "wild" insects feeding on plants is obviously overlooked.

As far back as 1906, Ross published on *Crithidia* in mosquitos in India (observed between 1895 and 1899) and in connection therewith stated (p. 107) "In fact, it was evident that they [*Crithidia*] had been already present in the insects before these were fed on the blood." The sanguivorous habit of an insect is no criterion as to whether a flagellate

occurring therein is a developmental form of a trypanosome, or a true parasite of the insect.

My own work on *C. pulicis* confirms the statement that natural flagellates can occur in sanguivorous insects, and in this case there is certainly no evidence to show that the results obtained on *C. pulicis* from "wild" *P. irritans* could be regarded as "almost guesswork." As with the *Tabanidae*, a natural flagellate of the host is present, and there is not one tittle of evidence to show that *C. pulicis* is other than a natural flagellate of *P. irritans*.

Arm-chair criticism is always easy and after five minutes of wild hypotheses, the march of progress may be endangered for as many years. This has recently been attempted by certain dogmatic individuals who have published compilations regarding trypanosomes,* *Crithidia* and *Herpetomonas* without really practical knowledge or first-hand investigation of some of the subjects discussed. But having formed an hypothesis, they then support it blindly. Further, though the hypothesis has been shown by recent research to be untenable, yet they continue to preach the same, and, as with the importunate friend, gain a hearing and obtain followers by their very insistence. The following so obtained is not of the highest order, needless to say, as witness a recent deluded beginner, who in his first essay in the investigation of the flagellates of flies, states of a man who has never published an original paper on *Herpetomonas*, and has merely confused the whole subject in a wordy text-book, that the history of *Herpetomonas* is related in greater detail by him! British Protozoology is in a bad way when original work is neglected, or confused with that of the mere compiler.

While writing this paper, my attention has been drawn to an account published by Mr J. S. Dunkerly on a parasite or parasites which he calls *Leptomonas muscae domesticae*. Most of his paper, however, is devoted to an attempt to discuss the genera *Leptomonas*, *Herpetomonas*, *Crithidia* and *Trypanosoma*. I would not trouble to notice the paper, except that in it the author attempts to establish that *Crithidia* is not a valid genus and he states (p. 649) that "It is with a view to the clearing up of at least one part of the vexed question [nomenclature] that I wish to re-state the following facts in their history." Indeed! This is rather an ambitious task to essay in one's first paper on such a complicated subject as the Flagellates of Insects. But Mr Dunkerly is nothing if not courageous. However, I beg to inform him that a much broader and more accurate view will have to

be taken than the one he has allowed himself to follow. To quote the views of Roubaud, Hartmann and Jollos and Prowazek in full on the one hand, while giving only meagre quotations from Patton, Swingle and Miss Mackinnon on the other, savours more of the methods of the "party politician" than of research for the advancement of Science. I cannot deal with this memoir in detail, for there is scarcely a statement in the paper that is accurate or not misrepresented. Practically all the authorities quoted by Mr Dunkerly have been discussed at length by Patton, Miss Mackinnon and myself, among others, *ad nauseam*.

But I must deal with one remarkable statement in Mr Dunkerly's paper, namely:—" *Crithidia* cannot be applied as a generic name to any form, as it has simply been the name given to two stages in the life history of a *Leptomonas* or to what in other cases are probably stages of Trypanosomes." This is amusing from one who has never published on parasites of the genus *Crithidia*, and, judging from his remarks, would not know a *Crithidia* if he saw it. For instance, he attributes a diagnosis of the genus to Patton, which diagnosis is inaccurate and in a form that Patton never used. According to Mr Dunkerly: "Patton... decided that all uniflagellate parasites of insects with the kinetonucleus anterior to the trophonucleus and without undulating membrane, are to be called *Herpetomonas* and that those having the kinetonucleus posterior to the trophonucleus and possessing an undulating membrane, should receive the generic name of *Crithidia*." Patton never used the terms "trophonucleus" and "kinetonucleus," and certainly did not say that in *Crithidia* the blepharoplast was always posterior to the nucleus.

Another statement made is also an unworthy one, namely:—"Lühe and Hartmann and Jollos have pointed out that Patton's failure to see the characters observed by Prowazek and others does not prove their non-existence." But Lühe (as listed) wrote in 1906 and Patton in 1908. Further, Patton never denied the existence of certain features in Herpetomonads, but disagreed with their interpretation as given by Prowazek and others—a very different thing. As to Hartmann and Jollos, I am sorry that I cannot seriously consider their second-hand views, for these authors, among other items, place *Piroplasma* (*Babesia*) and the malarial parasites in their "Flagellatenordnung Binucleata," despite lack of supporting evidence.

Mr Dunkerly appears to lay much stress on his edition of Léger's statement that *Crithidia* is "en form (sic) de grain d'orge." Leaving aside the words used in description, let us consider the figures of Léger. If Mr Dunkerly can find any important difference in the figures

of Léger and the figures of young forms of *C. gerridis*, *C. tabani* and *C. melophagia*, given by Patton, Flu, Swingle and myself, he is a mere quibbler. The whole discussion is one of words, words, *ad nauseam*. Mr Dunkerly further attributes to Léger a statement that *Crithidia* is "usually without an undulating membrane." This is an unfortunate inexactitude. Léger (1902) not only recognised "un rudiment de membrane ondulante," but figured it (Léger's Figs. 7-10). All reference also is omitted to Patton's explanation of the differences between Léger's account and his own, these being due to the fact that Léger had not described the fully developed flagellate when he first wrote. The mature *C. fasciculata* is stated by Patton (1908, p. 142) to be "very similar to the adult form (*C. gerridis*) I have described."

To lead up to the conclusion that *Crithidia* is not a valid genus, Mr Dunkerly favours us with what may be described as a crescendo of possibilities, probabilities and certainties, but not a particle of direct evidence or experiment. On p. 648 we read, "However, it *seems* from the evidence of the forms found in *Homalomyia* that the same organism *may be* without an undulating membrane at one stage of its life history, while possessing one at another stage" (the italics are mine). Later p. 651, we read "This...*seems certain* (1) that Léger's original pear-shaped *Crithidia* is only a stage of the *Leptomonas* life-history." Léger did not explicitly describe *Crithidia* as pearshaped in founding the genus. Further, it is a mere assumption that because a *Leptomonas* (in the sense of Chatton and Alilaire) is present in *Homalomyia*, there is really a *Leptomonas* in *Anopheles maculipennis* or in every insect from which *Crithidia* have been obtained.

By the end of his paper, Mr Dunkerly has become even more definite, for on p. 652 we read "*Crithidia cannot* be applied as a generic name to any form, as it has simply been the name given to two stages in the life history of *Leptomonas* or in other cases to what are probably stages of *Trypanosoma*."

In the first instance, there is no proof whatever that any true *Crithidia* is a stage or stages in the life history of a *Leptomonas*. Nor does Mr Dunkerly's work aid in that matter in any degree, especially as he is forced to admit (p. 649) that "the low percentage of infections have (sic) prevented the completion of it [the life-cycle] up to the present." Again, dealing with intermediate forms, a statement is made that all stages between the short, truncated forms of *Leptomonas muscae domesticae* and elongated forms with undulating membranes, occur. But the three figures quoted as illustrating intermediate

forms are inadequate and unsatisfactory, for they are all about the same length and merely illustrate, as Mr Dunkerly himself states: "the varying position of the kinetonucleus and the presence of an undulating membrane."

How can dogmatic statements based on the knowledge of part only of the life-cycle of a *Leptomonas* be regarded as finally settling the question of the validity or otherwise of a totally different genus, *Crithidia*? It is a presumption to set forth such statements on such hopelessly inadequate evidence.

With regard to the second part of the statement that *Crithidia* are probably stages of trypanosomes, no definite evidence in support of this statement is forthcoming. I can find nowhere that the conversion of a trypanosome into a true *Crithidia* and of a true *Crithidia* into a veritable trypanosome have been witnessed in the living organisms. *Herpetomonas* and *Crithidia*, considering their definition, have not necessarily anything to do with trypanosomes. To introduce the trypanosomes into the discussion of the genera *Herpetomonas* and *Crithidia* is merely and immediately to confuse the issue. Hypotheses as to whether a flagellate is primarily a parasite of an insect or of a vertebrate are purely speculative and can only be treated as such. The opinion of L. Léger in respect to the position of the trypanosomes, is, however, of interest:—"Les trypanosomes du sang, ne représentent qu'une adaptation partielle et secondaire d'un parasite primitivement intestinal ou enterocoelomique d'Invertébré." Having regard to this statement, the discussion of insect flagellates from the trypanosome point of view is commencing at the wrong end and so working backwards.

I think that there is little doubt that Mr Dunkerly has found a Herpetomonad in *Homalomyia canicularis*, belonging to the *Leptomonas* and leptotrypanosomes of Chatton and A. Léger. However, Mr Dunkerly has not got a *Crithidia*, for as Chatton and Léger say, "Elles [leptotrypanosomes] approchent de stade *Crithidia* sans toutefois l'atteindre." Leptotrypanosomes have the flagellum attached to the body, there is no membrane and the blepharoplast is posterior, conjoined characters which do not occur in *Crithidia*. However, the observations of Miss Mackinnon (1910) on a *Herpetomonas* from a *Homalomyia* are most instructive. Miss Mackinnon finds *Herpetomonas* therein which often exhibit the peculiarity of the flagellum being bent back and "sticking" to the body. May not this account for the phenomenon seen by Chatton and by Dunkerly? It is strange that Mr Dunkerly, although

he refers to Miss Mackinnon's paper, makes no mention of this observation to which the author draws special attention and illustrates by text-figures. That is only another example of the method of partial quotation pursued by those who wish—by methods analogous to those of the party politician—to destroy the genus *Crithidia*.

Again, in a short addendum Mr Dunkerly states that Flu in a recent paper on the parasites of the house fly "in the main" confirms "the chief points emphasised" by him. I am sorry that I cannot agree, for *Crithidia* is recognised by Flu as a genus, is defined by him and is kept separate from *Leptomonas*.

More earnest endeavour to work out the life history of flagellates such as *Leptomonas* is emphatically needed. At present, the flagellate form only is at all adequately dealt with, and until the complete cycle has been obtained, hypotheses built on knowledge of the flagellate stage only cannot but be somewhat insecure.

In conclusion, one is impelled to wonder whether research in Protozoology is to be conducted merely on the lines of party politics instead of the advancement of knowledge. Perhaps the seeming advocates of the former policy will re-consider their position. Apparently, Dr Woodcock, having written a thoroughly confused account of the Haemoflagellates, is now to be allowed to edit the work of other investigators more capable than himself, and to support Dr Woodcock we are to have the inexperienced efforts of his henchmen, like Mr Dunkerly, who seems in the domain of Protozoology to be essaying to fly in an aeroplane before he knows how to walk. The whole procedure is unworthy of British Protozoology and contributes nothing to the extension of knowledge.

Common sense dictates that the word of the practical worker is of more value than that of the "arm-chair critic." The statements of practical investigators like Patton, Flu and Swingle must be of infinitely more value than the hypotheses of Woodcock, Hartmann and others who have not themselves investigated *Crithidia*. It is amusing to note that apparently the only writers who wish to destroy the validity of the genus *Crithidia* are those who have not published researches on the same, and so cannot be expected to have a first-hand knowledge of the genus. We have to work out complete life-cycles if we are to go forward, and all the wrangling over nomenclature and evolution will be of no avail in the end. However, time will tell.

SUMMARY.

1. *Crithidia pulicis*, n. sp., is a parasite of the alimentary tract of the human flea, *Pulex irritans*.

2. The fleas used in this investigation were bred to the third generation in special "flea cages" on the human body. "Wild" fleas were examined and yielded the same parasite. Both larvae and adult fleas were examined.

3. *C. pulicis* exhibits pre-flagellate, flagellate and post-flagellate stages in its life history and these phases gradually develop, the one from the other.

4. The flagellate form is very active, movement being accomplished by means of the myonemes of the membrane and body.

5. Pre-flagellates (figs. 1-8) are oval bodies, from $2.3\ \mu$ to $7\ \mu$ long by $1.5\ \mu$ to $4.5\ \mu$ broad. The nucleus contains much chromatin and a small karyosome may be present. The blepharoplast is large. The flagellum develops from a chromatophile area. The rate of acquisition of flagellum and membrane varies in individual parasites. Division rosettes are frequent. The pre-flagellates have a somewhat frail appearance.

6. The flagellates of *C. pulicis* (figs. 9-17) are $26\ \mu$ to $65\ \mu$ long. Their cytoplasm is richly but finely granular, the nucleus well marked, the blepharoplast large, showing slight variations in shape. Chromidia may be present. The undulating membrane is large, well marked and possesses myonemes which can be detected in the living organism. The free flagellum is relatively short.

7. Post-flagellates (figs. 23-36) are from $3\ \mu$ to $6\ \mu$ by $2.0\ \mu$ to $4.6\ \mu$. They occur in the rectum and dejecta of the fleas. They are produced by concentration of the protoplasm round the nucleus and blepharoplast with absorption of the membrane and flagellum and finally the production of a thin varnish-like coat.

8. Longitudinal division (figs. 15-22) is the method of multiplication. It occurs in the pre-flagellate and flagellate stages. It may be symmetrical or asymmetric. Division is initiated by that of the blepharoplast, followed by that of the flagellum, membrane and nucleus and finally the body cytoplasm. Two types of asymmetric division have been observed.

9. The method of infection is contaminative, the post-flagellates in the faeces being the source of infection. There is no evidence of hereditary infection.

10. *Crithidia pulicis* is a member of the genus *Crithidia*, and is a true parasite of the insect, *Pulex irritans*.

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Further references will be found at the ends of some of the papers quoted.

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EXPLANATION OF PLATE X.

All figures were outlined with the Abbé-Zeiss camera lucida, using a 2 mm. apochromatic (Zeiss) or $\frac{1}{2}$ inch achromatic (Zeiss) objective, and compensating oculars 8 and 12. The magnification is approximately 1300 diameters except where otherwise stated.

Figs. 1-8. Pre-flagellate stages of *Crithidia pulicis*.

Fig. 1. Large pre-flagellate showing well-marked nucleus with karyosome and distinct blepharoplast.

Fig. 2. Smaller pre-flagellate $\times 1950$.

Fig. 3. Rosette of pre-flagellates, beginning to elongate.

Fig. 4. Pre-flagellate, showing commencement of differentiation of flagellum.

Fig. 5. Rosettes of pre-flagellates showing slight variation in the shape and position of the blepharoplast and in the extent of elongation.

Figs. 6, 7. Elongating forms.

- Fig. 8. Rosette, showing unequal rate of development of the pre-flagellates.
Figs. 9-17. Flagellates of *C. pulicis*.
Fig. 9. Small flagellate, showing nucleus, well-marked blepharoplast and body myoneme.
Fig. 10. Larger parasite, showing oblique blepharoplast and very chromatoid nucleus.
Fig. 11. Flagellate with broad posterior end.
Fig. 12. Broad form.
Fig. 13. Parasite showing twist of the posterior end of body. Such an appearance is often seen in life.
Fig. 14. Very narrow form, a division product of such forms as shown in figs. 15-17.
Figs. 15-17. Large flagellates, showing the very characteristic prominent membrane with myonemes. These forms are common.
Figs. 18-22. Stages in the division of *C. pulicis*.
Fig. 18. Broad form showing beginning of division of the blepharoplast.
Fig. 19. Shows parasite with dumb-bell shaped blepharoplast and flagellum beginning to split.
Fig. 20. Parasite showing complete division of blepharoplast and membrane, nucleus constricted.
Fig. 21. Separation of two daughter forms in symmetrical fission.
Fig. 22. Asymmetric division of *C. pulicis*. Separation of daughter forms almost complete.
Figs. 23-36. Post-flagellate stages of *C. pulicis*.
Figs. 23-25. Shortening forms showing concentration of protoplasm round the nucleus and blepharoplast and retraction of the flagellum.
Fig. 26. Cluster of four parasites, one of which has become the typical post-flagellate, the other three being not quite so advanced.
Figs. 27, 28. Almost complete assumption of the post-flagellate form. Fig. 28 $\times 1950$.
Figs. 29-33. Post-flagellates showing variation in size.
Figs. 34-36. Post-flagellates in which the contents are somewhat shrunken away from the thin varnish-like wall.

Annie Porter, del.

Crithidia pulicis, n.sp.