THE MONTANA STATE BOARD OF ENTOMOLOGY

SEVENTH BIENNIAL
REPORT

1927 - 1928

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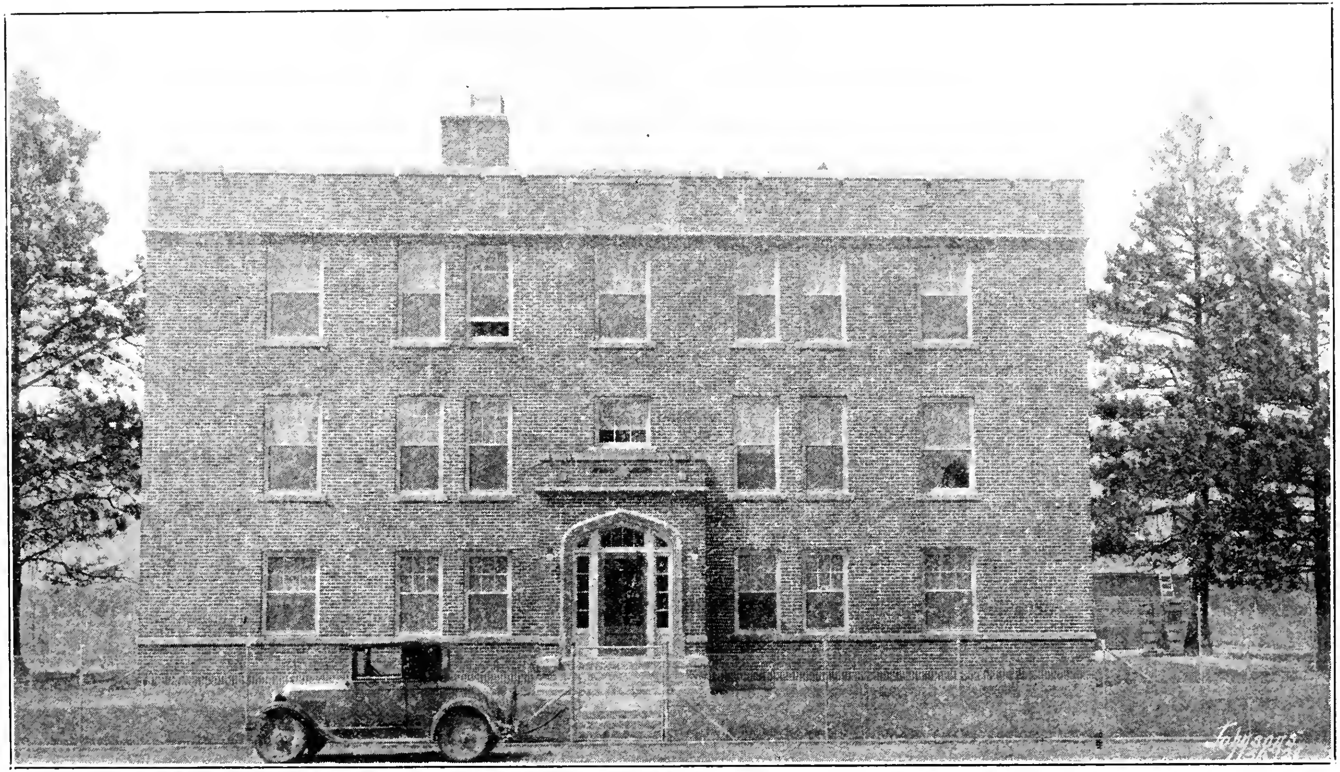

New Laboratory of The Montana State Board of Entomology
recently completed at Hamilton, Montana.

MONTANA STATE BOARD OF ENTOMOLOGY

W. F. COGSWELL, M. D., *Secretary State Board of Health*, Chairman, Helena, Montana.

W. J. BUTLER, D. V. S., *State Veterinary Surgeon*, Member, Helena, Montana.

J. R. PARKER, M. S., *Acting State Entomologist*, Acting Secretary, Bozeman, Montana.

LETTER OF TRANSMITTAL

Bozeman, Montana, December 15, 1928.

To His Excellency,
John E. Erickson, Governor,
Helena, Montana.

Dear Sir:

As acting secretary of the Montana State Board of Entomology I have the honor of submitting the Seventh Biennial Report which covers the calendar years of 1927 and 1928.

Outstanding developments of this period have been the completion of the new laboratory at Hamilton, the success of the Spencer-Parker vaccine in preventing Rocky Mountain Spotted Fever, and the rearing and liberation of the French tick parasite, *Ixodiphagus caucurtei.* Papers dealing with these developments will be found in the body of the report.

Rocky Mountain Spotted Fever has continued to spread throughout the state. Whereas the disease was formerly largely confined to Western Montana it has now been reported from 23 counties in Eastern Montana involving an area of more than 30,000 square miles. During the biennium 15 cases occurred in the western half of the state and 49 in the eastern half. The problem of Rocky Mountain Spotted Fever is therefore extremely vital to the welfare of the entire state and warrants the continued support of state funds.

Respectfully yours,

J. R. Parker, *Acting Secretary.*

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FOREWORD

J. R. Parker, *Acting Secretary*

According to the wording of the creative legislative act it is the duty of the Montana State Board of Entomology "to investigate and study the dissemination by insects of diseases among persons and animals, said investigation having for its purpose the eradication and prevention of such diseases." The study of insects which do not transmit disease and the control of insects attacking crops are therefore not within the province of this board but are taken care of by the Agricultural Experiment Station and the State Entomologist. The State Board of Entomology is composed of three members; the Secretary of the State Board of Health, the State Veterinary Surgeon and the State entomologist, and therefore brings together in one organization the three State Officials who are most concerned with the control of human diseases, animal diseases and insects which may transmit disease.

The field headquarters for the Montana State Board of Entomology is at Hamilton, Montana, where a modern, fire-proof, well equipped laboratory was built this past year with funds provided by the 1927 legislative session.

The laboratory is occupied jointly by two staffs of investigators, one employed by the Montana State Board of Entomology and the other by the U. S. Public Health Service. These two organizations have worked together under an informal plan of co-operation since 1921. The State Board of Entomology has been fortunate indeed in securing this co-operation; the U. S. Public Health Service has not only been generous in its financial support of the work but has also provided highly trained investigators whose researches have advanced the work far more rapidly than could have been expected if the State Board had continued to work alone.

The personnel of the State Board of Entomology remained unchanged from the time the Board was established in 1913 to March, 1928. These fifteen years of continuous service by all members of the Board have resulted in a continuity of effort and an effectiveness that could not have otherwise been obtained. In March, 1928, Professor R. A. Cooley, Secretary of the State Board of Entomology, was granted a year's leave of absence from his duties at the Montana State College and the Agricultural Experiment Station. He is spending the year searching for tick parasites in South Africa and studying ticks and tick parasites in England and France. The object of his trip is to obtain information that will be valuable to the State Board of Entomology in its efforts to control ticks in Montana by the use of parasites. Professor Cooley is making his trip without expense to the State, personally paying part of his travelling expenses, the remainder being paid from money contributed by organizations interested in the possibility of tick control by parasites. In this connection the Board of Entomology is pleased to acknowledge the financial aid extended by the Anaconda Copper Mining Company, the Montana Power Company, the Great Northern and the Northern Pacific Railroads. Professor Cooley will return to Montana in March, 1929, and will at that time resume his duties as Secretary of the State Board of Entomology. During his absence J. R. Parker, of the Montana Agricultural Experiment Station, is serving as Acting Secretary.

The main lines of work conducted by the State Board of Entomology staff during the past biennium were the study, rearing and liberation of the tick parasite, *Ixodiphagus caucurtei* duB., and the continuation of the tick control program carried on in the Bitter Root Valley since 1918. Studies in connection with the preparation and use of a vaccine to prevent Rocky Mountain Spotted Fever and investigations concerning the organism causing the disease, have largely occupied the attention of the U. S. Public Health Service workers. A more detailed account of the work together wth some of the results accomplished, will be found in the body of the report.

As acting secretary of the State Board of Entomology, I wish to gratefully acknowledge the aid given by Doctor R. R. Parker and Doctor R. R. Spencer in supplying material for the Biennial Report covering the work of the U. S. Public Health Service at the Hamilton Laboratory during the past biennium.

ROY KERLEE

To the list of those who have given their lives in the study of Rocky Mountain Spotted Fever, another name must be added. While employed as assistant bacteriologist in the Hamilton Laboratory, Roy Kerlee in some unknown manner contracted the disease and died on February 14, 1928.

Arthur LeRoy Kerlee was born at Darby, Montana, March 5, 1905, and received his early education in the Darby schools. He graduated from the Montana State College in 1927 with the degree of bachelor of science in Botany and Bacteriology. Following his graduation he was employed as assistant bacteriologist by the U. S. Public Health Service in the Hamilton Laboratory. While in college Kerlee was a fine student and a leader in many campus activities. Knowing full well the dangerous nature of the organism which he would be called upon to study, he entered upon his professional work with fearless enthusiasm. Although at the time of his death he had been engaged in research work for only a few months, he was looked upon as a young man of unusual ability who, if he had lived, would have gone far in his chosen profession.

TICK PARASITES

TICK PARASITES—EXECUTIVE REPORT.(1)

By

R. A. Cooley, *Secretary, Montana State Board of Entomology*

In the Sixth Biennial Report of the Montana State Board of Entomology it was stated that the so-called French tick parasite (*Ixodiphagus caucurtei* du Buysson) had been brought to Montana and that the work of colonizing it here had been started in the hope that in time it might be found to be an important factor in the destruction of the prevailing tick of the region (*Dermacentor andersoni* Stiles). It should be stated, clearly, that our experiences with the parasite so far have been distinctly encouraging, although some difficulties have appeared which will have to be overcome.

In order to lead to a clearer understanding of what is involved in an effort to introduce such a parasite, the necessary procedure will be outlined. The purpose of the experiment is to breed the parasites in quantity and to liberate them where the ticks occur in Nature, and by such methods as will make it possible for them to take up their habit of parasitizing the tick. The primary requirements for successfully carrying out the enterprise are the following:

1. Keeping the parasites alive in the laboratory and breeding large quantities when they are needed, particularly in the spring when they are to be liberated.

2. Growing a sufficient supply of non-infected ticks in which to feed the parasites.

3. Holding a sufficient supply of caged animals on which to feed ticks both in growing our supply of ticks and for use in feeding ticks while they are being parasitized.

4. Experimental work and study in order that we may better understand this little known parasite whose habits are perhaps not primarily adapted to the physical and biological conditions found in Montana.

5. Enough employees to carry out the tremendous amount of detail required in the work.

6. Equipment such as animal cages, cage racks, infesting bags, microscopes, as well as vials, pill boxes and scores of items of petty equipment.

7. Transportation to and from remote localities in the mountains, and to and from other parts of the state where effort is being made to colonize the parasite.

Previous to getting our initial stock of living parasites from Doctor Larrousse in July, 1926, no provision had been made for getting a supply of ticks in which to feed the parasites in the laboratory, and without such ticks the parasites could not be materially increased. However, at that season of the year we would not take steps to increase our supply, for it is necessary to begin with adult ticks in the spring. These considerations in turn made it impossible to liberate very many parasites in the spring of 1927. In the spring months of 1927, steps were taken to obtain a supply of fully fed female ticks, fresh out of nature and in a normal, vigorous condition. These were obtained

(1) Submitted by Professor R. A. Cooley on March 1, 1928, prior to his leaving for South Africa.

from a locality near Flathead Lake believed to be free of any infection with human diseases. These females laid eggs which were incubated in thermal cabinets, and the larval ticks which hatched were fed on laboratory animals. When they dropped, fully fed, they were recovered from the ''infesting bags'' and were incubated. About 250,000 nymphs were thus obtained from the fed larvae by September, and are our stock of ticks for maintaining the stock of parasites and for the quantity liberations in the spring of 1928.

In the season of 1928 it will be necessary to again rear a supply of nymphal ticks for 1929 and to rear large quantities of the parasites for liberation.

The procedure of rearing parasites is as follows:

In the early spring the nymphal ticks are brought in from the stock out of doors where they have been held in tubes or cages under conditions as nearly natural as possible, and put on laboratory animals together with living, adult parasites which lay their eggs in the ticks. The ticks continue to feed to repletion and drop off in the infesting bags. They are recovered and held at favorable temperatures in thermal cabinets. Those ticks which have been parasitized show up and are picked out and put individually in ''tubes''; the others, not parasitized, develop normally and produce adult ticks.

In order to make the best use possible of the French parasite a large amount of experimental work and study has been laid out. The principal subjects under investigation are the following:

1. Can the parasite endure the severe climate of Montana? A few parasites were placed out of doors in the fall of 1926 and were examined in the spring of 1927 and found to be all dead. In the light of later findings not much importance is attached to this, however, as we do not yet know what the normal stage of the parasite in hibernation may be. Parasites in a full state of nature, having fed in ticks in nature, may adopt means for hibernation of which we have no knowledge. In the summer and fall of 1927 numerous parasites in various stages of development were placed out for observation under a variety of field conditions, and in the spring of 1928 we may have results of value.

2. Can this parasite adapt itself to the habits of our wood tick? We cannot say positively what part of the world the parasite came from nor what its original tick host or hosts may have been. There is a considerable variation in the habits of ticks and much may depend on the adaptability of the parasite. We have given careful attention to the effects of various temperatures on the development of the parasite and to the responses to biological factors, and some observations of great interest have been made. The parasite feeds only in the fed nymph, so far as we have yet been able to discover. It follows, then, that the parasite can multiply in nature in Montana only during those spring and early months when the nymphs are feeding on their rodent hosts. This season is mainly over by July 15, though a lesser number may be found until about September first. It is possible that the parasites which, with favorable temperatures and a supply of nymphs to lay their eggs in, are capable of passing through several generations during a summer, may die off, wholly or in part, if unable to find ticks in which to lay their eggs. This, if true, would be very unfortunate, but already we have found substantial indications of an adaptation, hitherto unsuspected, which may wholly obviate this difficulty.

3. Is there another tick in Montana which the parasite can use as a host during the late summer and early fall, and what would be the effect of such a secondary host on the parasitism in the wood tick? Instances are common in nature in which a secondary host is of much value in completing the needs of a valuable parasite. We have other ticks in Montana the life histories of which should be studied in this connection. Dependent upon developments in the future we intend, if possible, to make such studies.

4. How rapidly may the parasite multiply and overcome the ticks in a given locality? It is of much practical interest to know not only if the parasite can survive our climate, both in summer and in winter, and adapt itself to the habits of our ticks, but also to know how rapidly it is able to increase to effective numbers. On this would depend to a considerable extent our decision whether to liberate more or less parasites in any one locality. If fewer are to be placed in one colony, more colonies can be started. Accordingly, experiments have been started the purpose of which is to determine from year to year the percentage of parasitism in each colony established.

5. How far and how rapidly can the parasites spread from a given center of introduction? Again, it will be of interest to know whether from a given center of distribution the parasites will spread by their own means over many miles in say five years, or remain rather closely confined to the locality where originally introduced. Field men will collect ticks at measured distances by shooting and trapping their rodent hosts, and bring them to the laboratory where they will be held under observation for parasitism. It is assumed that the rapidity and distances will vary with the kinds of animals which are tick hosts in the several localities. Some animals, such as rabbits, should spread the parasites rapidly and far.

6. What is the most effective or least expensive method of liberating parasites? The laboratory method of parasitizing ticks is to turn the newly-emerged parasites loose in a closed bag of unbleached muslin with a small animal, usually a rabbit or a ground squirrel, on which ticks in the nymphal stage are feeding. The parasites hunt the ticks in the fur and around the eyes and ears and lay eggs in them. It is assumed that the parasites have a similar habit on wild animals out in nature. In liberating the parasites it is our purpose to place them where they can find animals and ticks. It is, of course, necessary to place the parasites out at the right time of the year. At the start, there was very little information to guide us in determining the best method. A series of experiments has been started for the purpose of getting this information. Following are some of the methods that have been tried:

a. Capture wild rodents and infest them with nymphal ticks in the laboratory and after parasitizing the ticks turn the animals loose again in the same locality where they were captured. It is expected that in this way parasitized ticks will be dropped in the very localities where we want them to be, and that the parasites when mature and ready to lay eggs may find the ticks on other animals.

b. Mountain rats abound in the vicinity of the "goat rocks" where the mountain goats are abundant and where the ticks are plentiful. Mountain goats are undoubtedly a very important host of adult ticks. It is well known that mountain rats, or pack rats as they are sometimes called, delight in picking up and carrying off objects that human beings leave around. Ground squirrels also do this to some extent. A piece of rag or cotton is attractive to them. Vials or "tubes" containing parasites, so arranged that the parasites can escape when they emerge or when disturbed, are attached to small balls of cotton. In their night wandering the pack rats, and during the day the ground squirrels, find these and carry them to their nests.

c. During the winter and in the early spring rabbits often find difficulty in getting enough food. Another experimental method is to establish feeding places where the rabbits gather and form a habit of returning. After the rabbits have become accustomed to the place, parasites are liberated in the immediate vicinity surrounding the feeding place so that the parasites can find the ticks on any animals that come for food.

d. Place tubes about to yield adult parasites in the vicinity of the nesting places and haunts of the various rodent hosts of ticks, such as mountain rats, woodchucks, ground squirrels, pine squirrels, chipmunks, and rabbits.

e. Pull the cotton plugs and leave tubes of parasites in the grass in likely places where, by chance, rodents may pass.

Our work on ticks in previous years has taught us that in different localities the rodents vary in importance as hosts of the early stages, and hence as producers of adult ticks which are to feed on the larger animals, wild and domestic. Since, in the mountainous country in Ravalli County, mountain goats are abundant and undoubtedly feed very large numbers of adult ticks, it naturally is of interest to discover what the rodent hosts are and then to find a way to cause the parasites to attack the ticks on them in particular. Special attention is being given to this point and since we know that the mountain rat is one of these hosts of young ticks in the "goat rocks," and perhaps the principal one, parasites are being placed in their haunts.

What Progress Is Being Made With Parasites?

It should be stated quite plainly that it is too early to state what the results of the work so far done on parasites may be. It will be doubtless many years before the full results may be known. However, in a few years we may have enough information to indicate rather reliably what the eventual results are liable to be. It should be borne in mind that the work is new and that there is only a very little of experience to guide us; that the State is large; that it may require time for the parasites to become progressively adapted to the new conditions; that the labor of breeding the parasites in quantity is tremendous; and, finally, that these small insects which may die off largely each winter, as most other insects do, may require years to multiply and spread. We realize that since we are entrusted with the expenditure of public funds it is necessary that we be informed as early as possible regarding what the local results of parasite liberation may be, so that we may judge reliably whether or not progress may be expected.

Origin of Tick Parasites.

A study of all that is known regarding this very small and highly specialized group of tick parasites has shown it to be very probable that it originated in Africa. We believe that the three known species all came from that continent, and yet no special effort has ever been directed at finding tick parasites there, although much work on the ticks of Africa has been done. The tick parasites that we know of all have been discovered by chance in connection with other work. Plainly, it is desirable to make a search for other species of tick parasites in Africa. Having now to our advantage a knowledge of how to look for them, there is a reasonably good chance that by going to the right places and looking in the right way, we may discover other species that may be introduced into Montana. We cannot venture an opinion either as to what the habits of such a new species may be or how the habits may vary from those of the parasites we now have, but other species of parasites of different habits, in combination with those species we now have, may be of the greatest value to us.

Again, it is very desirable to study the habits of these parasites in their native environment and on the same hosts they have been using for countless years. The habits of their tick hosts should be studied and correlated with the parasite's habits. It is believed that information of this character may be of great value to us. We know that in 1909 Mr. C. W. Howard discovered what we have called the Texas species, in Portuguese East Africa, and I have recently learned through Mr. C. P. Lounsbury of Pretoria, Transvaal, that some years ago parasites were recovered in Natal from some ticks that were under observation. These parasites are now in a museum in South Africa and have never been studied. They may be new.

It is for these reasons that the writer in taking his sabbatical leave from the University, has decided to go to South Africa for the purpose of studying tick parasites. The expedition may be briefly outlined as follows: Leaving New York on April 12, 1928, the first stop will be made in England where some time will be spent with Doctor George H. F. Nuttall at Cambridge, a prominent authority and writer on ticks, who has very large collections and who has done field work on ticks in Africa. It is very desirable, so far as possible, to recognize ticks as they are collected and the time in England will be spent in studying Doctor Nuttall's collections. In France some time will be spent in the study of ticks with Doctor E. Brumpt in Paris, who also has extensive collections and has collected and done field work in Africa. Particular attention will be given to the French parasite of ticks for it was in Doctor Brumpt's laboratory that the species was studied. As much information as possible will be obtained on the habits of the parasite in nature, its seasonal history, and in correlating its habits with those of the tick it preys upon in France.

In South Africa it is expected that laboratory facilities will be obtained in one of the Government laboratories and that the work will be a combination of field and laboratory studies. The major part of the work will be in the field where the hosts of the early stages of ticks will be shot or trapped and examined for ticks. Any ticks recovered will be taken to the laboratory for study and an effort will be made to find a way to bring them to America alive.

A Second Tick Parasite.

Knowing that back in 1908 a tick parasite (*Hunterellus hookeri* Howard) had been discovered in Texas by workers in the U. S. Bureau of Entomology and described by Doctor L. O. Howard, a letter was written to Mr. F. C. Bishopp, who at that time was in charge of the branch laboratory of the Bureau at Dallas, Texas, asking if a supply of living parasites of the Texas species might be obtained through him. Mr. Bishopp very kindly sent me two lots of ticks (*Rhipicephalus sanguineus* Latreille) collected at Brownsville, Texas, stating that they would probably yield some parasites. A total of 192 nymphal ticks were received and 13 of these yielded adult parasites, or 6.7 percent. Thus we came into possession of a second living tick parasite, though our supply was small and precarious.

A laboratory study of this parasite was made by Mr. H. P. Wood of the Bureau of Entomology and an article was published in The Journal of Economic Entomology(2). This was the first published article on the habits of a parasite of this group and contained much information of value. A study of this article reveals that the habits and life history are remarkably like those of the French species, as later published by Doctor Brumpt, and as further studied by us. Mr. Wood was able to cause it to parasitize the American Dog Tick (*Rhipicephalus sanguineus* Latreille) but he was unable to cause it to develop in our northern tick (*Dermacentor andersoni* Stiles), or in any other tick. Mr. Wood also sent some of the living parasites to Mr. C. P. Lounsbury in Cape Town, South Africa; C. W. Howard, Lourenco Marquez, Portuguese East Africa, and to P. Silvestri, Portici, Italy. In no case were these workers successful in their attempts to rear the parasites on ticks.

Our supply of living parasites from Texas was sent over to the Hamilton laboratory. Mr. Glen Kohls, who was locally in charge of the parasite work, undertook to rear them on our wood-tick or Spotted Fever tick, adopting the same methods that are used in rearing the French species. For all time we must give credit to Doctor Brumpt for, with great labor and at much expense, having worked out a way of keeping the French species alive in the laboratory and causing it to breed and multiply. During something over thirteen years he must have kept a strain of this parasite alive, for he published on it in 1913 and sent living insects to Naushon Island, Massachusetts, in 1926.

Mr. Kohls' efforts were successful. While at first the insects did not lay eggs freely in the wood tick, successive generations took more readily to the new habit and now (February, 1928) in less than one year, there appears to be no more difficulty in keeping the species alive in the laboratory and in causing it to multiply at will than in the French species.

Clearly the two species are closely related and very much alike in their habits, and the fact that *H. hookeri* Howard has been accommodating enough in so short a time to modify its habits for us is very encouraging, for it is very likely that it may be very desirable to modify the habits of both species in other respects in order that they meet our needs.

We are now able to multiply both species at will in the laboratory, the only limitations appearing to be the supply of ticks and the labor necessary, and since the nymphal ticks can be reared in the laboratory if enough labor is employed, the number of parasites is limited only by the money expended.

We cannot emphasize too strongly the necessity for an extended study of all the factors that have a bearing on the parasite problem. We must know as much as possible about the habits of the parasites, the habits of the ticks, the habits of the animals that are tick hosts, effect of climate and numerous biological factors, and be constantly guided by new findings. Before liberating a second parasite in nature, it will be necessary to determine as accurately as possible what the effect will be on the first species liberated. Instances are

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(2) Wood, H. P. "Notes on the Habits of the Tick Parasite *Hunterellus hookeri* Howard." Journ. Econ. Ent. IV, 1911, pp. 425-431.

on record in which mistakes have been made by liberating parasites before determining their habits. We shall not at present liberate the Texas species in Montana but shall first make a careful study of it in the laboratory.

More Than One Parasite Desirable.

It is apparent that it is desirable to have more than one species of tick parasite operating on our wood-tick just as it is desirable to have more than one insectivorous bird feeding on our insect pests. Different parasites working in different ways or at different times of the year, or even all working in the same way, would be an advantage. Much experience on other insect pests in other states and countries confirms this opinion. The standard practice is to introduce as many as possible of the parasites that appear to promise benefit. Hawaii leads the world in the control of insect pests by the use of parasites, and, in an effort to control relatively very few troubles, has introduced upward of ninety parasites and predators. It is commonly said of Hawaii that "parasites have saved the Islands," which is literally true.

Montana, then, in attempting to bring the tick menace under control by the use of parasites, and in seeking for other species, even in Africa, is following standard methods and is backed by abundance of precedent.

Where Parasites Are Being Liberated in Montana.

The liberation of parasites was begun in the spring of 1927, in the mountain canyons bordering on the Bitter Root Valley in Ravalli County. As has been explained above, only a limited number of parasites was liberated during that year because we did not have the nymphal ticks on which to feed the parasites and could not get them without rearing them in the laboratory from adult ticks obtained in the spring. During the spring and summer of 1928 greatly increased numbers of parasites will be liberated in Ravalli County and colonies will be started in the lower Bitter Root Valley and in the mountains adjacent. As rapidly as conditions warrant and we are able to do it, other colonies will be started in various parts of the state.

The following localities have been provisionally decided upon for new work during 1928: In the upper part of the LoLo Valley; in upper O'Brien Creek, west of Missoula; in Owl Canyon four miles northeast of Bozeman; near Helena; in Musselshell County; near Miles City and Forsyth in eastern Montana; in Carbon County, and in either Fergus County or Garfield County. By referring to a map of the distribution of cases of Rocky Mountain Spotted Fever in another part of this report, it will be observed that the locations for parasite colonies have been selected where a considerable number of cases of Spotted Fever have occurred. It will be quite impossible to give as much attention to these scattered colonies as will be given to localities close by the laboratory. In the process we hope to learn what measure of success may be expected from such more or less hasty attempts in remote places. Until we have gotten much more information on the habits of the parasite and on methods, it will be necessary to concentrate our efforts mainly on the experimental colonies close by the Board's Laboratory in the Bitter Root Valley.

PRELIMINARY REPORT ON THE TICK PARASITE, IXODIPHAGUS CAUCURTEI DU BUYSSON.

By

R. A. COOLEY, *Secretary, Montana State Board of Entomology*[1]

Review.

The three known Chalcidid parasites of the Ixodoidea constitute a tribe, the Ixodiphagini, belonging to the sub-family Encyrtinae and family Encyrtidae. The genera and species are as follows:

Ixodiphagus texanus Howard
Hunterellus hookeri Howard
Ixodiphagus caucurtei du Buysson

In 1907 Doctor L. O. Howard[2] described the first known Chalcidid parasite of a tick, *Ixodiphagus texanus,* from specimens reared from nymphs of *Haemaphysalis leporis-palustris* Pack., which were received on March 15, 1907, from Mr. J. D. Mitchell, Victoria, Texas. The ticks had been taken from a cotton-tail rabbit and a jack rabbit.

In 1908 Doctor Howard described the second species, *Hunterellus hookeri*[3], from specimens reared by W. A. Hooker from numerous nymphal specimens of the Brown Dog Tick, *Rhipicephalus texanus* Banks, which has been collected by Mr. H. P. Wood from a Mexican dog on April 20, 1908, at Corpus Christi, Texas.

In 1912 the learned French entomologist, R. du Buysson[4], described *Ixodiphagus caucurtei* from specimens discovered by the eminent E. Brumpt, Professor of Parasitology of the Faculty of Medicine of the University of Paris. Doctor Brumpt's specimens were reared from nymphs of *Ixodes ricinus* Linn., which he had taken near Paris at Chantilly and Fontainebleau from deer.

Regarding the first species described (*I. texanus*) nothing has been contributed since the original publication. This insect has never been re-discovered.

Mr. H. P. Wood[5] in 1911 published the results of a laboratory study of *H. hookeri* and recorded important findings. He records the surprising fact that C. W. Howard had found the same species of parasite attacking nymphs of *Rhipicephalus sanguineus* Latr., at Lourenco Marquez, Portuguese East Africa, in 1909. By letter dated February 1, 1928, I have learned from Mr. C. W. Howard that his specimens of *H. hookeri* were reared from *Rhipicephalus evertsi* taken from a dog. The letter further states that the parasites emerged from the adult ticks. This is surprising and is the only case on record of tick parasites emerging from adult ticks. The parasites were very numerous and were found in only one lot of ticks. Identification as *H. hookeri* was made by Doctor L. O. Howard.

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(1) Submitted by Professor R. A. Cooley on March 1, 1928, prior to his leaving for South Africa.
(2) Howard, L. O. "A Chalcidid Parasite of a Tick." Ent. News XVIII, pp. 375-378, 1907.
(3) Howard, L. O. "Another Chalcidoid Parasite of a Tick." Can. Ent. XL, pp. 239-241, 1908.
(4) DuBuysson, R. "Un Hymenoptere Parasite des Ixodies." Archives de Parasitologie XV, p. 246, 1912.
(5) Wood, H. P. "Notes on the Life History of the Tick Parasite." Journ. Econ. Ent. IV, pp. 425-431, 1911.

Mr. Wood states that two nymphs of *Dermacentor parumapertus marginatus* Banks were taken "on a jack rabbit (or dog)" by Mr. McClure Lewis of Green Valley, California, and received at the Bureau's Dallas laboratory on June 18, 1910. One of these yielded *H. hookeri*. The author records the following additional localities where *H. hookeri* had been taken: Brownsville, Texas, and Monterey, Mexico. Of the Monterey specimens it is stated that the adult parasites were never obtained and that the identification had been made from immature specimens. If another species of tick parasite is ever found in that locality this identification must be brought in question.

To the records of geographical distribution of *H. hookeri* as given above are to be added the following, very kindly given to me by Mr. S. A. Rohwer of the U. S. National Museum:

1. Coimbatore, South India, February, 1921, on bull terrier dog (E. Ballard and Fullaway) det. Timberlake.
2. Two slides in tray with above, but slides not bearing determination. Indo-China. Bred from dog tick (E. Roubaud). Letter Feb. 17, 1913.
3. Vedado Havana, Cuba, ex nymphal ticks. (Dr. Etchegoyhen). Received from N. S. Mayo. Det. Gahan.

Mr. Wood found that mating takes place immediately after the parasites emerge and that the females are ready at once to lay eggs in fresh ticks. To larval ticks the parasites were found to pay no attention though the record does not state whether the larvae under observation had been engorged. Attempted oviposition was observed in a male and an unengorged female of *R. sanguineus*. It was believed that "all that the parasite requires is a nymph at least partly engorged". Adult parasites were seen to imbibe sweetened water. In eight to fifteen days a characteristic striped appearance in parasitized nymphs became evident and in about forty-four days the adult parasites emerged.

The number of parasites per tick was found to vary between three and seventeen and the females were more numerous. The number of days required to emerge varied between 20 and 67, though the temperature was not recorded.

The writer was successful in his attempts to rear the parasites artificially on *R. sanguineus*, though only one generation was reared. It was suspected that more than one egg was laid at a single insertion of the ovipositor as more parasites than the number of insertions were observed to emerge when mature. Attempts to parasitize other species of ticks were unsuccessful.

In a stimulating and very valuable paper published in 1913, Doctor Brumpt records certain of his studies on *I. caucurtei*. In a certain case 17 out of 100 nymphs of *I. ricinus* recovered from deer were found to be parasitized, and in another, 10 out of 100. At Fontainebleau 2 nymphs of *Haemaphysalis concinna* Koch were found parasitized. Experimentally he was able to parasitize 95 out of 100 of *I. ricinus*; 90 out of 100 of *R. sanguineus* and 100 out of 100 of *D. andersoni* Stiles. The last named species is our wood-tick of Northwestern United States.

In France the adult parasites are to be seen from April to November. This author also finds that the parasites attack only the nymphs of ticks, either unfed or engorged, and complete development takes place in 4 to 6 weeks. In captivity the life of the adult parasite is not to exceed 8 days. No hyperparasites were found. Above a certain amount humidity was found to be very unfavorable. It is stated that the tick, *R. sanguineus,* is rarest in

France where this parasite is present and the author believes that the parasite is responsible. Doctor Brumpt particularly discusses the wood-tick and keenly analyzes the biological factors concerned and urges the desirability of attempting to control it by the use of the French parasite.

The foregoing summarizes rather completely all that has been published regarding tick parasites, with the exception of brief notes published in the Sixth Report of this Board which gave an account of the introduction of *I. caucurtei* into America, and preliminary notes published by the present writer.(6)

Summary of Published Records.

The following primary facts, which throw a considerable amount of light on these valuable parasites, may be gleaned from the record. Three species of tick parasites have been discovered, two from Texas and one from France. All are alike in having been found feeding in nymphal ticks only and the parasitized nymphs were all found feeding on their host animals. One exception must be recorded, that of Mr. C. W. Howard's finding of parasites in adult ticks. The Texas species were both taken in the spring and the French species in the fall. The ticks from which the parasites were reared are *H. leporis-palustris, R. texanus, R. evertsi, R. sanguineus, I. ricinus* and *D. parumapertus marginatus*. Since being brought to America *I. caucurtei* has also been reared in *D. andersoni* and *Dermacentor variabilis* Say. The host animals of parasitized ticks taken out of nature are cottontail rabbits, jack rabbits, dog and deer, exclusive of laboratory animals at the Hamilton Laboratory.

The Egg and Egg Laying.

So far as the printed records show, the eggs of tick parasites have never been observed. As a preliminary to a search for eggs in parasitized nymphal ticks, the ovaries removed from females of *I. caucurtei* were smeared on a slide and stained with Giemsa's stain when the very numerous eggs became apparent. The egg is broadly ellipsoidal, 0.06 mm. in length and without markings. To one end is attached a stem or egg-stalk. This stem is not always evident in smears but may be coiled against the end of the egg where it appears as a small eccentric protuberance. See figure 6. The dorsal walls of the several slightly fed nymphs were removed, cleared and mounted. An examination showed the punctures made by the ovipositors but we have not been able to demonstrate the eggs in the ticks.

Our method of rearing parasites in the laboratory was learned from Doctor Larrousse on Naushon Island in 1926, and essentially the French method has been used ever since with some modifications. The newly emerged adult parasites are liberated from very small vials on laboratory animals which have been previously infested with nymphal ticks. Some observations have been made on the habits of the parasites in depositing their eggs. The parasites appear to be well adapted to finding the ticks in the fur of the host animal and in such nearly bare places as in the ears and around the eyes. The objects of their search are recognized at once and generally the ovipositor is inserted almost immediately on finding a victim. The act of ovipositing takes but a few seconds, and a parasite after ovipositing once in a tick generally goes away in search of another, though rarely the act is repeated in the same tick. Two parasites have been observed ovipositing in one nymph at

(6) Cooley, R. A. "Tick Parasites." The Med. Sentinel, XXXV, (1927), pp. 805-815. (Portland, Oregon).

the same time, and it is not uncommon for more than one female to lay eggs in one nymph. Mr. Kohls[7] placed one female parasite with one unfed nymph in a small vial and watched them for two and one-half hours, during which period he observed the insect to insert the ovipositor in the tick forty-seven times. The observation was then discontinued. Doctor Brumpt found that at each insertion of the ovipositor the female lays from 5 to 10 eggs. Mr. Wood in his study of *H. hookeri* reports having found 13 parasites in a nymph that "had been parasitized only about three times—in no case as many as 13 times." No exact work has been done to determine how many eggs a female may lay but the number must be large for she works rapidly and the supply of eggs in the ovaries is very large.

Egg Laying in Fed Larvae.

In the summer of 1927 Mr. Kohls conducted an incidental experiment that yielded very interesting results. The supply of nymphs of *D. andersoni* is produced by feeding larval ticks on laboratory animals. There is thus a plentiful supply of fed larvae on hand during the summer months. One pill box of recently fed larvae, which had dropped on August 20 and amounting to about 350, were placed in a small glass jar with about 90 adult *I. caucurtei* (males and females mixed) and set in the sunshine at room temperature for a period of 3 hours. Following the exposure to parasites they were placed in a thermal cabinet at 19°C., which is the usual procedure for rearing ticks. By September 22 all of these fed larvae had "molted" to the nymphal stage. They were then put in a longevity tube and placed out of doors in the "tick yard" under conditions simulating nature. On November 11 these nymphs were put on a rabbit in the laboratory for feeding. In order to activate the nymphs they had been brought into the laboratory two weeks earlier and held at room temperatures. Seventy-eight fed nymphs recovered from the rabbit were placed at 19°C. and were examined from day to day. On December 5 it was noted that several showed the characteristic mottling indicating parasitism and eventually 32 nymphs, or 41.02 per cent of the whole number, were isolated as parasitized. From these, 9 yielded adult parasites in due time. While 9 out of 32 is a small number to come through to the adult condition, there may be and there may not be special biological significance in the fact for it has been found that occasionally a lot of parasites which appears to be normal may fail to emerge and we attribute the failure to faulty conditions of humidity or temperature.

The foregoing shows that under the conditions stated;

a. *I. caucurtei* will lay eggs in freshly fed larvae of *D. andersoni.*

b. While it is true that in the case of eggs deposited in nymphal ticks, development of the parasite begins promptly in suitable temperatures and will proceed to the maturing of the parasites, in the case of eggs deposited in fed larvae the development is delayed.

c. The living parasite is carried through the quiescent period of the fed larva and is alive in the next stage.

d. The parasite remains alive through a resting period of the unfed nymph prolonged for 50 days (September 22 to November 11).

e. Upon the nymph being fed the parasites will develop to maturity.

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(7) Mr. Glen M. Kohls, a Junior in Entomology at Montana State College, was employed by the Board of Entomology for one year beginning April 1, 1927, and was placed in local charge of parasite work at the Hamilton Laboratory.

Egg Laying in Other Tick Stages.

It is well established that, under laboratory conditions at least, adult females of the French species will lay eggs freely on nymphs of various species of ticks while the latter are attached to host animals. It may be the usual habit of the parasite to seek out tick hosts and to search them for nymphal ticks. Such a habit is strongly indicated as the normal one. In the practical work of establishing colonies of parasites, however, it would appear to be unwise to neglect the possibility that there are other ways in which eggs are normally laid. In fact, the information so far gained points to the desirability of experiments and studies that will reveal much more regarding the habits of these insects. It is particularly desirable to determine if there are adaptations by which the species may survive periods of the year when nymphal ticks are not to be found on their hosts. We now know that when confined in vials the female will lay eggs in fed larvae, flat nymphs and fed nymphs. Wood has observed attempted oviposition "in both a male and unengorged female of *R. sanguineus.*" Mr. Wood does not state whether these adults of *R. sanguineus* were later fed and observed for parasitism. It seems to be reasonable to assume that in a state of nature the parasite may occasionally lay eggs on unattached ticks, yet when one considers the habit which the nymphs have of waiting on the vegetation for rodents to pass within reach, in many species at least, it is realized that the parasite may have better success in finding ticks in considerable numbers by imitating the ticks in searching for rodents or other animals.

It should be borne in mind also in further work on the biology of these exceedingly interesting parasites, that egg laying in stages other than the nymph may be connected in some way with special adaptations which may be necessary in the full round of the insect's biology.

Development of the French Parasite.

The act of ovipositing appears to have little or no effect on the health and vigor of the tick. Feeding on the host continues as usual and when fully fed the tick drops off. In the rearing of parasites unit lots of flat nymphs of about 250 each are used on each laboratory animal, and on an average of about 75 fully fed nymphs are recovered from each cage bag. The feeding of these ticks is done in wire cages enclosed in bags of unbleached muslin, and the process is carried on in a room held at about 75°F. This method was devised by Doctor R. R. Parker some years ago when it was found that a high room temperature caused the ticks to feed more successfully.

The following table is condensed from the records made in rearing ten "Series," or generations, of parasites:

Series No.	No. Nymphs in Series	Host	No. Nymphs Parasitized	No. Ticks Produced Parasites	Per Cent Parasitized	Per Cent Producing Adult Parasites	No. Adult Ticks
1	124	Rabbit	112	90	88.7	83.5	6
2	228	C't'ntail Rbt.	224	190	98.2	84.8	1
3	81	Rabbit	34	28	41.9	82.3	21
4	46	Rabbit	0	0	0	0	41
5	254	Rabbit	166	123	62.9	74.0	68
6	154	Rabbit	26	16	16.8	61.5	106
7	139	Rabbit	55	37	41.9	67.2	53
8	98	Rabbit	22	10	22.4	45.4	58
9	37	Mtn. Rat	2	0	5.4	0	28
10	128	Mtn. Rat	75	57	58.5	76.0	57

These figures show that of the total engorged nymphs recovered from the bags an average of 48.53 per cent were parasitized, the per cent varying from 0 per cent in Series 4, in which a domestic rabbit was used, to 98.2 per cent in Series 2 in which a cottontail rabbit was used. The table further shows that the percentage of the fed nymphs to produce adult parasites varies from 0 per cent in Series 4 to 84.8 per cent in Series 2, and an average percentage of 71.8. With more experience we have been able to raise the percentage of parasitism, and 80 per cent is now common.

The parasitized and non-parasitized engorged nymphs cannot be distinguished up to the time they finish feeding nor for some time thereafter. In fact, the number of days between dropping and showing visible signs of parasitism varies greatly. We have no data on the length of this period out of doors but numerous rearings in which the parasitized nymphs were held at constant temperatures in thermal cabinets have shown the following results:

At 17°C., minimum 22 days, maximum 43 days.
At 22°C., minimum 12 days, maximum 48 days.
At 27°C., minimum 12 days, maximum 25 days.
At 32°C., minimum 12 days, maximum 31 days.

The highest percentage of nymphs to successfully complete their development and produce adult parasites is at 22°C., at the temperatures tried up to this time. This is not very far from the most favorable temperature for the parasite and an average of long series of rearings shows that the number of days from dropping to the first indications of parasitism at this temperature to be 18.49 days.

In the experiments summarized above we also included a parasitized lot at 12°C. This experiment was started on July 18, 1927, and at the date of this writing (March 1, 1928), none of the nymphs have yet yielded adult parasites. Signs of parasitism began to show up on September 28 and examinations made from time to time have shown others to show parasitism. On January 4, 1928, additional nymphs were found to be parasitized and on this date (March 1) the parasites are still alive and 12 more have just been "isolated" as parasitized. It is doubtful whether the parasites of this series (12°C.) will ever mature, but it is clear that there is great vitality in the species and it is indicated that it may survive exposure to lowered temperatures for a prolonged period.

At these same temperatures the minimum and maximum days from dropping to emergence of the adult parasites are:

At 17°C., minimum 76 days, maximum 97 days.
At 22°C., minimum 40 days, maximum 51 days.
At 27°C., minimum 33 days, maximum 44 days.

At 33°C., no adults emerged, the temperature being too high. Averaging the duration periods for all of the parasites reared at 22°C. we have 45 days, and in our experience in rearing very many parasites at this temperature in the practical work at the Hamilton Laboratory, we have found that we may depend upon the bulk of the parasites to emerge in close to 45 days.

The signs of parasitism, to which reference is made above, is a very characteristic mottling or striping shown in figure 8. Laying open the nymph just at this stage, by fastening to a small block of paraffin and running the very fine point of a small scalpel around the border, one finds that the appearance is caused by the larvae showing through the now translucent body wall. The larvae at this time are very dark from the contents of the intestinal tract, which is made up very largely of the blood which the tick had drawn from its host. The entire contents of the tick is now within the parasites and the entire cavity is occupied by the larvae. The tick must have died some time earlier, or generally in something less than 18 days at 22°C. The ticks are alive and able to crawl for some days after dropping, which for our purpose is fortunate for thereby the nymphs are able to hide away when in a state of nature and provide places more suitable for the parasites than would be found on the bare surface of the ground. The tick is seeking a suitable place for passing the quiescent period before emerging to the adult tick stage but in so doing it is providing a measure of protection for the parasites. The adult ticks and adult parasites thus arise from the same places.

A parasitized nymph opened before the external mottling has appeared reveals the larvae imbedded in the semifluid visceral mass from which they may be separated if the mass is spread out. Their size naturally depends upon their age, and the difficulty of finding them is proportional to their size. We have as yet been able to discover almost nothing of the processes of development and the early effect on the tick host up to the time that the larvae may be easily seen.

Beginning at the time the larvae may be found in the wet mass of the viscera the process, as observed, is as follows: The larvae continue to feed, gradually reducing the body contents until it is all gone, only the empty shell remaining. Up to this time the larvae apparently do not void anything from the intestinal tracts. The body is semi-transparent, excepting the intestinal tract, and under a moderately high-powered binocular microscope the brown or blackish granular intestinal contents may be plainly seen through the body wall. During several days the shell of the dead tick contains only the distended larvae; then the larvae commence voiding the intestinal contents in the form of numerous dark brown pallets, each one of which is enclosed in a clear, glistening membraneous covering. Among these are to be found a smaller number of clear white or translucent pellets, otherwise like the dark ones. These pellets are piled all in one place, generally, though rarely the pile is divided. The mass of fecal pellets is most often piled in the anterior end of the cavity. The process continues until the intestinal tracts are emptied, when the larvae appear as whitish or translucent maggot-like grubs disposed in one part of the cavity with the mass of feces in the other part. The larvae next arrange themselves in parallel positions with their anterior ends directed away from the pellets, in which position they pupate leaving the pupae similarly arranged.

The size of the larvae is inversely proportional to their number in a single tick and the size of the resulting adults correspondingly varies. It is evident that the larvae must consume all of the semifluid mass in the host tick in order to be successful in completing their transformation and expanding their wings. Clearly there is here a very nicely adjusted adaptation and one is impressed with the neatness of the operation. The pure white pupae occupy one part of the cavity while the pile of fecal pellets occupies another, and when the pupae emerge to adults there is space in which they may expand their wings. The mass of moist pellets probably also assists in maintaining a proper degree of humidity in the cavity. In a few rare instances we have observed small amounts of unconsumed viscera in the cavity of the tick's body.

Many times on opening nymphs in which the development of the parasites had been delayed, we have observed the larvae to be below the average in number, though the duration of the period of development is apparently not just inversely proportionate to the number of parasites.

The appendages of the insects, legs, wings and antennae are evident in the naked pupae as would be expected in this group of insects. See figure 3. While at first white, the pupae gradually darken and a reddish pigmentation appears in the eyes and ocelli. A single emergence hole is gnawed through the body wall and the mature insects file out and preen themselves. Mating takes place immediately.

It is possible that the adaptation in the species whereby all of the contents in the tick's body is consumed before further development proceeds will explain why the parasite is found only in nymphal ticks. If it is a fact that a considerable amount of the wet visceral mass left over in the tick would be an interference in preparing a clear space in which to pupate, or would upset the balance in the humidity, the species may have adapted itself to a food mass just large enough to be consumed. It is quite conceivable that the food mass in a fed larva is too small and that in a fed adult is too large.

The Number of Parasites per Tick.

Numerous counts of the number of parasites to emerge from a single nymph have been made. It should be borne in mind that these counts have been made only in connection with the laboratory rearing of parasites. The number per nymph in nature may be very different. The number of parasites to emerge from a single tick under these conditions, as based on 100 nymphs taken at random from routine rearings, was found to vary from 4 to 66 and averaged 21.19. The lowest number ever found was 2 and the highest 73. The size of the insects naturally varies inversely in proportion with the number to derive their food from one tick.

Seasonal History of the Parasite.

Having as yet had no experience with *I. caucurtei* in nature we can venture no opinion on what the seasonal history may be. However, certain experiences in the laboratory appear to have a bearing and are important.

In the Fall of 1926, soon after we obtained our initial supply of the living parasite, we attempted to hold our stock at various lowered temperatures hoping that it would become inactive and survive until spring when the adults might be liberated out of doors in our experiments. We did this to conserve our limited supply of nymphal ticks and to save labor. By December it was found that the parasites, which were principally in the pupa stage, were dying off rapidly. Much care had been taken to maintain the correct humidity and yet the parasites were found to die if held too long at a low temperature (12° and 17°C.), or they would go on with their development and emerge if held at higher temperatures. Since that time we have found it to be more expedient to keep the parasites breeding all winter. In more recent experiences, as reported on an earlier page, we have found that development goes on, though slowly, at as low a temperature as 12°C.

Up to the present time it appears that the species is primarily adapted to continuous development throughout the year and that it has not developed a hibernation adaptation, at least of the more usual kind. If this is true it is indicated, in turn, that the insect is adapted primarily to a tropical or sub-tropical climate in which case it was probably introduced into France instead of being strictly native there.

Longevity of Adult Parasites.

Doctor Brumpt found that in captivity the life of the adult parasites does not exceed eight days. In our work on *I. caucurtei,* which has been done in considerable volume, we have frequently noticed that the adult dies at room temperature in two to four days. On September 24, 1927, 14 parasitized nymphs which had been held during development at 17°C. were, upon emergence, placed at 2° to 6°C. and observed for longevity from day to day. Under these conditions the period varied from 12 to 19 days.

In an experimental liberation in Blodgett Canyon started June 28, 1927, in which 50 tubes of parasites were attached individually to small balls of cotton and so arranged that when disturbed by rodents the parasites would be liberated, one vial was dropped from the cotton by the animal that disturbed it and the small cotton plug failed to come out. Just two weeks after the tube had been put out, a field man discovered the tube where it had been dropped and found all the parasites to be alive. These parasites in a small vial on the ground had survived for 14 days and might have lived longer. This happened in a small ''cave'' in the rocks inhabited by mountain rats, and the temperature at 11 a. m. on the day of the discovery, was determined as 9°C. This experience plainly indicates that the life of the parasites in a state of nature may be much longer than in the laboratory.

Correlation Between the Seasonal Histories of Dermacentor andersoni Stiles and Ixodiphagus caucurtei duBuysson.

In an attempt to colonize the French tick parasite on the prevailing tick of the Northwest region, a primary consideration of necessity must be a suitable relationship between the life histories or seasonal requirements of both species. The ticks in the proper stage or stages of development must be available to the parasites at the egg-laying periods of the latter species. Laboratory studies of the parasite, as already stated, have indicated that the species is not primarily adapted to a period of hibernation in the usual sense. Moreover, we have as yet discovered no indication that the parasite is adapted to a period of aestivation during unsuitable temperatures for development in the late summer months.

From the data at hand it appears that the parasite requires a continuous supply of nymphs in which to lay eggs during the summer months and that as cold weather comes on in the fall, the species will be required to become inactive without a special adaptation for hibernation through a protracted period of unfavorable temperatures.

Climate of Paris and of Montana.

Since *I. caucurtei* is known to have come from near Paris, at Fontainebleau and Chantilly, where it must be more or less well adapted, although it may have come originally from some other part of the world, it is of interest to compare the climate of Paris with that of points in Montana. Hamilton, Helena and Miles City are chosen for comparison. These points are fairly representative and parasite colonies are to be established near these points, if possible. The Board's laboratory is located at Hamilton and the principal experiments in colonizing the parasite are located nearby, in the mountains. Miles City is located some 475 miles eastward, in the eastern end of the state, well into the plains area. Helena is intermediate geographically and the climate is intermediate. The winter atmospheric temperatures in the mountains near Hamilton are lower than at the town which is in the Bitter Root Valley, but there is more snow in the mountains which has a very marked influence on temperatures at or near the surface of the ground, where the parasite must pass the winter. Precipitation records for the winter months at Hamilton and Miles City show similarity and yet it is a fact that more snow lies in the mountains near Hamilton than on the plains surrounding Miles City.

The normal climographs herewith presented are based on records obtained through the Weather Bureau of the United States Department of Agriculture and from "Handbuch der Klimatologie" by Julius Hann, 1911. Each climograph is compiled from records covering a considerable number of years, in no case less than twenty years.

The temperature is given in degrees Fahrenheit and the precipitation in inches. It will be observed that Paris has the mildest winter climate while Hamilton and Helena are intermediate and Miles City is coldest. Paris has a higher precipitation during the fall and winter months, while the points in Montana have a higher precipitation in May and June. In general, the climate of Hamilton is more like that of Paris than is that of Helena or Miles City.

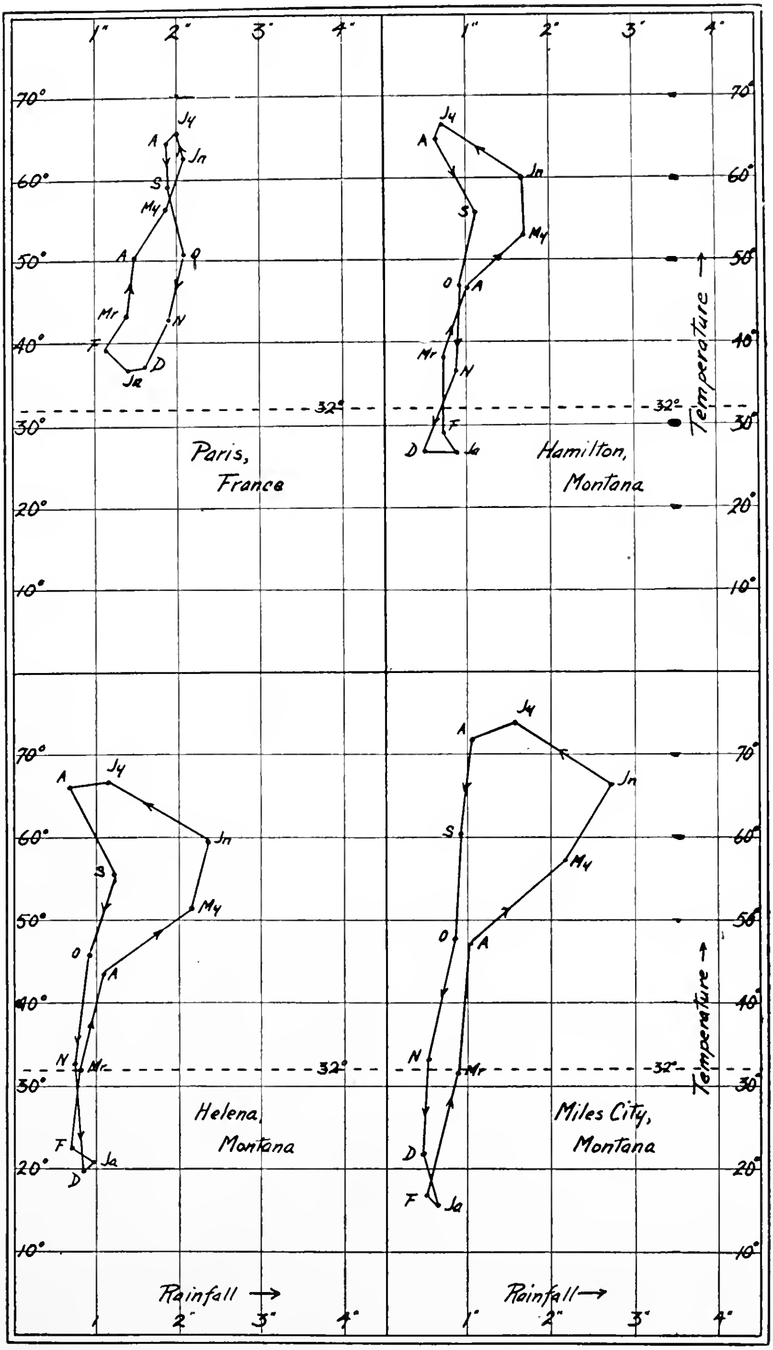
A table is presented which gives also the mean minimum temperatures and the lowest temperature records for the points covered in the climographs. It will be observed that the points in Montana have much lower air temperatures in winter than does Paris.

	Mean Minimum Temperatures				Lowest Temperatures Recorded			
	Paris (20 yrs.)	Hamilton (20 yrs.)	Helena (42 yrs.)	Miles City (41 yrs.)	Paris (47 yrs.)	Hamilton (20 yrs.)	Helena (42 yrs.)	Miles City (44 yrs.)
Jan.	32.4	17.5	12.1	3.4	1	–33	–42	–65
Feb.	32.7	18.9	14.5	5.3	4	–28	–41	–49
Mar.	36.3	26.9	23.0	19.6	12	–13	–20	–38
Apr.	40.5	33.6	33.4	34.7	26	8	6	– 7
May	46.2	39.4	41.0	44.1	29	18	22	17
June	52.0	46.3	48.3	53.7	36	30	31	33
July	55.4	50.4	54.1	58.9	42	36	36	31
Aug.	54.3	49.6	53.1	56.3	42	32	29	32
Sept.	50.0	42.1	44.3	45.5	33	28	20	16
Oct.	43.9	33.7	35.5	34.2	23	11	– 1	– 5
Nov.	37.4	26.8	24.3	21.2	5	– 9	–22	–26
Dec.	35.1	18.1	17.4	11.0	–14	–20	–40	–52

Extent of the Problem in Montana.

The wood tick is found more or less scatteringly throughout the greater part of Montana and cases of Rocky Mountain Spotted Fever have occurred in 32 out of a total of 57 counties, though in most of these counties the disease exists only in the milder form. It is desirable then that parasites may destroy the ticks throughout the State.

The State of Montana is as large as an empire, being the third largest of all the states. It is 560 by 310 miles in extreme limits. Within this are found numerous types of country with widely varying physical features, including rugged mountains, foothills and plains. Ticks are to be found in the most inaccessible mountain fastnesses, the mountain valleys and basins, the agricultural valleys and grazing lands, and in the plains country which is used for stock raising, or in some cases, left unused. If once started, ticks can find more or less favorable conditions wherever rodents occur as hosts of the early stages, and large animals, whether wild or domestic, as hosts of the adult ticks.


Paris,
France
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The problem of the use of parasites as destroyers of ticks is therefore a large one and moreover, the results of our study and experiments are applicable also in the states of the northwest and in Canada.

Description of Plates.

Plate 1.

Fig. 1. Adult parasites that came out of one tick, and the killed tick. (Magnified about 6 times.)

Fig. 2. Adult parasites magnified about twelve times. The variation in the size of the parasites is well shown.

Fig. 3. Pupae about ready to emerge as dissected from a killed tick. Magnified about twelve times.

Plate 2.

Fig. 4. Two dead nymphal ticks cut open to show the parasites in a natural position. The one at the left shows the fecal pellets at the lower end. The pupae at this stage are pure white.

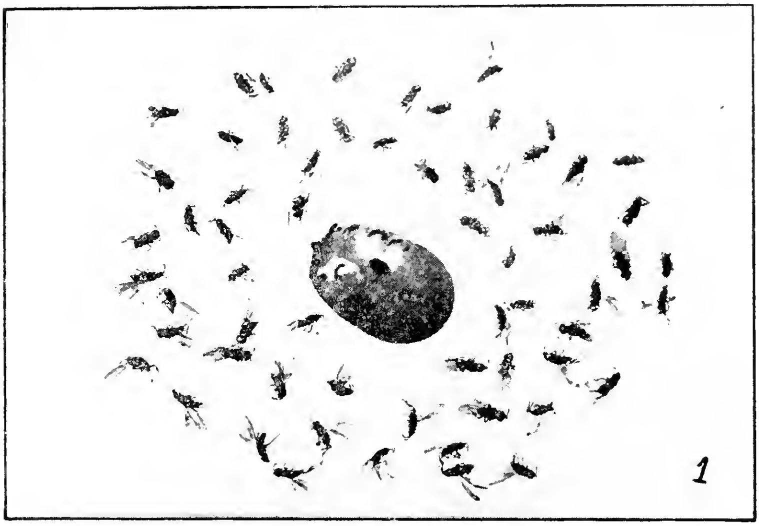
Fig. 5. Nymphal tick cut open and showing the entire body cavity of the tick occupied by the larvae of the parasite. The viscera of the tick have been completely consumed.

Fig. 6. Eggs of the parasite as they appear when removed from the ovaries of the adult female.

Plate 3.

Fig. 7. The larval parasites removed from one killed tick.

Fig. 8. Nymphal ticks showing "signs of parasitism." The mottling is due to the parasite larvae showing through the thin body wall of the tick. This stage soon passes after which the killed nymphal tick is brown in color.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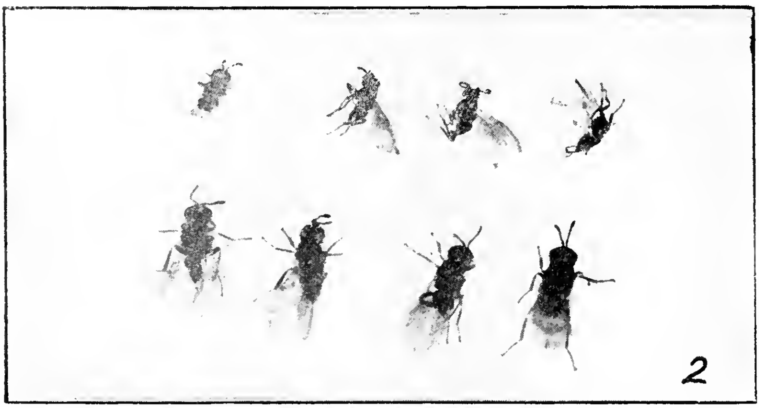
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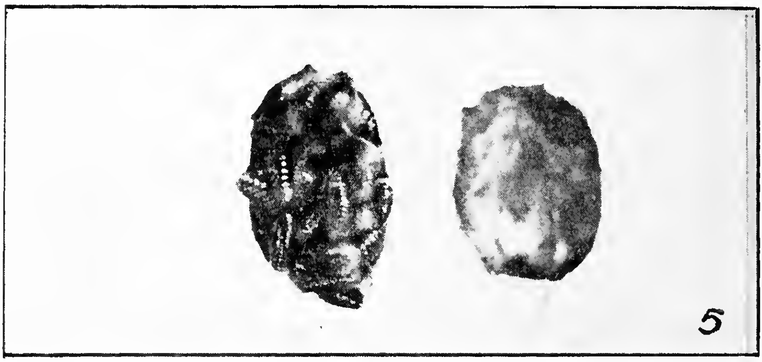

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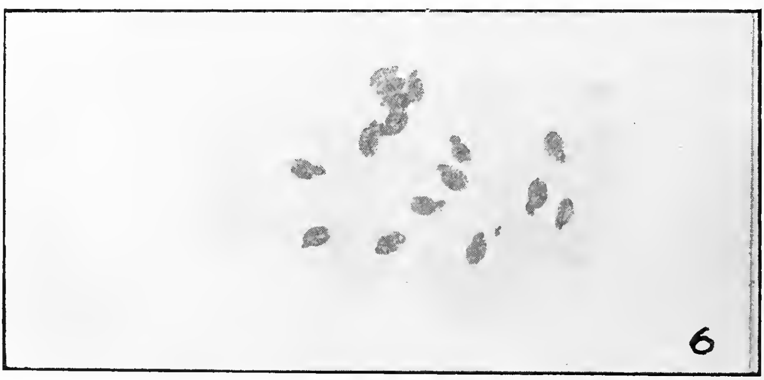

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QUANTITY PRODUCTION OF TICK PARASITES

By

FRED A. MORTON, *Laboratory Assistant, Montana State Board of Entomology.*

The Montana State Board of Entomology, in its laboratory at Hamilton, Montana, is rearing large numbers of the French tick parasite (*Ixodiphagus caucurtei* du Buysson) for liberation throughout the state. It is a Chalcidoid parasite (sub-family Encyrtinae) which attacks nothing but ticks, and so far as is known, develops in only the nymphal stage of ticks. Consequently, rearing the parasite necessitates rearing a suitable tick to the nymphal stage, and parasite production is naturally limited by nymphal tick production.

Technique of Tick Rearing.

In this laboratory the Spotted Fever tick, *Dermacentor andersoni*, is reared as a host for the parasite and since that requires rearing the tick through all stages of its development before parasites can be produced, the technique of tick rearing will be considered first, under the following headings: (1) The Adult Stage, (2) The Egg Stage, (3) The Larval Stage, and (4) The Nymphal Stage.

1. THE ADULT STAGE.

(a) Pre-engorgement Period. The flat adults are quite susceptible to both excessive and insufficient humidity, but may be kept for about a year with comparatively low mortality if placed under satisfactory conditions. The method now in use is to place the ticks inside a "longevity tube," which is a pyrex tube (1½ in. by 8 in.) plugged with clay at one end for a distance of about 2 in., a piece of muslin being tied over the other end. A few pieces of dried leaves or shavings are placed inside the tube so the ticks will not be forced into direct contact with the wet clay plug. The tube is then placed in a 5 in. vertical hole in the tick yard, which keeps the clay plug in the tube always moist, and allows the ticks to seek the humidity within the tube which is most satisfactory to them.

Usually, after the ticks have been held in the longevity tube for 3 or 4 months, they will be found ready to feed but sometimes it is necessary to incubate them. In this event they are placed at a high temperature (30°-35° C.) for several days, and then at a lower temperature (15° C. or colder) for a day. After several such alternations of temperature the ticks will generally engorge quite rapidly. This type of incubation is nearly always required for ticks less than 2 months old.

(b) Method of Engorgement. For a period of several months in the spring of the year, it is possible to purchase engorged female ticks from stock owners who reside in localities where ticks are not known to be carriers of human disease. These ticks lay eggs quicker than those engorged in the laboratory, but they lay fewer eggs, probably because the laboratory stock can be engorged to a larger size. However, it is not safe to depend on being able to purchase engorged ticks, and it is necessary to feed a considerable number of adult ticks in the laboratory.

Rabbits are used as host animals in engorging adults. The rabbit's belly is clipped closely and two screen capsules are fitted to it by means of adhesive tape. The capsules are circular, about 3 inches in diameter, and are stamped from 20 mesh brass screening by a die, in such a fashion that the finished capsule has a circular depression ⅜ inch by 2 inches, surrounded by a ½-inch rim, somewhat the shape of a low crowned hat. The rim fits tightly against the rabbit's belly and the ticks are fed within the depression. About 40 flat ticks are put into each capsule, approximately half of which will be females.

(c) Post-engorgement Period. After the ticks have fed for 10 days the capsules are removed from the rabbit's belly and the ticks picked. Using the two capsules we get an average of between 25 and 30 engorged females per rabbit. Each female is placed in a shell vial (½ inch by 1 inch) which is then plugged with cotton and held in a humid thermal cabinet at 20° C., pending oviposition, which, at this temperature, occurs within 1 or 2 weeks. This time may be lengthened or shortened without injury to the tick, by using any desired temperature between 12° and 30° C. Greater extremes of temperature might not prove harmful, but they have not been used.

2. THE EGG STAGE.

The average engorged female tick lays between 5,000 and 10,000 eggs and at 20° C. has an oviposition period of about 3 weeks. The first eggs hatch when they are about a month old and the female dies soon after. Maintenance of the proper humidity is quite important with eggs, although a certain percent of them remain viable under considerable desiccation.

3. THE LARVAL STAGE.

(a) Pre-engorgement Period. The vials of eggs are examined every three or four days and those in which the eggs have started to hatch are transferred to another tray and a record kept of the date on which the larvae, or seed ticks, were noticed. They may be fed from two to six weeks later, but if only two weeks old, it is preferable to incubate them for several days at 30° C. prior to engorgement. Seed ticks are more susceptible to the lack of humidity than any other stage of the tick and will die within a few days if subjected to ordinary room humidity at room temperature. With a high humidity they will survive at 30° C. for a month or more.

(b) Method of Engorgement. Rabbits are used as hosts for seed ticks and are infested in the following manner: The rabbit is placed within a 12-inch by 15-inch muslin "infesting" bag suspended by hooks, and the flat seeds from two or three vials deposited in his ears, the favorite site of attachment. This is easily accomplished by tapping the vial with a small block of wood; the seeds cling to each other and the vial may be easily emptied, as the mass will come out almost as a unit. A piece of tape is then placed around the rabbit's ears to prevent his shaking the seeds out before they become attached; the bag is tied with a piece of string and then placed in a cage. The cage rests in a frame, around which another bag is tied. The tape is removed from the rabbit's ear 24 hours later and the infesting bag removed and burned. On the third or fourth day some of the seeds, having become completely engorged, detach themselves from the rabbit's ears and drop to the bottom of the bag surrounding the cage. Just before the seeds start dropping the bags are cleaned out, and they are picked by turning the bags inside out within a galvanized sheet metal container 27 inches square by 6 feet high, and shaken clean. A 14-mesh removable screen inside this device separates the engorged seeds from the trash, and the seeds roll down the funnel shaped bottom into a removable cup which has a screen, with meshes just sufficient to retain the seeds, soldered across the lower end. After all the bags are thus picked, the cup is removed and shaken to clean the seeds of small debris. It is necessary to pick these bags of seeds twice, either on the third and fifth or the fourth and sixth days. We obtain approximately 3,000 engorged seeds per rabbit.

(c) Post-Engorgement Period. After picking, the seeds are measured in cubic centimeters and poured into longevity tubes, 12 or 15 cc. to the tube. These tubes are kept for about three weeks at room temperature with their clay plugs resting on moist sand. At the end of this time most of the seeds have molted to nymphs and may be transferred out doors to the tick yard.

4. THE NYMPHAL STAGE.

(a) Pre-engorgement Period. The flat nymphs are kept in longevity tubes for periods ranging from two to eight or nine months. They have been found to overwinter better out doors than when kept inside, although the mortality is quite high in either case, a 30% mortality being considered average when kept out doors. Nymphs present an added difficulty when it is desired to engorge them; they frequently refuse to feed even when several months old, and they are extremely resistant to known methods of incubation. The present system is to place the nymphs (which are at least six weeks old) at 30° C. for two or three days, and then at 6° C. for a day. This procedure is repeated until a fair number of them crawl up on to the cloth which is tied over the end of the tube. An average of about 15% of these will feed, while practically none of those will feed which remain in the trash at the bottom of the tube.

(b) Method of Engorgement. A longevity tube is selected which has a suitable number of active flat nymphs on the cloth. The cloth is removed and the nymphs on it smeared in the ears and around the eyes of a rabbit in an infesting bag. The rabbit is then treated as described under "Method of Engorgement of Seeds," and the engorged nymphs are picked in the same device as was used for the seeds, using, of course, a screen of larger mesh. Nymphs are picked on the fourth and sixth, or fifth and seventh days.

(c) Post-engorgement Period. The nymphs are measured in cubic centimeters and then put in cardboard pill boxes, about 50 nymphs to the box. In about a month they start molting to adults, and just prior to molting are transferred to longevity tubes, about 400 to the tube. They complete molting in the tubes and are then put out in the tick yard.

Technique of Parasite Rearing.

When parasites are to be reared the ticks are reared to the nymphal stage as already described, and are then parasitized by the following method:

From 12 to 24 hours after the rabbit has been infested with flat nymphs, 50 to 100 parasites are shaken into the rabbit's ears and around the head, and he is tied up in the infesting bag for 4 or 5 hours longer. The parasites immediately attack and lay eggs in the engorging nymphs.

These parasitized nymphs engorge, drop, and are picked exactly as normal nymphs and after being put in pill boxes are placed at 22° C. Three or four weeks later the parasitized nymphs may be easily distinguished from the others by the fact that the parasitized ones have turned brown, while the normal ones retain their blue color. The normal nymphs are allowed to molt to adults, and are placed in the tick yard with the other flat adults, while the parasitized nymphs are removed and the majority of them sent to locations throughout the state where it is desired to establish the parasite. A few are retained to keep a continual supply on hand for parasitization, and these are placed individually within a 2-inch piece of 6-millimeter glass tubing, cotton plugged at both ends. These are then placed at any desired temperature between 18° and 23° C., which are known to be safe limits. The tubes are looked over daily, and those containing adult parasites are either used immediately or placed at 6° C., where they may be kept with low mortality for about a week.

We obtain an average of 20 parasites per nymph, and a 70% parasitization. Parasites emerge from approximately 80% of the parasitized nymphs under conditions of fairly high humidity, but are unable to emerge if the nymphal shell becomes dry and brittle.

TICK PARASITE LIBERATION IN MONTANA DURING 1928.

By

J. R. Parker, *Acting State Entomologist, and*

W. J. Butler, *State Veterinary Surgeon*

The fight on parasites is universal; no country is free from them. Some are of lesser, others are of greater importance. Some are agricultural pests, others attack man and animals. Naturally, those parasites which are the most insidious and hardest to control have an intricate and complex life cycle. In such cases, control by medicinal or mechanical agencies is very difficult and ineffective. New methods continually have to be devised. Today, one of the most advanced methods of fighting and controlling many of our parasites is with parasites.

The Montana State Board of Entomology is commissioned with the duty of investigating and studying the dissemination by insects of diseases among persons and animals, with the purpose of eradicating and preventing such diseases. Thus far the study of the Board has been confined largely to the tick, *Dermacentor andersoni*, which is known to be a carrier of the causative agents of Spotted Fever, Tick Paralysis and Tularaemia.

The Montana State Board of Entomology has naturally been greatly interested in the tick parasite studies of Doctor Brumpt, Professor of Parasitology at the University of Paris, and particularly in the efforts of Doctor Brumpt's associate, Doctor Larrousse, to introduce the French tick parasite (*Ixodiphagus caucurtei* du Buysson) into the United States. In 1926 through the efforts of Doctor S. B. Wolbach, of the Harvard Medical School, Doctor Larrousse came to the United States and established a tick parasite colony on Naushon Island. Professor R. A. Cooley, representing the Montana State Board of Entomology, proceeded to Naushon Island and there reviewed and studied with Doctor Larrousse the methods used in rearing the tick parasite *I. caucurtei*. Through the courtesy of Doctor Larrousse, Professor Cooley obtained and brought to Montana 27 of these parasitized nymphs. This small initial stock has gradually been increased to the extent that during the summer of 1928 we were able to release 381,190 parasites and still hold enough in reserve to provide a much larger supply of parasites in 1929.

The tick parasite and its habits are described in considerable detail by Professor Cooley elsewhere in this report. It is sufficient to say at this time that it is a tiny wasp-like insect about 1/16 of an inch in length. The mature parasite does not sting or bite and is harmless to man and beast. The adult female's only concern is to lay eggs in the nymphal stage of the tick while the male's whole sphere in life is to fertilize the female's eggs.

When matured parasites are liberated they seek rodents and other small animal life that harbor the nymphal stage of *D. andersoni.* They then skirmish through the hair, or fur, of the animal in search of nymphs. Upon finding one, the ovipositor is inserted almost immediately and in most instances 5 to 10 eggs are laid. The act of ovipositing takes but a few seconds. The female parasite may repeat ovipositing in the same nymph, but ordinarily goes in search of another victim. Two parasites have also been observed ovipositing in one nymph at the same time, and it is not uncommon for more than one female to lay eggs in one nymph.

Mr. Kohls, at the Board of Entomology Laboratory in Hamilton, reports having observed one female parasite insert its ovipositor in one nymph 47 times. The eggs of the parasite hatch into tiny maggots which feed upon the tissues of the nymphal tick finally killing it. After the maggots or larvae are full grown they transform to pupae within the body of the tick where they remain until they emerge as adult parasites.

In distributing the tick parasite and in the endeavor to establish tick parasite colonies, the two methods most commonly used are as follows:

Ticks for the rearing of the parasite are gathered in districts where Spotted Fever, Tularaemia or Tick Paralysis have never been known to occur. Nymphs from these ticks are used for the breeding of parasites.

Parasitized nymphs are placed in glass vials open at both ends. Over the open ends wire gauze is attached. The mesh in the wire gauze is large enough to permit the escape of the parasites when they emerge from the nymphs. The wire gauze is simply an added precaution to prevent the loss of the nymphs, or their escape should one happen to be not parasitized and remain alive. These glass vials containing the parasitized nymphs are taken out into the country and placed at the mouth of rabbit holes, mice and rat runs and in and around the nests and holes of other rodents that harbor the nymphal stage of *D. andersoni.* Nymphs are not found on the common domestic animals. In some cases and especially in a pack or mountain rat country, cotton and bright articles are attached to the glass vials so as to attract the pack rats and have them carry the vials to their nests.

The second method is to trap rodents, take them to the laboratory and allow newly parasitized nymphal ticks to attach themselves to the rodent. The latter are then returned to the locality where they were caught and set free. After the nymphal ticks have completed feeding they drop from the rodents, crawl to protected places in trash or in rodent burrows. In such places conditions are ideal for the development of the parasites and when the adult parasites emerge they generally will have little trouble in finding tick infested rodents close at hand.

Areas in which tick parasites (*I. caucurtei*) were to be liberated were selected on the basis of previous distribution of Rocky Mountain Spotted Fever in Montana, with the end in view of establishing colonies of parasites in those areas where the disease had been most abundant. The selection of points within these areas at which the actual liberation of parasites was to take place was based on the abundance of adult ticks and various small rodents which act as hosts for nymphal ticks. In the selection of liberation points in Eastern Montana, Doctor R. R. Parker of the U. S. Public Health Service, who had formerly spent two years studying ticks in that section of the state, accompanied the State Board workers in their search for localities most suitable for the development of the parasite. His knowledge of Eastern Montana conditions saved many days of travel and gave assurance that the most desirable places were selected. Doctor Baldwin of Miles City and Doctor Smith of Billings, representatives for the Livestock Sanitary Board, aided the tick parasite work in every possible way. Their knowledge of local tick abundance was most helpful and they stood ready at all times to either transport the State Board workers or to receive and release shipments of tick parasites from the Hamilton Laboratory.

In the laboratory the number of parasites emerging from one nymphal tick varies from 2 to 73, the average of 100 nymphs taken at random in one experiment being 21. In estimating the number of parasites liberated we have thought it better to be conservative and have taken 14 as the average number of adult parasites emerging from each parasitized engorged nymphal tick. Localities, dates of liberation and the estimated number of parasites released are shown in the following table:

Tick Parasite Liberation During 1928.

Place	Dates and Number of Parasites Released	Total
	Bitter Root Valley	
Lake Como	May 24, 250; June 7, 4,920; June 22, 1,600; July 9, 2,400; Oct. 6, 30,000	39,170
Blodgett Canyon	May 22, 250; May 29, 2,680; July 10, 7,200; Oct. 30, 20,000	30,130
Lo Lo Canyon	Oct. 6, 30,000	30,000
Lick Creek	June 22, 2,400; July 13, 4,800; Oct. 18, 75,000	82,200
O'Brien Creek	June 15, 4,000; June 29, 4,800; July 13, 9,600; Oct. 6, 30,000	48,400
	Total	229,900
	Central and Eastern Montana	
Helena	April 30, 1,400; June 5, 700; Oct. 3, 15,000	17,100
Bozeman	June 2, 1,120; June 9, 700; Oct. 2, 14,000; Oct. 15, 7,000	22,820
Edgar	April 22, 336; May 9, 840; May 21, 280; July 16, 644; Aug. 20, 700; Sept. 23, 20,000	22,800
Musselshell	May 7, 420; May 22, 280; June 15, 2,000; July 4, 2,000; July 15, 644; Aug. 15, 560; Aug. 20, 700; Sept. 26, 40,000	46,600
Forsyth	May 8, 210; May 23, 280; July 16, 644; Aug. 20, 700; Sept. 24, 15,000	16,830
Miles City	April 21, 336; May 8, 420; May 23, 140; June 1, 700; June 16, 3,500; July 15, 644; Aug. 24, 700; Sept. 25, 18,000	24,440
Garfield County	June 2, 700	700
	Total	151,290
	Grand total for season	381,190

With reference to over-wintering, it is pleasing to report that Mr. Fred Morton, supervising parasite rearing work for the Board of Entomology at the Hamilton Laboratory, has advised the Board that he has recovered parasites from the Mill Creek hibernation experiment. The data in this experiment are as follows:

Twenty-eight nymphs dropped September 20, 1927, and 20 dropped September 23, 1927, were placed in two longevity tubes containing a few leaves and pine needles. They were still active and were held at 19° C., prior to being placed out doors on October 1, 1927. These tubes were placed on the ground and both ends covered with leaves. These tubes were brought into the laboratory on April 11, 1928. They were held at room temperature, and on May 23, 1928, matured parasites emerged from three of these nymphs. In this particular experiment parasites under out door conditions carried over the winter.

It is also encouraging and pleasing to note that Doctors Larrousse, Wolbach and King report that *I. caucurtei* survived the winter under natural conditions and were able to reproduce on Naushon Island. (Science LXVII, No. 1735.)

Professor Cooley, representing the Montana State Board of Entomology, but financed by private subscriptions, is now in Africa in an endeavor to secure other parasites for *D. andersoni*, and also to secure additional data on the present known parasites which may aid the Board in its work of eradicating, or at least controlling, in our most infested districts, the tick *D. andersoni*.

The work so far has been encouraging. With added experience which will come as the work progresses, together with the adequate laboratory facilities, which the State of Montana has so generously furnished, and with the data and parasites which Professor Cooley may bring home, we are justified in the hope that we may so reduce the tick population by the use of parasites as to make Spotted Fever, Tick Paralysis and Tularaemia rare diseases in the State of Montana.

ROCKY MOUNTAIN SPOTTED FEVER

ROCKY MOUNTAIN SPOTTED FEVER [1]

By

R. R. Parker, *Special Expert, United States Public Health Service*

Rocky Mountain spotted fever was recognized as a distinct disease entity by many of the early physicians of the Rocky Mountain region for many years before any attempt was made to determine either its etiology or mode of transmission. Since the first studies by Wilson and Chowning (1902-1904) there have been many field and laboratory investigations relating to epidemiological, etiological, serological, immunological and entomological phases. The most important of these, because of their fundamental significance, were: First, those of Ricketts and his associates of the University of Chicago (1906-1909), who showed that the disease is primarily an infection of small mammals among which it is transmitted by the wood tick (*Dermacentor andersoni* Stiles) and that large mammals are not susceptible, who proved to be fact the wood tick theory of human infection advanced by Wilson and Chowning and who made important observations and deductions concerning the maintenance and behavior of the virus in nature; second, the entomological and zoological studies by Prof. R. A. Cooley, State Entomologist of Montana, in association with Hunter, Bishopp and King of the federal Bureau of Entomology and representatives of the Bureau of the Biological Survey (1909-1911), which resulted in demonstrating the two-year life cycle of the wood-tick—larva, nymph and adult engorging in successive years—and the relative importance of many mammals as tick hosts; and third, those of Dr. S. B. Wolbach of Harvard Medical School (1916-1919), who showed the rickettsial nature of the virus.

The most comprehensive investigation of Rocky Mountain spotted fever, however, was begun by the Bureau of the Public Health Service in the fall of 1921 under the direction of Surgeon R. R. Spencer, with whom the writer has since been associated. This investigation has been carried on in co-operation with the Montana State Board of Entomology, which was created in 1913 for the specific purpose of dealing with the spotted fever problem in Montana. To the persistence of this Board[2] (the membership has fortunately remained intact since its organization) under often discouraging conditions, is due much of the progress that has been made.

Despite these various investigations, however, and the resultant continued additions to our knowledge there are still many unsolved and puzzling problems

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(1) This paper was read before the Section on Medical and Veterinary Entomology of the Fourth International Entomological Congress at Ithaca, N. Y., August 13, 1928.

(2) Professor R. A. Cooley, State Entomologist, Secretary; Dr. W. F. Cogswell, Secretary of the State Board of Health, chairman; Dr. W. J. Butler, State Veterinary Surgeon, member.

relating to etiology, local variations in the virulence of the virus and its maintenance and spread in nature, and the incidence, distribution and prevention of human cases.

In this paper it is the purpose of the writer to discuss these general phases, particularly from the standpoint of the data secured by Surgeon R. R. Spencer and himself, in the hope that he may convey a reasonably comprehensive general conception of the present status of the problem. This has seemed desirable not only because no paper relating to spotted fever has been read before an entomological meeting for nearly ten years (never one of a general nature), but also in deference to our visitors from other lands to whom the subject, which is one of our outstanding problems in medical entomology, is less familiar than to ourselves.

Maintenance and Perpetuation of Rocky Mountain Spotted Fever Virus in Nature.

In 1909, after four years of investigation, Ricketts supposing a one year life cycle of the wood tick, conceived that the virus of Rocky Mountain spotted fever was maintained in nature in the following manner: The virus was carried over the winter in infected female ticks, from about fifty per cent of which infection was passed on through the egg to the resultant larvae. The latter were able to infect susceptible rodents upon which they chanced to engorge, the virus being then picked up by non-infected immature ticks that were infesting such rodents, and by stage to stage transmission, which he had shown to occur, was passed on to adults of that generation. The process repeated in each generation, resulted in perpetuation. He felt that if dependent on hereditary transmission alone, which at first thought would seem sufficient, it would gradually die out.

Except for slight modifications to fit the later-proven two year life cycle of the wood tick, subsequent investigations in the main have verified Rickett's findings and ideas, but have also extended them and have introduced into the problem of maintenance a new factor, the transmission of the virus by the rabbit tick, *Haemaphysalis leporis-palustris* Packard, which was demonstrated by Parker in 1921. This tick infests all species of rabbits and is common on game birds. It was first suspected as the result of two years' observations (1916-1917) of ecological relationships in spotted fever maintenance in Eastern Montana. These studies suggested that wood tick transmission alone was insufficient to account for the survival of the virus over periods of several years, common in some sections of eastern Montana, during which wood ticks become very scarce. The evidence at hand indicated the rabbit tick as the most logical parasite to be studied as a possible second carrier. Whether or not the premise were well taken, studies made in the Bitter Root in 1921, several years later, resulted in showing that this tick is a vector and that infected individuals could be recovered in nature. Our later observations have shown that stage to stage and hereditary transmission of the virus occur in the same manner as in the wood tick.

Evidence thus far secured, therefore, points to stage to stage transmission of the virus within each successive generation of the tick carriers, its hereditary transmission from one generation to the next, and the starting in each generation of new lines of tick infection each year through the medium of tick-infested small mammals, as the three apparent channels along which infection flows to achieve perpetuation. In the light of present knowledge in any given focus of infection the successful flow of the virus along these channels is dependent upon factors of several orders; each separate factor being a variable both from year to year and focus to focus.

The factors of the first order are: Tick abundance, and those which affect the survival and virulence of the virus in ticks. Of the latter, except that they exist, we must confess almost complete ignorance. Presumably, however, they may be meteorological, they may have to do with the kind of animal blood ingested or even with the idiosyncrasies of individual ticks, or conceivably all three may be concerned. Of tick abundance we can speak with more confidence. Provided conditions are favorable for the survival of virus in the ticks, it is the element of chance that controls the flow of infection along the channels indicated, and the likelihood that continuity will be maintained obviously increases or decreases as ticks increase or decrease in abundance.

Whether ticks increase or decrease, that is, relative tick abundance, is controlled by factors of the second order which resolve into two groups: (1) meteorological, (2) those which concern host availability. Of the first group we again confess a far from satisfactory knowledge, but we do know that the activity and survival of ticks is unfavorably affected by extreme dryness or moisture, and by abrupt or continued changes of temperature. The latter group of factors consists of the abundance, actual or relative, the habits and the distribution of each separate species of larval, nymphal and adult tick host, whether or not that species is susceptible to infection.

The species of host animals present, their abundance and distribution, and even their habits, are in their turn predetermined by a third order of factors which are: Floral associations and their distribution, the local topography, meteorological conditions again, and the character and degree of development of local agriculture. These factors interweave to form a tangled skein which may appropriately be called "the spotted fever complex". As entomologists trained to realize the importance of detail, we can readily appreciate that the untangling of this complex so that each separate factor may be given its proper relative value in maintaining infection, is both desirable and essential from the standpoint of intelligently directed and successful control. This is a problem in ecology—epidemiology, if you prefer—but as applied to any disease complex the writer conceives the two terms as synonymous.

An investigation of this complex was begun by the writer in 1921 when with the Montana State Board of Entomology. The demonstration of rabbit tick transmission was the chief accomplishment of that season. The same line of work was continued on a far more comprehensive scale after the entry of the Public Health Service into the investigative field that fall; the general plan of study involving the keeping, for a period of years, for a circumscribed area, of as accurate and detailed a yearly record as possible of the variations of all the known and likely factors of maintenance for correlation with annual case incidence and the yearly prevalence of virus-containing ticks. After two years it was necessary to discontinue this line of effort in order to concentrate on the virus investigations which later led to the production of a vaccine. Although the main purpose of the investigation was thereby prevented, much new, interesting and valuable information was secured. This concerned new host relationships; tick habits that aid them in securing hosts; animal habits, daily and seasonal, that determine their importance as tick hosts and explain how they function in spreading and maintaining the virus; the distribution and prevalence of virus-containing ticks; variations in the percentage of ticks containing virus; the functioning of the rabbit tick in the spotted fever complex; a more accurate method than feeding alone of testing ticks for presence of spotted fever virus and the occurrence in ticks of an immunizing phase of the virus and the phenomenon of "reactivation."

This information has resulted in a much broader conception of the spotted fever problem and a better, though far from complete, understanding of ecological relationships in the natural maintenance of spotted fever virus both in the Bitter Root region and in other localities. Lack of time, however, forbids presentation of these results except for a brief discussion concerning the rabbit tick and the incidental working in of certain other findings in other sections of the paper.

Incidental to the above studies was the discovery of tularaemia infected wood ticks in nature and the subsequent proof that both wood ticks and rabbit ticks are natural carriers of *Bacterium tularense,* and that the former tick is an important agent of human infection.

There are three possibilities which may be concerned in maintenance that have not thus far been mentioned. One is that of the existence of chronic infection in one or more species of susceptible tick hosts, which would likely result in a longer period during which infection could be acquired by ticks. Little work along this line has been done and thus far the evidence is negative. The second is the chance that infected male ticks may transmit the virus to females during the act of copulation. That this is a definite possibility is indicated by Wolbach's finding of spotted fever rickettsia in spermatozoa. If it occurs, then hereditary transmission may be a more important consideration in maintenance than heretofore supposed. The third is the possibility that other parasites, besides the two ticks that have been studied, may be carriers of the virus. Very few observations have been made in this connection. Ricketts showed transmission by adult *Dermacentor albipictus* Packard[3] infected as nymphs, and Maver, transmission by nymphal *D. variabilis* Say, infected as larva and by adult *D. parumapertus marginatus* Banks infected as nymphs. No infective *albipictus* have been recovered from nature in spite of repeated tests. The few tests that have been made with *variabilis* have also proven negative, but this tick must be viewed with suspicion as a possible potential carrier, since the present western limit of its range overlaps the easternmost occurrence of *D. andersoni. Parumapertus marginatus* occurs throughout the southern portion of the range of *andersoni* and must be considered as a possible factor in maintenance in that section of the Rocky Mountains.

The Rabbit Tick in Relation to Spotted Fever.

The full significance of rabbit tick transmission of the virus in the spotted fever complex is not yet apparent. Thus far, it has been studied only under Bitter Root Valley conditions, and incidentally rather than intensively. The

(3) These ticks were collected from horses during the winter months and Ricketts at the time supposed them to be D. andersoni.

data there secured, however, suggest that it is a factor in the maintenance of the virus, as already noted; that it plays an important part in the intensive distribution of the virus in nature; and that, indirectly, it is of consequence in human infection by increasing the number of virus-containing wood ticks.

The more important findings concerning this tick are as follows:

(1) Rabbit tick infestation of its hosts is often very heavy, the habitat adaptations of each host species determining the degree to which it is infested. Cottontail and snowshoe rabbits are especially heavy carriers. In some seasons one hundred per cent of snowshoe rabbits examined have been found infested; and seasonal averages may show as high as several hundred ticks per animal examined. The percentage of grouse infested has varied from eighteen to seventy-five, depending on the species and year. They are less heavily infested than rabbits, but it is not uncommon to find from twenty-five to fifty or more ticks on a single bird.

(2) Relatively high percentages of infested hosts have been found carrying infected ticks. In 1922, of infested snowshoes it was 36.84; of cottontails, 27.27; and of jackrabbits, 33.33. The percentage of grouse has varied between twenty and fifty.

(3) Heavy tick infestation and high percentages of infested hosts carrying infected ticks, in conjunction with the daily and seasonal habits of movement of both rabbits and grouse, make the rabbit tick an important medium for intensive distribution of spotted fever virus.

(4) Rabbits, of all species thus far studied, are hosts of both wood ticks and rabbit ticks. The resultant potential opportunity for wood ticks to acquire infection is apparently greater than for any other species of wood tick host.

(5) A higher percentage of virus-containing wood ticks has been found in unfed adults collected in rabbit habitats and in animal habitats in which rabbits are associated with other small mammals, than from those habitats from which rabbits are absent. In 1922, of such ticks from Columbian ground squirrel habitats, 1.05 per cent contained spotted fever virus; 1.48 per cent from habitats in which ground squirrels and cottontails were associated; 2.40 per cent from cottontail habitats, and 5.20 per cent from ground squirrel-snowshoe rabbit habitats.

(6) Perhaps the most interesting fact concerning this tick is that, *as recovered from nature,* it commonly carries, in contradistinction to the wood tick, a mild strain of spotted fever virus, so mild indeed that it seldom produces typical or even recognizable lesions in experimental animals. Fever is often absent, or if present, low and irregular, and diagnosis must usually be based on the outcome of a subsequent immunity test with the active virus. The reason why this tick carries a mild strain in nature is not at all clear. When infected with laboratory strains, the virus content more often produces characteristic infection.

An interesting and seemingly important subject for investigation is whether wood tick infection or rabbit tick infection is the more fundamental. This involves such questions as: Whether or not the presence of infection in any given locality is dependent upon primary infection in the wood tick, or in the rabbit tick? Or, can each species maintain infection independently of the other? If the rabbit tick were the original vector of the virus and the wood tick secondary, then since the rabbit tick has by far the wider distribution, we would expect to be able to recover infection from this tick at points well beyond the range of the wood tick. No attempt to do this has thus far been made. The limits of the present endemic area, as delimited by geographical distribution of human cases, is well within the range of the last species.

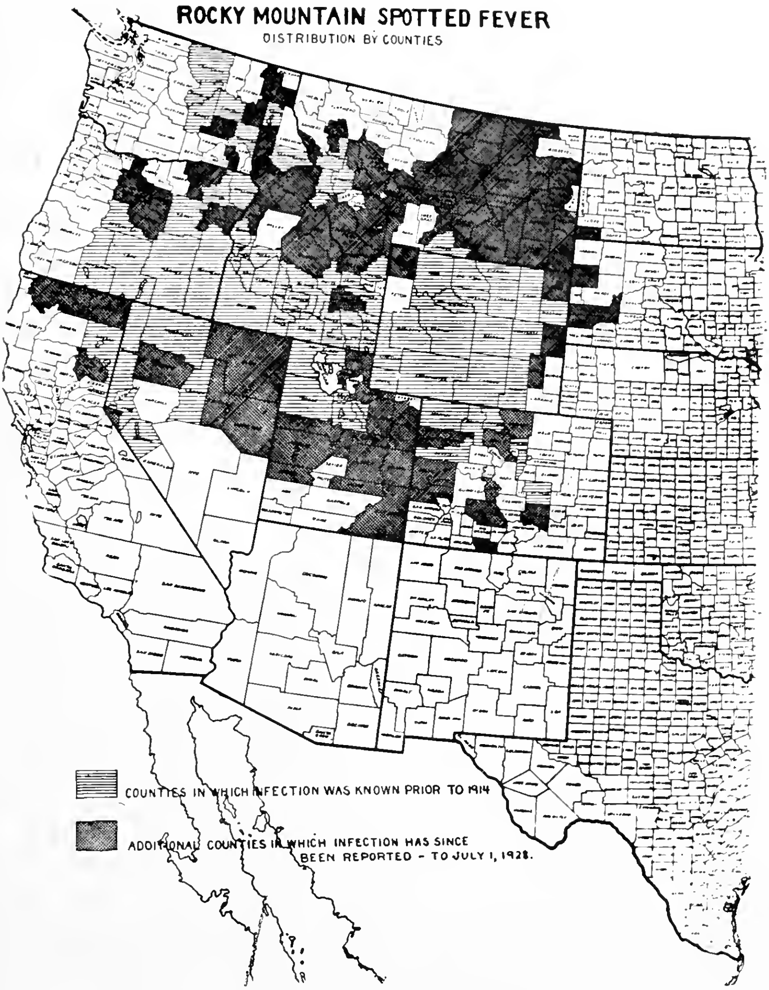
The fact that both wood ticks and rabbit ticks are natural vectors of spotted fever virus sets aside the often made assertion that each species of rickettsia is specific for one species of insect or arachnid host.

Distribution and Spread.

Rocky Mountain spotted fever occurs in parts of the states of Washington, Oregon, California, Nevada, Utah, Idaho, Montana, Wyoming, Colorado, and Nerth and South Dakota. Reports of occurrence in Arizona, New Mexico and British Columbia have never been definitely verified. The total area involved is perhaps one-eighth of the continental United States. Sporadic cases have been reported from points far from the Rocky Mountain region and far beyond the known range of *Dermacentor andersoni*. Most of these, however, have occurred either in persons who have been recently exposed in, or who have had opportunity for contact with tick infested domestic animals recently shipped from, a known endemic area. Of the few that cannot be thus accounted for the only one concerning which there has been any really suggestive evidence was reported in Indiana in 1924. The serum of this patient, after recovery, when mixed with virus and injected into guinea pigs, conferred a degree of protection fully as good as the sera of many recovered definite spotted fever cases. The possibility of making a diagnosis of Rocky Mountain spotted fever that is incorrect in areas far from the known endemic region is well illustrated by a case seen by Spencer in Norfolk, Virginia, in 1925. Clinically this could have been either spotted fever or typhus but the results of blood injection into guinea pigs and of other laboratory tests were entirely negative for both, and its etiology still remains a mystery.

The shaded parts of the accompanying map show the counties within the known endemic region in which infection had been reported up to July 1, 1928. The lighter shading indicates those counties, seventy-eight in number, in which infection is known to have been present prior to 1914; the heavier shading shows an additional ninety-three counties in which infection has since been reported. To what extent it shows recent better recognition or reporting of cases can not be accurately judged. However, cases are being reported from new localities each year to a sufficient extent to justify the belief that both intensive and extensive spread is taking place. Extensive spread has certainly occurred in eastern Montana, North Dakota, and probably South Dakota. In Montana, until 1914, infection was definitely known in but four counties; Missoula, Ravalli and Granite, west of the continental divide, and in Carbon county, bordering Wyoming in the south portion of the eastern part of the state. It has now been reported from thirty-five. The first known occurrence in South Dakota was in 1915; in North Dakota, 1919.

The mechanism by which spread, as indicated by the occurrence of cases in man, is accomplished is unknown. The obvious explanation is that infection has been established in new areas by infected ticks, either wood ticks or rabbit ticks, carried on animals coming from old foci. Although this seems reasonable and may be the correct explanation, yet, if spread of infection is thus easily accomplished, it is difficult to understand why it has not long ago



crossed from the west to the east side of the Bitter Root Valley, or why within two years' time it suddenly appeared over an area of approximately 30,000 square miles in eastern Montana. In the former instance, host-borne infected ticks have unquestionably been carried from the west to the east side of the valley for many years; yet, if we accept the occurrence of human cases as an index, infection has never become established, although ticks are numerous. In the latter instance, opportunity for the introduction of infected ticks from either of several old Carbon County foci or from Wyoming has existed for many years. If spread from either of these established areas of infection had been responsible it is difficult to understand why infection should have appeared at many points over so vast an area in so short a time, and particularly at points so far removed from both.

In the above discussion, the word "spread" has been used on the supposition that occurrence of human cases in a new area is the result of more or less recent introduction of the virus. But does this necessarily follow? Is it not possible that spotted fever virus in an avirulent non-symptom producing phase(4) has been more generally distributed throughout the Rocky Mountain fever region than human occurrence would lead us to suppose, and that apparent new foci, as indicated by first case occurrence, may in reality be sections in which changing environmental conditions have changed an avirulent type of virus to one infectious for man? In this connection the persistent occurrence in the Bitter Root Valley of a mild type of the virus in the rabbit tick, in spite of a contemporaneous highly virulent type in the wood tick, has suggested to us that in non-wood tick sections a mild or even non-symptom producing type of virus may be maintained by the rabbit tick, and that, following wood tick introduction or increase, as has apparently occurred in many sections of eastern Montana, passage of the virus through a greater variety of species of host animals (rabbit ticks only passing the virus through rabbits) may result in an increase of infectiousness to a point at which the virus is capable of infecting man. If this hypothesis be true it implies virus maintenance by the rabbit tick independently of the wood tick, and as noted elsewhere, is presumably susceptible to proof by the testing of rabbit ticks from points far beyond the Rocky Mountain endemic region.

The idea of the existence in nature of a non-symptom producing type of spotted fever virus is not entirely new, and was first suggested by Ricketts in a somewhat different connection. As a matter of fact, we have frequently encountered such a type of spotted fever virus in experimentally infected wood ticks. Moreover, the known regional variation in virulence from the highly fatal western Montana type to the mild southern Idaho type, suggests that the idea of still further differentiation to an even milder form is not unreasonable.

This discussion of spread would not be complete without further reference to the idea, so long held, that infection is absent on the east side of the Bitter Root Valley. While it is true that no cases have originated there, nevertheless the virus is present in nature. As thus far observed, however, it occurs only in a non-symptom producing phase such as has been referred to above. This has been shown with unfed adult wood ticks collected on the east side in 1922, 1923, and 1928. When tested for the presence of spotted fever virus by the feeding and inoculation method,(5) none have caused frank spotted fever, but for certain localities an immunizing phase of the virus has been demonstrated in fully as high a percentage of the ticks as is usually found carrying virulent infection on the west side.

The question at once suggests itself: Whether infection in this form has long been resident on the east side, or is it the result of the more recent

(4) By "non-symptom producing phase" is meant one in which the specific agent invades the tissues of the host without the causation of clinical evidence of infection.

(5) By this method ticks to be tested are first fed on a guinea pig for three days, then removed, and if no fever develops within ten days, the same ticks are eviscerated in salt solution and injected intraperitoneally into the same animal. If fever does not develop in another ten days, an immunity test of blood virus is given. If the virus is present in the ticks it will be shown: (1) by fever following feeding, or (2) by fever following tick-viscera injection, or (3) if the ticks have immunized instead of causing frank infection, by absence of fever following each of three operations.

introduction of highly virulent west side strains which have abruptly lost their virulence under the changed ecological conditions? Present evidence on this point is inconclusive. Suffice it to point out, therefore, that the problem of spread from the west to the east side is not one of gradual encroachment into contiguous favorable areas, but involves the bridging of a strip of unfavorable tick country which in some parts of the valley is several miles in width. This means that although many thousands of tick-bearing animals must have left infected west side sections during the several decades that west side infection has definitely been known, yet it would only be the adult ticks carried by large animals (not immature ticks borne by rodents) that would be likely to reach favorable east side sections. While a considerable number of infected adult ticks must thus have been carried across the barrier zone, the number during any one season is necessarily small. Furthermore, of this small number, only the females, from some of which the virus is transmitted hereditarily, are dangerous as possible virus establishing agents. For reasons too detailed to discuss in this paper, the mathematical chance of thus establishing infection seems to be very small.

Incidence.

Annual incidence in any locality, area or region of infection, is directly dependent on the abundance of infective ticks and those meteorological conditions that determine the duration and degree of their activity. Indirectly it is affected by all the factors noted above as concerned in virus maintenance in nature. This results in a more or less definite though irregular cycle of case abundance. This cycle occurs both in local foci and endemic areas but the peaks of occurrence in the separate foci within a given area are not necessarily chronologically coincident.

The effect of meteorological conditions is most apparent in areas of considerable case incidence like the Snake River valley of southern Idaho, where a marked decline of cases occurs during periods of prolonged dry weather, incidence again increasing following precipitation.

Agricultural development may be mentioned as another factor affecting annual incidence. Cases often begin to appear soon after settlement in localities not before known infected. This results from more favorable tick conditions due to resultant increase in rodents and the bringing in of domestic animals. The course of subsequent incidence depends largely on the type of agricultural development, whether extensive or intensive, or a combination of the two. If intensive, a point of development is finally reached beyond which conditions become increasingly unfavorable to virus maintenance. Extensive farming, however, or a combination of extensive and intensive operations, favor continued prevalence.

Seasonal incidence is delimited by the period of adult tick activity. Most cases occur from March to June and are usually most numerous in April and May. They have, however, been reported during all the months in the year. Winter cases, except under unusual conditions, come from mountainous areas.

Occupationally, the greatest incidence is among those engaged in agricultural pursuits. In some areas farmers are most affected; in others, men engaged in the sheep industry, and in still others persons handling cattle, as determined by the local type of agriculture. Cases also occur among prospectors, miners, hunters, trappers, railroad section hands, forest service employees, oil field workers, fishermen, picknickers, tourists and others whose pleasure or business pursuits take them into infected districts. A surprising number of cases occur among townspeople who are often in infected areas for only a few hours.

Difference in occupational exposure results in many more cases among men than women. A considerable proportion of cases among the latter are due to ticks brought home by the husband or other male members of the household.

Age incidence is also largely the result of occupational exposure, at least as between children and adults. Incidence among children is much higher where they actually live in infected districts (e. g., Bitter Root Valley), than in sections where danger is mainly beyond the limits of cultivable land (e. g., in parts of the Snake River valley in southern Idaho).

Table I shows the known case prevalence in infected states by years from 1909 to 1926. The Montana record is the only one at all accurate. Some records, for example those for recent years in Idaho, have been omitted because the data is so incomplete as to give an entirely false impression. In fact, except for Montana the tabulation is so incomplete that it gives but a poor idea of the incidence or of the relative prevalence in the different states. Two good illustrations of this have recently come to our attention. One is a letter received from a physician in northeastern California, advising that he has treated about a dozen cases each year during his several years of residence; that is, he alone has treated as many cases each year as have been officially reported in any one season from the entire affected area within the state. The second is a letter from a physician in Wayne County, Utah, a supposed uninfected area, advising of the common occurrence of Rocky Mountain spotted fever and of seventeen cases that he has treated in 1928.

Virulence.

Rocky Mountain spotted fever, so far as we are aware, is unique in the fact that virulence, in any local endemic focus, as represented by death rate, is reasonably constant year after year.

The Bitter Root Valley in western Montana has long been known as the focus of greatest virulence. Accurate records for the ten-year period, 1917 to 1926, show that the mortality rate approximates seventy-five per cent. It is about ninety per cent in adults, and fifty per cent in children. There are, however, other sections in the northwest where the death rate is equally or nearly as high, but less is known of them because of lesser economic importance. One is a small area on Rock Creek, twenty-five miles east of the Bitter Root Valley. There are two in Wyoming, one on Kirby Creek near Thermopolis, the other in the Wind River Mountains. In southern Idaho, near Soda Springs, an area of one hundred per cent mortality was formerly known, but no cases have been reported since 1903.

The mildest type of the disease prevails in the Snake River valley in Southern Idaho, where mortality seldom exceeds five per cent. Ambulatory cases are not infrequent according to local physicians. Within this large area, however, there are small local portions in which cases are of greater than the average severity, though by no means approaching the extreme viru-

TABLE I.

ROCKY MOUNTAIN SPOTTED FEVER PREVALENCE.

STATES	Earliest Record	1910	1911	1912	1913	1914	1915	1916	1917	1918	1919	1920	1921	1922	1923	1924	1925	1926
California	1903		3	4	2	2	12	11		3	3	3	11	5	3	6	3	3
Nevada	1880		14	11	7	9	8	20	14	4	2	4	9	1	13		11	
Idaho	1870	223	334	197	258	386	360	151	154	103	165	170	204	168				
Oregon	1887				6	6	53	27	22	13	32	36	45	68	32	13	23	29
Utah							35	34	15	5	10	9	8					
Colorado	1905				1		14	5	6	1	2	8	2	11	6	6	4	
Wyoming	1877					26	61	27	15	4	6	20	3	77	27	34	37	110
Montana	1873				11	16	42	21	21	10	12	25	27	58	48	44	31	32
Washington	1904			3	6	2	8	3	5			2	3	5	1	5	1	1
South Dakota	1915						3							3	1		2	2
North Dakota	1919										1		1					1
New Mexico	REPORTED BUT NOT VERIFIED																	
Arizona	REPORTED BUT NOT VERIFIED																	

MORTALITY.

California	1903-1926	76 Cases	8 Deaths	Death Rate	10.53 Per Cent
Nevada	1911-1926	127 Cases	15 Deaths	Death Rate	11.81 Per Cent
Idaho	As computed by Maxey (1908)			Death Rate	4.86 Per Cent
Oregon	1913-1926	405 Cases	70 Deaths	Death Rate	17.28 Per Cent
Utah					
Colorado	1913-1926	66 Cases	12 Deaths	Death Rate	18.18 Per Cent
Wyoming	1922-1926	285 Cases	65 Deaths	Death Rate	22.80 Per Cent
Montana	1917-1926	369 Cases	89 Deaths	Death Rate	40.34 Per Cent
Washington	1912-1926	45 Cases	5 Deaths	Death Rate	11.11 Per Cent
South Dakota	1915-1926	11 Cases	2 Deaths		
North Dakota	1919-1926	3 Cases	1 Death		

lence of the Bitter Root Valley type. In other sections of the northwest the mortality ranges between ten and twenty-three per cent, being highest in Wyoming. In some states, however, the death rate is undoubtedly less than that shown on Table I, since records of morbidity in most affected states are far less complete than those of mortality.

With respect to this question of virulence an interesting condition exists in three infected canyons near Mountain Home, Idaho. The cases from each canyon exhibit a different and quite constant degree of virulence, those from one being quite severe, those from the next of intermediate severity, while those from the third are mild. This condition has been noted by all the doctors who have practiced in Mountain Home over a considerable period of years, and apparently offers an exceptional opportunity for the study of factors affecting virulence.

Periodic variations in virulence are believed to occur by some physicians who have attended cases from the same focus or locality over a considerable number of years. This is quite possible, but thus far the evidence is inconclusive, since it is impossible to eliminate the personal complexes of patients and also because of the likelihood that difference in age incidence in different years (the mortality rate increases with age), in some instances at least, offers an explanation of varying death rate.

The cause of local or regional difference in virulence as manifested in human cases, is obscure. Wolbach has stated that "rapidly repeated passages of the virus during a long period in a single species of animal, peculiar to or particularly abundant in a given locality, would conceivably modify the virulence in man." This seems to be a reasonable possibility and is perhaps not an unlikely explanation of the low virulence which prevails in areas such as the sagebrush desert country of southern Idaho, in which susceptible host species are few and rabbits are the dominant hosts of early stage ticks. We have given some thought to this phase of the disease complex, but have arrived at no definite conclusion. The prolonged life cycle of the tick (seldom completed under two years) and the fact that in nature the successive stages feed at intervals approximating a year, and the numerous possible factors involved, have thus far stood in the way of laboratory experiments along this line. Of the possible explanations which have occurred to us, we have felt that the most reasonable one and the one which best fits the known epidemiological data is, that virulence varies with the number of susceptible species of small mammals through which infection is being passed in any given endemic focus. This checks well with the present knowledge of animal life in various foci, but for conditions outside Montana, this is admittedly scanty. The idea is purely hypothetical. It is possible that there are localities in which, due to local ecology, the mild type of infection borne by the rabbit tick affects the virulence of the virus as carried to man by the wood tick, but no such relationship exists in the Bitter Root Valley where both species are abundant.

Etiology.

The virus of Rocky Mountain spotted fever belongs to the rickettsiae which Wolbach (1925) is certain represents a new group of micro-organisms. The spotted fever rickettsia was first observed by Ricketts in blood smears but it was through Wolbach's carefully controlled experiments (1916-1919) checked by human autopsy findings that it was definitely established as the casual agent of the disease. For it he proposed the name, *Dermacentroxenus rickettsi*. His results have since been confirmed by Nicholson, Cowdry, Noguchi and ourselves.

We can find no common meeting ground with Woodcock, who believes that rickettsiae are not micro-organisms, but products of cellular digestion.

Observations on the Virus.

Although the virus of Rocky Mountain spotted fever has been studied by several investigators, their results, for the most part are not essential to the purpose of this paper, and it is only intended to discuss the more significant findings of Surgeon R. R. Spencer and the writer.

Of greatest significance, because it in part led to the later production of a vaccine, was the observation that there are definite differences between the virus as it occurs in the cold blooded intermediate host, i. e., tick virus, and in the blood of man and laboratory animals, i. e., blood virus. The more important of these are:

1. In our experience and that of other investigators the properties of blood-virus remain essentially constant throughout an indefinite number of animal passages. It is always an infectious and never an immunizing virus. In ticks, on the other hand, it alternates between a non-symptom and immunity producing phase that is present in aestivating and hibernating ticks and an extremely virulent, infectious phase, which, in each successive stage of the tick, follows the ingestion of fresh blood. This transition we have termed "reactivation" but what happens in the tick to bring it about following the ingestion of blood we do not know. It can scarcely be a simple multiplication of rickettsia since they are often present in abundance before blood is ingested.

2. Tick-virus is much more concentrated than blood-virus. An average c.c. of the latter contains 500 M. I. D. for a guinea pig. One c.c. of reactivated tick-virus, on the other hand, which is equivalent to the pooled viscera of one hundred adult ticks that have fed three days, may contain from 100,000 to 1,500,000 M. I. D. That is, volume for volume, tick-virus is two hundred to three thousand times more concentrated. Single ticks usually contain from 1,000 to 15,000 M. I. D.

3. The third difference is that whereas blood-virus will not infect through the unabraded skin, tick-virus will do so readily. For lack of an exact term—since we do not know the controlling factor—we have called this quality of tick-virus "aggressiveness."

4. Blood-virus alone, whether killed or attenuated by heat or chemicals, does not immunize. Tick-virus when killed by either phenol, formalin, chloroform or alcohol, has high protective value.

Other observations made on the virus in recently infected adult ticks have thrown some light on what happens in previously non-infected ticks following the ingestion of blood-virus. There first appears to be a transition period during which the virus loses its blood-virus characteristics and takes on those of tick-virus. This is followed by a period of invasion during which the parasite becomes generally distributed throughout the tick tissues. During this time, which may be a matter of days or weeks, the tick is unable to transmit infection by feeding. This is in spite of the fact as noted below that the virus content of individual ticks during the period of invasion may be as high as 15,000 M. I. D. per tick, and is evidently because the virus has not yet reached the salivary glands. This explains the difficulty that has been experienced by some investigators in transmitting infection with recently infected adult ticks, and which has not necessarily been due, as some have supposed, to failure of the tick to acquire infection. Since ticks cannot infect by feeding during these two periods their occurrence has been demonstrated by the daily inoculation into guinea pigs of the viscera of the recently infected adults. The same phenomena of transition and invasion must also occur if the infective blood meal is ingested by the larva or nymph, but in the early stage ticks they are not so easily demonstrated because of the difficulty in getting immature ticks to feed a second time. In them, however, invasion is probably more rapid, because the tissue changes accompanying transformation undoubtedly facilitate and speed up the generalized distribution of the parasite.

Thus far no one has succeeded in showing any morphological cycle of the virus, although it seems quite possible that one occurs. Wolbach has described three morphological forms and the sequence in which they appear in nymphal ticks. While we are confident that these and other types which have been described are all manifestations of the disease agent, we feel that our own studies show that the virus is also frequently present in a form that is either ultra-microscopic or is non-demonstrable by methods that have been used. Our experimental evidence to this effect consists first of results of the testing of individual ticks of numerous laboratory reared lots, each lot consisting of ticks from the same parent female, infected as larvae on the same host, and of subsequent identical history until tested as adults. Each tick was tested by the examination of smears of six tissues for the presence of rickettsia and the inoculation of the remaining viscera into a guinea pig to test infectiousness of the virus content. Of each lot thus tested, the larger portion of the ticks showed correlation between abundance of rickettsia and infectiousness, but there were always two smaller groups in one of which the ticks were non-infectious although rickettsia were present and often numerous, while in the other the ticks were infectious but rickettsia could not be demonstrated. The second line of evidence is from ticks infected as adults. In these it frequently happens that titration will show a virus content of 10,000 to 15,000 M. I. D. for a guinea pig per tick before any rickettsia invasion of the tick tissues can be shown. The same lot of ticks tested later, however, will show rickettsia in the greatest abundance, even though the virus content in M. I. D., as sometimes happens, may have become considerably reduced. We also consider the well-known scarcity of rickettsia in blood-virus as further evidence in support of our contention.

Other results of our virus studies have either been detailed in earlier papers or will be discussed in others which will appear in the near future.

Kuczynski and Brandt claim to have cultivated the rickettsia of Rocky Mountain spotted fever and believe it to be a variant of *Proteus* X-19. On this relationship we have no opinion to express. Their culture work has not been verified and until this is done it must be accepted that Wolbach and Schlesinger are the only ones who have approached successful cultivation of the virus. Their results, however, were secured by a tedious process of tissue culture, which is not practicable for routine work.

Studies of the virus of Rocky Mountain spotted fever are potentially of more than specific importance. Of the several recognized rickettsia dis-cases, it is the only one that is satisfactorily reproduced in laboratory animals (human infection is almost perfectly reproduced) and the only one in which the use of the intermediate host for experimental purposes is not markedly restricted. In spotted fever studies, therefore, a much wider range of experimentation is possible and it offers consequently the best avenue for the study of the characteristics of this group of micro-organisms.

Mechanism of Human Infection.

Ricketts stated that human infection "would seem to consist of the injection of salivary secretion, laden with virus, into the cutis of the host." Certainly we must agree with this view in the majority of cases.

Most persons becoming infected with spotted fever give definite history of tick bite from two to ten or twelve days before onset. A minority, however, perhaps five per cent, deny the possibility of having been infected in this way. While in most instances it is safe to assume that they have been mistaken, yet the fact that tick bite has almost certainly not occurred in four of nine cases originating in our own laboratory has caused us to give credence to some of those negative reports. Heretofore, our tick-bite-negative laboratory cases were assumed, for lack of a better explanation, to have become infected by contamination of an abrasion with blood-virus, a well recognized danger. The explanation was unsatisfactory in several of these cases, however, because of the apparent absence of any demonstrable abrasion.

Recent experiments are of some interest in this connection. It has long been known that blood-virus of experimental animals would not pass the unabraded skin, nor would it infect if ingested. The possibility that tick-virus would act differently was not considered until a short time ago. In an exceptionally clear-cut series of tests it was shown that tick-virus (which our previous experiments have shown volume for volume to contain many times more infectious doses than blood-virus) would infect if merely dropped among the hairs on to the unabraded skin of guinea pigs; also if dropped into the eye. In another series of tests it has been demonstrated, contrary to previously accepted ideas, that tick excrement is frequently infectious, but tests to determine whether it will cause infection by contamination of the unabraded skin have not yet been made.

These findings, we believe, furnish an explanation of our hitherto not satisfactorily explained laboratory cases, all but two of which have occurred in laboratory attendants who were engaged in rearing infected ticks for making vaccine. They also suggest that spotted fever may occasionally at least be acquired in nature without tick bite. Opportunity for this is afforded by the custom of hand-picking ticks from domestic animals. In so doing, ticks—engorged or partly engorged females especially—are not infrequently ruptured by pressure of the fingers, or the fingers may come in contact with the tissues of ticks ruptured while on the host. The fingers thus contaminated may conceivably transfer highly infectious tick-virus to some easily penetrated portion of the skin or to the conjunctivae, as is known to happen frequently in tularaemia.

Prevention.

There are two avenues along which the problem of prevention has been approached; (1) wood tick control with the object of keeping the agent of human infection at a point of minimum abundance, and (2) personal prophylaxis.

Wood Tick Control. In most sections of the northwest spotted fever has been prevalent in more or less sparsely settled sagebrush areas of low value, largely used for range purposes, and territorially infection has been dilute. Although the disease has been greatly dreaded, economic pressure in most sections has not been acute. The Bitter Root region of infection, however, is a fertile mountain section of high priced agricultural land and denser population. From this valley came an incessant demand for investigation and control, which finally resulted in the beginning of research work in 1902, and in 1911, of tick control operations which have been continued to the present time. From a small beginning, they have gradually extended until control work, as now carried on by the Montana State Board of Entomology, covers a strip of land approximating seventy miles long and from three to fifteen miles in width along the west side of the valley. The measures used have been dipping, hand-picking and grazing control of domestic stock, and the destruction of rodents, the first three methods being directed against female ticks to prevent egg deposition, the last against the immature stages to prevent them from becoming adults. After eighteen years of work it must candidly be admitted that the results have been far from satisfactory. This has been due in part to inadequate operating funds, in part to lack of permanency of results, and in part to the fact that procedure has been based on a too superficial knowledge of the relative importance of the separate factors of the local complex that controls maintenance. The last deficiency has in considerable measure been removed by the recent ecological studies of the Public Health Service. These have indicated that the most fundamental factors of maintenance that affect the prevalence of infection in the valley have their seat in the mountains that border it on the west. From these infection overflows each year to pour down into the valley. The first aim of control, then, should be shutting off the flow of infection, and in the light of this knowledge the efficiency of present control methods, which only apply to valley conditions, may be compared to that of one who attempts to extinguish a forest fire from the windward side, or to relieve the flooded valley of the Mississippi through operations at New Orleans.

To this end our studies have further indicated that the elimination of the Rocky Mountain goat is the most important single step to be taken. The ranges of this animal, because of its habits and its efficiency as an adult tick host, are areas of heavy wood tick concentration and are veritable reservoirs of the virus. For the present, however, no changes in control methods are contemplated by the Montana State Board of Entomology pending determination of the value for control purposes of the tick parasite, *Ixodiphagus caucurtei* Du Buysson. Work with this parasite which was brought into the state in 1926 through the courtesy of Drs. Wolbach and Larousse is being carried on by Professor R. A. Cooley, Secretary of the above Board.

No attempt to control Rocky Mountain spotted fever in any other section of the Rocky Mountain region has yet been made. This has been due to the great expanse of country involved, to the absence in other sections of so acute an economic condition as that in the Bitter Root Valley, and to inability to recommend practical methods of attack. Unfortunately, the siutation is further complicated because the factors of maintenance differ so widely in different localities that on the basis of present knowledge no one control program could generally be applied. Indeed, in many localities, specific research problems would be involved. Still another complication is introduced by the lack of evaluation of the exact part played by the rabbit tick in maintaining infection. If this tick is capable of maintaining infection independently of the wood tick, it is a point of considerable importance, especially in respect to the degree of permanency of control results.

Personal Prophylaxis. Attention has just been directed to the fact that in the present status of our knowledge the initiation of wood tick control measures has not been feasible outside the limits of the Bitter Root Valley and that even there the results have been unsatisfactory. Personal prophylaxis must therefore be relied upon for protection against infection. The most effective measure is the avoidance of known tick infested or infected areas. Next best is the examination of the clothing and body for ticks. Field workers on control and investigation work do this at frequent intervals, but for the person exposed to average danger, twice a day would be usually sufficient. This results from two fortunate facts, (1) that ticks usually crawl about the body or clothing for some time before attaching and (2) that ticks are seldom able to infect until some hours after feeding has begun, the virus as we have shown, not being infective until reactivation has taken place. The shortest known interval was recorded by Ricketts, one hour and forty-five minutes, only once. Probably ten hours or longer is more frequent and we have often known of ticks, later proven infective that fed on guinea pigs for three days with negative results.

Except for the wearing of some type of apparel that prevents ticks from crawling up inside the trousers legs, the idea of wearing tick proof clothing is not approved.

These measures of prevention, however, are far from one hundred per cent effective. In spite of careful examination of clothing, ticks may hide in seams or folds of the cloth and later become a source of danger to the person himself, his family or others. Cases originating in this manner are not uncommon. An extreme illustration is the case of a woman who, at time of infection had been confined to bed for over a month. There are numerous records of double infection by the same tick in persons occupying the same bed, and the writer knows of one instance of three persons, who were infected at points fifteen and twenty miles apart by the same tick. Unsuspecting persons have also been infected by ticks from stock shipped from infected districts.

Because of the impossibility of one hundred per cent protection against tick bites, vaccination is now being recommended. An effective vaccine has long been sought. The first experimental vaccine was prepared by Ricketts. This was a live virus-immune serum mixture, but because of its live virus content, it could not wisely be used for human prevention. In 1923, Noguchi announced the production of another sero-vaccine in which the virus content had been killed by heat. While this successfully protected guinea pigs, it was later proven valueless in monkeys and man. The vaccine which is now being used is one prepared by the Public Health Service, and which during the past four years has been sufficiently tested to justify the statement that it is of definite value.

In a preceding section it has been noted that spotted fever virus in ticks passes through an immunity-producing stage. Although immunity production by the injection of tick tissues or eggs had previously been observed (Ricketts, Davis, Fricks, Parker) no one had happened to encounter it on a sufficient scale to fully realize the potentialities. This realization came to us as the result of mass work. In the spring of 1922 thousands of *unfed* adult ticks were collected from nature and injected intraperitoneally into guinea pigs to test for the presence of spotted fever virus. If infected they consistently immunized and never caused clinical spotted fever. This was followed by the observation that other thousands of *fed* ticks, collected from animals and similarly tested, invariably produced typical infection. The deduction of the phenomenon of reactivation naturally followed. Next came the demonstration of the high concentration of the reactivated tick-virus and the discovery that chemically killed tick-virus, in contradistinction to similarly killed blood-virus, stimulated the formation of protective bodies in guinea pigs. Subsequent tests showed that it would also protect rabbits, monkeys, and finally man.

The method of vaccine production is unique. At present it is prepared as follows: Larval ticks are engorged on infected rabbits and reared to the adult stage. As adults they are fed several days on guinea pigs to reactivate the virus, are then ground and thoroughly comminuted with sterile sand, and suspended in salt solution which contains four ticks per c.c. and two per cent of a virus killing agent and preservative that consists of four parts phenol and one part formalin. Subsequently two dilutions are made. After seven days the tick tissue and sand are thrown down by centrifugation. The supernatent salt solution containing the killed virus constitutes the vaccine, one c.c. of which contains the killed virus content of one tick and five tenths of one per cent of the preservative. The better portion of a year is required for preparation.

However, tests made within the last year suggest that the tedious and dangerous process of rearing hundreds of thousands of infected ticks from larva to adult, which has been responsible for most of our cases of laboratory infection, can be abandoned and the vaccine prepared from ticks infected as adults. For this method unfed adults will be collected from nature in the spring (previous infection does not matter), fed two days on infected guinea pigs, left for two weeks at room temperature to insure generalized invasion of the tick by the virus, and then placed in refrigeration. In the fall they will again be placed at room temperature and shortly given a second, or virus-reactivating blood meal, then made into vaccine. If this method proves as successful as now indicated, both time and expense will be saved and the danger to our laboratory workers materially lessened.

The value of this vaccine for human prophylaxis has been under test for the past four years. One test was made in the Snake River valley in Southern Idaho, against the mildest known type of infection. The area selected was one of relatively high case incidence in which a large proportion of the cases occurred in men handling sheep on the range. This made it possible to restrict the test to this one industry. During 1926 and 1927, 193 sheepherders were vaccinated, 364 non-immunes serving as controls. A single case occurred among the vaccinated men, one in 193. This was a man who took but one injection of vaccine. Among the 364 controls there were 22 cases, 1 in every 16.55 men.

For the past four years a second and contrasting test has been under way in the Bitter Root Valley against the most virulent known type of infection. Here a total of 2,049 persons have been vaccinated, some one, some two, some three and some four years. This group of vaccinated persons is divisible into two subgroups; one consisting of 59 highly exposed laboratory and control workers, the other of residents exposed only to ordinary danger. In the former highly exposed group eight cases of infection have occurred with seven recoveries and one death. This is in marked contrast to the fact that before the vaccine was in use, five cases and five deaths—100 per cent mortality—had occurred in laboratory workers, all of whom were working with Bitter Root valley strains. Among the larger group of 1,990 persons exposed to ordinary danger four cases have occurred, all of which have recovered. This is again in marked contrast to the occurrence of 18 cases and 15 deaths among non-vaccinated persons of equal exposure in the same area.

During 1927 and 1928 the vaccine has been quite widely used outside the test areas especially in Montana and Wyoming. Of 2,104 persons so vaccinated only two have become infected. These were both from the highly virulent Kirby Creek focus in Wyoming and have been the only recoveries of seven cases originating there in the past three years.

These results suggest that the vaccine commonly confers full protection against the milder types of spotted fever and ordinarily affords a sufficient degree of protection against the virulent types to ensure recovery. The Bitter Root results further suggest that a better degree of protection is given against strains from nature than against laboratory strains.

The duration of any considerable degree of protection rarely exceeds one year, but accumulating evidence suggests there is an accelerated immunity in persons vaccinated in successive seasons, such as often results from repeated use of smallpox vaccine. Reactions from vaccination, for the most part, are quite mild.

Thus far, the demand for the vaccine has exceeded the supply. While it evidently has a definite field of usefulness, because of high cost and the need of yearly vaccination we do not consider it as an ultimate major factor in the solution of the spotted fever problem.

Conclusion.

In the preceding pages an attempt has been made to convey a comprehensive conception of the progress that has been made toward the solution of the Rocky Mountain spotted fever problem. Direct discussion of the economic importance of this disease has thus far been omitted, in part because this has to some extent been indicated in the context under the various headings, but chiefly that it might be pointed out in concluding that Rocky Mountain spotted fever in reality, is but one phase of a still larger problem that confronts the affected states, namely, what, for lack of a better designation may be called the "wood tick problem."

Spotted fever, though the most important, is but one of several ills that *Dermacentor andersoni* inflicts upon the Rocky Mountain region. In man it also causes tick paralysis, is responsible for numerous cases of tularaemia, and in some parts of Wyoming and Colorado, at least, transmits apparently a still different infection, an intermittent type of fever, of unknown etiology. There are also on record numerous cases of septic infection and other pathological conditions which are credited to secondary infection at the site of tick bite. Losses among domestic animals are far from a small item. Sheep and cattle are most commonly affected, especially the former. Tick paralysis in sheep has been known since 1914 when first described by Hadwen. Recently the writer with the co-operation of Dr. James Dade, chief sheep inspector of the Idaho State Sheep Commission, and Dr. W. J. Butler, State Veterinary Surgeon of Montana, was able to prove wood tick transmission of tularaemia to sheep. The losses from this cause are considerable, and are doubtless occurring over a far wider area than we have knowledge of at the present time. In some sections cattle are commonly affected by a condition similar to tick paralysis as recognized in sheep. And last, but by no means least, is the mental worry to which tens of thousands of families in the affected region are subjected each year during the tick season, and to a greater and greater extent each year the dread of tick borne infection is preventing the enjoyment of the opportunities for outdoor life which constitute one of the greatest natural assets of the Rocky Mountain region.

Montana has been forced to an active recognition of this tick problem not only by the acute situation in the Bitter Root Valley, but also by the recent rapid spread of spotted fever over a large part of the state. As a result this state has but just completed at Hamilton, in the Bitter Root Valley, a $60,000 laboratory erected for the sole purpose of accommodating the state and federal agencies that are working on wood tick-borne and wood tick-caused pathological conditions of man and domestic animals. Although the wood tick problem has been less acute in other affected states, it has nevertheless been serious, and is becoming increasingly so as agriculture continues to push out and develop new sections continually creating more favorable tick conditions and new areas of infection.

As viewed from the standpoint of our present knowledge the only satisfactory solution of this larger tick problem is a practical means by which ticks can be kept at a point of minimum abundance. Considering the unsatisfactory status of tick control procedure and the vast areas of tick infested mountainous or otherwise noncultivable land that are involved it is to be earnestly hoped that the tick parasite work now being conducted by the Montana State Board of Entomology will, in some measure at least, aid in the solution of the problem.

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ROCKY MOUNTAIN SPOTTED FEVER IN MONTANA, (1) 1927-1928.

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During the biennium since the publication of the Sixth Biennial Report of the State Board of Entomology, 64 cases of Rocky Mountain Spotted Fever have occurred in Montana, 35 during the season of 1927 and 29 during that of 1928.

In the old western Montana area of virulent infection which includes most of the west side of the Bitter Root Valley and the major portion of the Bitter Root Mountains, there have been 10 cases, 2 less than during the preceding biennium.

Of the four persons that were infected in 1927, three in Ravalli and one in Missoula County, only one died, a boy of 12 years of age, whose family had but recently moved into the infected district. He had not been vaccinated. Of the three recovered cases, two had received the Public Health Service vaccine and had relatively mild courses of infection. The third was a boy living on the east side of the valley, near Missoula. He was presumably infected by a tick brought from west of the river by a relative who was working on the recently completed Lo Lo grade which traverses the edge of the infected area.

Of the six cases in 1928, three originated in Missoula County and three in Ravalli. All the former were fatal. Two of them were infected in the very dangerous O'Brien Creek section, a few miles west of Missoula. The third was the young son of a non-commissioned officer at Fort Missoula just east of the Bitter Root River. The source of the tick was uncertain but it is likely that it was picked up within the limits of the military reservation. If so, it could have been one brought across the river from the nearby O'Brien Creek country, or it may be that there has been an actual spread of infection to the east side. A similar fatal case occurred in the same locality in 1922, the patient claiming to have neither crossed the river to the west side nor to have had contact with any domestic animals moved from that side. Of the two cases definitely known to have originated from O'Brien Creek ticks, one was the five-year-old child of parents who had recently arrived from Wisconsin. The child, who was staying at an east side ranch, was apparently infected by a tick brought from O'Brien Creek by one of the parents. The other case was that of a school teacher who made a short trip into O'Brien Creek Canyon and was there but a few hours. Of the three recovered cases in Ravalli County, two had been vaccinated. Of these one was extremely mild, the other moderately severe. The third case, not vaccinated, was a farmer who had been back in the mountains to turn water into an irrigation ditch.

Of the above 10 cases only three were persons who had resided in the infected districts for some time. Two, possibly three, were due to ticks

(1) Contribution from the Rocky Mountain Spotted Fever Laboratory of the U. S. Public Health Service at Hamilton, Montana.

carried from the west to the east side of the river, two were boys who had lived in the infected section but a few weeks, one was a person who visited an infected country for only a few hours, and one was a Government inspector who, during a period of a few weeks, made an occasional trip into dangerous areas. Although this proportion of cases among recent arrivals, and persons having short or indirect contact with the infected area, is unusually high, it serves to direct attention to the fact that many such cases occur, 50% of those occurring during the past 12 years falling in this category.

The mortality rate in the Bitter Root area during the biennium concerned (40 per cent) has been less than in recent years, presumably due in part to the recovery of all four persons who had been vaccinated, and in part to the fact that 60 per cent of the cases were children, among whom the mortality rate is usually about 50 per cent. The death rate among not-vaccinated persons for the past 12-year period has been 76.81 per cent of all cases, but 84.91 per cent among adults.

In addition to these naturally infected cases, four have occurred among laboratory workers who were in contact with even more virulent strains than those met with in nature. All were vaccinated. Three recovered, one died.

There have been no infections during 1927 and 1928 in the equally dangerous but much smaller focus known as the old Rock Creek area, which includes the southeastern corner of Missoula County and an adjoining section of Granite County along Rock Creek.

In the remainder of the mountainous western half of Montana, there have been but five cases, one less than during the previous two years. One was in Gallatin, one in Jefferson, one in Lake, and two in Lewis and Clark County. No cases occurred in Madison, Meagher, Broadwater, Mineral or Powell Counties, from all of which infection has previously been reported.

Cases were reported from two new points in Lewis and Clark County, one near Craig, the other near Nelson. One from Jefferson County was apparently infected near Boulder. If so, this is also new territory, since all previous records from this county have come from the northern portion near the Lewis and Clark County line.

Spotted Fever was first reported from Lake County in 1928, but incidental to the securing of data relative to this case it was learned that another, which was fatal, had been diagnosed in 1926. Both were infected in the same general locality on Mission Creek, east of St. Ignatius.

In the half of Montana east of the Rocky Mountains there have been 49 cases and seven deaths, one more than during 1925 and 1926. Infection in this section was unknown, except in Carbon County, prior to 1914, but beginning with that year has since been reported in 23 counties involving an area of more than 30,000 square miles. Each year infection has occurred at new points. In 1928, a case occurred for the first time in Roosevelt County. In counties previously known to have foci of infection, spotted fever has been reported from new localities as follows: Near Kirby in Big Horn County, near Mosby in Garfield County, near Clara in Golden Valley County, on Beaver Creek in Phillips County, near Roscoe in Carbon County, south of Bundy in Musselshell County, on West White Beaver Creek in Stillwater County, near Hinsdale in Valley County, and from near Birney and on Armell's Creek in Rosebud County.

These data show that during the biennium just past, Rocky Mountain spotted fever has continued both the intensive and extensive spread which began in 1914.

Years in which case occurrence will be considerably in excess of that of the past few seasons are to be expected.

Table I shows the prevalence of the disease since 1911 in the areas of highly virulent infection in Missoula, Ravalli and Granite Counties.

Table II shows similar data for other sections of the state. Besides the 1927 and 1928 cases, several other additions have been made to this table since its publication in the Sixth Biennial Report. These were 1925 and 1926 cases that originated in Montana but were treated at points in Wyoming and South Dakota. Cases originating in Carter County frequently go to Bellefourche, South Dakota, for medical attention. Others from southern Rosebud, southern Powder River and southeastern Big Horn Counties go to Sheridan, Wyoming.

Map I shows in solid black the three areas of infection known in 1914 in Missoula, Ravalli, Granite and Carbon Counties. The black dots and crosses show the subsequent spread. Each dot and cross indicates the approximate point of origin of one of the 352 cases that have occurred outside the three old foci since 1914. The crosses are for cases during 1927 and 1928.

TABLE I

ROCKY MOUNTAIN SPOTTED FEVER CASES IN MISSOULA, RAVALLI, AND GRANITE COUNTIES

From 1913 to 1928.

County—	1913	1914	1915	1916	1917	1918	1919	1920	1921	1922	1923	1924	1925	1926	1927	1928
Missoula	3	5	5	1	1	1	2	2	3	4	2	2	6	3	1	3
Ravalli	8	7	3	5	5	2	3	2	8	3	8	2	0	3	3	3
Granite	0	0	0	1	0	0	0	0	0	0	1	2	0	0	0	0
Totals	11	12	8	7	6	3	5	4	11	7	11	6	6	6	4	6

Laboratory and control work cases not tabulated have occurred as follows: 1922, 1; 1923, 1; 1924, 1; 1925, 1; 1926, 3; 1927, 1; 1928, 3.

TABLE II

ROCKY MOUNTAIN SPOTTED FEVER CASES IN COUNTIES OTHER THAN MISSOULA, RAVALLI, AND GRANITE

Beginning in 1914, the First Year that the Disease Showed a Wide Distribution Within the State. (See footnotes 1, 2, 3 and 4.)

County—	1914	1915	1916	1917	1918	1919	1920	1921	1922	1923	1924	1925	1926	1927	1928	
Beaverhead											1	1				2
Blaine									1							1
Broadwater							1	1		2	1		1			6
Big Horn		2				1	1		1	1	1	3	3	1	1	15
Carbon	2	3	1	4	2	1	1		3	3	2	3	1		3	29
Carter		4								2	3	1	1	1		12
Custer		6							1		1	2	3			13
Daniels						1										1
Dawson									1							1
Fallon									2		3	1	1	2		9
Fergus			1	3		2	3	3	7	3	3	1		1	1	28
Gallatin		2							1						1	4
Garfield		5	3	1			1	1	7	11	4	1	3	5	1	43
Golden Valley								1	1	1	1		1	1		6
Jefferson									1	1		1	1	1		5
Lake													1		1	2
Lewis & Clark							3	1		2	1	1		1	1	10
Madison				2	1											3
McCone								2								2
Meagher										1						1
Mineral													1			1
Musselshell	1	2	6	1	2	1	10	2	8		2	1		1	6	43
Prairie		1		1							2			1		5
Phillips													2	1		3
Powell									1							1
Powder River		4									2		1			7
Richland	1															1
Roosevelt															1	1
Rosebud		3			1	1	1	5	4	8	3	5	4	9	2	46
Stillwater				2					1		3				2	8
Treasure		2	1						2	1		1		1		8
Yellowstone			1	1	1				9		5	2	2	4	2	27
Valley			1								1	2	1	1	1	7
Wheatland													1			1
Totals	4	34	14	15	7	7	21	16	51	36	39	26	28	31	23	352

1. There are records of 17 cases in Carbon County between 1894 and 1904. None are reported from 1905 to 1914.
2. In 1904 single cases were reported from each of the following counties: Fergus, Park, Gallatin, and Beaverhead. These diagnoses are of doubtful reliability.
3. Cases imported from other states have been treated in Montana as follows: 1915, one Idaho case in Cascade County; 1917, one Idaho case in Golden Valley County, and two Wyoming cases in Yellowstone County; 1921, one Nevada case in Custer County; 1922, one Wyoming case in Yellowstone County.
4. Records of Montana cases treated in other states as follows: 1915, one from Fallon County treated in South Dakota; 1922, one from Stillwater County treated in Wyoming; 1923, two from Carter County treated in South Dakota; 1924, one from Powder River County treated in Minnesota; one from Carter County treated in South Dakota; 1925, one from Carter County treated in South Dakota; 1927, one from Big Horn County and one from Treasure County treated in Wyoming.

MAP I.

SHOWING DISTRIBUTION OF ROCKY MOUNTAIN SPOTTED FEVER IN MONTANA IN 1913 AND IN 1928.

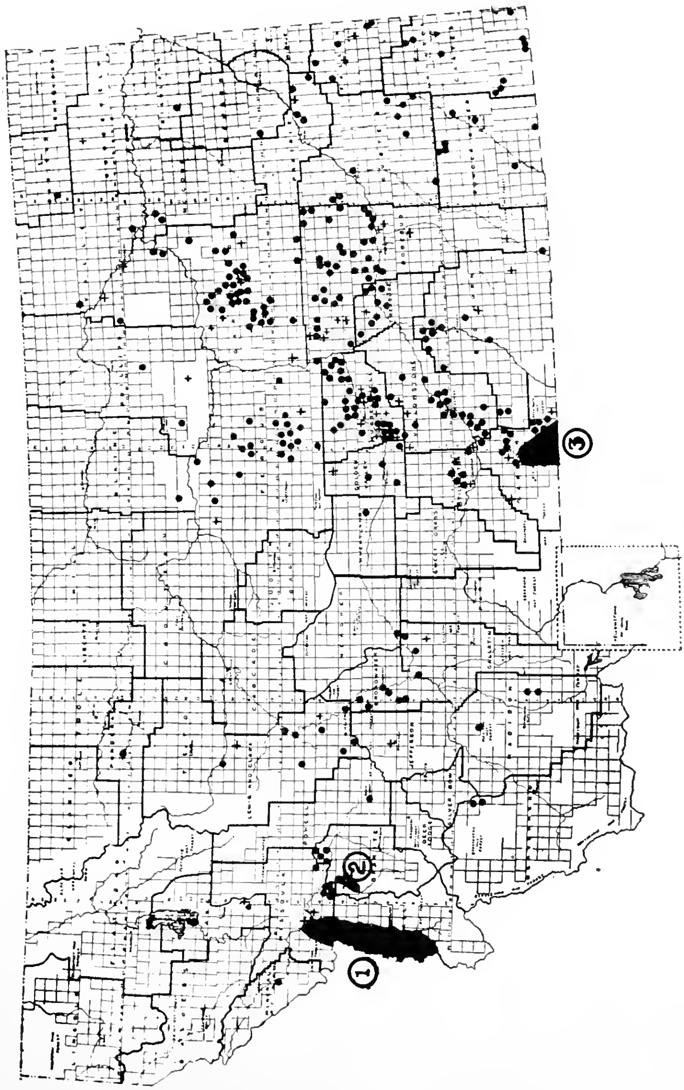
Old foci known prior to 1914:

1. Bitter Root area.
2. Rock Creek area.
3. Carbon County area.

Cases outside old foci since 1913:

● Points of origin of cases, 1914-1926.

+ Points of origin of cases, 1927-1928.



VACCINATION AGAINST ROCKY MOUNTAIN SPOTTED FEVER[1]

By

R. R. Parker, Special Expert

United States Public Health Service

In the Sixth Biennial Report of the State Board of Entomology, Spencer and Parker[2] reported the results of the first two years' use (1925 and 1926) of the Public Health Service vaccine for the prevention of Rocky Mountain spotted fever as distinctly encouraging. Additional data secured during the seasons of 1927 and 1928 now justify the statement that definite prophylactic value has ben demonstrated.

Two areas have been used for test purposes; one a small section of the Snake River valley in southern Idaho where the type of infection is the mildest known, the other, the Bitter Root Valley in Western Montana, where the disease is prevalent in its most virulent form. The Idaho test was conducted for two years, during the seasons 1926 and 1927, while in the Montana area observations have been made during four seasons, 1925 and 1928. The surplus of vaccine not used in these test areas has been distributed upon request to physicians in other sections of the Rocky Mountain region, particularly in eastern Montana, Wyoming and Idaho.

The Idaho test was confined to persons handling sheep on the range (herders, camptenders, and others). A part of these were vaccinated while those refusing vaccination served as controls. During the two seasons concerned, a single case occurred among 193 vaccinated men; this in a herder who had refused to take more than the initial injection. Among the 364 not-vaccinated controls there was one case in every 16.55 men.

During the four-year period of the Bitter Root Valley test over two thousand persons have been vaccinated. Part were residents of the infected districts on the west side of the valley, and part were persons who more or less frequently visited the danger zone. Of thirty cases of infection that have occurred, twelve were in vaccinated and eighteen in not vaccinated persons. Of the former, eleven recovered and one died. Of the latter, three recovered and fifteen died. The death rate among the vaccinated persons, therefore, was 8.33 per cent, and among those not vaccinated 83.44 per cent.

These data indicate that the vaccine commonly confers full protection against very mild types of spotted fever and usually sufficient protection to insure recovery against the highly virulent types.

In parts of Montana outside the Bitter Root Valley area, 401 persons have been vaccinated, most of them during 1928.

The demand for the vaccine, both from Montana and other sections of the northwest, has been greatly in excess of the surplus that has been available for use outside the test areas.

How Vaccine May be Secured.

The vaccine is prepared only at the Field Station of the United States Public Health Service, at Hamilton.

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(1) Contribution from the Rocky Mountain Spotted Fever Laboratory of the United States Public Health Service at Hamilton, Montana.

(2) Spencer, R. R., and Parker, R. R. Prophylactic Vaccination against Rocky Mountain Spotted Fever. Montana State Board of Entomology, Sixth Biennial Report, p. 29.

The first of the output for the season of 1929, which it is expected will be larger than in previous years, will be available about the middle of February or shortly thereafter. It will be distributed directly to physicians upon receipt of requests directed to, The Officer in Charge, U. S. Public Health Service, Hamilton, Montana. The earlier such requests are forwarded the more likely it is that they can be filled promptly. Each request should state the number of persons for whom vaccine is needed.

Persons wishing to be vaccinated should make their desire known to the physician concerned in sufficient time to allow the filing of his request for vaccine well before the beginning of the tick season.

Full directions for administration will accompany each lot of vaccine.

A SHORT REVIEW OF THE WORK OF THE UNITED STATES PUBLIC HEALTH SERVICE ON ROCKY MOUNTAIN SPOTTED FEVER

By

W. F. Cogswell

Secretary, State Board of Health

On the departure of Dr. R. R. Spencer, Surgeon of the U. S. Public Health Service, after six years' extensive investigational work on Rocky Mountain spotted fever in Montana, it seems to be an opportune time to briefly review the work of the Service, as far as it pertains to Rocky Mountain spotted fever, and to give expression to Montana's appreciation of the work.

It was in 1903 that Dr. T. D. Tuttle, who was at that time State Health Officer, requested the Service to make an investigation of a peculiar disease which was then occurring in the Bitter Root Valley. Dr. John Anderson, Surgeon,, was detailed for this work and spent the 1903 season in Montana. In 1904 Dr C. W. Styles made an investigation for the Service. Dr. Edward Francis was sent out in 1905, and in 1906 Dr. W. W. King of the Service, made an investigation. In 1911 Dr. T. B. McClintock and Dr. W. C. Rucker did investigational work for the Service. All of these men did valuable work and their reports were of value to the men who succeeded them.

For the first ten years it seemed to be the policy of the Service to detail a different man to Montana each year to study the Rocky Mountain spotted fever problem, but in the year 1913 this policy was changed, when Dr. L. D. Fricks was detailed for the work. Doctor Fricks spent four successive seasons in Montana. It was during Doctor Fricks' time that the State Board of Entomology tick control program was worked out. This consisted in killing the rodents on the west side of the Bitter Root Valley and adopting a plan for the dipping of cattle. This program is still being continued. The rodents are the hosts of the nymphs of the wood tick and cattle are the hosts of the adult ticks.

In 1917, on account of conditions arising from the world war, the U. S. Public Health Service discontinued its investigational work in Montana, but in 1921, on the urgent request of the State Board of Health, the State Board of Entomology and various civic organizations, the Service again entered the field and since that time has done intensive work. Dr. R. R. Parker, who had been in the employ of the State Board of Entomology for about five years and who was thoroughly familiar with the entomological end of the work, was taken over by the Service, and in 1922 Dr. R. R. Spencer was sent out from Washington and put in charge of the work. These men have worked together for six years, and one of the many results of their work is the development of the Spencer-Parker vaccine for the prevention of Rocky Mountain spotted fever.

Montana appreciates the work of the Service in investigating this disease, as do all of the Rocky Mountain states in which the disease occurs. This appreciation was eloquently expressed by the last legislature of Montana when it voted $60,000.00 for the construction of a proper laboratory for the Service to conduct its work, and thus throwing around the investigators every protection possible against the disease.

The voluntary retirement of Doctor Spencer from active work in Montana does not mean that the work is to be discontinued. Surgeon General Cumming visited the laboratory in August of this year, where he met the state health officers of Minnesota, Utah, Oregon and Montana, who are all greatly interested in the work. Assurances were given that the Service would produce the Spencer-Parker vaccine on a much larger scale in the future and would no doubt be able to supply the demand, not only from Montana but from the other Rocky Mountain states.

The United States Public Health Service has aided health work in Montana in many other ways, and it is hoped that Congress will make adequate appropriations so that the good work may be continued and developed more fully.

CONTROL WORK: ROCKY MOUNTAIN SPOTTED FEVER CONTROL DISTRICTS, BITTER ROOT VALLEY, 1927-1928

By

F. J. O'Donnell

Field Agent, Montana State Board of Entomology

Spotted fever control work in Ravalli and Missoula counties for the biennium 1927-28 has been conducted along the same general lines as were followed in the two preceding bienniums (1923-1926) and as was reported in the Sixth Biennial Report.

The work was, therefore, confined principally to rodent control and stock dipping, for the reason that these measures up to the present time, have proved to be the most effective means of control.

Rodent Control Work. Rodent control work during the past two years has been very satisfactory both as regards the acreage covered and results obtained. The total acreage in both Ravalli and Missoula counties for the biennium 1927-28, as shown in Table I, is somewhat less than for the preceding two-year period. This is due, in part, to the marked reduction of rodents in certain areas in some of the control districts, thus reducing the work required in those areas and the elimination of some other areas where the sparse rodent population did not warrant the expenditure of the time and money necessary to treat the land. Also, there was considerably less work done on the National Forest lands for the reason that the financial assistance given to us in previous years by the Bureau of Biological Survey, for treatment of these lands, was discontinued in 1926. Any work done on the federal lands the past two seasons was, therefore, done with State and County funds, and at the expense of other areas within the control districts. The accompanying table also shows that the expenditures for the years 1927-28 are slightly in excess of the expenditures for the preceding biennium. This is due to a greater concentration of effort in those areas where, in the past, because of the peculiarities of the land, rocky, brushy, etc., it has been difficult to reduce or even control the rodent population, and which serve as a reservoir for the infestation of the adjoining lands. Thus far we have been only partially successful in controlling these areas. Additional time and renewed effort seem to be the required factors necessary in attaining any satisfactory degree of success.

However, the rodent control work in the districts as a whole, has, as above stated, been very satisfactory. The field work is well organized and directed by competent and experienced district supervisors and we believe that we are getting the best results obtainable by this method of spotted fever control.

Dipping. The dipping vats in the Florence, Stevensville, Victor and Hamilton districts have been repaired and filled with dipping solution and placed at the service of the stockmen each year. The Gold Creek vat was filled in 1927, but because of cracks in the concrete walls, the solution was diluted with ground water and after several unsuccessful attempts to make the necessary repairs it was decided to abandon the use of this vat until such time as title to the vat site could be acquired by the State, and then to reline the vat with heavy galvanized iron, similar to the lining recently built into the Stevensville, Victor and Hamilton vats. A galvanized lining of proper weight and strength will cost about $350.00 installed and it was thought inadvisable to expend this amount on vat as the site was not State-owned land.

During the summer of 1927, an attempt was made to acquire title to the Gold Creek vat site, but at that time the land on which it was located was in litigation and a good title could not, therefore, be secured. Dipping during 1927-28 was not as satisfactory as during the preceding two years, that is, the number of stock dipped was somewhat less, due to unfavorable weather during the dipping season. This condition could be remedied to a great extent if, as has been emphasized in previous reports, there were a sufficient number of vats so located that it would not be necessary to drive stock long distances and also in stormy weather stockmen would take advantage of the occasional clear days if the vats were more conveniently accessible.

Stock Poisoning. During the active rodent poisoning each year, complaints are frequently made to either the field men or direct to the station concerning the death of range stock and farm animals which have died allegedly as a result of eating rodent poison distributed by agents of this department.

During the past five years thirty-four such complaints were received at the Station as follows:

1924..................10	1925.................. 4	1926.................. 6
1927.................. 8	1928.................. 6	

In each instance an investigation was made immediately and when possible, and circumstances warranted, a specimen of stomach contents was procured and

SUMMARY COLUMBIAN GROUND SQUIRREL CONTROL RAVALLI AND MISSOULA COUNTIES.

RAVALLI COUNTY CONTROL DISTRICTS.

Table I.

Year	County Revolving Fund	County Appropriation	Poisoned Once Acres	Poisoned Twice Acres	Poison Quarts	Labor Hours	Number of Baits Used	Average Baits Per Acre	Avg. Cost per Acre per Treatment
1923	$ 2.500		42,498.61	35,321.32	3,886.57	4,172.85	280,925.6	3.61	$ .0334
1924	2,000	$ 1,500	56,405.10	41,215.42	3,233.27	5,455.64	258,661.6	2.65	.03209
1925	2,000	1,500	68,029.80	50,133.67	6,704	5,883.02	480,200	4.063	.0343
1926		3,000	85,043.28	65,388.31	8,178.37	7,840.08	654,270	4.32	.0334
1927		3,000	86,093.40	48,131.12	5,910.75	6,968.83	472,860	3.52	.0315
1928		3,000	91,851.50	21,514	5,700	7,421.92	456,000	4.022	.0386
RAVALLI COUNTY—Farms Between Control Districts and Bitter Root River.									
1923			790		161	48.48	12,880	16.30	.0676
*1924			7,091.18		309.50	278.25	24,760	3.49	.0252
MISSOULA COUNTY CONTROL DISTRICTS.									
1923		$ 2,300	66,739	54,432	2,570	3,543.50	205,600	1.614	.01704
1924		2,000	69,915	51,431	1,370	3,039.50	109,600	.8207	.0132
1925		2,000	58,520	36,042	2,189	4,130.50	157,000	1.66	.0242
1926		2,000	65,280	51,538	1,586	3,979.50	161,248	1.37	.0283
1927		2,000	55,650	56,823	1,874.50	4,154.50	149,960	1.33	.0194
1928		2,000	72,310	51,580	1,898	4,058.50	151,840	1.22	.0173
	Federal Appropriation	**RAVALLI COUNTY—Federal Lands in Control Districts.**							
1923	$ 190.16		6,840	6,480	235.50	327	18,740	1.30	.025
1924	213.13		6,920	1,440	162.50	390	13,000	1.79	.024
1925	49.87		2,360	1,080	137.50	188.75	11,000	3.19	.0319
1926	31 60		960	560	76.50	81.50	6,120	4.00	.0334
NATIONAL FOREST—Ravalli County									
1923	$ 1,063		24,936		1,017	1,802	81,360	3.25	.0397
1924	344.87		17,894		534.50	983.75	42,760	2.39	.03
1925	488.88		32,400	2,580	810.50	1,211.25	64,840	1.85	.0226
1926	465.40		22,360		813	1,136	65,040	2.90	.0295

*Placed in Ravalli County Control Districts, March, 1925.

sent to Dr. Emil Starz, Chemist, Live Stock Sanitary Board, Helena, with a request that it be tested for strychnine, which is the active ingredient in our grain poison. When the test was positive for strychnine and other circumstances indicated that the animal had eaten rodent poison distributed by agents of this department, and through no fault of the owner of the animal, then settlement was usually made, based on the assessed valuation of the animal. Of the ten complaints received in 1924, two were of cattle poisoning which occurred in 1923. One, a case of three heifers which had strayed into and were trapped in the Stevensville vat corrals, where they probably got some arsenic. The other was a case of three head of dairy stock which were killed probably as a result of eating rodent poison. In both of these cases settlement was made, the former for $45.00 and the latter for $50.00, or a total of $95.00. In only four of the remaining eight cases did it seem likely that the animals died of rodent poison. These four cases involved five horses and seven pigs and settlement was made for $210.00, or a total of $305.00 for 1924.

The four complaints in 1925 included nine sheep, two cows, two pigs and four geese, and settlement was made for $270.00.

Of the six cases reported in 1926, which involved 5 cows and two horses, none was due to rodent poison and in only one of the four cases reported in 1927 did circumstances justify payment for the animal, a horse, for which settlement was made for $25.00.

None of the cases reported in 1928 indicated rodent poison as the cause of death.

We therefore made settlement in only one case and for only one animal during the past three years, at a total cost of $25.00, as compared with settlement made in ten cases involving 19 animals and 4 geese, at a total cost of $575.00 for the preceding two years.

This marked reduction in the loss of range stock, farm animals and poultry incident to the distribution of rodent poison is due principally to a change in the method of placing the squirrel baits at or near the squirrel burrow. Up to and including 1925 the bait (about a teaspoonful) was placed at the side or on the mound to the rear of the burrow in a small pile. This method of placing the bait made it possible for a farm animal to more easily get the poison in amounts sufficient to cause death. For the past three seasons we have been using the same amount of poisoned grain per bait, but it is scattered over an area eight or ten inches square, thus making it difficult for an animal to procure a lethal dosage of poison without also picking up sand, soil, etc., in the attempt to get the grain.

Another factor in the decrease of the loss of stock from rodent poison is the use of calcium cyanide in pastures, particularly hog and sheep pastures, where it is not always convenient to remove the stock while the land is being treated and also the use of this material near farm dwellings where grain fed animals, and geese and ducks, are more likely to get at the grain poison. The calcium cyanide is placed into the burrow and where deemed necessary the burrow is closed with a shovelful of earth, thus all danger to stock, geese, ducks, etc., is entirely eliminated.

Cooperation of the counties concerned has been very encouraging. We have had the moral support as well as such financial assistance as was requested from the Commissioners of the respective counties. Ravalli County has each year appropriated $3,000.00 and Missoula County has given $2,000.00 each year to be expended for rodent control work under the direct supervision of the Board of Entomology.

Our dealings with the Commissioners and other officials of both Missoula and Ravalli Counties have, as in the past, been very satisfactory. These officials have always shown a keen interest in the work and have assisted in every way possible.

Acknowledgment is also made of the cooperation and assistance of the Live Stock Sanitary Board, and especially Dr. Emil Starz, for the laboratory work done for this station during the past two years.

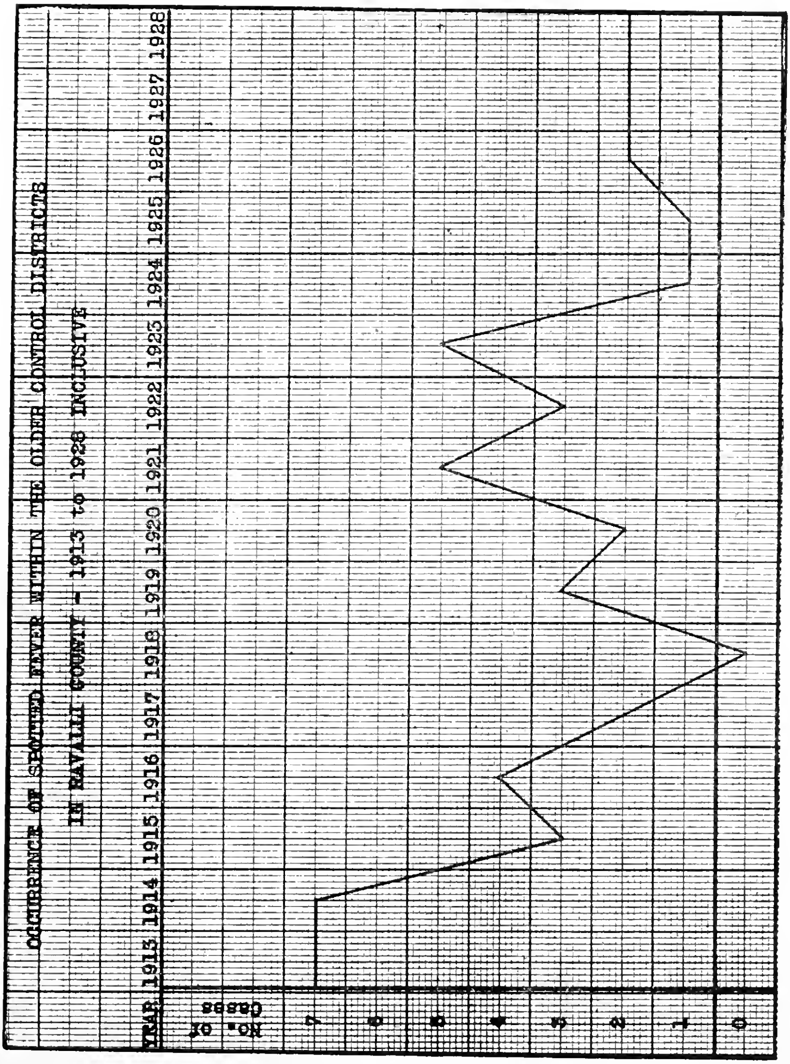
Occurrence of Spotted Fever Within the Older Control Districts in Ravalli County—1913 to 1928 Inclusive.

The accompanying graph has been prepared for the purpose of showing the incidence of spotted fever within the boundaries of the older control districts in Ravalli County from 1913 to 1928, inclusive. Actual control work was first begun in a small area west of Victor by McClintic of the United States Public Health Service in 1911, and 1913 by King of the Federal Bureau of Entomology in an area west of Florence.

These Federal Bureaus gradually extended the control work until 1917, when all of the territory between the Missoula-Ravalli County line on the north to Lost Horse Creek on the south, was included within the boundaries of the control districts. From 1913 to 1917 the Board of Entomology acted in a cooperative capacity only. At the close of the season of 1917 both Federal agencies were obliged to withdraw from the field.

In the spring of 1918 the Board took charge of the work and since that time has had full supervision. Under the direction of the Board additional area was included in the districts and new districts created, until now the control area includes all of the territory from Burnt Ridge south of Darby to the Big Flat north of Missoula, a distance of about 69 miles, varying in width from 3 to 7 miles and embracing an area of 212,180 acres on which active control work is done.

The graph, however, shows only those cases which have occurred in the Florence, Stevensville, Victor, Hamilton and Gold Creek districts, as the control work has been more or less continuous over the greater portion of this area since 1913. The districts south of Gold Creek and north of Florence were created subsequent to 1921. It will be noted that there has been a total of 49 cases during the 16-year period covered by the graph, or a rate of 3.06 cases per year, also it will be noted that there is a downward trend in the curve and that for the past three years there have been only two cases per year. To what extent the control work is responsible for the decrease in the incidence of the disease is a matter of conjecture. There are several other factors which probably have more or less influence in controlling the disease, as for instance the circular letter outlining the season's control program which is each spring addressed to every resident in the control districts and a short time later the appearance of the rodent crews in the field, no doubt serve, to some extent at least, as a seasonal reminder, and to emphasize the potential danger from ticks and tick-infested areas. Another factor is the relatively large number of persons living in the control districts who have received the Spencer-Parker vaccine during the past three years. It is evident, however, that much good has been accomplished by the control work. The great reduction in rodents over large areas and the resultant and very apparent decrease of ticks in these areas fully justify the expenditures of money and effort necessary to properly carry on the control program.



TULARAEMIA AND TICK PARALYSIS

INTRODUCTION

In the Sixth Biennial report a general account of tularaemia and a statement of what is known of its occurrence in Montana was presented in a paper by Dr. R. R. Parker of the Public Health Service. The first evidence that this disease was prevalent in the State was the recovery of infection from specimens of *Dermacentor andersoni* which were being tested for the presence of the virus of Rocky Mountain spotted fever at the Field Station of the Public Health Service at Hamilton, in 1923. This led to the demonstration by the local staff of that service that the wood tick is a frequent carrier of this disease. In the above noted paper, Dr. Parker gives an account. (1) of how it is transmitted to man, (2) of how the disease is presumably maintained in nature among our native species of rodents (at least eleven of which are known to be susceptible, (3) of its then known distribution in the State and (4) of 55 cases which were reported in 1925 and 1926 from Garfield, Custer, Powder River, Rosebud, Musselshell, Fallon, Dawson, Yellowstone, Gallatin and Madison Counties.

Dr. Parker advises that since the sixth report, sixteen additional cases have been reported, nine in 1927, five in 1928, and one each for 1924 and 1925, the diagnosis of the last two having been made by means of agglutination tests.

Of the 1927 and 1928 cases, one was in Fallon County, two in Yellowstone, one in Powder River, one in Garfield, one in Dawson, one in Fergus, one in Rosebud, one in Lewis and Clark, two in Silver Bow, one in Custer and two in Broadwater. The 1924 case was in Flathead County, and the 1925 in Garfield. In addition to these instances of human infection, ticks infected with tularaemia were collected near Menard, Gallatin County, in June, 1928, by members of the staff of the State Board of Entomology.

In the paper which follows, Drs. Parker and Butler give the results of preliminary investigations which they have made in Montana of pathological conditions in sheep following wood tick bites. Their data, secured in 1923 and 1925, suggest that tularaemia may have been responsible, at least in part, for the losses that have been incurred.

RESULTS OF PRELIMINARY INVESTIGATIONS IN MONTANA OF PATHOLOGICAL CONDITIONS IN SHEEP DUE TO THE WOOD TICK, DERMACENTOR ANDERSONI STILES(1)

By

R. R. Parker, *Special Expert, U. S. Public Health Service,*

and

W. J. Butler, *State Veterinary Surgeon of Montana.*

Hadwen's description in 1914 of tick paralysis in sheep in British Columbia[2] was the first evidence of pathology in sheep resulting from the bite of the wood tick, *Dermacentor andersoni* Stiles. This condition has since been observed in the same region by Bruce,[3,4] but until 1923 there was no evidence of suspected wood tick-caused disease in sheep from any other section within the range of this tick. In the spring of that year, losses attributed to *D. andersoni* were reported from both eastern Montana and southern Idaho. This resulted in an arrangement between the Field Station of the U. S. Public Health Service at Hamilton, Montana, on the one hand, and Doctors W. J. Butler and James Dade, respectively the State veterinary surgeon of Montana and the chief inspector of the Idaho State Sheep Commission, on the other, for the comparative study of future outbreaks in their respective states.

The results of the studies with Dade indicate that tularaemia infected ticks have been largely, possibly wholly, responsible for the southern Idaho trouble. On account of these studies (Parker and Dade) either has recently appeared in the Public Health Reports.

The details of the Montana observations are incorporated in the present paper. Unfortunately, the results have been less definite than those secured in Idaho. This has been due to the very incidental character of the field observations and the lack of opportunity to make bacteriological examination of the tissues of affected animals. It seems probable, however, that similar conditions have been of more frequent and widespread occurrence than present data show, and it is hoped that the publications of this preliminary data will stimulate more careful observation and reporting by sheep owners of disease conditions that occur in wood tick areas shortly after sheep are placed on the spring range. There is apparently a feeling among many owners that a certain percentage of loss is to be expected, and, unless it is unusually heavy, there is no desire to ascertain the cause.

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(1) Contribution from the Rocky Mountain Spotted Fever Laboratory of the U. S. Public Health Service at Hamilton, Montana, and the office of the State Veterinary Surgeon at Helena Montana.

(2) Hadwen, Seymour. On "Tick Paralysis" in Sheep and Man following Bites of **Dermacentor venustus;** with Notes on the Biology of the Tick. Parasitology, Vol. 6, pp. 283-287, 1914.

(3) Bruce, E. A. Tick Paralysis in British Columbia. Health of Animals Branch, Can. Dept. Agr. Bull. No. 28, 1920.

(4) Bruce, E. A. Tick Paralysis. Jour. Amer. Vet. Med. Assoc., Vol. 48, N. S. 21, No. 2, Nov. 1925.

The field observations herein reported were made almost entirely by Doctor A. F. Baldwin of Miles City, local deputy for the Montana Livestock Sanitary Board; neither of the writers being able to visit any of the affected bands during the acute stage of the trouble.

G. B. Pope Band, 1923.
(Near Miles City, Custer County.)

On April 30, Doctor Baldwin was called to investigate deaths of yearling sheep in four bands belonging to G. B. Pope of Miles City. The owner attributed his losses to ''poison weeds.'' Doctor Baldwin reported that affected sheep tended to walk ''in circles to right or left'' would ''stagger and fall'' and were unable to rise. He noted that the affected animals were infested with wood ticks and removed the larger ones (i. e., the engorged females) from several of them, not, however, supposing that these ticks were in any way concerned with the condition of the sheep. Much to his surprise, when he again visited these same sheep a few hours later, those from which the ticks had been removed were on their feet, while the others were still ''down.'' The engorged females were therefore picked from the latter with the same result as before. This experience appeared to justify a diagnosis of ''tick paralysis'' and was the first suggestion of the possible occurrence of this condition in Montana. Dead jackrabbits were noted in the locality in which the affected bands were being grazed.

The total loss of sheep in the four bands exceeded 100 head, the heaviest loss being 75 yearlings of a band of 3,800 which had recently been shipped in from Big Timber. In each of the other three smaller bands, about 50 yearlings and ewes were affected.

Trumball and Taudy Band, 1923.
(Near Miles City.)

On May 5, a few days after visiting the Pope band, Doctor Baldwin was called to see the Trumball and Taudy band, which was found similarly affected, except that in addition the sick ewes were aborting their lambs. The sheep concerned had just been shipped in from Ekalaka (Carter County) and Ismay (Custer County), reaching Miles City on May 3. A number of sheep were down and unable to rise. In his inspection report, Doctor Baldwin stated that ''the sheep from which *all* large ticks (engorged females) were removed made recovery. On those that died, other *large* ticks were found that had been missed on the first examination. The ticks were attached under the jaw and on the neck, belly and legs.'' These further observations seemed to strengthen the idea of tick paralysis. The dying of rabbits was reported from the section of Custer County from which part of the sheep had been shipped.

Several ticks from affected sheep of this band were sent to the Hamilton laboratory. Five partially engorged females were emulsified in salt solution and each inoculated intraperitoneally into a guinea pig in the same manner as in routine tests which were being made for Rocky Mountain Spotted Fever in fed ticks[5].

All five guinea pigs died, two in 3 days, two in 6 days, and one in 7 days. Four were afebrile, but the temperature of one reached 41.0°C. The gross pathology as noted at necropsy was not specifically suggestive, and because

(5) Spencer, R. R. and Parker, R. R.; Rocky Mountain Spotted Fever: Infectivity of Fasting and Recently Fed Ticks. Pub. Health Rpts., Vol. 38, No. 8, pp. 333-339, Feb. 23, 1923.

the circumstantial evidence so strongly suggested tick paralysis, the tests were not carried further. Later in the season, after the demonstration of the prevalence of tularaemia-infected wood ticks in nature and of the transmissability of *Bacterium tularense* by *D. andersoni*,[6] the above tests were recalled. They were entirely comparable to many subsequent tests of known tularaemia-infected ticks and this fact coupled with the knowledge of the dying off of jackrabbits in localities where the bands became affected makes it seem almost a certainty that the guinea pigs died of acute tularaemia. Had tissue transfers been made to other guinea pigs as in subsequent tests (see 1925 data below) it is felt that typical lesions would have resulted.

Danford Band, 1925.

(Near Calabar, Custer County)

The same condition as observed in 1923, was again met with by Doctor Baldwin in the Danford band, during lambing in early April, 1925. A high percentage of the band had been affected, and according to Doctor Baldwin, the sheep "act peculiar," stagger around, go down, and abort their lambs.

Ticks collected April 10 from two ewes that were "down" and had aborted were received at the Hamilton Field Station April 13. These ewes had been left behind the band at the outskirts of Miles City. Five groups of ticks, each consisting of two partly engorged females, were emulsified in salt solution and injected intraperitoneally into five guinea pigs. Four of these five tests were positive for tularaemia, the fifth was terminated by intercurrent infection.

Guinea Pig 1 died in five days; the spleen was slightly suggestive of tularaemia and transfer by spleen emulsion was made intraperitoneally to two guinea pigs, one of which died in three days, the other in seven. The gross lesions were typical in both.

Guinea Pig 2 died in six days with typical lesions of tularaemia. A spleen emulsion inoculated into two guinea pigs also caused typical death, respectively, in four and five days.

Guinea Pigs 3 and 4 remained apparently well and were killed and autopsied 13 days after inoculation. Both showed no lesions other than slightly enlarged spleens. The spleen of Guinea Pig 3 was transferred by salt solution emulsion to a second guinea pig which died in 8 days with suggestive lesions. Transfer to a third guinea pig caused death of typical tularaemia in 8 days. The spleen of Guinea Pig 4 was similarly transferred through a series of three additional guinea pigs. There was necrosis of spleen and lungs in the third guinea pig which died four days after inoculation. The fourth also died in four days with typical lesions of spleen, liver and glands.

Guinea Pig 5 died 13 days after inoculation. Necropsy showed evidence of intercurrent infection, and no transfer was made.

Serum was collected from one of the above ewes April 27, 17 days later than the removal of the ticks, both ewes having recovered in the meantime. This serum agglutinated *Bacterium tularense* in all dilutions up to and including 1:640.

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(6) Parker R. R., Spencer, R. R., and Francis, Edward. Tularaemia Infection in Ticks of the Species Dermacentor andersoni Stiles in the Bitter Root Valley, Montana. Pub. Health Rpts., Vol. 39, No. 19, pp. 1057-1073, May 9, 1924.

The spleen of one of the transfer guinea pigs from Guinea Pig 2 was forwarded to Surgeon Edward Francis at the Hygienic laboratory at Washington, and with it he infected guinea pigs from which *Bacterium tularense* was recovered in pure culture. This culture was agglutinated by the immune serum of a rabbit which had been immunized against a culture of *B. tularense* recovered from a human case in Utah by Doctor Francis in 1920.

Cusick Band, 1925.

(Near Powderville, Powder River County.)

In the spring of 1925 the Cusick band of 1,570 sheep was being ranged on Stump creek, a few miles east of Powderville and about 70 miles south of Miles City. These sheep had been dipped in the fall, had wintered in unusually good condition, and were free of sheep ticks (*Melophagus ovinus*). They remained in fine shape until they became heavily infested with wood ticks in late March, after which the whole band lost condition. One ewe was "down" April 1, two April 2, three April 3, five April 4 and ten April 5. On this date the owner came to Doctor Baldwin for advice and was told to look for and remove engorged wood ticks. Doctor Baldwin visited the band April 17, on which date he found that over a hundred head had been down, but that after removal of the engorged females all had recovered except seven of the original ten, from which the ticks had not been removed until after they had been down several days. Tick infestation was heavy, some sheep having more than a hundred. They were attached to the legs, belly, neck, around the base of ears, above the eyes, and on top of the head. Many dead jackrabbits were seen and many sick ones that were heavily infested with ticks.

Ticks were secured from three ewes, A, B and C, two of which, B and C, were "down," the third recovered. A blood sample was also taken from the recovered ewe, A. This material was forwarded to the Hamilton Field Station and was received April 23.

On May 11, one of the writers, Butler, accompanied Doctor Baldwin on a second visit to the band. Blood samples were secured from three more ewes, D, E and F, as well as ticks from one of them, ewe D. All were reported as recovered animals. A third visit was attempted in late May by Butler, Baldwin and Parker, but impassable creeks, due to high water, prevented reaching the band.

Most of the engorged and partly engorged females collected on April 17 and May 11, from the four ewes, A. B. C and D, were tested for tularaemia infection. Seven guinea pigs were inoculated subcutaneously with two partially engorged females each; 21 partially fed females were fed on four other guinea pigs and of the females that thus completed engorgement, six were further tested by inoculating them separately and three by inoculating their eggs; 14 engorged females recovered directly from the ewes were kept for egg deposition and after this was well advanced, nine were tested by injecting them subcutaneously into guinea pigs and five by the injection of their eggs. The four sera from ewes A, D, E and F were tested for ability to agglutinate *B. tularense*.

The results of the tick inoculations showed that of a minimum of five tularaemia-infected ticks, three were from ewe B and two from ewe C. The number of ticks actually infected was unquestionably considerable in excess of five, but many of the tests which resulted in suggestive lesions in the initial test animals were not completed because of disorganization of the station staff by laboratory acquired tularaemia infections. The four sera from the supposedly recovered ewes (A, D, E and F) all failed to agglutinate *B. tularense*, but unfortunately none of the samples were from the acutely ill ewes B and C, from which the definitely tularaemia infected ticks were recovered.

The above test results are not of *direct* significance as regards the responsibility or lack of responsibility of tularaemia-infected ticks for the condition in the Cusick band. The typically positive tests will be detailed, however, both because they were of value as showing that tularaemia infection was present in ticks infesting affected sheep, and because they illustrate so well the advisability of not ascribing definite tularaemia-negative significance to apparent tularaemia-negative pathology in guinea pigs injected with suspected infection bearing material.

Ewe B.

Engorged Female from Ewe. Kept for egg deposition, and on May 7 when this was nearly complete, tick was emulsified in salt solution and injected intraperitoneally into Guinea Pig 1. The latter was dead May 20. Necropsy showed enlarged spleen, enlarged and haemorrhagic inguinals and a suggestive lung condition. A transfer by lung emulsion caused the death of Guinea Pig 2 on May 29. Autopsy showed typical tularaemia lesions of spleen liver and glands.

Engorged Female from Ewe. Inoculated intraperitoneally April 25, into Guinea Pig 2. The later was apparently well when killed and autopsied May 8. The only gross pathology was a slightly enlarged spleen and several small grayish white necrotic foci in the fatty tissue at the distal poles of the testes. Transfer by spleen-polar fat emulsion was made to Guinea Pig 3, which was dead May 20. Inguinal glands were slightly enlarged and haemmorrhagic, spleen was four times enlarged with a few scattered foci of necrosis, liver pathological but not macroscopically necrotic and was adherent to the stomach. Spleen emulsion transfer to Guinea Pig 4 caused death of the latter May 23. There was a generalized lung involvement and spleen two times enlarged. Guinea Pig 5 was injected with a spleen emulsion and died May 26. The spleen emulsion transfer to Guinea Pig 6 resulted in a typical recognized tularaemia infection, the guinea pig dying May 31, and showing ulceration at site of inoculation, caseous inguinal buboes, enlarged caseous pelvics and retrosternals, spleen five times enlarged, and it and liver typically necrotic.

Partially engorged Females that were refed. Nine partially fed females from ewe B were placed on a guinea pig for feeding April 28, and removed May 7. Five had completed engorgement, and three had fed little, if at all. The host guinea pig died afebrile May 9, with indefinite lesions and was not transferred.

The three partially engorged ticks were immediately inoculated intraperitoneally into a guinea pig which died May 20. The spleen was three times enlarged, the inguinals enlarged and the right haemorrhagic. Transfer by spleen emulsion to a second guinea pig caused the latter's death May 31 of typical tularaemia. There was ulceration of site in inoculation, consolidation and necrosis of the lungs, spleen and liver were typically necrotic, and retrosternal and pelvic glands were enlarged and caseous and the retropancreatic enlarged. Of the five fully engorged females four were tested May 16, egg deposition being nearly complete, by injection into guinea pigs, and one by inoculation of its eggs. One of the guinea pigs that received tick viscera was dead May 25. The spleen was but slightly enlarged and of normal color; the omentum contained grayish white necrotic foci and there was a serous peritonitis. Transfer by an omentum emulsion was made subcutaneously to a second guinea pig which died typically on May 31.

The above tests showed at least three ticks from ewe B to have been infected with *B. tularense*. Although four tests were definitely positive, the possibility cannot be excluded that one of the two refed ticks may not have acquired infection through the medium of the guinea pig on which they both fed. Definite infection was not recovered from any of the companion ticks, however, but this may have been because transfers were not made from the initial test animals.

Ewe C.

Partially engorged Females that were refed. On April 28, four slightly to one-third engorged females were placed on a guinea pig, which was dead May 4, but without typical pathology. From this guinea pig a series of five transfers were made. The test animals for the second and subsequent transfers all died of typical tularaemia. The spleen of the second transfer guinea pig, preserved in glycerine, was forwarded to Francis and from it he infected four guinea pigs from one of which he recovered a pure culture of *B. tularense.* This was agglutinated by a 1 : 1280 dilution of the same immune serum previously used.

All the ticks recovered from the above guinea pig were injected into a second, which died May 17 with very slightly enlarged spleen, injected inguinals and evidence of pneumonia. It is of interest to record that in spite of a series of three transfers typical diagnostic lesions were not produced, although one or more of these ticks had been responsible for the infection of their host. The lesions in each of the three transfer animals were suggestive, however, and a note was made to the effect that had transfers been continued typical lesions might have resulted.

Partially fed Females that were refed on a Yearling Sheep. On April 28, 25 ticks were placed on a yearling ewe purchased locally. This sheep had always been pastured under fence in a tick-free country. Thirteen days later, on May 11, eleven fully engorged females were recovered and were inoculated in five guinea pigs, four receiving two ticks each and one three ticks. Two test animals died respectively May 15 and May 16 with indefinite lesions. Tissue transfers from both resulted in typical tularaemia infections. The other three initial test animals showed equally suggestive lesions, but were not transferred.

Samples of serum secured from the yearling host on May 12 and 19, respectively 14 and 21 days after infesting it with ticks, both failed to agglutinate *B. tularense.*

The data of the above tests show that at least one of the four ticks fed on the guinea pig and at least one of those fed on the sheep, were infected.

Summary of Test Data for Cusick Band.

The material received from the Cusick band consisted of four lots of ticks from four ewes and four samples of sera.

Two of these lots of ticks were collected from supposedly recovered ewes, two from ewes actually ill. All four samples of sera were from recovered animals.

Of 16 ticks tested from the two recovered ewes none were shown definitely to be infected with tularaemia. However, several initial test guinea pigs showed necrosis of liver or spleen or suggestive lesions of other tissues, and had tissue transfers been made it is probable that several positive tests for tularaemia would have resulted.

Of fifty ticks tested from two acutely affected ewes a minimum of five, i. e., at least 10 per cent, were shown infected with *B. tularense,* three from one ewe, two from the other. That the actual number of tularaemia-infected ticks was greater is very probable.

None of the four sera from supposedly recovered animals agglutinated *B. tularense.*

Barley Bros. Bands.

(Northwest of Miles City, Custer County)

On April 21, Doctor Baldwin was called to see affected sheep of a band belonging to Barley Bros., on Sand Creek, 25 miles northwest of Miles City. A band of 1500 yearlings which were ranging on the East Fork of Sand Creek had become heavily infested soon after going on the range. Symptoms were noted in the sheep in early April but the herder thought them due to eating poisonous weeds. However, in a few days so many sheep were sick that he went to headquarters for help.

When the band was visited by Doctor Baldwin April 21, a hundred sheep had been down, 25 had died and many others had been affected to greater or less degree. Hides recently moved from sheep that had died showed ''many dark haemorrhagic'' spots where ticks had been attached.

The band was again visited May 11 by Doctors Butler and Baldwin. Fourteen engorged and four partly fed females and six males were taken from a sheep that was down. This sheep was reported to have been down before, but had recovered after removal of the wood ticks and the soaking of points of attachment with dip. Ten of the above ticks were tested in the same manner as before. One of the engorged females caused suggestive lesions in the initial test animal and spleen emulsion transfer to a second guinea pig caused death of typical tularaemia. In guinea pigs representing three other tests necrosis and other suggestive lesions were present. For example, one of the autopsy records reads as follows: ''Lungs typically necrotic; spleen four times enlarged, both it and liver suspicious.''

During the period that the above band was affected another was being grazed within a fenced area in which no stock had been grazed for several years and ticks were few. No trouble was experienced, but later on, after removal to open tick infested range on Coal Creek, this band also was severely affected. The loss amounted to 60 yearlings and 40 ewes.

Dead jack and cottontail rabbits were extremely numerous on both ranges concerned.

In 1926 ticks were again numerous in the areas concerned but no trouble was experienced. However, one of the owners, J. Barley, himself became infected from tick bites, the specific character of his illness being verified by an agglutination test.

RESULT. Of ten ticks tested from a yearling down for the second time, one was definitely shown infected and suggestive lesions were caused by several others.

Discussion.

There seems to be no reasonable doubt of the initiating part played by the tick, *D. andersoni,* in the disease outbreaks in sheep reported above, but the relative extent to which tick paralysis and tularaemia are to be blamed for the resultant pathology is less clear. The possibility that some third tick-caused condition may occur in sheep cannot be denied, but if such exists there is no evidence to suggest it in the data thus far secured.

The possibility that tick paralysis may have been concerned rests solely on Doctor Baldwin's observations as to actions of affected sheep and their more or less rapid recovery providing infesting female ticks were removed soon after symptoms were noted.

Relative to the possible responsibility of tularaemia there is more data, but, except in the case of the two Danford ewes, it is entirely circumstantial and does not justify any opinion as to the extent to which *B. tularense* may have been involved. Although the writers feel that tularaemia infected ticks were undoubtedly present on affected sheep at the time of the 1923 trouble there is no incontrovertible evidence to this effect. For that matter, there was at the time no knowledge that tularaemia infection was present in the state, nor was it until later that season that the Public Health Service began experiments which resulted in demonstrating the part played by *D. andersoni* in its dissemination. However, judging from our present knowledge, it seems highly probable that the death of jack rabbits, which was reported coincident with the trouble in sheep in the areas concerned, was due to this disease. If this hypothesis be correct, our present knowledge would then justify the assumption that tularaemia infection must have been present in ticks; and, as has previously been noted, it is now felt that the death of the five guinea pigs, each injected with a single female tick taken from affected sheep of the Trumball and Taudy band, was caused by this infection. While this is not susceptible to proof, yet the familiarity of one of us (Parker) with the results of injecting thousands of wild wood ticks into laboratory animals justifies at least the conclusion that some infectious agent was responsible. Furthermore, the fact that all five ticks, which were selected indiscriminately from those received, caused death, suggests that *part* of them at least may have acquired this infectious agent from the sheep on which they were feeding. That so high a percentage of unfed ticks similarly selected at random should be infected with disease organisms is extremely unlikely.

In 1925, tularaemia infected ticks were recovered from affected sheep of each of the three bands concerned. This fact alone, however, lacks significance because, as reported by Parker,[7] we know that tularaemia was quite prevalent that year in ticks, in rabbits and other rodents, and in man in the part of eastern Montana concerned, and it is certain that infection could have been recovered equally well from unfed wood ticks collected from the sagebrush. In other words, though we know that sick sheep were infested with an agent infected with, and capable of transmitting, *B. tularense,* we never-

(7) Parker, R. R. Tularaemia and its occurrence in Montana. Mont. State Board of Entomology, Sixth Biennial Report, pp. 30-41, January, 1927.

theless have no actual evidence to show that such transmission really occurred. However, in the case of the two Danford ewes that aborted, we do have the additional evidence that the serum of one of them, secured after recovery, agglutinated *B. tularense* in dilution of 1 : 640. Furthermore, as in the case of the Trumball and Tandy band in 1923, so high a percentage of the infesting ticks were found to be infected with tularaemia that it seems probable that *part* of them may have become infected from the sheep on which they were feeding. However, this one positive agglutination test would mean little, were it not for the results of the Idaho tests, in the spring of 1928.[8] We feel that these combined data justify the opinion that in this instance *B. tularense* was the specific disease factor concerned. On the other hand, we do not feel that even the Idaho data warrants any assumption as to the extent to which tularaemia may have been responsible for the trouble in the other eastern Montana bands. For example, although tularaemia infected ticks were recovered from sheep of the Cusick band, sera of four supposedly recovered ewes failed to agglutinate *B. tularense* even in low dilutions. Furthermore, the symptoms as observed by Doctor Baldwin in the affected eastern Montana bands do not agree with those observed by Dade in southern Idaho. Therefore, although the evidence for the Danford ewes suggests that tularaemia-infected ticks were at least in part responsible for the trouble in that particular band, there is nothing but circumstantial evidence of doubtful value on which to base any more inclusive opinion.

The lack of critical and differential data on which to base any definite conclusion is due to insufficiently complete field observations of affected bands and failure to determine whether or not *B. tularense* were present in the tissues of animals acutely ill or recently dead. These deficiencies can readily be supplied in case of future outbreaks. Satisfactory field data, however, will require the observation of affected bands for several days or longer and, in addition, a considerable number of the affected animals should be isolated for detailed observations which should be correlated with bacteriological and serological laboratory studies.

The possibility has been considered that tick paralysis and tularaemia in sheep may be one and the same condition. While this should be borne in mind, present evidence is admittedly against it.

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(8) See introductory part of this paper.

MONTANA'S LABORATORY FOR THE STUDY OF INSECT-BORNE DISEASES

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Probably there does not exist another research laboratory like the one of the Montana State Board of Entomology which has recently been completed at Hamilton. The field of work as laid down in the Act which created this Board in the year 1913, is "to investigate and study the dissemination by insects of diseases among persons and animals, said investigation having for its purpose the eradication and prevention of such diseases," and the new laboratory is designed and equipped, so far as is possible within the available fund, to perform the necessary studies connected with insect-borne diseases. The Act authorizing the erection of this Laboratory, signed by Governor Erickson on March 16, 1927, appropriated the sum of $60,000. The building is a brick faced, reinforced concrete structure of three stories, 40 feet by 66 feet on the ground. A one-story wing 88 feet in length is attached to the west and is designed for the care of stock laboratory animals. The building is made without a basement in order that it may be as sanitary as possible, and to have as few places as possible where insects or rodents may hide away. The construction throughout is such as to avoid cracks and sharp corners. The boiler burns oil and is automatic and certain of the rooms have automatically controlled temperatures. While the building is planned generally for the field of medical entomology, and closely associated lines, it is laid out particularly for the needs immediately before the state. The common wood tick of the region, *Dermacentor andersoni* Stiles, and the diseases of man and the losses in livestock of which it is the cause, constitute the immediate problem.

The work on Rocky Mountain Spotted Fever is being conducted under an informal plan of co-operation between the U. S. Public Health Service and the Montana State Board of Entomology. At the present time the U. S. Public Health Service is concerned mainly with a further study of the Spencer-Parker vaccine. This vaccine, made from the bodies of infected ticks, was originated two years ago at the old, improvised laboratory at Hamilton and at the Hygienic Laboratory at Washington, D. C., by the two U. S. Public Health Service workers whose name it bears. It now has been used on about 4,000 persons in Montana, Idaho and Wyoming, with uniformly encouraging results.

A feature of the Spotted Fever studies in Montana is that they are planned and carried out with careful reference to field conditions. It is for this reason that the new laboratory was erected at Hamilton in close proximity to regions where Rocky Mountain Spotted Fever is found. Field conditions may be thus easily studied, materials for study may be brought in, and parasites may be bred and quickly liberated.

Laboratory work on Spotted Fever has always been dangerous for the workers. Up to the time that the Spencer-Parker vaccine was developed there had been 4 cases of laboratory infection with the disease in Montana (3 at the field laboratories and 1 in Helena). All of these cases were fatal. Since we have had the vaccine there have been 7 laboratory cases and 6 of these have recovered. It is now indicated that the vaccine gives full protection against all strains of the disease excepting the more virulent one in the region near the laboratory, and sufficient protection in the majority of cases to insure recovery from infection with the more virulent strains. It is hoped the new laboratory with its modern conveniences will afford much better protection for the workers than was possible in the one being abandoned which was a vacated schoolhouse.

The new building provides an incinerator for the disposal of dead animals and all waste; a specially adapted laundry and drying equipment for the cleaning and drying of the numerous cloth bags used to confine the ticks on laboratory animals; dressing, locker and shower rooms for the use of workers, who are expected to make a change of clothing on coming from the field and on leaving the laboratory; a shop for the making of cages and other equipment; a poison room for mixing and drying poisoned grain to be used in the destruction of certain rodents; and quarters for the custodian. A special feature of the equipment is a series of three refrigerator rooms cooled by a Frick ammonia machine. These rooms are cork walled, two are automatically maintained at 40°C., and the third at 0°C. In these rooms are fifteen thermal cabinets automatically maintained at graded temperatures ranging from 0°C. to 37°C. These cabinets are put to a variety of uses including storage and incubation of cultures, virus, and vaccine, and experimental work on ticks and parasites.

The building provides several rooms in which laboratory animals such as guinea pigs and rabbits, and wild rodents such as ground squirrels, mountain rats, and wild rabbits of various kinds, are held in cages stacked in steel frames. The production of vaccine in quantity involves the feeding of a very large number of infected ticks, as well as numerous tests on guinea pigs. Many of the experiments on the virus and the studies of the organisms require the use of laboratory animals. In the parasite work it is necessary not only to parasitize the ticks while they are on animals but also to rear the ticks which are to be parasitized. To prove that parasites have been established in the experimental field areas and determine the percentage of parasitized ticks, wild rodents must be trapped and held in cages until the ticks drop off.

It is thus necessary to hold a very large number of animals in cages in the screened and guarded laboratories. This requires the daily feeding of animals, often the taking of their temperatures, and at all times the keeping of the cages and rooms in a clean and sanitary condition.

There are also provided two general laboratories equipped with facilities for chemical and bacteriological studies. For the grinding of infected ticks and further preparation of the vaccine, a special room with suitable facilities is provided. There are also furnished office and record rooms and a photographic room. The rabbit hutches in the animal wing deserve special mention as they are made in keeping with the established practice of rabbit breeders in the Northwest. These hutches are built in four tiers facing a hollow square or court. While the four walls of the building are complete, the roof is left off in the portion over the court thus practically exposing the animals to the open air, which exposure, growers have found, keeps rabbits in a healthier condition. The guinea pig room is of tighter construction and is given a small amount of heat. A root cellar under this room provides ample space for the storage of vegetables used in feeding the laboratory animals.