Studies in Fascioliasis in Oregon Sheep and Goats



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SUMMARY

- 1. Fluke ova hatched in varying lengths of time from 14 days to 13 months and 20 days.
- 2. Freezing killed embryonated ova but did not affect those not embryonated.
- Miracidia lived as long as twenty-four hours after hatching.
- 4. Miracidia attacked four species of *Lymnaea* and one of *Physa*.
- 5. Lymnaea (Galba) bulimoides Lea was the first definitely determined host for Fasciola hepatica on the North American continent. L. (G.) ferruginea Halderman has been added to the list of hosts.
- 6. Lymnaeae are widespread throughout Oregon, even at very high elevations.
- 7. Snails lived at 37° F. for a month without food when water was provided.
- Cercariae are capable of living in snails and being discharged for more than six months from date of infestation.
- 9. Individual snails are capable of living with large numbers of cercariae in them, 580 having been discharged from one specimen.
- 10. Carbon tetrachloride in 1-c.c. doses destroyed mature and nearly mature flukes in livers of sheep and goats.
- 11. Snails were destroyed by use of copper sulfate broadcast on habitat.
- 12. The destruction of snails did not prevent their reappearance on the same pastures the following year.
- 13. Cost of application was \$5.25 per acre.
- 14. Copper sulfate broadcast in twice the amount necessary to kill snails was not poisonous to sheep.

Studies in Fascioliasis in Oregon Sheep and Goats

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INTRODUCTION

The first studies made with this disease by this Station were made in 1919 when attempts were made by use of copper sulfate to control snails that might act as hosts of the parasite. Dr. Asa C. Chandler has worked out apparently very efficient methods of snail destruction. Attempts to use his methods in destroying snails on fluke-infested land failed, and work on the problem was discontinued until 1926.

Shortly after Montgomerie² reported his success with carbon tetrachloride, opportunities for its use arose in the vicinity of this Station. Since September, 1926, studies of this disease have been one of the department problems.

This disease being very common throughout the state all phases have been studied. Since a new treatment had been found, our first efforts were in that field. Later a need for information on other phases of the problem was felt, so studies were begun, the results of which are included in this publication.

LIFE-HISTORY

The life-history of this parasite (Fasciola hepatica) had been known for a great many years. All of the different steps have been determined in Europe, but the intermediate host for this continent had not been discovered before 1928 when, on August 20, snails infested with Fasciola hepatica were found. On attempting artificial infestation of snails considerable information on fluke ova was obtained.

FLUKE OVA

Ova were gathered generally from the gall-bladders of infested sheep and cattle, although some were obtained from feces. These eggs were thoroughly washed and kept under ordinary room temperatures exposed to ordinary room lights. Some ova hatched in as short a time as 14 days and some hatched continuously for 13 months and 20 days after collection.

The effect of cold on fluke ova was determined by placing newly collected ova and ova well embryonated in a cold plant at a temperature of 10° F. for twenty-four hours. Those newly collected hatched after thawing, but those having live embryos had been destroyed.

		_		
Source	Date col- lected	Date first embryonated	Date hatched	Days
Sheep	5/11/27	5/25/27	6/ 9/27	28
Cow	5/11/27	No embryos	Did not hatch	
		6/24/27		
		Not noted		
		Not noted		
		Not noted		
Sheep	3/13/29	Not noted	9/23/29	6 months 10 days
				13 months 20 days
Sheep	3/25/29	4/ 4/29	Placed with snails	
				2 months 19 days
		Not noted		
Sheep	4/13/29	5/ 2/29	5/28/29	1 month 15 days
		4/16/29		
		Not noted		
Sheep	4/25/29	5/28/29	7/25/29	3 months
Cow	11/ 8/29	12/ 2/29	12/20/29	1 month 12 days
Cow	4/21/30	Not noted	5/ 5/30	14

HATCHING TIME OF FLUKE OVA

Two groups of ova were of interest because of the long periods of time through which they hatched.

One group collected February 22, 1929 was placed out of doors under an unenclosed porch March 13, 1929, exposed to ordinary daylight at temperatures ranging between 31° and 72° F. On May 5 they were taken to the laboratory and kept under room temperature exposed to ordinary room lights. Free-swimming miracidia were found on June 16, 1929 and at bi-weekly intervals until April 10, 1930.

A second group of ova collected early in March, 1929, handled under the same condition as the group collected February 22, 1929, continued to hatch until November 18, 1929, at which time water in which they were hatching dried up.

Ova collected from cows' feces received from a packing plant November 8, 1929, washed and stored at room temperature, nearly all hatched by December 20, 1929. A second group of ova from these same feces washed December 20, 1929, were all hatched by February 22, 1930. A third group from the same feces washed February 22, 1930 had not hatched up to April 21, 1930.

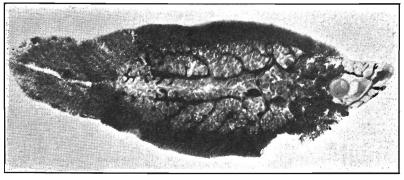


Fig. 1. Mature fluke stained and injected. Enlarged 4 times.

METHODS OF FINDING OVA IN DROPPINGS

The concentration of fluke ova by the use of sugar, salt, and calcium chloride solutions produced only the destruction of the ova. Dr. Robinson's method proved useful only where it was desirable to make counts.

The method described by Damaso de Rivas⁵ gave far better results in concentration of ova in feces of carnivora than in herbivora. This method did not destroy the ova, but so much debris accompanied the ova when thrown to the bottom that the method did not prove entirely practical.

For diagnostic purposes several smears and the use of a binocular served very well.

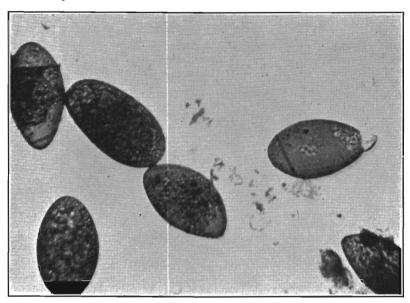


Fig. 2. Fluke ova. Magnified about 200 times.

MIRACIDIA

Miracidia seemed to be more active in morning hours after short exposure to sunlight through ordinary window glass. Water containing embryonated ova but no free-swimming miracidia was swarming with miracidia one hour after exposure to sunlight. Miracidia isolated were found to stay alive for 24 hours after hatching. This was the maximum time as the majority died after 8 hours.

Miracidia were seen to attack the following snails: Physa triticea Lea, Lymnaea (Galba) bulimoides Lea, L. (G.) ferruginea Haldeman, L. (G.) bulimoides techella Haldeman, and one other Lymnaea not yet identified. Cercariae were found developing in Lymnaea (G.) bulimoides Lea only.

Lymnaea (Galba) bulimoides Lea about one-third grown, exposed to limited numbers of miracidia on October 14, 1929, discharged cercariae January 23,

1930, three months and nine days after infestation. Another group of snails from a different source, identified by some conchologists as L. (G.) bulimoides. Lea and by other conchologists as L. (G.) ferruginea Hald, were exposed in the same manner as the foregoing group of Galba bulimoides. No cercariae swarmed from this group and upon examination they failed to show infestation.

SNAILS

In the studies made of fresh-water snails some 23 different species have been collected. Identifications have been made by Dr. H. A. Pilsbry, Dr. J. R. C. B. Tomlin, Mr. A. W. Hanham, Mrs. Ida S. Oldroyd, Dr. W. B. Marshall, Dr. Paul Bartsch, Mr. W. J. Clench, Dr. G. Dallas Hanna, and Mrs. D. Sinitsin. Owing to the fact that Lymnaea truncatula Müller had been incriminated by Europeans as host for Fasciola hepatica, most of the attention was given the Lymnaeae found in this region. Of these Lymnaea Galba bulimoides Lea appeared to be the most common. At different times specimens of Lymnaeae collected from fluke-infested pasture were variously identified by different authorities, and the results were very confusing. Apparently several species of Lymnaeae exist together on some of these infested pastures, although on others one species will predominate. Specimens from one pasture have been repeatedly identified as Lymnaea (Galba) bulimoides Lea, and such snails have been found infested many times. Specimens from this pasture are the only Lymnaea that have been artificially infested successfully. Lymnaeae from another flukeinfested pasture have been identified as Galba obrussa Say, Galba humilis Say, Galba ferruginea Hald, Galba bulimoides Lea, Lymnaea truncatula Draper, Galba desidiosa Say, Lymnaea parva Lea, and Lymnaea (Galba) palustris var michiganersis Walker. Only twice have infested snails been found on these pastures even after examination of large numbers of specimens at every season of the year. These parasitized snails seem to agree with Baker's description of G. ferruginea.

In one collection of five groups of specimens from five quite widely separated fluke-infested pastures all were returned Galba bulimoides Lea. Examination of these specimens revealed apparent differences even to the eyes of the authors. Mrs. D. Sinitsin, working with Dr. Sinitsin, Associate Zoologist of the Bureau of Animal Industry, U. S. Department of Agriculture, who was at this laboratory during the summer of 1929, made studies of the animal, jaw, radula, and genitalia of such specimens as had been found on infested pastures. The results of her studies we hope will soon be made available by publication.

LYMNAEA (GALBA) BULIMOIDES LEA

A good description of this snail has been given by Baker. His type specimen was obtained in Oregon. His measurements (up to 13.75 mm. in length) are apparently for mature specimens. This size is not common except in spring and summer months. At other times half-grown specimens are more numerous. Specimens that compare to his measurements apparently do not have as many whorls as he describes.

Habitat. Lymnaea (Galba) bulimoides can be found chiefly along the banks of small streams that come down from the hills. Some of these streams increase in size until they flow into larger streams, but a good many might be considered only springs as they flow for a short distance

and then sink into the ground. These seepy places on the hillsides are generally wet the year around. Such places are pretty well tracked over and the tracks fill with water; here it is that we find snails in great numbers. Such a stream is usually made up of several springs and these springs form excellent breeding grounds for snails. Along the streams that divide and converge are found so-called tule grasses, and these are quite typical of fluke-infested pastures. Water that becomes quite stagnant does not seem desirable to bulimoides as no specimens have been found in such water.

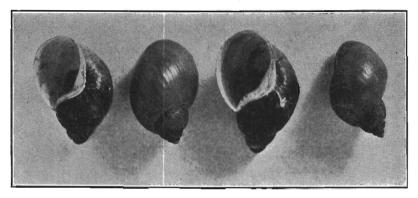


Fig. 3. Shells of Lymnaea (Galba) bulimoides Lea. Enlarged 3 times.

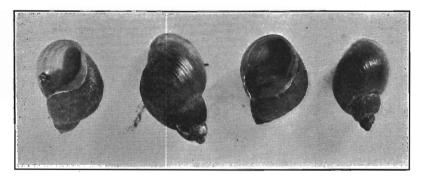


Fig. 4. Shells containing Lymnaca (Galba) bulimoides Lea. Enlarged 3 times.

In the spring these snails move from place to place. One year in February large numbers of half-grown snails were found in the ruts of a wagon road normally dry six months of the year. In a pasture snails were found in great numbers two hundred yards from any source of summer water. Possibly these snails had burrowed into the ground during the dry season. On one ranch where water was being piped from springs that had been dug out snails were found in the trough built to receive this water. This trough was several inches off the ground.



Fig. 5. Infested spring and bog hole.

Generally we think of fluke infestations as occurring only on very low land or at least chiefly on low lands. In this state the snail has been found at rather high elevations. Galba bulimoides Lea was collected from a spring that drains into the John Day river near Prairie City in Grant county at an elevation of 3,400 feet. Flukes were found in large numbers in native cattle

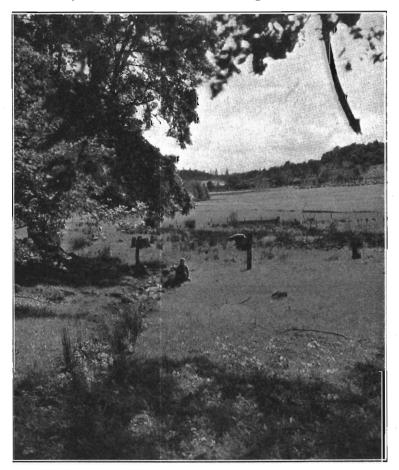


Fig. 6. Typical infested stream and pasture.

at Fort Klamath at an elevation of 4,200 feet. Some of the high meadows of the Malheur National Forest in Grant and Baker counties are notorious for their possibilities for fluke infestation. Some of these meadows are 5,600 feet above sea-level. Apparently the snail host is present over much of this state as flukes have been found in the following counties: Tillamook, Lincoln, Lane, Douglas, Curry, Coos, Josephine, Linn, Marion, Benton, Polk, Hood River, Klamath, Grant, Baker, and Union. Flukes

have been reported from Harney, Lake, Jackson, Malheur, Clatsop, Columbia, Multnomah, Washington, and Clackamas counties. *Lymnaeae* have been collected in Lincoln, Lane, Douglas, Coos, Josephine, Linn, Marion, Benton, Polk, Baker, Grant, and Klamath counties.

In some of these counties it gets as cold as 40° F. below zero for long periods. During the winter of 1929-30 snails were collected in Western Oregon on December 20 from water that was cold enough to freeze ice on the edges of a small creek. Snails have been gathered from this particular pasture every month in the year. In December, 1929, as many as 18 halfgrown snails were found in one horse track. Infested snails have been found as early as March 2 and as late as October 30.

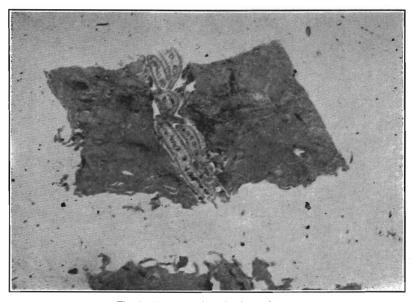


Fig. 7. Immature fluke in liver of lamb.

Effect of temperature on snails. On June 21, 1929, five snails varying in size from 3 to 5 mm. were placed in a refrigerator at a temperature remaining at 37° F. These snails were partly covered with water. On June 28, 1929, one snail had died, but on July 23 the remaining four were still alive. These snails lived more than one month in a watch-glass with no food.

Snail ova. Snails one-half grown (7 mm. long) are capable of depositing ova. Ova are deposited several times during the year. Collections have been made in October, November, March, April, May, June, and September. This would indicate the possibility of ova being deposited from March until late November.

Ova are deposited in fairly firm gelatinous masses, some round and some curved ovate in shape. These masses vary in size according to the ova contained. The number of ova contained varies from 2 to 70.

The size of the ovum when deposited is about 500 microns with an animal pole measuring 125 microns. Hatching time varies. Ova collected November 3, 1928 and March 25, 1930 hatched in 16 days' time. These young snails, hatched April 10, 1930, had grown to 2½ mm. in 20 days. Some variation in growth was apparent even in snails hatched the same date, some of the smaller snails measuring only 1.5 mm. after 20 days. In 32 days some were 3 mm.

Artificial infestation. At first much difficulty was met in keeping snails alive in aquaria. An attempt was made to accomplish this by putting them in wire baskets and arranging the baskets in the mud and water beside a small stream partly in the water leading from an irrigation ditch. Some of these snails were exposed to miracidia, but the mortality rate in both exposed and check snails was very high. Snails from a ranch three miles west of Corvallis were exposed to numerous miracidia on July 25, 1929, and kept in baskets as described above until October 7, 1929. During this time the temperature of the water varied from a maximum of 62° to a minimum of 52° F. Eight baskets were used with varying numbers of snails in each basket. On date of removal from creek, live snails were found in only two baskets. One basket contained three mature snails and the other baskets contained only immature snails. No infestation could be found in the three remaining snails.

It was found that by obtaining mud and plant life from native habitat and placing these in aquaria with snails and adding fresh water every other day or so, snails could be kept alive for more than six months. The material placed in the aquarium with the snails provided food and material upon which to crawl.

The first successful attempt at infestation was in snails collected from a pasture near Jefferson, Oregon, October 10, 1929. Specimens from this pasture had been repeatedly identified as Galba bulimoides Lea. On October 14, 1929, these snails, about one-third grown, were exposed to limited numbers of miracidia hatched from eggs collected March 13, 1929. On January 23, 1930, these snails were isolated in watch-glasses. On January 24, 1930, cercariae encysted on walls of the watch-glasses. These snails were placed in wire baskets on mud and on April 17, 1930 again isolated in watch-glasses. During the night 7 cercariae were discharged. It was 14 weeks from the time of infestation until the first cercariae were found to have been discharged and 6 months and 3 days until the second collection of cercariae. Curiously enough some of these snails, still alive on May 23, 1930, had made very little apparent growth.

Another attempt at infestation was made with specimens raised from ova collected and kept in wire baskets, the snails being at least two months of age but exact date of hatching not noted. These snails were exposed to great numbers of miracidia March 22, 1930. They were apparently all right for several days when the losses began. Of the 61 snails infested all but 3 had died at the end of 34 days. One of these was accidentally destroyed in changing material in the aquarium. Examination of this specimen April 15, 1930 revealed rediae in the various stages of development varying in size from 1 mm. to 2 mm. These rediae contained developing cercariae in numbers from 1 to 12. One redia contained 2 daughter rediae about 250 microns long.

On the same date as the foregoing attempt, March 22, 1930, snails variously identified (see page 8), obtained from another pasture, were exposed to miracidia in great numbers. They did not suffer the same mortality and many of them were alive May 1, 1930.

The first artificially infested snails examined by the authors were Lymnaea truncatula Müller received from Dr. R. F. Montgomerie, University College of North Wales, Bangor. These snails were packed in clay and were 12 days en route, arriving October 8, 1928. Nearly all the larger snails were dead, but one live one was quite thoroughly infested.

OTHER SNAILS

Physa and Polygyra are two very common genera of snails on fluke-infested pastures. Miracidia attacked one of these Physa, probably Physa triticea Lea.

Succinea stretchiana Gld. has been found on fluke-infested pastures but not in any great numbers. Other snails are present in the small streams that flow down from the hills. One that is especially common is Goniobasis plicifera var silicula Gld. This snail is nearly always thoroughly parasitized. Some five different cercariae had been found in this snail previous to 1929, but none were Fasciola hepatica.

REDIAE AND CERCARIAE

The description by W. Rees Wright' of Rediae and Cercariae is so complete that it is quoted.

"The Redia. The mature redia is an elongated sac approximately cylindrical in cross section, about .4 mm. in diameter, and varying in length from less than 1 mm. to more than 3 mm.; the mean length of a number of specimens containing mature cercariae was over 2 mm. The wall of the sac is very thin, generally less than 10 microns. For the greater part of its surface it is only one cell thick; in surface view it appears to be striated both longitudinally and transversely, this appearance being due to the presence of very delicate muscle fibres external to the principal layer of cells (these fibres can not be detected in sections). A massing of the transverse fibres, and to a lesser degree of the longitudinal fibres near the oral end gives rise under certain conditions of contraction to a well marked ring or collar as described and figured by Thomas; the appearance has rarely been met with among the very numerous specimens that I have examined. Thomas seems to think that the function of the collar is to maintain the cylindrical form of the body, but its position very near the anterior end of the body hardly supports this view; one would expect it to be situated nearer the middle of the body if it served this purpose.

"In the posterior half of the sac are two out-growths, situated close together. These were termed by Thomas the posterior processes. I suggest the term procruscula (from pro, in place of, and crusculum, a little leg) as being briefer and more indicative of the supposed function of these organs, which is to assist in the locomotion of the redia. There is a simple digestive system consisting of a mouth pharynx and a blind, thin walled digestive sac.

"There is also an excretory system, consisting of numerous flame-cells and their ducts which has not yet been completely elucidated. The main groups of flame-cells occur near the oral extremity and again near the procruscula, as Thomas pointed out.

"In none of the specimens that I have examined have I seen any trabeculae or strands of cells across the lumen of the sac, as described and figured by Thomas. Though indicated by Thomas, many subsequent authors have not pointed out that the redia is more than a passive parasite; in the case of very thin shelled snails it is often easy to see the rediae both devouring the tissue of the liver of the snail and also moving through it."

He makes note of the fact that the collar described by Thomas appeared only rarely. This collar has been seen here only in live specimens containing undeveloped cercariae.

Nothing can be added to the descriptions of the cercaria. Observations at this laboratory agree in all deatils with those of Wright.

The first cercariae collected from Galba bulimoides Lea were obtained August 20, 1928. The infested snails were obtained from pastures where sheep had died from so-called acute liver rot. These cercariae were obtained by crushing.

The measurements and description made of first rediae and cercariae at this time were as follows: cercariae body, 300 by 250 microns, tail 650 to 700 microns long, unarmed and having two suckers, ventral sucker somewhat larger than oral. Presence of material in body made determination of excretory pore impossible. Tail attached to body without notch. This cercaria moved with a rapid lashing of its long tail and seemed to encyst on contact. Cysts had milk-white appearance to naked eye.

The rediae measured $2\frac{1}{2}$ mm. by 360 microns and had at least one procrusculum. The mouth part was easily seen. The constriction or collar was present in some rediae. One out of about 100 snails was infested.

Naturally infested snails containing mature cercariae have been collected from pastures in Western Oregon as early in the year as March 2 and as late as November 3. A single snail discharged 580 cercariae in 48 hours.

ARTIFICIAL INFESTATION

The first guinea-pig was fed cysts October 1, 1928, destroyed fourteen days later, and several very small flukes were found. One was on the outside of the liver. The others had burrowed in, leaving well-marked tracts behind them. The fluke found on the surface of the liver measured 2½ mm. long by 1 mm. wide. Anterior sucker measured 200 and the ventral 250 microns. Measurements were made with the parasite under a cover-glass.

On November 7, 1928, 102 cysts were fed to a mature guinea-pig. These cercariae had encysted after swarming from snails. The guinea-pig was destroyed fifty-three days later, December 28, 1928 and two flukes recovered, one from the gall-bladder and one from the duct leading to the gall-bladder. Ova were found in the bile. Measurements were as follows: flattened under slip, 19 mm. by 6 mm. wide; not flattened, 12 mm. by 3 mm.

A mature rabbit was fed 120 cysts October 25, 1928, and 100 cysts November 2, 1928; destroyed December 28, 1928 and 10 flukes recovered, 5 mature and 5 immature. Ova were found in the bile ducts but not in the gall-bladder. The liver was very much enlarged and cirrhotic.

On November 2, 1928 a yearling ewe was fed 80 cysts. January 14, 1929 the ewe was destroyed and 11 flukes were found. No notes were made on the condition of the liver.

Cysts collected April 16, 1929 and kept in the dark on moist tissue paper were fed to guinea-pigs July 29, 1929. One pig was killed at fourteen days; no flukes were found in the liver and none could be obtained upon washing out peritoneal cavity with cold water. The other pig was destroyed after one month. No flukes were found in liver.

Cysts collected July 24 and 25, 1929 were fed to guinea-pigs, 25 cysts each, September 19, 1929. No flukes were recovered at fourteen days. On October 18, 1929 cysts that were collected July 24 and July 25, 1929, were fed to guinea-pigs. No flukes were recovered on autopsy. These cysts had been kept in water in corked vials at room temperature in the dark.

Wright noted that the cercaria developed a double wall when encysting. The outer wall is quite easily removed, leaving the encysted fluke transparent. The outer wall, at first milky white in appearance, changes later to a yellowish color. Wright suggested that the outer wall was developed merely as a method of attachment, but all encysted cercariae have this double covering, even those encysting on the surface of the water. The inner wall is very tough. Both artificial gastric juice and artificial pancreatic juice failed to digest this wall.

ATTEMPT AT NATURAL INFESTATION

Eight 6-months-old Angora goats were enclosed on a three-acre pasture where infested snails had been found. These goats were placed in this corral September 23, 1928, after fecal examinations had indicated that they were free from fluke infestation.

On November 4 and December 10, 1928, 2 goats were killed. No evidence of fluke infestation was found, but other parasites were very numerous.

On January 29, 1929, the 6 remaining goats were removed from the infested pasture.

On February 1, 1929, and February 12, 1929, 2 goats were destroyed. No evidence of flukes was found.

The 4 remaining goats were used in treatment work with the lungworm. Autopsies were held on all but one. No flukes or evidences of flukes were found.

SYMPTOMS AND LESIONS

These have been well described by several authors. In a good many cases symptoms have been emphasized as characteristic that are common to other diseases.

One symptom often noted is the one variously known as bottle jaw, water ball, or wattle. This is an edema that appears in the submaxillary space and often on the lower face. This symptom has been found in sheep suffering from infestation with immature liver-fluke only and also in animals infested with only mature parasites. It is commonly seen in sheep suffering from gastritis and enteritis, resulting from nematode infestation.



Fig. 8. Wattle under jaw caused by immature and mature flukes, also by stomach-worms and intestinal worms.

Very little has been said about the presence of pot-bellies during the early stages of infestation. These seem to be quite characteristic of immature fluke infestation when parasites are present in large numbers. The condition later disappears and is not present after parasites have developed to maturity. These pot-bellies are the result of fluid in the abdominal cavity. This fluid is generally clear but occasionally colored with blood. As many as five gallons have been removed from the abdomen of an average-sized ewe. Autopsies of such animals have revealed extensive fibrinous patches on both the posterior surface of the diaphragm and the surfaces of the liver. Such lesions are apparently only temporary as they have not been found in animals infested with mature parasites only.

Microscopic studies of livers in which large numbers of immature flukes were present in the tissue have frequently shown from two to five parasites in a single tract. No evidence that these were bile ducts could be detected. It is not known whether these flukes entered through the same wound in Glisson's capsule or whether one made the tract and the others followed.

In this state it is a popular opinion that scours is one of the most common symptoms of fluke infestation. It is often referred to as "the black scours." This is not a constant symptom, as many badly infested flocks have been observed which were not scouring. In one flock of badly infested goats this symptom was universally present. These goats also showed a marked stiffness that has not been noted in sheep. Autopsies revealed ascites, with both mature and immature flukes in the livers of these goats.



Fig. 9. Under-surface of liver showing small bile ducts and rough, discolored liver surface caused by immature flukes.

The lesions common to the four different stages of the disease as described by Neumann¹⁰ have all been noted in sheep. Often sheep will die suddenly in the first stage when only a few parasites are present. This has led to the belief that death was due to secondary invaders as reported by Dodd.¹¹ Our attempts to prove this by giving pathogenic anaerobes together with cysts of Fasciola hepatica have so far failed. In these experiments cultures of B. oedematiens, V. septique, B. novyi, and B. welchii were used, being administered both with the cysts and ten days afterward.

Lesions common to the first stage of fluke infestation have been observed in 5-months-old lambs, which were autopsied in July. Similar lesions have been found in 2-year-old ewes in December. Some of the parasites found at this time were less than 1 mm. in length, indicating according to Sinitsin¹² an age of not more than ten days. These particular sheep were dying without showing any symptoms. Only in some instances were lesions gross enough to indicate cause of trouble.

Livers of treated animals have shown remarkable progress in regeneration. Just how complete this regeneration might be has not been definite-

ly determined, but livers containing large numbers (more than 150) of mature flukes have shown very little gross pathology other than enlargement and thickening of the bile ducts and a few scars on the surface.

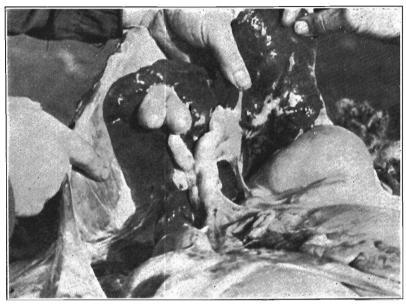


Fig. 10. Enlarged bile ducts filled with mature flukes. From this liver 179 flukes were taken.

The liver tissue is nearly back to normal.

TREATMENT

Experimental work with treatment has been entirely with the use of carbon tetrachloride. Montgomerie² demonstrated the value of carbon tetrachloride against fluke in sheep. Later Shaw and Simms⁸ proved its worth against fluke infestation in goats.

The first sheep treated for fluke by this Station were range sheep on winter feed of only hillside pasture. They were old ewes and in quite poor flesh. Occasionally losses occurred; autopsy proved that they were infested with fluke. On November 11, 1926, weight was obtained on the entire flock of 410 head. This weight of 35,500 pounds was obtained while sheep were dry. Treatment of 1 c.c. carbon tetrachloride was given. Two losses occurred, but on December 27, 1926, 408 head weighed 38,353 pounds. The sheep were dry on this day also and had been on pasture only. This gain in weight shows an average of 6.97 pounds.

Effect of treatment on immature fluke. Ewes and their ewe lambs that had been dropped in January and February were turned on to pasture in March. Both ewes and lambs began dying of immature fluke infestation in June. On July 19, 1928, a positive diagnosis was made by finding great numbers of immature flukes in each of two yearlings and three spring

lambs. The owner was told that treatment with 1-c.c. capsules of carbon tetrachloride would not be effective² and was advised to market the lambs which were in good condition. In spite of this advice the entire flock was treated with 1-c.c. doses and changed to a pasture with negative fluke history. Of this flock 250 were sold as fat lambs. The commission firm handling these reported that they were badly infested with flukes. Of the remaining 250 lambs 52 died by August 14 or within 26 days. At this time autopsies of two showed severe infestation with immature flukes.

By September 14 only 168 lambs remained. They were in very poor condition but were not scouring. A few showed edematous swellings of the submaxillary region. At this time a single lamb with severe symptoms was given 1 c.c. of carbon tetrachloride. Within three days the "wattle" had disappeared and the lamb showed marked improvement.

After selecting 12 of the poorest and weakest for laboratory experiments the rest were treated with 1 c.c. of carbon tetrachloride on September 17.

On September 20 the flock was observed, and marked improvement was noted. Treatment apparently had destroyed 3 lambs as they were dead the day following. The remainder of these lambs continued to improve with the exception of a few, and at the end of thirty days another treatment was given. No losses occurred following this. With no further treatment they developed into ewes of excellent quality.

Of the 12 lambs taken to the laboratory, 6 died without treatment by September 28, 1928. In 5 of the 6 that died the bile duct leading to the gall-bladder had ruptured and flukes were everywhere in the peritoneal sac. One lamb was treated six hours before death, and live flukes were found upon autopsy.

Fecal examinations were made of 2 of the remaining lambs on September 25. Ova were found in only one specimen. They were present at the rate of 156 per gram of feces. This lamb was given 1 c.c. of carbon tetrachloride the day of the examination. The other received the same-sized dose the following day. Both died on September 29. Autopsy of the first one treated revealed 150 partly decomposed mature parasites with gall-bladder containing a large number of poorly shaped ova. The other lamb revealed about 90 immature living parasites all in the main bile duct. No mature parasites or ova could be found.

Treatment of ewes with repeated doses of carbon tetrachloride. The ewes from the above-reported flock were treated on September 18, 1928. Losses had been severe and of the flock only 261 remained. These were in very poor condition, but only a few were showing so-called characteristic symptoms of fluke infestation. An occasional wattle could be seen and some had pot-bellies but no scours. Autopsy of one still warm revealed a ruptured bile duct, and 150 mature parasites were counted. Fecal examinations were made on 8 specimens and only a few ova could be found. Thinking to kill what parasites were mature and to cause the destruction of others as they developed it was decided to treat as nearly as possible every three weeks. On September 18, 1928, 261 were treated. Treatment disturbed the appetite somewhat, and 6 died in the next ten days. The rest showed marked improvement.

On October 20, 1928 the second treatment was given. There were no symptoms at this time and only a few ova in the feces. Three ewes had died in the past few days.

On October 24, 1928, an autopsy on a treated ewe killed in moving revealed only a few immature parasites. There was some fluid in the abdomen. The bile ducts were filled with a dark-colored material containing many ova.

Treatment with 1 c.c. carbon tetrachloride not repeated. Eleven goats, 17 bucks, and 63 ewes were treated October 18, 1928. Autopsy of a yearling ewe revealed 700 flukes in the liver, large enough to be mature. One animal in very weak condition died that night. All others made good recovery. The owner reported that more than 30 percent of the original band of sheep had died before treatment.

Effects of repeated dosing with 1 c.c. carbon tetrachloride in healthy animals. Beginning June 4, 1928, nine sheep and one goat were given 1 c.c. carbon tetrachloride daily until June 11, 1928. No ill effects were noted.

Clinical observations have led to the practice of trying the treatment on a few of the suspected animals to determine any sensitivity. All of the animals treated by the department have been sheep raised in the Willamette Valley and pastured on grass grown on acid soils. No effort was made to increase calcium content of the blood before treatment, and no indications of sensitivity to carbon tetrachloride have been noticed. Sensitivity has been reported in sheep maintained entirely on alfalfa hay and grasses grown on soils high in mineral content.

SNAIL DESTRUCTION

The work of Chandler¹ and later that of Walton and Jones⁹ demonstrated methods of snail destruction.

BLUESTONE AND LAND-PLASTER

Owing to the character of some of the infested pastures, it was realized that the destruction of the snail might be practical only under certain conditions. In order to demonstrate its practicability under these conditions the following attempts were made.

Plot 1 was 126 feet by 6 feet, mostly covered with water about 3 inches deep in which grass was growing abundantly and in which were numerous oak leaves. This low wet place contained large numbers of snails varying in size but apparently all of one species. This plot was treated April 18, 1929 with instant bluestone or copper sulfate and land-plaster (calcium sulfate), 1 part of bluestone to 8 parts of land-plaster. The materials were mixed and broadcast, the land-plaster outlining the treated area. The mixture was applied at the rate of 270 pounds per acre.

Observations on Plot 1, April 20, 1929. Of 200 snails collected 15 proved to be still alive when placed in fresh water. Most of the snails in the treated area were dead. Some live snails on mud or grass near water's edge may have crawled there since treatment. Rain fell during night of April 19, 1929. Observations on April 21 and April 23, 1929 showed a few live snails. Broadcast-

ing here killed 93 percent. The 100 check snails from untreated water were all alive.

Plot 2, April 18, 1929. This plot was the same as Plot 1 in size but was only partly covered with water. Partly bare, damp places of dark soil were fairly well covered with snails. Bluestone and land-plaster were applied at the same rate and same manner as on Plot 1.

Observations on Plot 2, April 20, 1929. Of 200 snails collected, mostly from moist land, 36 were found alive upon being placed in fresh water, but these snails were all dead April 21, 1929. No live snails were found on the plot April 23, 1929. There was thus an 82-percent kill in 48 hours and a 100-percent kill in 72 hours.

The 100 check snails from neighboring untreated land were all alive.

Plot 3, April 23, 1929. A ditch 1 foot wide by 1 foot deep containing much vegetable material such as leaves and grass was used. The water flowed rather swiftly, and moisture extended for about 1 foot on each side. The flow of water was stopped and one-half of the ditch was treated as soon as it had run dry, using the same mixture and in the same manner as in plots 1 and 2.

Observations on Plot 3, April 25, 1929. Of 100 snails collected, 4 were alive, representing thus a 95-percent kill. Other animal life in the ditch was destroyed.

The 100 check snails from the upper end of the ditch were all alive.

Plot 4, April 23, 1929. In this test 325 square yards of wet-land over-flow from a ditch was treated the same as the other plots. Snails were very numerous, together with snail ova.

Observation on Plot 4, April 25, 1929. Of 200 snails collected, 6 were alive, but none of them survived 24 hours, representing a 97-percent kill in 48 hours, and a 100-percent kill in 72 hours.

The 100 check snails were all alive and most had crawled from the container during the night.

COST OF PASTURE TREATMENT

The cost of pasture treatment, not including labor, was \$5.25 per acre. Instant bluestone is more expensive than crystals but goes into solution very readily and is more easily mixed with land-plaster. These plots, with the exception of Plot 3, contained fewer snails in 1930 than before treatment in 1929, which might have been due to directing the supply of water into a neighboring creek during the summer months.

DANGER OF PASTURE TREATMENT TO LIVESTOCK

Plot 5, April 30, 1929, 216 square yards in area, was treated with copper sulfate and land-plaster 1 to 8 at the rate of 270 pounds per acre.

One yearling sheep that had been on poor feed was placed on the treated land. The grass was plentiful, and the sheep began eating.

Observations were made daily. The sheep remained in apparent good health. Rain fell twice in 6 days.

Plot 5, May 6, 1929. On this date the grass was cut and retreated with twice the amount first used.

Observations were made daily. No ill effects were apparent.

THE USE OF DUCKS TO DESTROY SNAILS

In one pasture where water in the creek was more or less confined to one channel, one female duck and eight half-grown ducks were confined. They were constantly in the stream and seemed to make a difference in the number of snails during summer, although snails were found in the stream ten feet from where the female duck was confined, and the following winter and spring snails were very plentiful where the ducks had been kept.

DISCUSSION

It seems easily apparent that this problem demands considerable more work, especially on the bionomics of Fasciola hepatica and the different Lymnaeae apparently so common in this state. Observations seem to indicate that G. bulinoides Lea is more commonly and easily infected with fluke than is G. ferruginea Hald. With more knowledge of the possible snail hosts will come increased possibilities for destruction. It would seem entirely possible to eliminate these hosts, but in so doing much damage might be done to other animal life that might be having some beneficial influence on plant life.

Much work needs to be done with the treatment in order to determine just what makes sheep susceptible to carbon tetrachloride. According to the character of feeds used, sheep in the eastern part of the state should not be showing any evidence of calcium deficiency; but it is in this part of the state that sheep have shown a susceptibility to carbon tetrachloride. The use of the chemical might point the way to the cause of so-called pregnant-ewe paralysis, as certain ewes heavy with lamb have shown a definite susceptibility.

More work is needed on the possibility of secondary invaders playing a role in fluke infestation losses. Observations would lead one to believe in such a possibility, but so far experimental evidence at this Station is negative.

No word has been said here of the fluke problem in cattle, but thousands of dollars are lost annually because of liver condemnations, and cattle are also of importance as a source of snail infestation. No one can hope to eliminate this parasite from sheep and goats without considering the role cattle play in its spread.

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