

Lungworms (*Dictyocaulus Filaria* *Rudolphi*) in Sheep and Goats



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FOREWORD

This bulletin is of value to Oregon sheepmen, especially sheepmen of Western Oregon and those of Eastern Oregon using irrigated pastures, because it brings out at least four main points regarding lungworms infesting sheep, as follows:

1. The parasite lives but a short time after reaching maturity.
2. Effects of parasite are indicated by lack of growth even when symptoms are not pronounced.
3. Popular remedies against this parasite prove not only ineffective but dangerous.
4. Apparent resistance to infestation was built up by good feed and shelter. The visible effect of infestation was more apparent in animals on poor feed and those suffering from other disease.

From these four points, it is recommended (1) that lambs be watched closely enough to detect the first effects of these parasites; namely, loss of weight and bloom; and (2) that lambs be furnished better and more liberal feed.

Illustration on Cover

Figure 1. Eastern Oregon yearling artificially infested.

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SUMMARY

1. Artificial infestation of Guinea pigs, sheep, lambs, and one kid with *Dictyocaulus filaria* (Rud.) were successfully carried out, and information on the length of life of the parasite after reaching maturity was thus obtained. Parasites lived but a short time in animals on good feed and comparatively free from other parasites. Check animals, however, made the best gains.

2. Attempts to destroy this parasite in watch glasses proved the parasite to be quite resistant, apparently protected by the mucus secreted by the lining of the air passages.

3. Attempts to destroy the parasite in lungs of infested animals resulted in the death of hosts in cases where materials were used that successfully destroyed parasites *in vitro*.

Lungworms (*Dictyocaulus Filaria* Rudolphi) in Sheep and Goats

By

J. N. SHAW

INTRODUCTION

FROM time to time in the past, various treatments have been recommended as being efficient against the sheep lungworm, *Dictyocaulus filaria* (Rud.). The use of such treatments in practice, however, failed to reveal any such efficiency; and it was with the hope of finding a successful treatment that this investigation was started. Soon after beginning the project it was found that naturally infested animals could not be depended upon for experimental purposes, because of the scarcity of such animals during some years and because of the apparent seasonal appearance of the disease. Because of these facts, artificial infestation was attempted. At the time the work began, the complete life-history of the parasite had not been worked out. The migrations between the digestive tract and the lungs of the host were unknown until the Hobmaiers (1) determined that some of the stages were spent in the lymphatics. All the different stages found by these workers have been produced in the station laboratory. The life cycle of this parasite is quite simple, no intermediate host being necessary for its reproduction. The mature worms from two to three inches long are found mostly in the ends of the air passages in the lungs. Embryonated eggs are deposited by the females and hatch in the lungs. After hatching, the young worms are coughed up and swallowed by the host and are then passed with the droppings. After going through several changes or molts, the larvae are ready to infest the sheep. These molts are complete in ten days' time.

ARTIFICIAL INFESTATION

Cultural methods. Until the efforts of Romanovitch and Slavine (2) and Guberlet (3) were successful, no one had been able artificially to infest sheep with this parasite. Their results in four and two lambs respectively indicated a very high susceptibility.

Cultures used at this laboratory were obtained both from eggs derived from females removed from lungs of animals slaughtered for the purpose and from feces of infested animals. Tap water proved to be the most satisfactory media. Sterilized soil was used with different grasses growing in it but migration of the infective stage larvae took place only to a slight extent. All cultures were made at room temperature, and it was found that under these conditions larvae reached the infective stage in a maximum of ten days. In cultures made from eggs obtained from females in normal saline and in tap water, the larvae sometimes failed to develop to the infective stage.

In some cases these cultures decomposed and it was thought such decomposition was responsible for the lack of development to the infective stage. Washing of the larvae in the first stage was tried, but so many were lost during the process that it was felt other methods of obtaining cultures should be used.

A modified Baermann apparatus was used to collect larvae from feces. This apparatus consisted of an 8-inch glass funnel in which was placed a 5-inch soil screen. The stoppered funnel placed in a ring stand was filled with water until the fecal material in the soil screen was all in contact with water. After a period of three hours, the stopper at the tip of the funnel was opened and larvae removed to suitable containers. The feces were collected twice daily from clean straw in the small pen enclosing the infested sheep. As no other larvae were present in such fresh feces and as practically all larvae were able to free themselves from the feces in as short a time as three hours, there was practically no contamination of cultures. If the feces were allowed in contact with the water longer than three hours, other larval forms were present. Using the feces in pellet form without crushing, and removing the feces from the water at the end of three hours, gave a culture usually almost free of fecal debris and color. While no counts were made of larvae discharged from a sheep during a day, they were numerous enough to give the first 5 c.c. of fluid drawn from the Baermann apparatus a milky appearance. Third-stage larvae migrated to a very slight extent and were never found on the sides of containers. For the most part they remained coiled on the bottom but became very active when agitated. No growth took place, and both first-stage larvae and infective-stage larvae measured 500 microns. The cephalic button in the first-stage larvae is its outstanding characteristic. This disappears in the second and third stages, but in these stages the sheaths are present generally to aid in determining extent of development, although in some cases the first sheath is discarded. In the third stage or infective stage, the larvae are mostly at rest. Larvae of the fourth stage removed from the mesentery lymph glands were unshathed but measured the same as other larvae.

Infestation of mature sheep. Attempts have been made to infest 19 mature sheep of various ages, using from 400 to 26,000 infective-stage larvae per sheep. Parasites were counted for feeding purposes by counting the numbers of larvae in five different drops of a drop pipette and taking the average. Larvae were given to sheep either in capsules or by stomach tube. Parasites developed to maturity in 3 of the 19 sheep. In one other ewe, destroyed for other reasons, six half-grown parasites were found. This ewe had a very severe case of mastitis. (Ewe fed May 24, 1933, killed June 16, 1933.)

Two of the three sheep in which mature parasites developed were suffering from a progressive pneumonia peculiar to the Northwestern states. Parasites reached maturity in only one normal mature animal. In one ewe, in which many immature parasites were found upon autopsy, the parasites were fed over a period of thirty-one days, a fact which might account for the many immature parasites.

Infestation of lambs. Records have been kept on attempts to infest 27 lambs and 1 kid. With only two exceptions, all of the lambs and the kid

showed evidence of infestation either by coughing or by the presence of larvae in the feces. Autopsies were held on 10 lambs and 1 kid. In 6, worms were found. In the other 4, definite lesions of lungworms were found but



Figure 2. Infested lambs.



Figure 3. Non-infested lambs—sixteen pounds difference in weight.

no worms, although the hosts were killed at from 22 to 35 days after larvae had been fed. Apparently the lambs had been successful in throwing off the worms before they had reached maturity.

In another group of 8 lambs on which weights were kept, the 4 not receiving larvae made a gain of 8.4 pounds more than the 4 fed larvae over a three-month period, although at the beginning the fed lambs aver-

aged 1.9 pounds more than the checks. Two of the lambs coughed somewhat one month after being fed, but no larvae were ever found in the feces of the four lambs fed. Lambs were weighed when three months of age.

In still another group of 5 wether and 8 ewe lambs two months of age, the ewe lambs made an average gain of sixteen pounds more than infested wether lambs on same feed and separated only by panels. In the group of wether lambs, symptoms were pronounced and larvae appeared in the feces for a period of 9 weeks from date of infestation. Lambs were last weighed when 4 months of age.

Apparently under some conditions lungworms do not live long after reaching maturity. In two lambs fed infective-stage larvae May 17, 1932, larvae appeared in the feces on June 24, 1932, and could not be found on July 11, 1932. Autopsy on one lamb July 22, 1932, revealed lungworm lesions but no worms. The other lamb killed August 3, 1932, revealed five worms, 3 females and 2 males, none of which appeared mature, being only half the usual length of mature worms. The females did not deposit eggs when placed in warm normal saline solution. In these lambs, larvae were discharged for a period of 17 days only. They were on grass hay and oats with ewe's milk, but kept confined indoors.

Table 1. INFESTATION OF LAMBS

Lamb	Larvae fed	Larvae in feces	Symptoms	Autopsy
1	629	*	No coughing	Alive
2	1,140	*	Coughing	Alive
3	912	*	Coughing	Alive
4	798	*	No coughing	Alive
43	5,400	Killed	No coughing	Larvae in mesentary lymph glands
44	5,400	†	Coughing	Recovered, no autopsy
45	5,400	†	Coughing	Recovered, no autopsy
48	5,400	†	Coughing	Recovered, no autopsy
52	3,463	† Killed	Coughing	Pneumonia—no worms
53	3,463	† Killed	Coughing	Pneumonia—5 mature worms
86	13,000	Killed or died before larvae could appear in feces	Coughing	Pleural adhesions—no worms
87	13,000	Killed or died before larvae could appear in feces	Coughing	Slight pneumonia—no worms
88	13,000	Killed or died before larvae could appear in feces	Coughing	Pneumonia—no worms
89	13,000	Not examined	Coughing	No autopsy
90	13,000	Not examined	Coughing	No autopsy
93	18,900	*	Coughing	Alive
94	18,900	*	Coughing	Alive
97	22,000	*	Coughing	Alive
98	22,000	Died	Coughing	Pneumonia—immature worms
101	13,500	*	Coughing	Alive
102	18,900	*	Coughing	Alive
104	18,900	Died	Coughing	Slight pneumonia—1 mature worm
105	18,900	*	Coughing	Alive
A.H.	10,260	Killed	No coughing	Larvae in mesentary lymph glands
Toby	13,000	†	Coughing	Alive
Blind lamb ..	10,000	Died	Coughing	Pneumonia—many worms
White lamb ..	10,000	†	Coughing	No autopsy
Goat kid	1,640	Died	Coughing	Pneumonia—many immature worms

* No larvae in feces. † Larvae in feces.

Artificial infestation of guinea pigs. In order to check on the effectiveness of the third-stage larvae, nine guinea pigs were fed. Of the nine pigs fed, larvae proved infective in six. One pig lived long enough for the parasites to reach maturity. The other five died as the result of pneumonia produced by the worms. Large numbers of larvae were given but no difference could be detected in the severity of the disease in those fed what might have been considered large doses and those fed much larger doses. Parasites developed to maturity more than a week earlier in guinea pigs than in lambs, the shortest time being twenty-eight days, while in lambs it was thirty-five days before larvae appeared in the feces.

TREATMENT EXPERIMENTS

Treatments of worms in vitro. Realizing that any material used to destroy lungworms in the lungs would very probably kill by contact, it was thought advisable to try out various treatments on worms *in vitro*. Materials were applied to worms in watch glasses and in lungs of sheep recently destroyed. In watch-glass preparations, worms were in some cases washed in saline solution before being placed with materials to be used in treating. Unless washed, worms seemed to be somewhat protected by surrounding mucus. The following materials were used with the results indicated:

- Beech wood creosote—killed immediately.
- Carbon tetra-chloride—killed immediately.
- Germifectant—killed immediately.
- Normal saline and carbolic acid .5%—killed immediately.
- Carbofuchin—killed immediately.
- Normal saline solution—lived 48 hours.
- Mag. sulfate saturated solution—lived several hours.
- Mercurochrome 2% solution—lived 12 hours.
- Neo Arsphenamine .3 gms. to 4 liters—lived 1 hour.
- Metaphen 1 to 500—lived 1 hour.
- Mercuric salicylate with quinine and Urea Hydrochloride $\frac{1}{2}$ %—lived $\frac{1}{2}$ hour.
- Emetine hydrochloride 2 gr. in 1 c.c. water—lived 45 minutes.
- Oil of pine tar; tar oil $\frac{1}{2}$ casine and sodium bicarbonate for emulsion—killed in 15 minutes.
- Pinol (Coopers' product) 2% solution—killed in 10 minutes.
- Pinol (Wm. Coopers and Nephews) 1% solution—killed in 10 minutes.
- Exposed for 2 minutes to 2% solution and removal to normal saline—did not kill worms.
- Oil from Pinol (distilled from Coopers' Pinol)—killed immediately.
- Procaine 1% solution—no effect.
- Tetrachlorethylene—killed immediately.
- Glycerin and creosote, equal parts—killed immediately.
- Creosote and alcohol 1 to 10—killed immediately.
- Hexylresorcinal—killed immediately.
- Hexylresorcinal in lungs removed from carcass—alive after 3 hours.
- Iodine fumes in NaCl solution—killed immediately.
- Iodine fumes in water—killed immediately.
- 4 $\frac{1}{2}$ % Pyrethrins in alcohol—killed immediately.
- 4 $\frac{1}{2}$ % Pyrethrins and olive oil, equal parts—killed in 10 minutes.

- 4½% Pyrethrin and olive oil (dipping and washing)—no effect.
4½% Pyrethrin (olive oil extract)—killed almost immediately.
Olive oil—alive after 4½ hours.
Hexylresorcinol crystals ¼ gm. and olive oil 5 c.c.—killed in 4 minutes.
Hexylresorcinol and olive oil, 5 c.c.—alive after 3 hours.
Oil of Chenopodium and olive oil, 1 to 20—alive after 2½ hours.
Oil of pine tar and olive oil, 1 to 5—alive after 2½ hours.

Treatments of worms in vivo. Many materials have been credited with the ability to destroy lungworms in the lungs of sheep and goats. In most cases such materials have been injected into the trachea or inhaled in the form of a gas. It was thought possible that materials injected intravenously might prove efficient. With this in view and with the hope of finding something of value, the materials listed in the following table were tried. It was realized from the first that an effective material must be non-irritating to the mucus linings of the air passages and at the same time lethal to lungworms. All materials used with the exception of olive oil proved to be irritating to the mucus membranes of the air passages. Olive oil in as large a quantity as 60 c.c. was injected without producing irritation. Measuring the capacity of the air passages of the average-size sheep it was found that such passage would hold about 2 ounces of fluid. For this reason it was thought advisable to give fairly large amounts of materials so as to assure the possibility of contact with the worms, which are usually in the small ends of the air passages.

Materials used are listed in Table 2 with results as indicated.

DISCUSSION

While the results of these experiments are mostly negative so far as finding a successful treatment is concerned, some interesting facts have been brought to light. First of all, it seems very apparent that other factors are involved in the infestation of sheep with this parasite besides the number of infective larvae. In most cases where natural infestation was great, other parasites were present in large numbers. In one flock where infestation was severe, flukes were causing losses and the infested animals were on poor rations. Just how much influence feed has on the susceptibility of animals has not been definitely determined, but the experiments of this laboratory indicate that feeds are far more important than has been supposed. Young animals that have been successfully infested have maintained good condition on good feed, while yearlings on poor clover hay lost condition when infested. Two lambs and a ewe on poor grass hay developed symptoms, and both lambs discharged larvae in the feces. The ewe coughed somewhat and lost weight very rapidly. The loss of weight was very probably due to this poor hay although the parasites might have been partly responsible. The better gains made by check animals in two experiments indicate the pathogenicity of the parasites.

Guinea pigs on a deficient diet developed symptoms and died one week sooner than pigs on usual diet given guinea pigs.

Age seemed to be a very important factor, as it was impossible to infest older animals except in cases where the animal suffered from some

other disease. In spite of large numbers of larvae given, no symptoms could be produced with the exception of some coughing, and no larvae could be found in the feces except in the cases mentioned.

Table 2. RESULTS WITH VARIOUS MATERIALS FOR DESTROYING LUNGWORMS

Material	Animal	Amount	How used	Results
Chloroform	Sheep	1 c.c.	Intravenously	Killed by injection, worms alive
Chloroform	Sheep	5 c.c.	Intravenously	Killed by injection, no worms
Carbon tetrachloride	Sheep	1 c.c.	Intravenously	Died 3 days later, live worms
Cresote 5 c.c., alcohol 15 c.c., glycerine 5 c.c., water 25 c.c.....	Sheep	5 c.c.	Intravenously	Killed 1 month later, live worms
Cresote 5 c.c., alcohol 15 c.c., glycerine 5 c.c., water 25 c.c.....	Sheep	10 c.c.	Intravenously	Killed 8 days later, live worms
Cresote 5 c.c., alcohol 15 c.c., glycerine 5 c.c., water 25 c.c.....	Sheep	10 c.c.	Intratracheally	Pneumonia
Cresote 5 c.c., alcohol 15 c.c., glycerine 5 c.c., water 25 c.c.....	Sheep	15 c.c.	Intratracheally	Pneumonia
Cresote 5 c.c., alcohol 15 c.c., glycerine 5 c.c., water 25 c.c.....	Sheep	10 c.c.	Intratracheally	Killed 2 days later, live worms
Cresote 5 c.c., alcohol 15 c.c., glycerine 5 c.c., water 25 c.c.....	Sheep	5 c.c.	Intratracheally	Killed 2 hours later, live worms
Nearsphenamine	Sheep	.15 gm.	Intravenously	Killed 2 months later, live worms
Nearsphenamine	Sheep	.3	Intravenously	Died 2 months later, live worms
Hexylresorcinal crys. in olive oil	Sheep	$\frac{1}{2}$ gm. in 5 c.c.	Intratracheally	Killed 4 days later, live worms
Hexylresorcinal	Sheep	1 gm. in 5 c.c.	Intratracheally	Died next day, live worms
Hexylresorcinal.....	Goat	60 c.c.	Intratracheally	Killed goat in 15 minutes, no worms
Hexylresorcinal.....	Goat	30 c.c.	Intratracheally	Goat recovered
Tetrachlorethylene ..	Sheep	30 c.c. in olive oil	Intratracheally	Killed sheep
Tetrachlorethylene ..	Sheep	5 c.c.	Intratracheally	Sheep killed, no worms
Tetrachlorethylene .. NaCl solution + 5% carbolic acid	Goat	30 c.c.	Intratracheally	No apparent ill effects
NaCl solution + 5% carbolic acid	Goat	60 c.c.	Intratracheally	No apparent ill effects
NaCl solution + 5% carbolic acid	Goat	225 c.c.	Intratracheally	Killed without struggle, live worms
Chandlers iodine, vermicide	Sheep	60 c.c.	Intratracheally	Died that night, live worms
Chandlers iodine, vermicide	Goat	30 c.c.	Intratracheally	Died in 20 days, pneumonia
Iodine fumes	Goat	Fumes	Inhaled few seconds	No apparent ill effect
Iodine fumes	Goat	Fumes	Inhaled few seconds	Died 11 days later, live worms
Iodine fumes	Goat	Fumes	Inhaled 5 seconds	Died 3 days later, no worms
Chlorine gas	Sheep	Gas	Three minutes	Died that night, live worms
Chlorine gas	Sheep	Gas	Two minutes	Marked discomfort
Sterile olive oil.....	Sheep	5 c.c.	Intratracheally	No distress or coughing
Sterile olive oil.....	Sheep	10 c.c.	Intratracheally	No distress, some coughing
Sterile olive oil.....	Sheep	20 c.c.	Intratracheally	No distress, some coughing
Sterile olive oil.....	Sheep	60 c.c.	Intratracheally	No distress, some coughing
Sterile olive oil and carbo fuchin..	Sheep	30 c.c.	Intratracheally	Killed in 17 days, no worms Much distress, coughing, difficult breathing

Reinfestation of susceptible animals proved possible to a limited extent. Symptoms of coughing developed in five out of eight yearlings, and larvae appeared in the feces of three of the animals used for this purpose. In

these cases where reinfestation took place the infective stage larvae were given in smaller doses over a period of four weeks. Under natural conditions it would hardly be conceivable that sheep would be exposed to larvae in any such doses as used in these experiments.

Treatments used did not prove successful in destroying worms and were in all cases very irritating to the lining of the air passages. Parasites seemed to be protected by the mucus. Here again the mucus seemed to have the same power of protection as in the case of the worms treated *in vitro*, showing the futility of attempting to destroy worms in the air passages where the mucus is abundant because of the parasites. The location of the parasites in the air passages would make the application of a treatment seem highly impractical. The use of colored solutions in a few cases indicated that solutions did not succeed in reaching all parts of the air passages.

No effort has been made to describe the various stages in the development of this parasite as the descriptions published by Daubney (4) are considered very complete.

Much work has yet to be done to learn the reasons for the low degree of susceptibility of animals to artificial infestation and what factors are responsible for the ability of the host animal to rid itself of this parasite in such a short time. Kauzal (5) found that some animals remained infested 84 to 98 days. In all our artificial infestations, no animals remained infested longer than 56 days.

Mature parasites proved to be very resistant even *in vitro*. In normal salines mature worms remained alive 3 days, while worms in the lungs of a dead sheep remained alive 6 days. Full-strength hexylresorcinol failed to kill worms in air passages of lungs from dead sheep in 3 hours.

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