STATION BULLETIN 367 OCTOBER 1939

Pulpy Kidney Disease in Oregon Lambs

(Infectious Entero-toxemia)

J. N. SHAW, O. H. MUTH, and L. SEGHETTI

Oregon State System of Higher Education
Agricultural Experiment Station
Oregon State College
Corvallis

TABLE OF CONTENTS

Pulpy Kidney Disease in Oregon Lambs

(Infectious Entero-toxemia)

J. N. SHAW, O. H. MUTH, and L. SEGHETTI
Department of Veterinary Medicine

INTRODUCTION

For many years the sheep breeders of Curry County have reported heavy losses in their best lambs. These losses have occurred on lush pastures after the lambs are six weeks to two months of age. In this particular county, losses usually begin in early April and end not later than July. Some breeders report losses as high as 25 per cent, although such heavy losses have not been experienced since our investigation started. No popular common name has been given to this disease. Most animals were found dead and so thoroughly devoured by buzzards that nothing characteristic could be detected by the owners, except that death usually occurred rather suddenly. After the investigation started, it was found that in most of the cases examined the kidneys were dark and very soft; hence the common name "pulpy kidney." This is now the accepted common name among the workers with this disease in England, Australia, and New Zealand.

This disease was first brought to the attention of members of the Department of Veterinary Medicine in 1933, at which time it was believed that some poisonous plant was involved. It was then recognized that the disease was similar to sudden losses occurring on rape pastures in the Willamette Valley. Such losses were in older lambs, but lesions were quite similar. In the Valley, the breeders believed the disease to be hemorrhagic septicemia, and, in spite of unfavorable results, continued to use vaccine for protection against this disease. Efforts to isolate hemorrhagic septicemia organisms, however, met with failure.

During the spring of 1939, typical losses occurred early, not only in Curry County, but in Benton, Douglas, Lane, Linn, Marion, Morrow, and Lake Counties. One typical case occurred in the Station flock.

This year, the spring being early, the lambs were on lush pastures at about the same age throughout the state, and typical pulpy kidneys were found in lambs two weeks old. Pulpy kidney disease assumes much more importance when we study the losses and their distribution this past spring. On one farm near the Station a 12.5-per-cent loss was experienced on cultivated pastures, where heretofore with the use of permanent pastures the disease had apparently not been experienced.

Intensive investigation of this disease was made possible by legislative appropriation of special state funds, and such investigations were started in April, 1937.

SYMPTOMS

Opportunities to study symptoms in lambs dying under natural conditions have not been very great, owing to the suddenness with which lambs die. In those cases that have been studied, the following symptoms have been noted. Weakness, lack of muscular control, frothing at the mouth, convulsions, and evidences of distress. Owners have remarked that lambs seemed to be in extreme pain. In some cases lambs able to stand would stand with head against the fence. In many cases evidences of scouring were noted. Lambs killed by the injection of toxins or intestinal washings died usually in convulsions and in considerable pain.

Figure 1. Type of pasture where this disease has existed for years.

LESIONS

No doubt the most characteristic lesion is the pulpy condition of the kidney. The kidneys usually show pathology, although this is not always present. In well marked cases, the consistency is much like that of raspberry jam. Dr. E. T. Bell of the University of Minnesota states in a personal communication that this "suggests a gas 'bacillus' infection." Notable also are the lesions of the heart. The pericardial sac often has an excess of fluid, and small hemorrhages are found under the epicardium and endocardium. The subendocardial hemorrhages are usually in the left ventricle.

The mucosa of the abomasum is congested and a similar condition is found in the last portion of the small intestine. In one lamb recently dead, twelve intussusceptions were found. Intestinal washings from this lamb were very toxic.

TOXICITY OF INTESTINAL FILTRATES

Since the toxicity of intestinal contents of animals dying from a similar disease has been proved by others, one of the first things attempted in these investigations was to determine the toxicity of the intestinal content from animals dying from this disease. Since it was not always possible to obtain lamb carcasses immediately after death, examination of the intestinal content was not regularly carried out.

Material from the posterior two-thirds of the small intestine was usually selected. This was mixed with an equal quantity of physiological salt solution, centrifuged for from thirty minutes to one hour at 2,000 r. p. m., decanted and passed through a Seitz E. K. filter. It was then injected intravenously.

INTESTINAL FILTRATE FROM LAMBS DYING FROM THE DISEASE

Experimental Animal	Amount given	Results
Lamb	20 cc.	Death in 5 minutes
2 yr. wether	20 cc.	Death in 1 hour
2 yr. wether	20 cc.	Death in 1 hour
Lamb	40 cc.	Death in 2 hours
4 yr. wether	45 cc.	Death over night
Lamb	20 cc.	Death over night

INTESTINAL FILTRATE FROM NORMAL LAMB

Experimental Animal	Amount given	Results
Lamb	35 cc.	Lived

An attempt was made to neutralize the toxic substance in intestinal filtrate from a typical case. The filtrate was prepared in the same manner as above. It was then divided into two equal quantities, to one of which was added 12 cc. of Wellcome Lamb Dysentery Antitoxin. Both were well shaken and allowed to stand for one hour at room temperature. A sixty-pound lamb was then injected with each lot of material.

INTESTINAL FILTRATE FROM LAMBS DYING FROM THE DISEASE

Lamb	Amount given	Results
1	87 cc. intestinal filtrate	Dead in 5 minutes
2	87 cc. intestinal filtrate plus 12 cc. antitoxin	Lived

The symptoms shown by the animals dying as the result of receiving intestinal filtrates were similar to those observed when toxin was administered. Necropsy findings were also similar.

The neutralizing effect of antitoxin on intestinal filtrate was also tried, using rabbits as test animals. Intestinal filtrate was prepared from a typical case in the usual manner. It was then divided into two portions, to one of which was added an equal quantity of Wellcome Lamb Dysentery Antitoxin. After mixing and standing one hour at room temperature, a quantity of each was injected intravenously into a rabbit.

INTESTINAL FILTRATE FROM LAMBS DYING FROM THE DISEASE

Rabbit	Amount given	Results
1	1.5 cc. intestinal filtrate	Dead in 1 hr. 20 minutes
2	1.5 cc. intestinal filtrate plus 1.5 cc. antitoxin	Lived

CAUSE

Although there may be several unknown contributing factors to this disease, Bennetts (1) has concluded that the rapid growth of *Clostridium perfringens* (Type D),* with the elaboration of its toxin in

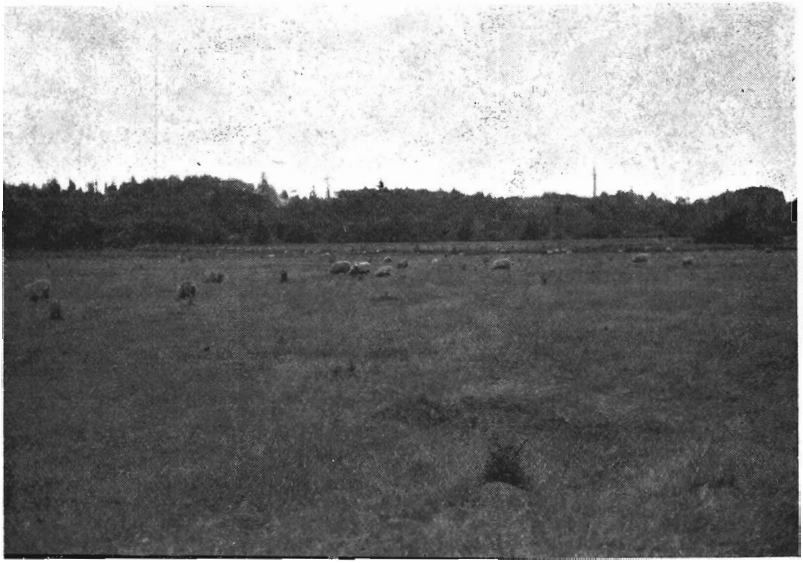

Figure 2. Losses also occur on coast bottom land.

the small intestine under conditions wherein the toxin is rapidly absorbed, accounts for the immediate symptoms and death in lambs. The following work was carried out with the object of determining whether this disease of Oregon lambs was due to the same cause.

BACTERIOLOGICAL STUDIES

Isolation of organism. Microscopical and cultural examinations of tissues and blood made immediately after death from naturally occurring cases of the disease were negative. Smears from the ileal portion of the intestine showed a preponderance of *Cl. perfringens*-like organisms, as compared with

* Bergey's Manual of Determinative Bacteriology, Fifth Edition, approves the terminology *Clostridium perfringens* in reference to that group of bacteria previously referred to as *Clostridium welchii*. It appears logical and convenient to retain the serio logical type classification of Wilsdon; namely, types A, B, C, and D.

smears taken at various times from normal lambs or lambs dying of other causes. Investigators who have worked with this disease have consistently isolated *Cl. perfringens* Type D as the pathogenic organism. In 1937 *Cl. perfringens* Type A was recovered from the intestinal flora of natural cases.

Eleven strains of *Cl. perfringens* Type D (*Cl. welchii* Type D) were isolated in pure culture from the small intestine of natural cases of this disease. The procedure employed in the isolation of these strains was based on that described by Bennetts (1) and others who have worked with the *Cl. perfringens* group.

A primary culture was obtained from the ileum and inoculated into horse-flesh infusion containing egg-meat (Difco) pH 7.4. The cultures were incubated for 24 hours at 37° C. and then heated to 80° C. for 20 minutes to destroy non-spore-forming organisms. Rapid subcultures were then made through a series of glucose broth media and then made into litmus milk. The "stormy fermentation" reaction produced in this medium was tentatively accepted as evidence of the presence of the *Cl. perfringens* group. One loopful of this culture was transferred to a deep tube of 1 per cent veal infusion agar pH 7.4. The contents were then well mixed and a loopful was transferred to a second tube of the same medium. This was repeated, until a series of four tubes was inoculated. These cultures were incubated at 37° C. for 24 hours. The tube showing the most discrete colonies was then selected. The culture was transferred to a sterile Petri dish by means of a sterile capillary pipette inserted to the bottom of the tube. The cylinder of agar was seared with a hot spatula, and isolated colonies were picked to horse-flesh infusion egg-meat medium, which had been previously boiled and cooled. The process was repeated until a pure culture was obtained. After 24 hours incubation in meat broth, subcultures were again made into agar shakes to note any differences in colony type that might indicate the presence of more than one species. Cultures were judged to be pure on the basis of similarity of colony types together with the morphology of the organism in the meat cultures and deep colonies.

Morphology. The morphology of these strains as shown by the Gram stain were characteristic of the *perfringens* group. They were Gram-positive rods of varying sizes, usually occurring singly. Spores were not easily demonstrated, but have been observed in cultures using coagulated egg albumin (Difco) and also in material taken directly from the intestine. They are oval and either central or subterminal in position. Organisms recovered from animal tissue frequently are encapsulated. No motility has been observed in hanging drop preparations.

Cultural characteristics. The bacilli are anaerobic, no growth occurring under aerobic conditions. The alkaline pyrogallic method of Buchner was used to secure anaerobiosis and all liquid media were boiled and cooled to 40° C. previous to inoculation. All cultures were incubated at a temperature of 37° C. All the strains isolated closely resemble *Cl. perfringens* (*B. ovitoxicus* described by Bennetts) and agree in the following cultural characteristics: the shape and form of colonies, the "stormy fermentation" developed in milk, and the softening of gelatin. Gas and acid are produced in dextrose, maltose, lactose, saccharose, glycerol, and levulose, but mannite, salicin, dulictol, or inulin are not fermented.

The various biochemical reactions characteristic of the *perfringens* group can scarcely be depended upon to differentiate these specific strains. While

there are some characteristic reactions for the classification of *perfringens* strains, there is also much ambiguity. In view of what has been established in the critical study of these organisms, the emphasis laid on the antigenic components of the different strains is the underlying principle in the differentiation of strains. These antigenic properties can best be demonstrated by toxin-antitoxin reactions.

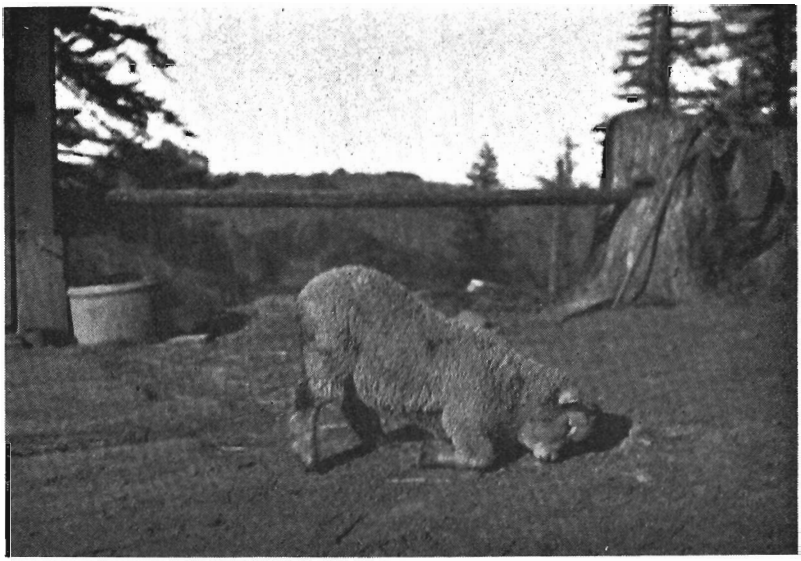

Figure 3. Lamb showing symptoms.

Pathogenicity. Cultures of the different strains were tested for pathogenicity by intramuscular injections into guinea pigs and sheep. Two-tenths of a cubic centimeter and 5 cc. of a 24-hour meat-broth culture respectively, were fatal to guinea pigs and sheep over night. Sheep on necropsy showed extensive swelling of the injected quarter. The subcutaneous tissue of the injected leg showed a blood-tinged emphysematous edema accompanied by muscular necrosis. The mesentery showed a generalized congestion, the most marked lesions being congestion and softening of the kidneys. The heart contained numerous subendocardial hemorrhages in the left ventricle.

Preparation of toxin. The medium used for the preparation of toxin was horse-meat infusion to which egg-meat medium (Difco) was added. The reaction was adjusted to pH 7.6 and a sterile solution of glucose was added at the time of inoculation to make the final concentration 0.2 per cent. The medium was heated and cooled to approximately 40° C. before inoculation. The flasks were inoculated by removing aseptically at the time of death a portion of the pectoral muscle of a pigeon previously inoculated with 5 cc. of a 24-hour culture. The cultures were incubated at 37° C. for 60 hours. At this time they were filtered through cotton, centrifuged at 2,500 r. p. m. for 30 minutes and the supernatant fluid

decanted. This method proved as satisfactory as filtration through a Seitz filter and especially more useful in dealing with a number of cultures.

No attempt was made to prepare toxin from each strain by this method, since the supply of immune sera obtained for cross-neutralization tests was limited. This method was used because it yielded toxin of greater potency than did other methods. The individual strains showed some fluctuation in their toxin-producing power. Small quantities of all strains, however, proved toxic to laboratory animals.

Adopting the method outlined above, strains of EW and L3 were selected as typical strains for further study of the properties of the toxin and for type determination.

Properties of the toxin. Small quantities of the toxin produced by these strains when injected intravenously were lethal to laboratory animals and sheep. The interval before death varied with the size of dose, in some cases being only a few minutes. The lowest lethal dose for mice was 0.005 cc., while for a 90-pound sheep 7 cc. proved fatal. The toxin is thermolabile. Animals injected with toxin that previously had been heated for 30 minutes at 70° C. remained normal. Toxin that was stored in the refrigerator at a temperature of 40° C. for two weeks showed no appreciable diminution of toxicity. The hemolysin produced varied with the period of incubation. After 60 hours of incubation, the hemolytic power of the toxin in vitro was almost negligible. Intradermal injections produced necrosis of the surrounding tissue. Toxin was rendered atoxic when incubated one week at 37° C. in the presence of 0.2 per cent formalin.

Toxin-antitoxin tests. A number of workers in different parts of the world have studied the *Cl. perfringens* group of organisms. Wilsdon (2) in an extensive study of this group showed that the toxins of the *Cl. perfringens* group could be divided into four distinct serological types.

Type A strains produce Type A toxin and the antitoxin neutralizes Type A toxin only.

Type B strains produce Type B toxin and the antitoxin neutralizes Types A, B, C, and D toxins.

Type C strains produce Type C toxin and the antitoxin neutralizes Types A, B, and C toxins.

Type D strains produce Type D toxin and the antitoxin neutralizes Types A and D.

In order to classify the type of toxin produced by the strains EW and L3, antitoxic sera corresponding to Types A, C, and D were obtained through the courtesy of Dr. R. F. Montgomerie of the Wellcome Physiological Research Laboratories. Type B (Lamb Dysentery Antitoxin) was obtained from Wellcome Physiological Research Laboratories.

The toxin and antitoxin were mixed in appropriate quantities and allowed to stand in the dark for one hour at room temperature. In carrying out the tests, each of the four types of sera (A, B, C, and D) was added in the proportion of three parts of toxin to two parts of antitoxin. The dose of toxin employed was many times the lethal dose for mice. The maximum quantity of the total volume of each mixture used did not

exceed 0.5 cc. Mice were used throughout the experiments, and all tests were carried out in duplicate and confirmed twice before being accepted.

It was found that Type D and Type B antitoxin afforded complete protection against the unknown toxins. The neutralizing effect of Type A or Type C antitoxins was negligible. The animals receiving these sera died nearly as quickly as those receiving toxin alone. Wilsdon (2) and Bennetts (1) have shown that Type B antitoxin is capable of neutralizing Type D toxin, but Type D antitoxin does not neutralize the toxin of Type B. Since Type B cultures were not available, no attempt to test the action of Type D antitoxin on Type B toxin was made. The symptoms and lesions at necropsy in affected lambs are evidence that the organism dealt with is not the lamb dysentery bacillus.

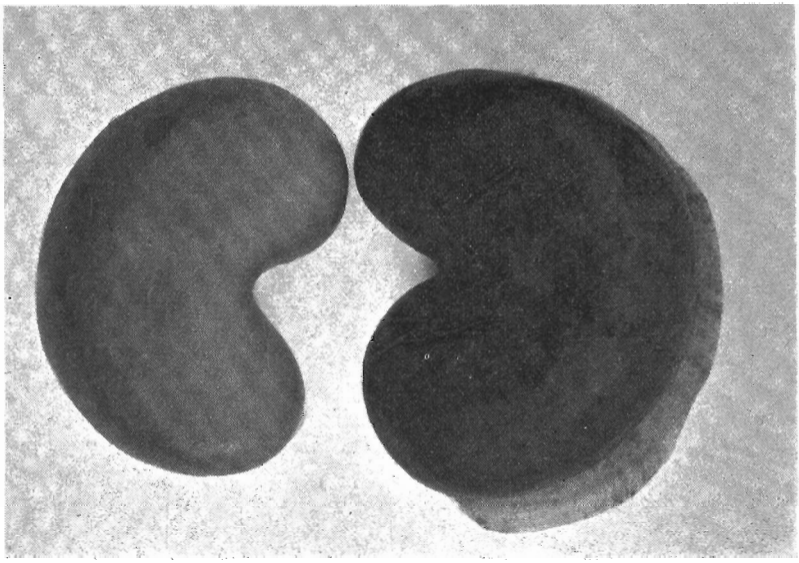

Figure 4. Normal and pulpy kidneys.

Immunization experiments. Rabbits were immunized with the supernatant fluid of formalized culture L3 and EW. After immunization, the rabbits were bled and the serum mixed with equal quantities of toxin. The amount of toxin was several times the lethal dose for a guinea pig. A moderate oedematous swelling appeared in the guinea pigs receiving the toxin-antiserum mixtures and animals recovered in a few days. The controls receiving 0.5 cc. of toxin died over night. Two sheep injected with toxin-antiserum mixtures developed oedematous swellings and recovered in several days. A control sheep receiving 5 cc. of toxin died in ten hours.

A preliminary experiment to determine the immunizing value of a single dose of formalized culture was attempted with lambs. Several

lambs were injected in the axillary region with 5 cc. of the formalized culture. Twenty-six days later two lambs were tested by intramuscular injections with 5 cc. of toxin. This amount of toxin was lethal to the control animals in 4 hours. Apparently a single dose of formalized culture is not sufficient to produce any appreciable immunity, for the injected lambs died in approximately the same interval of time as the controls. Further immunization experiments are being carried out to determine the dosage and efficacy of immunization with formalized cultures.

Reproduction of the disease per os. Bennetts reproduced the disease by drenching opium-treated lambs with culture. Oxer (3) failed to reproduce the disease by using Bennetts' method. Roberts (4) produced the disease by feeding cultures in conjunction with a large feed of "adjusted milk" to young lambs. At this Station several attempts to reproduce the disease per os with varying amounts of *Cl. perfringens* culture resulted in consistent failures. Five three-months-old lambs were drenched with 100 cc. of a 5-per-cent solution of sodium bicarbonate immediately followed by 100 cc. of a 48-hour culture, with negative results. A second experiment was carried out with month-old bottle-fed lambs. Two lambs were given 50 cc. of 5-per-cent solution of sodium bicarbonate immediately followed by 100 cc. culture in cow's milk. Another lamb was given 250 cc. of culture in 500 cc. of cow's milk. The lambs remained healthy.

To determine if *Cl. perfringens* Type D in an unaltered state proliferates and if toxin is absorbed in the small intestine of normal sheep, a laparotomy under local anesthesia was performed on a lamb and a yearling wether. An incision was made in the pyloric portion of the abomasum and a capillary pipette introduced through the pylorus into the small intestine. Thirty cubic centimeters and 160 cc. of highly toxic cultures, respectively, were injected into the small intestine. Results were negative. The results are in contradiction to the present view of proliferation and subsequent absorption of toxin in the alimentary tract.

Comparison of the effect of artificially produced toxin. In order to compare the effect of injection of intestinal filtrates and of artificially produced toxin with the naturally occurring disease, early in this work a quantity of desiccated toxin (*Cl. perfringens* Type D) was obtained from the Wellcome Physiological Research Laboratories. Two hundred mgs. were dissolved in 85-per-cent NaCl and injected into the jugular vein of a 30-pound lamb. Symptoms were as follows: labored breathing, weakness, convulsive movements, coma, and death in thirty minutes.

Upon necropsy, abnormal quantities of peritoneal and pericardial fluids were found. There was a marked congestion of the vessels of the small intestine and mesentery, with some evidence of hemorrhage in the mesenteric lymph nodes. The abomasum appeared congested. There were a few ecchymotic hemorrhages under the endocardium.

Cultural filtrates obtained from strains isolated from natural cases were highly toxic when injected intravenously. The time of death varied from one-half hour to three and one-half hours after injection, depending upon the size of animal and amount of toxin given. Lesions in such cases were not always so pronounced as in animals dead on pastures.

OTHER ETIOLOGICAL STUDIES

Comparison of the effect of the injection of histamine. During the study of this disease it was suggested that histamine produced in and absorbed from the digestive tract might be the lethal factor of the disease. A quantity of histamine hydrochloride was obtained and after being dissolved in 85-per-cent NaCl was injected into the jugular vein as follows:

Number	Sheep	Amount of histamine	Results
1	30-lb. lamb	180 mgs.	Dead in 11 minutes
2	25-lb. lamb	100 mgs.	Dead in 25 minutes
3	90-lb. wether	50 mgs.	Improved after 1 hour
4	150-lb. wether	500 mgs.	Dead in 17 minutes

The symptoms occurring in the sheep injected with histamine were similar to those observed in the sheep injected with toxin. There was labored breathing, convulsions, trembling, and coma.

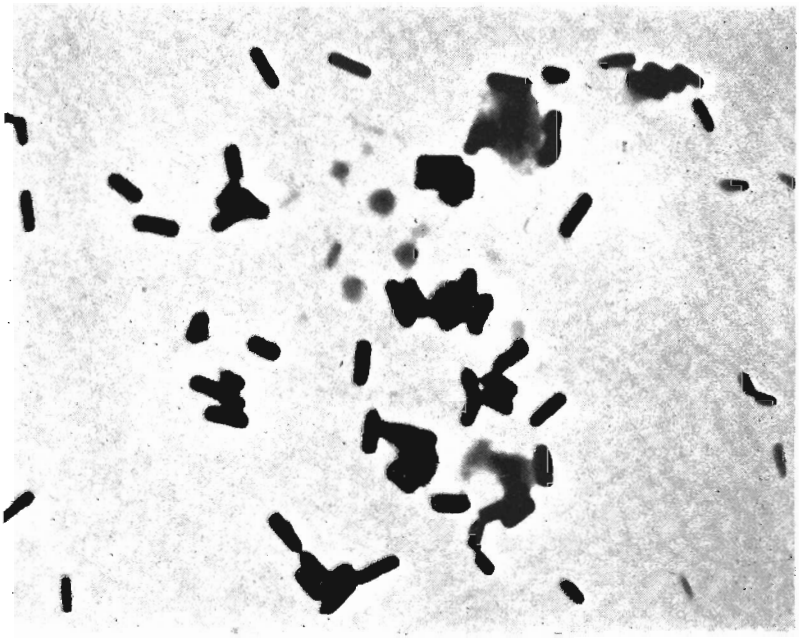

Figure 5. *Clostridium perfringens* Type D, cause of the disease.

Necropsy revealed severe subendo- and subepicardial hemorrhages, and also ecchymotic hemorrhages of the lungs, bronchial, urinary bladder, and lymph nodes. The kidneys appeared normal.

Experiments with tapeworms. In one area where this disease occurs every year, tapeworms (*Monesia expansia*) infest lambs in large numbers. In

spite of the fact that many of the fat lambs going to market are severely infested, it has been the opinion of some that these parasites were instrumental in the severe losses that occur in these lambs. Consequently, efforts were made to determine the toxic properties of these tapeworms.

Worms collected from an animal recently dead of the disease were washed, ground in a mortar, diluted with 85-per-cent NaCl and preserved with 0.5-per-cent chloroform over night. The following morning 10 cc. of the supernatant fluid was injected intravenously into a 40-pound lamb. There was slight increase in the respiratory rate for a short time, but no ill effects followed. No effort was made to determine if the injected lamb harbored tapeworms, but this lamb had been raised in a locality where *Monesia* are not common.

This experiment was repeated. A 50-pound lamb known to be infested with *Monesia* was injected intravenously with 22 cc. of similarly prepared extract. The results were similar to those of the first experiment.

Other attempts to reproduce the disease in sheep have consisted of the intravenous injection of various extracts prepared in the School of Pharmacy of Oregon State College. These extracts were prepared from the rumen content of naturally occurring cases, from rape, and from pasture grasses of the affected areas. These experiments have proved negative.

PREVENTION

Since sick lambs are seldom seen, preventive medicine becomes necessary. The first efforts to prevent losses were made with the use of hemorrhagic septicemia vaccine. Careful checks were not made with this product, but the sheep breeders reported that it did not stop losses.

Breeders formed the opinion that tapeworms were in some way responsible for losses and began treating the lambs with copper sulphate combined with nicotine sulphate. Many reported this treatment stopped losses at least for a time. They also reported that the treatment often resulted in death.

Workers in New Zealand reported that losses stopped following yarding for several hours. This keeping the lambs off feed perhaps had the same effect as starving lambs before dosing for tapeworms with the copper sulphate-nicotine sulphate mixture. Yarding was tried in Curry County, but did not seem to give results. Breeders reported that it also slowed up the lambs and therefore was objectionable.

FIELD TRIALS WITH ANTITOXIN

In the spring of 1938, field trials to determine the protection given by the injection of *Cl. perfringens* antitoxin were started. Two lots of antitoxin were used. One was obtained from Dr. I. B. Boughton, Sonora, Texas, the first worker to isolate *Cl. perfringens* Type D in the United States (5). The other was obtained from the Wellcome Physiological Research Laboratories, Beckenham, Kent, England, and is sold under the trade name of Wellcome Lamb Dysentery Antitoxin. This product is believed to contain antibodies capable of protecting against toxins of all types of *Cl. perfringens*.

These antitoxins were administered subcutaneously in the axilla in doses of 5 and 10 cc. In each instance an equal or greater number in each flock was left untreated as checks.

TREATMENT OF LAMBS WITH *Clostridium perfringens* ANTITOXIN, 1938

Ranch number	Treated with antitoxin									Untreated		
	Texas 5 cc.			Texas 10 cc.			Wellcome 5 cc.					
	Treated	Lived	Died	Treated	Lived	Died	Treated	Lived	Died	Number	Lived	Died
1	36	36	0	30	30	0	66	66	0	208	193	15
2	....	....	..	....	....	..	100	100	0	150	146	4
3	....	....	..	....	....	..	155	153	2	175	169	6
4	55	55	0	....	....	..	40	40	0	1,100	1,081	19
5	....	....	..	....	....	..	130	130	0	130	129	1
6	30	30	0	....	....	..	.....	.....	..	30	30	0
Total	121	121	0	30	30	0	491	489	2	1,793	1,748	45

Percentage treated lambs died 0.3i
Percentage untreated lambs died 2.41

TREATMENT OF LAMBS WITH *Clostridium perfringens* ANTITOXIN, 1939

Ranch number	Treated with 5 cc. antitoxin			Untreated		
	Treated	Lived	Died	Number	Lived	Died
1	50	50	0	53	52	1
2	60	60	0	62	62	0
3	120	120	0	120	117	3
4	157	157	0	108	103	5
5	200	200	0	207	184	23
6	150	150	0	136	128	8
Total	737	737	0	686	646	40

Percentage treated lambs died 0.00
Percentage untreated lambs died 5.83

These trials were continued in 1939, Wellcome Lamb Dysentery Antitoxin being the only product employed. It was administered in the same manner as in 1938 in 5 cc. doses.

DISCUSSION

Many phases of this disease problem are not satisfactorily understood. Ordinarily, the toxins produced by the organism causing botulism, and by certain staphylococci, are the only ones absorbed from the normal digestive tract, and just what conditions are present to cause the absorption of the *Cl. perfringens* toxin are not well understood. British workers were able to kill lambs by feeding them toxins only after slowing up the bowel with drugs. It is quite possible that the high protein-low fiber diet of the lambs on lush pastures produces a similar condition to that produced by the drugs used by the British workers. Doctor Robert Jay of the Bureau of Animal Industry has advanced the theory that absorption takes place because the lambs are in a state of sensitization to the grasses. This theory we have not been able to prove or disprove. Some authorities (6) have suggested that absorption of histamine might be responsible, and we have proved that the symptoms and lesions produced by the injection of histamine hydrochloride are somewhat similar

to those found in the lambs that die on pasture. Histamine was not recovered from the blood of one lamb killed with injected toxin.

Feed no doubt plays a very important part in this disease, as we know that the disease seldom occurs except on lush pasture, seems worse after a rain, and affects mostly the fattest wether lambs. The disease appeared this year on one farm where cultivated pastures were used for the first time. These pastures were produced for the purpose of providing more and better early feed for lambs. Yet with this practice losses rose to 12½ per cent. That all lambs lost died of entero-toxemia was not proved, but the owner was certain they all died of the same disease.

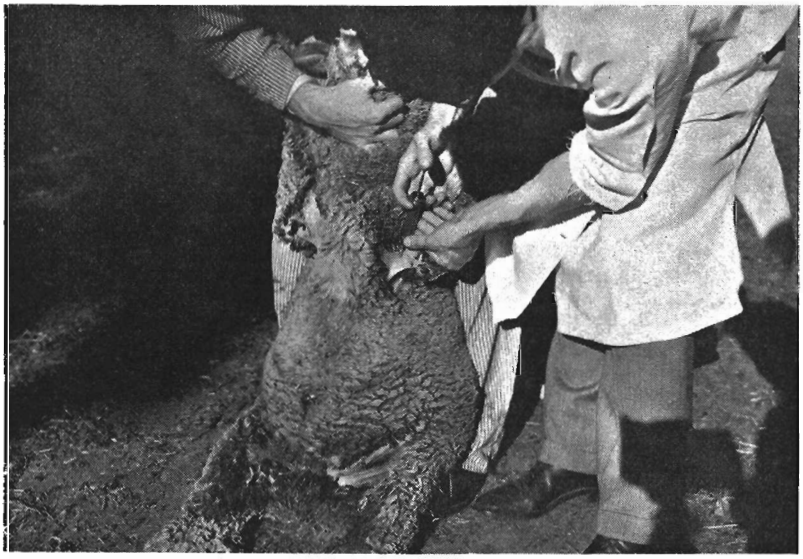

Figure 6. Method of administering antitoxin.

In spite of the beliefs of the sheep breeders, we are unable to see how the tapeworm can play a part in causing this disease. This year we saw many cases in which the parasite was not present. Now that Dr. Stunkard has worked out the life cycle of the tapeworm, however, we should be able to prove its importance to the lamb industry of Curry County. Mites of the same genus as those incriminated by Dr. Stunkard have been found on the pastures where the disease has existed year after year.

Considering the results obtained in two laparotomies, they indicate that when a quantity of culture and toxin is introduced directly into the small intestine no injury or absorption of toxin takes place. In order to bring about a pathological effect, the organism, toxin, or both, must in some way produce a localized injury for the portal of absorption of the toxin. The process may depend either on chemical or physical affinity for the tissue cells. While there is no question that a similar

condition can be reproduced by intramuscular or intravenous injections, it appears that only under certain conditions the gastro-intestinal tract becomes permeable to the toxin. As the problem now stands, one can not reach a decision as to how the toxin of *Cl. perfringens* Type D is absorbed from the small intestine.

From a comparative study of the relatively long incubation period of the cultures (60 hours) for maximum potency of the toxin, together with the morphological, cultural, and toxin-antitoxin reactions, the evidence obtained as to the cause of death of lambs dying from pulpy kidney disease in Oregon incriminates *Cl. perfringens* Type D.

A method of prevention, if practical, must be low in cost and easily applied. In some areas where losses have occurred year after year, sheep breeders will be willing to pay a high price for anything that will protect their lambs. This is not true where the disease does not appear regularly. Methods of producing a protective substance against *Cl. perfringens* organisms have not been too successful, and therefore the cost of producing a satisfactory protection against pulpy kidney disease is likely to remain high.

SUMMARY

1. The cause of sudden death losses in Oregon lambs has been found.

2. The cause of these losses is an anaerobic bacterium, *Cl. perfringens* Type D.

3. This organism is responsible for similar losses in Texas (1), England, Australia, and New Zealand.

4. Because of sudden death, treatment is not successful.

5. Methods of prevention by the use of antitoxins have been tried and proved successful.

LIST OF REFERENCES

1. Bennetts, H. W. *Infectious Entero-Toxaemia (the So-Called Braxy-Like Disease) of Sheep in Western Australia*, Coun. Sci. Ind. Res., Aust. Bul. 57, 1932.
2. Wilsdon, A. J. *Observations on the Classifications of Bacillus welchii*, Second Report of the Director of the Institute of Animal Pathology, Univ. Cambridge, 1931, pp. 53-85.
3. Oxer, D. T. *"Pulpy Kidney" or Acute Infectious Entero-Toxaemia of Suckling Lambs Due to B. ovitoxicus (Bennetts)*, Coun. Sci. Ind. Res., Pamphlet 35, 1932.
4. Roberts, R. S. *Braxy-Like Diseases of Sheep*, Vet. Rec., Vol. 50, No. 21, 1938, pp. 591-604.
5. Boughton, I. B., and Hardy, W. T. *Infectious Entero-Toxaemia of Young Lambs*, Forty-ninth Annual Report, Texas Agric. Exp. Station, 1936, pp. 278-279.
6. Duke, William W. *Allergy, Asthma, Hay Fever, Urticaria and Allied Manifestations of Reactions*, C. V. Mosby Co., St. Louis, Second Edition, 1926, p. 34.

Publications that have been consulted, but to which no specific reference has been made:

Bosworth, T. J., and Glover, R. E. *A Differential Character of Clostridium welchii Type D*, Proc. Roy. Soc. Med., Vol. 28, Part 2, 1935, pp. 1004-1006.

Boughton, I. B. *Immunization Against "Milk Colic"*, The National Wool Grower, Vol. 27, July, 1937, p. 36.

Dalling, T., and Ross, H. E. *Clostridium welchii: Notes on the Relationship Between the Types of Cultures and the Production of Toxin*, Jour. of Comp. Path. and Therap., Vol. 51, Part 4, 1938, pp. 235-248.

Eggerth, Arnold H., Littwin, Ralph J., and Deutsch, Joyce V. *The Determination of Histamine in Bacterial Cultures*, Journal of Bacteriology, Vol. 37, No. 2, Feb., 1939.

Gill, D. A. *Infectious Entero-toxemia and Cl. welchii Group with Special Reference to So-Called Pulpy Kidney in Lambs*, New Zealand Jour. Sci. and Tech., Vol. 18, No. 2, 1936, pp. 106-119.

Ibid. *"Pulpy Kidney" Disease of Lambs*, New Zealand Jour. Agric., Vol. 45, 1932, pp. 332-341.

Kendall, A. I., and Gebauer, E. *The Production of Histamine by Certain Strains of the Gas Bacillus*, Jour. Inf. Dis., Vol. 47, p. 261.

Montgomerie, R. F. *"Strike" of Sheep in North Wales*, Vet. Jour., Vol. 91, No. 8, 1935, pp. 350-358.

Ibid. *Some Diseases of Sheep*, Vet. Jour., Vol. 94, No. 4, 1938, pp. 165-171.

Montgomerie, R. F., and Rowlands, W. T. *Pulpy Kidney Disease of Young Lambs in North Wales*, Vet. Jour., Vol. 90, No. 10, 1934, pp. 399-403.

Newsom, I. E., and Thorp, Frank, Jr. *The Toxicity of Intestinal Filtrates from Lambs Dead of Overeating*, Jour. Amer. Vet. Med. Assoc., Vol. 93, N. S. 46, 1938, pp. 165-167.

Stewart, W. L., and Henderson, D. W. *Anaerobic Infections in Lambs*, Jour. of Comp. Path. and Therap., Vol. XLII, No. 1, March, 1929, p. 240.

Tunnicliff, E. A. *A Strain of Clostridium welchii Producing Fatal Dysentery in Lambs*, Jour. Infect. Dis., Vol. 52, May-June, 1933, pp. 407-412.

Watts, P. S. *Observations on the Bacterial Flora of the Intestine of Normal Sheep*, Vet. Journ., Vol. 94, No. 2, 1938, pp. 60-74, and Vol. 94, No. 3, 1938, pp. 112-127.

Wilsdon, A. J. *The Relationship of Bacillus Ovitoxicus (Bennetts) to the Cl. welchii Group*, Third Report of the Director of the Institute of Animal Pathology, Univ. Cambridge, 1932-33. pp. 46-51.

OREGON STATE BOARD OF HIGHER EDUCATION

Edward C. Pease ..... *The Dalles*
F. E. Callister ..... *Albany*
Beatrice Walton Sackett ..... *Marshfield*
C. A. Brand ..... *Roseburg*
E. C. Sammons ..... *Portland*
Robert W. Ruhl ..... *Medford*
Edgar William Smith ..... *Portland*
Willard L. Marks ..... *Albany*
Herman Oliver ..... *John Day*
Frederick M. Hunter, Ed. D., LL.D. ..... Chancellor of Higher Education

STAFF OF AGRICULTURAL EXPERIMENT STATION

*Staff members marked * are United States Department of Agriculture investigators stationed in Oregon*

Geo. W. Peavy, M.S.F., Sc.D., LL.D. ..... President of the State College
Wm. A. Schoenfeld, B.S.A., M.B.A. ..... Director
R. S. Besse, M.S. ..... Vice Director
Esther McKinney ..... Accountant
Margaret Hurst, B.S. ..... Secretary

Division of Agricultural Economics

E. L. Potter, M.S. ..... Agr'l. Economist; In Charge, Division of Agri. Economics

Agricultural Economics

W. H. Dreesen, Ph.D. ..... Agricultural Economics

Farm Management

D. C. Mumford, M.S. ..... Economist in Charge
G. W. Kuhlman, Ph.D. ..... Associate Economist
W. W. Gorton, M.S. ..... Research Assistant
H. L. Thomas, M.S. ..... Associate Agricultural Economist Soil Conservation*
J. C. Moore, M.S. ..... State Land Planning Specialist, Division of Land Utilization*
V. W. Baker, B.S. ..... Assistant Agricultural Economist, Division of Land Utilization*

Division of Animal Industries

P. M. Brandt, A.M. ..... Dairy Husbandman; In Charge, Division of Animal Industries

Animal Husbandry

R. G. Johnson, B.S. ..... Animal Husbandman
O. M. Nelson, B.S. ..... Animal Husbandman
A. W. Oliver, M.S. ..... Assistant Animal Husbandman
B. W. Rodenwold, M.S. ..... Assistant Animal Husbandman

Dairy Husbandry

G. H. Wilster, Ph.D. ..... Dairy Husbandman
I. R. Jones, Ph.D. ..... Associate Dairy Husbandman
H. P. Ewalt, B.S. ..... Assistant Dairy Husbandman
Arless Spielman, B. S. ..... Research Fellow (Dairy Husbandry)

Fish and Game Management

R. E. Dimick, M.S. ..... Wildlife Conservationist in Charge
F. P. Griffiths, Ph.D. ..... Assistant Conservationist*
A. S. Einarsen, B.S. ..... Associate Biologist, Bureau Biological Survey*
Frank Groves, B.S. ..... Research Assistant (Fish and Game Management)

Poultry Husbandry

H. E. Cosby ..... Poultry Husbandman in Charge
F. L. Knowlton, M.S. ..... Poultry Husbandman
W. T. Cooney, B.S. ..... Research Assistant (Poultry Husbandry)

Veterinary Medicine

I. N. Shaw, B.S., D.V.M. ..... Veterinarian in Charge
E. M. Dickinson, D.V.M., M.S. ..... Associate Veterinarian
O. H. Muth, M.S., D.V.M. ..... Associate Veterinarian
R. W. Dougherty, D.V.M. ..... Associate Veterinarian
A. S. Rosenwald, B.S., D.V.M. ..... Assistant Poultry Pathologist
O. L. Searcy, B.S. ..... Technician
Roland Scott, D.V.M. ..... Research Assistant (Veterinary Medicine)
C. R. Howarth, D.V.M. ..... Research Assistant (Veterinary Medicine)
Marion Robbins, B.S. ..... Technician in Poultry Pathology

Division of Plant Industries

G. R. Hyslop, B.S. ..... Agronomist; In Charge, Division of Plant Industries

Farm Crops

H. A. Schoth, M.S. ..... Agronomist: Division of Forage Crops and Diseases*
D. D. Hill, Ph.D. ..... Associate Agronomist
R. E. Fore, Ph.D. ..... Assistant Agronomist*
Elton Nelson, B.S. ..... Agent, Division of Fiber Plant Investigations*
Louisa A. Kanipe, B.S. ..... Junior Botanist, Division of Seed Investigations*
H. H. Rampton, M.S. ..... Assistant Agronomist: Division Forage Crops and Diseases*
L. E. Harris, M.S. ..... Assistant Agronomist
H. E. Finnell, M.S. ..... Assistant Agronomist
A. E. Gross, M.S. ..... Research Assistant (Farm Crops)

Food Industries

E. H. Wiegand, B.S.A. ..... Horticulturist in Charge
T. Onsdorff, M.S. ..... Assistant Horticulturist

STATION STAFF—*(Continued)*

Horticulture

W. S. Brown, M.S., D.Sc. — Horticulturist
H. Hartman, M.S. — Horticulturist (Pomology)
A. G. B. Bouquet, M.S. — Horticulturist (Vegetable Crops)
C. E. Schuster, M.S. — Horticulturist, Div. Fruit and Vegetable Crops and Diseases*
W. P. Duruz, Ph.D. — Horticulturist (Plant Propagation)
G. F. Waldo, M.S. — Ass't. Pomologist, Div. Fruit and Veg. Crops and Diseases*
E. Hansen, M.S. — Assistant Horticulturist (Pomology)

Soil Science

W. L. Powers, Ph.D. — Soil Scientist in Charge
C. V. Ruzek, M.S. — Soil Scientist (Fertility)
M. R. Lewis, C.E. — Irrigation and Drainage Engr., Bureau Agric. Engineering*
R. E. Stephenson, Ph.D. — Soil Scientist
E. F. Torgerson, B.S. — Associate Soil Scientist (Soil Survey)
James Clement Lewis, B.S. — Research Fellow in Soils

Agricultural Chemistry

J. S. Jones, M.S.A. — Chemist in Charge
R. H. Robinson, M.S. — Chemist (Insecticides and Fungicides)
J. R. Haag, Ph.D. — Chemist (Animal Nutrition)
D. E. Bullis, M.S. — Associate Chemist
M. B. Hatch, M.S. — Assistant Chemist
L. D. Wright, M.S. — Assistant Chemist

Agricultural Engineering

F. E. Price, B.S. — Agricultural Engineer in Charge
H. R. Sinnard, M.S. — Associate Agricultural Engineer (Farm Structures)
C. I. Branton, B.S. — Assistant Agricultural Engineer
W. M. Hurst, M.A. — Agricultural Engineer, Bureau Agricultural Engineering*

Bacteriology

G. V. Copson, M.S. — Bactertologist in Charge
J. E. Simmons, M.S. — Associate Bacteriologist
W. B. Bollen, Ph.D. — Associate Bacteriologist
C. P. Hegarty, Ph.D. — Research Assistant (Bacteriology)

Entomology

D. C. Mote, Ph.D. — Entomologist in Charge
J. C. Chamberlin, Ph.D. — Asso. Ento. (Div. Truck Crops and Garden Insects)*
A. E. Bonn, B.S. — Junior Entomologist (Div. of Truck Crops and Garden Insects)*
H. A. Scullen, Ph.D. — Associate Entomologist
B. G. Thompson, M.S. — Assistant Entomologist
S. C. Jones, M.S. — Assistant Entomologist
K. W. Gray, M.S. — Assistant Entomologist
W. D. Edwards, M.S. — Assistant Entomologist
H. E. Morrison, M.S. — Assistant Entomologist
Joe Schuh, M.S. — Research Assistant (Entomology)
G. R. Ferguson, B.S. — Research Assistant (Entomology)

Home Economics

Maud Wilson, A.M. — Home Economist

Plant Pathology

C. E. Owens, Ph.D. — Plant Pathologist in Charge
S. M. Zeller, Ph.D. — Plant Pathologist
F. P. McWhorter, Ph.D. — Plant Pathologist*
B. F. Dana, M.S. — Plant Pathologist (Div. Fruits and Veg. Crops and Diseases)*
F. D. Bailey, M.S. — Associate Plant Pathologist (Insecticide Control Division)*
P. W. Miller, Ph.D. — Assoc. Pathologist (Div. of Fruit and Veg. Crops and Dis.)*
G. R. Hoerner, M.S. — Agent (Division of Drug and Related Plants)*
R. F. Grah, B.S. — Agent (Division of Drug and Related Plants)*
R. Sprague, Ph.D. — Associate Pathologist (Div. of Cereal Crops and Diseases)*
John Milbrath, Ph.D. — Research Assistant (Plant Pathology)

Publications and News Service

C. D. Byrne, M.S. — Director of Information
E. T. Reed, B.S. — Editor of Publications
D. M. Goode, M.A. — Editor of Publications
J. C. Burtner, B.S. — Associate in News Service

Branch Stations

Leroy Childs, A.B. — Supt. Hood River Branch Experiment Station, Hood River
F. C. Reimer — Superintendent Southern Oregon Branch Experiment Station, Talent
D. E. Richards, B.S. — Supt. Eastern Oregon Livestock Br. Expt. Sta., Union
H. K. Dean, B.S. — Supt. Umatilla Br. Expt. Sta. (Div. W. Irrig. Agri.), Hermiston*
Obil Shattuck, M.S. — Superintendent Harney Branch Experiment Station, Burns
H. B. Howell, B.S. — Supt. John Jacob Astor Branch Experiment Station, Astoria
R. G. Johnson, B.S. — Acting Supt. Squaw Butte Regional Range Experiment Station
G. A. Mitchell, B.S. — Supt. Pendleton Branch Station (Dry Land Ag.), Pendleton*
G. G. Brown, A.B., B.S. — Horticulturist, Hood River Br. Expt. Sta., Hood River
Arch Work, B.S. — Supt. Medford Sta. (Asso. Irrig. Engr., Div. of Irrig.), Medford*
E. S. Degman, Ph.D. — Asso. Pomologist, Div. Fr. & Veg. Cr. & Dis., Medford*
Bruce Allyn, B.S. — Junior Irrigation Engineer (Division of Irrigation), Medford*
L. G. Gentner, M.S. — Associate Entomologist, So. Ore. Br. Expt. Station, Talent
J. F. Martin, M.S. — Junior Agronomist, (Div. Cereal Crops and Dis.), Pendleton*
M. M. Oveson, M.S. — Superintendent Sherman Branch Experiment Station, Moro*
R. W. Henderson, B.S. — Research Assistant, Sherman Branch Experiment Sta., Moro
R. E. Hutchinson, M.S. — Asst. to Supt. Harney Branch Experiment Station, Burns
J. R. Kienholz, Ph.D. — Jr. Pathologist, Div. Fr. & Veg. Cr. & Dis., Hood River*