

TABLES XIII AND XIV.

Strong Colonies,						Fair Colonies.						Larvae on Walls.						No Larvae on Walls.					
Sheep	Oes. Col.	H.C.	Tric.	Rhab.	Body Wght.	Sheep.	Oes. Col.	H.C.	Tric.	Rhab.	Body Wght.	Sheep.	Oes. Col.	H.C.	Tric.	Rhab.	Body Wght.	Sheep.	Oes. Col.	H.C.	Tric.	Rhab.	Body Wght.
D	0	50	40	10	64.8	J	2	80	8	10	65.3	A adult	0	50	30	20	90	—	—	—	—	—	—
I	0	50	30	20	62.13	K	0	50	30	20	53.3	B adult.	0	45	30	25	78	—	—	—	—	—	—
G	0	70	20	10	53.11	C	1	85	14	0	72.7	E	0	60	25	15	60	—	—	—	—	—	—
H	0	50	40	10	62.9	F	0	50	40	10	47.1	—	—	—	—	—	—	—	—	—	—	—	—
L	4	80	10	6	41.7	M	0	80	0	20	39.	—	—	—	—	—	—	—	—	—	—	—	—
N	0	55	25	20	48	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—

Average body weight, 9/6/24: Lambs 62.13; Adults 84.  
 Proportion of "positive" to "negative" infection 11.3.

Proportion of "positive" to "negative" infection 11.3.  
 Species pe.centage ..... Oes. Col 0.5 % H.C. 59 % Tric. 24 % Rhab. 16

Strong Colonies,						Fair Colonies.						Larvae on Walls.						No Larvae on Walls.					
Sheep	Oes. Col.	H.C.	Tric.	Rhab.	Body Wght.	Sheep.	Oes. Col.	H.C.	Tric.	Rhab.	Body Wght.	Sheep.	Oes. Col.	H.C.	Tric.	Rhab.	Body Wght.	Sheep	Oes. Col.	H.C.	Tric.	Rhab.	Body Wght.
N	10	70	20	0	49.8	M	12	70	18	0	44	B adult	8	50	25	17	81.2	K	—	—	—	—	59.8
D	7	70	20	3	68.8	—	—	—	—	—	—	E	5	60	20	15	54.2	F	—	—	—	—	51.12
H	0	85	15	0	67	—	—	—	—	—	—	L	0	45	30	25	45.12	I	—	—	—	—	67.2
—	—	—	—	—	—	—	—	—	—	—	—	G	0	50	30	20	58.0	A adult	—	—	—	—	91.10
—	—	—	—	—	—	—	—	—	—	—	—	C	9	70	21	0	76.8	J	—	—	—	—	59.8

Proportion of "positive" to "negative" infection 4.10  
 Average body weight, 19/9/24: Lambs 58.8.  
 Adults 86.6. Species Percentage Oes. Col. 4 % H.C. 81 % Tric. 15 % Rhab. Frac.

*Notes on Tables XIII and XIV.*

(1) For the incubation of the faeces, a fruit jar of about 1,000 c.c. capacity was used and faeces were placed about two inches deep at the bottom.

(2) For the purpose of record, the result of the cultural test was classified under the following headings:—

*Strong colony*: when the mature larvae migrating on to the walls had invaded about a quarter of the surface in between the faeces and the lid. This would mean a strong and probably fatal infection in the host.

*Fair colony*: when a thick patch of larvae about 1 to 2 inches in height and 1 to 2 inches broad was seen above the level of the faeces. This would mean an infection that would not produce clinical symptoms in the sheep.

*Larvae on walls*: when individual, scattered, rather rare larvae were seen on the walls of the jar. This would indicate the presence of a few worms in the host, but which could not be described as a verminosis.

*No larvae on walls*: when no larvae were present the sheep was considered clean.

For the purpose of record, the "strong colonies" and the "fair colonies" were classified as "positive infection" cases and the "larvae on walls" and "no larvae on walls" were called "negative infection." The proportion of "positive" to "negative" cases is quoted in the tables as well as the percentages of the various larvae present.

By comparing the two tables given above, it can be seen that three month's feeding of bone meal plus G.W.W.R. reduced the positive cases of infection (strong and fair colonies) from 11 to 4, and the negative cases (larvae on walls and no larvae on walls) increased correspondingly from 3 to 10, notwithstanding the intravening artificial infection with 12,000 larvae.

The average body weight of lambs, however, was reduced from 62.13 lb. to 58.8 lb. This could be explained by the fair infection of adult *Oesophagostomum columbianum*, but more so by the very severe nodular infection of the intestines, which at that time was in the acute caseous stage. This observation would tend to show that the lick does not affect the larvae, as these are still in the nodule-producing stage, but that it does have an effect on the adult worm. To give in full the systematic record of body weights throughout the whole experiment would occupy too much space, but it is of interest to select the weights recorded at different dates, and Table XV shows the comparisons desired.

TABLE XV.

PERIODICAL WEIGHING OF PREVENTIVE FEEDING BATCH. (See Tables XIII and XIV.)

Date.	Tobacco Leaf Lick.	Bone Meal Lick.	Bone Meal—Sodium Arsenite Lick.	Bone Meal + G. W. W. R.	Loogas Lick.	Chenopodium Lick.	Control Lot.
<i>7th May, 1924—</i>	Lb	Lb.	Lb.	Lb.	Lb.	Lb.	Lb
Two months before infection.....	58	48·12	51·5	62	49·2	47·9	51·12
<i>9th June, 1924—</i>							
One month before infection.....	57·3	49	48·8	62·13	46·5	46·9	50
<i>29th July, 1924—</i>							
About one month of infection: total, 1,200 larvae.....	57·4	42·14	47·1	63	42·12	45·9	4·51
<i>19th September, 1924—</i>							
Thirty-nine days after completion of infection.....	53	39·11	40·14	58·8	41·6	40·7	40·14

*Notes on Table No. XV.*

(1) It must be mentioned that at each weighing the body weight of the lambs in each batch was totalled up and divided not by the number of lambs that were present, but by the number of lambs that were utilised at the commencement, the reason being that the mortality of lambs was considered to be the factor that should affect the results above all others.

(2) Dates of weighing:—

*May 7th, 1924*, is the body weight of lambs when taken from the veld and put into the experiment. At that date the lambs showed generally from fair to light infection of Wire Worms, *Trichostrongylus* and Nodular worms, but their condition was not much affected.

*June 9th, 1924*: the lambs had then had access to the lick for one month with the exception of two out of the three bone meal batches. They showed a loss in weight evidently due to the natural worm infection.

*July 29th, 1924*: At this date each of the lambs had already received 12,000 mixed larvae. The body weight average shows that all lambs lost in weight with the exception of the bone meal and G.W.W.R. batch.

It is also shown that bone meal lick is an important factor for the growth of lambs, but the presence of a concurrent worm infection strongly handicaps the animals.

The cultural test of faeces carried out at the same date showed that the Nodular worms were not yet mature, but the Wire Worms and the *Trichostrongylus* were in full oviposition in all batches with the exception of bone meal + G.W.W.R., in which the larvae were rather scarce.

*September 19th, 1924*: It was then 39 days since the artificial infection was completed. The majority of lambs showed a severe verminosis at the cultural test. The number of severely infected against the lightly infected was as follows:—

Bone meal, G.W.W.R. ...	Strong infection	4 lambs ; light infection	8 lambs.
Tobacco leaf ... ..	" "	7 " "	5 "
Bone meal ... ..	" "	7 " "	5 "
Bone meal Sod. ars. ...	" "	9 " "	3 "
Chenopodium leaf ...	" "	11 " "	1 "
Loogas ... ..	" "	11 " "	0 "
Controls ... ..	" "	7 " "	5 "

The great majority of lambs showed *Oesophagostomum* infection with the exception of the tobacco leaf batch in which there was practically no infection of Nodular worm, although the infection of the other worms was severe.

One lamb in the Loogas batch died of verminosis.

The body weight readings of the 19th September showed that the artificial infection given to the experimental lambs was of a very severe nature, as the lambs lost from a maximal of about 11 lb. in Control and Bone meal Sodium arsenite batch to a minimum of 4½ lb. and 5 lb. in Bone meal plus G.W.W.R. and tobacco leaf respectively.

At the beginning of October, 1924, it was found that the infection with 15,000 larvae was too severe for a fair trial of the licks as the mortality of lambs would be too high. Accordingly, all sheep were

dosed for Wire Worm infection, which, according to the cultural test of faeces, seemed to be the most severe and dangerous, and the experiment was discontinued.

*Conclusions :*

(1) The artificial infection of 15,000 mixed larvae proved to be too severe for the purpose of the experiment.

(2) None of the licks used showed to be completely effective against *Haemonchus contortus*, *Oesophagostomum columbianum*, *Trichostrongylus* or *Strongyloides papillosus*, either at the larval or at the adult stage.

(3) Lambs in all batches showed stunted growth and the bone meal plus G.W.W.R. batch showed the best average body weight.

(4) From the general experience gained during the running of the experiment, it was concluded that the Bone meal plus G.W.W.R. lick was the most effective and had a specific action on the Wire Worm. Tobacco lick came second and was more effective against the Nodular worms, although it seemed to be but slightly effective against the Wire Worms.

(5) At post mortem examination of the sheep that died after the conclusion of the experiment it was found that the nodular infection of the intestine was exceptionally severe. It was thought at the time that this finding confirmed the statements made in the "General considerations on the longevity of larvae of *Oesophagostomum columbianum*," viz., that sheep and lambs infected artificially and kept under stable conditions were more susceptible to the nodular worm than those running on the veld and infected under natural conditions, and it was decided that in further observations, experimental sheep should not be secluded in a stable for any lengthy periods.

VI<sub>G</sub>.—*Second Experiment.*

From the experience gained in the first serial experiment, a second one was planned in December, 1925, with lambs averaging six months of age.

It must be borne in mind that the monthly dosing with the G.W.W.R. is the treatment proved to be the most efficacious against *Haemonchus contortus* and is recommended by the Laboratory as a monthly dose to sheep grazing on infected farms.

The object of the second experiment was to compare the effect of the G.W.W.R. administered in a small dose every day in the form of a lick plus bone meal, against the monthly dose of G.W.W.R. on sheep plus a daily lick of bone meal. Additional to the two mentioned licks, a new test was given to the tobacco lick and a control lot was also included.

*Technique of the Experiment.*—Ten lambs about six months of age and two adult sheep were put in each lot, and a flock of 56 head was combined which was sent to graze during the day and was kraaled during the night in four separate camps where the lick was available in troughs. The grazing camp was known to be infected with the usual round worms of sheep, and in addition, an artificial infection was also given.

Accustoming the sheep to the respective licks was started on the 12.12.25 and by the 22.12.25 practically every lamb was licking

freely. Sixteen hundred mixed larvae were given through the mouth to each animal at the rate of 200 larvae every 48 hours during the period from the 22.12.25 to the 8.1.26.

On the 8.1.26 it was noted that several lambs were infected with *Moniezia* and a specific chemotherapeutic treatment was attempted in order to prevent a state of infection extending beyond the scope of the experiment.

On the 9.2.26 a second infection was started lasting until the 1.3.26, when each lamb received about 200 larvae every second day, or a total of 2,000 larvae per head.

The individual cultural test of the faeces was carried out every month and the body weight of each sheep was also recorded monthly.

The tables showing the monthly results with regard to the strength of the colony of larvae for each sheep, and the percentage of each species of larvae, is filed in the records of the experiment. As it would be impossible to reproduce 40 tables in this report, it is thought that the following résumé will convey some idea of the state of the infection of the sheep during the experiment.

(1) *Oesophagostomum columbianum*.—Bearing in mind the few larvae detected by the cultural test, it has to be concluded that the infection with this worm was never severe. With the exception of three positive cases in January, 1927, the tobacco lot showed practically no infection during the year. The Bone meal, G.W.W.R. showed the highest number of infected sheep in June, 1926, when three sheep out of nine showed a small percentage of larvae of *Oesophagostomum columbianum*. The bone meal plus monthly dose of G.W.W.R. showed the highest number of infected cases, with six positive cases in May and nine positive cases in June, 1926, and January, 1927.

The number of adult *Oesophagostomum columbianum* found at post mortem examination was higher than would be expected when judged by the cultural test, suggesting that the licks inhibited oviposition.

(2) *Haemonchus contortus* was present throughout the experiment in the positive cases of tobacco, bone meal, G.W.W.R. and controls and practically absent during all the regular monthly dosing of bone meal plus G.W.W.R. The proportion, however, of *Haemonchus contortus* in the three infected lots showed some interesting variations. The percentage of larvae reached its maximum in the months of January, February, March and April with an average of 40-50 per cent. A gradual decrease in the percentage was noticed from May to June, July, August and September, when the percentage was about 10. There was a slight increase in October, and a decided increase in November. In December a percentage of 40 was noticed again in the three batches. The increase in the percentage in October and December seems to be due to the natural infection due to larvae picked up by the sheep on the veld; the temperature and rainfall were favourable for the migration of larvae that were stored in the depth of the soil from the previous summer.

(3) *Trichostrongylus* was present throughout the experiment, and in large numbers in the positive cases of all batches.

(4) *Strongyloides papillosus* was present throughout the experiment in fairly large numbers in all batches.

(5) The month of May was apparently the most severe on all lots, reduction in weights being noticed all round, and with the exception of the batch receiving the monthly dose of G.W.W.R., each lot lost one lamb due to verminosis.

As the second infection of 2,000 mixed larvae was completed in March, the worms grew to maturity during a comparatively cold season (end of summer and beginning of winter) and required a period longer than for the first infection which took place in the middle of summer; furthermore, the lambs were older than at the time of the first infection, and the second artificial infection did not show up clinically until May. It must also be noted that since the beginning of the experiment, the lambs grazed on a comparatively small area of veld and infected the pasture heavily by passing out faeces containing worm eggs. The lambs were consequently victims of a severe natural infection during March, April and May.

(6) The writer feels justified in dividing the curves under discussion into two phases. The first phase starts in January and ends in May, with the effect of the first artificial infection plus the natural infection from the grazing camp. This phase is considered as the most typical for the object of the experiment as it shows the result of the two contrasting factors: the worm infection on one side and the effect of the preventive licks on the other, during the critical season for sheep infected with worms, namely the summer. The results of the body weight reading offered some interesting conclusions which are given under paragraph "3" of Conclusions.

(7) The second phase of the curve extending from June to October gives a graphic picture of the preventive lick experiment during the winter period, when several new factors came into action. The dry cold condition of the winter restricted the natural infection of the lambs that would otherwise have resulted from the reservoir of larvae in the pasture, it decreased the vitality and the number of the surviving worms in the lambs and hence enabled the lambs to pick up condition and weight. For instance, the initial ten lambs in each lot showed during the winter an increase of 11.8 lb. for the bone meal plus monthly G.W.W.R., of 17.8 lb. for the bone meal mixed with G.W.W.R., of 13.9 lb. for the tobacco lots and 4.11 lb. for the controls.

It may be added that the reason why the lambs in the bone meal plus monthly G.W.W.R. lot showed a lower increase in body weight than those in the other two licks could apparently be found in the result of the monthly cultural tests, where it is shown that this lot had the highest number of sheep infected with *Oesophagostomum columbianum* with six positive cases in May and nine positive cases in June, 1926. As a matter of fact, it seems that the anthelmintic principles intimately associated with the bone meal plus G.W.W.R. (Arsenic and Copper) and with the tobacco leaf (Nicotine) are carried in a sufficient amount to the large intestine where they act against the *Oesophagostomum columbianum*, whereas in the case of the monthly G.W.W.R. the dose is apparently absorbed before a sufficient quantity reaches the large intestine.

(8) The continuation of the body weight curves during November and December could be considered as an appendix, or as the beginning of a new yearly curve, as the new summer conditions with high temperature and rain exposed the lambs to a new natural infection with a drop in body weight in consequence.

(5) Taking into consideration the mixed infection of the four mentioned species, the total number of positive cases varied from 30 to 50 per cent. during the entire course of the experiment.

The monthly record of the body weight is given in Chart No. 5.

*Notes on Chart No. 8.*

(1) During January, the best result is shown by the bone meal G.W.W.R. lick where there is an increase in the average body weight of 6 lbs. The tobacco lick batch showed an increase of 3.6 lbs. in body weight, although one lamb out of ten was lost as a result of acute wire worm infection. In the batch allowed access to pure bone meal lick, the loss averaged 1 lb. per lamb, a result that was anticipated seeing that previous experiments showed that bone meal has no anthelmintic value and only acts as phosphorus supplier and stimulant of digestion. In this batch favourable results can only be expected after administration of the monthly dose of G.W.W.R.

The control lot which was left exposed to the artificial infection without any lick lost 1.11 lb. per head and two deaths occurred due to acute wire worm infection. It may be added here that the monthly cultural test that was carried out in between January and February after the dosing of the third lot showed that all the lambs belonging to bone meal plus G.W.W.R., tobacco, and control were infected with wire worms, whereas the bone meal plus monthly dose of G.W.W.R. did not show wire worm larvae.

(2) During the month of February, the bone meal plus G.W.W.R. and the tobacco lick batches did not show the same increase in body weight as in the previous month. This is probably due to the fact that the surviving worms were now adult and caused more damage to the host than when in the larval stage.

Sheep dosed with bone meal plus monthly G.W.W.R. dose showed an average increase of 5.7 lbs. against the loss of 1 lb. in the previous month, thus showing the beneficial effect of the G.W.W.R.

One lamb died in the control lot.

(3) It must be mentioned here that under farming conditions lambs are subjected to new infections of larvae with each rainfall, and it was consequently decided to subject the four lots of lambs to a new infection of 2,000 mixed larvae.

It can be seen that at the end of March the bone meal plus G.W.W.R. had lost 1 oz. per individual and that the tobacco lot lost 1.6 lb., whilst two lambs died as a result of verminosis.

The control lot lost two other lambs bringing down the average body weight by another 6.4 lbs.

The batch in the monthly dose of G.W.W.R. was the only one that improved in body weight, showing an increase of 12 ozs. per lamb.

(4) During April a considerable increase in the body weight was noticed in the lot under the monthly G.W.W.R. dose, a slight increase was shown in the lot allowed access to bone meal plus G.W.W.R., whereas both the tobacco and the control lots lost in body weight; in addition to which two control lambs died from verminosis.



*Conclusions:—*

(1) The results obtained in January tend to show that lambs heavily infected with mixed verminosis may lose considerably in body weight during the month elapsing in between two consecutive dosings with G.W.W.R.

(2) The results obtained at the end of March show the importance of periodical monthly anthelmintic treatment in order to enable the sheep to acquire more resistance against the new infections that take place with each rainfall.

(3) The average body weight obtained in May expresses the result of licks on various infected sheep during a period corresponding to the summer months. The bone meal plus monthly G.W.W.R. dose gave the best results as no lambs were lost and there was an increase of 7.3 lbs. in the average body weight. Bone meal mixed with G.W.W.R. gave the next best results with an increase of 5.4 lbs. in body weight and the loss of one lamb. The tobacco lot came third with the loss of 1.6 lb. per lamb in body weight, and the death of four lambs. The control lot lost 15.8 lbs. from the initial average body weight, and 8 out of 10 lambs died. The result of the control lot shows the extreme severity of the worm infection with which the lambs in this experiment had to contend, but such severe conditions are very rarely noted by farmers of South Africa in their flocks.

(4) The G.W.W.R. embedded in bone meal and the tobacco leaf lick seemed more effective against the *Oesophagostomum columbianum* than G.W.W.R. administered monthly.

(5) Bone meal plus the monthly dose of G.W.W.R. kept the lambs clean of wire worms but did not give protection against nodular worms, *Trichostrongylus*, *Strongyloides papillosus* and *Moniezia* infection. A severe infection of wire worms produced a decrease in body weight during the time that elapsed between two consecutive monthly dosings, but no mortality was observed.

(6) Bone meal mixed with G.W.W.R. and the tobacco lick did not appreciably prevent the infection of wire worms, *Trichostrongylus*, *Strongyloides papillosus* and *Moniezia* infection, but it seemed that the nodular worm infection was not too severe.

(7) The lot of lambs fed with bone meal and dosed monthly with G.W.W.R. gained 22.1 lbs. body weight per head, the lot allowed to bone meal mixed with G.W.W.R. gained 22.10 lbs. body weight per head, the lot allowed to tobacco lick gained 11.9 lbs. per head, and the control lot lost 10.13 lbs. per head.

(8) As a provisional deduction from the experiment under discussion, it would appear safe to recommend for practical stock raising in farms infected with Strongylidae, the monthly dosing with the G.W.W.R. and a daily lick. Until more evidence is obtained, such lick should have as a principal ingredient the tobacco leaf as, by administering the G.W.W.R. lick, too great an accumulation of arsenic may result.

#### VII.—*Third Experiment.*

From the experience gained in the two previous experiments, a new experiment was started on the 15th November, 1926, with four lots of lambs.

The first lot received 2 parts bone meal, 2 parts crushed dry tobacco leaf, 1 part kitchen salt (Sodium chloride) and the proportion according to weight was 234 parts bone meal, 52 parts crushed dry tobacco leaf and 100 parts kitchen salt. The mixture was slightly moistened when placed in the troughs.

The second lot received bone meal daily and in addition they were dosed once a month with G.W.W.R.

The third lot received the same lick as the first lot (bone meal, tobacco leaf, salt) and were dosed once a month with G.W.W.R.

The fourth lot (controls) received a bone meal lick daily.

The lambs received 1,000 mixed larvae in about 20 days and were run on the same camp used for Experiment No. 2.

The objects of the experiment were:—

(1) To compare the anthelmintic efficiency of tobacco leaf lick *versus* monthly dosing with G.W.W.R. on worm infected sheep allowed daily access to bone meal lick.

(2) To ascertain the effect of a combination of these two measures on sheep infected with wire worm and nodular worm.

The experiment is still in progress and from the data so far on record, it seems that the artificial infection plus the natural infection proved to be too severe, especially as clean lambs which were allowed to run in the same camp died of verminosis without having been artificially infected.

The experimental evidence gained from the three experiments referred to admits of the following provisional conclusions:—

(1) Preventive licks containing one of the following ingredients, tobacco leaf, G.W.W.R., Sodium arsenite, Chenopodium plant, Loogas, used according to the technique described, do not completely prevent or eradicate the infection of *Haemonchus contortus*, *Trichostrongylus*, *Oesophagostomum columbianum* and *Strongyloides papillosus*.

(2) The regular monthly administration of G.W.W.R. keeps sheep clean from *Haemonchus contortus*, but does not act as a preventive, and is the best measure against wire worms that this Division could recommend in the light of our present knowledge about anthelmintics.

(3) The administration of tobacco leaf or G.W.W.R. mixed with bone meal in the form of a lick is an important measure for reducing the infection in sheep and keeping up the body weight.

(4) The best method of protecting sheep against verminosis seems to be the monthly dose of G.W.W.R. and allowing them access to a lick composed of bone meal, tobacco leaf and salt.