

## ARSENICAL COMPOUNDS IN THE TREATMENT OF BLACKHEAD IN TURKEYS.

By ERNEST EDWARD TYZZER, M.D.

(From the Department of Comparative Pathology of Harvard Medical School, Boston.)

(Received for publication, January 20, 1923.)

In a previous investigation<sup>1</sup> an attempt was made to prevent blackhead infection in turkeys by the addition of an amebicide, *Chaparro amargosa*, to their daily ration. While this procedure resulted in an almost complete disappearance of *Entamoeba gallinarum*, a non-pathogenic ameba commonly found in the ceca of the turkey, it failed to prevent blackhead infection.

A study of the blackhead parasite, at this time, furnished morphological evidence indicating that it is a flagellate rather than a true ameba.<sup>2,3</sup>

The discovery that blackhead may be produced experimentally in young turkeys by feeding them large numbers of the ripe eggs of *Heterakis papillosa*, the common intestinal worm of poultry,<sup>4</sup> and also the demonstration that a fatal form of the disease may be produced in turkeys by the inoculation of active blackhead lesions,<sup>5</sup> have furnished reliable experimental methods of producing the infection, which may be utilized for therapeutic tests. In the course of successive seasons various drugs generally accepted as specifics in the treatment of protozoan diseases in man have been tested.

Tartar emetic, of recognized value in leishmaniasis, a flagellate infection, was injected intravenously at intervals, in toxic doses, into a large turkey inoculated with blackhead, but it failed to prevent the development of the disease or its fatal termination.

Large doses of quinine hydrochloride injected either into the veins or deep in the breast muscle also failed to check the course of the inoculated disease.

Emetine hydrochloride given subcutaneously in large doses at 24 and 48 hour intervals failed to prevent the development of the inoculated disease, but of two treated turkeys one eventually recovered.<sup>6</sup> No special significance was attached to this result, for one recovery had already been noted in a series of untreated cases of the inoculated disease.

---

<sup>1</sup> Tyzzer, E. E., *J. Med. Research*, 1919-20, xli, 211.

<sup>2</sup> Tyzzer, E. E., *J. Med. Research*, 1919, xl, 1.

<sup>3</sup> Tyzzer, E. E., *J. Parasitol.*, 1920, vi, 124.

<sup>4</sup> Graybill, H. W., and Smith, T., *J. Exp. Med.*, 1920, xxxi, 647.

<sup>5</sup> Tyzzer, E. E., and Fabyan, M., *J. Infect. Dis.*, 1920, xxvii, 207.

<sup>6</sup> Tyzzer, E. E., Fabyan, M., and Foot, N. C., *J. Infect. Dis.*, 1921, xxix, 268.

Subsequently Wegeforth and Wegeforth<sup>7</sup> reported success in both the prevention and the cure of natural blackhead in turkeys by the addition of ipecac to the daily ration. On account of the widespread interest in this report and since it is possible to be misled with respect to both the incidence of the disease and the outcome of natural blackhead in a flock, it seemed important to subject this drug to experimental test. We found that ipecac could be administered to young turkeys in amounts considerably larger than those used by the above investigators without any untoward effect.<sup>8</sup> A group of twenty-three turkeys was furnished with infectious material in the form of hen yard dirt mixed with dry mash. Eleven of these received ipecac daily, while the others served as controls. Ipecac in the large doses employed failed wholly to prevent blackhead, but appeared to have some effect, in that the onset of the disease was delayed in the treated birds. None of the eleven turkeys to which the ipecac was administered escaped infection, and the only one of these to recover, failed to develop normally. From these results, it did not appear that ipecac should be accepted as a specific for blackhead.

Precipitated sulfur added in large amounts to the food seemed also to retard slightly the onset of blackhead.<sup>8</sup> As sulfur is relatively inert and does not prevent the development of either intestinal worms or the protozoon of blackhead, it is probable that any benefit obtained from either ipecac or sulfur is due to its evacuant action.

#### *Spontaneous Blackhead.*

During the present season (1922) fifteen broods totalling 154 turkeys were hatched between May 6 and August 26. All were hatched in an incubator, from which they were transferred to a brooder, and about a week later were placed out of doors in cages each furnished with a warm hover. The broods of approximate ages were subsequently united into three flocks, each of which was provided with a yard and shelter.

The first case of blackhead appeared on June 8, and new cases continued to develop with considerable frequency up to the end of October, but only three cases occurred in the 2 following months. Since these turkeys were being raised primarily for market purposes, the continued appearance of new cases of infection together with the high mortality was not promising. The disease was so generally distributed throughout the various broods that to move them to a

<sup>7</sup> Wegeforth, H. M., and Wegeforth, P., *J. Pharmacol. and Exp. Therap.*, 1921, xvii, 249.

<sup>8</sup> Tyzzer, E. E., and Fabyan, M., *J. Exp. Med.*, 1922, xxxv, 791.

new range, while it might lower the incidence of the disease, would presumably contaminate more ground. Most of the following observations were made in the course of attempts to cure turkeys naturally infected with blackhead. This preliminary work served to indicate the drugs which were most effective, and towards the end of the season an experiment was carried out for the purpose of putting the more promising ones to a rigid experimental test in the treatment of inoculated blackhead.

Throughout the summer any turkey showing symptoms of the disease was placed in a special well grassed yard, the "hospital," in order to avoid its being abused and possibly killed by the healthy turkeys. Spontaneous recovery occurred early in the season in one case diagnosed as blackhead, but subsequently through June and July each case terminated fatally. Late in the season a 7 pound male bird, after showing mild symptoms for several days, also recovered without treatment. There were therefore only two recoveries in sixteen untreated cases of the disease, a mortality of 87.5 per cent.

*Neoarsphenamine Treatment.*—Beginning early in August neoarsphenamine (sodium 3,3'-diamino-4,4'-dihydroxy-arseno-benzene-*N*-methylene sulfinate) was injected into each case as soon as possible after the appearance of characteristic symptoms. Both American and French products were employed. The powder was dissolved in a small amount of sterile distilled water and the concentrated solution injected at once into the wing vein. It was evident that this solution caused severe pain when introduced into fixed tissues. Injection into the breast muscle produced extensive local edema which evidently hastened the death of young birds. Large doses were at first employed, in some instances as much as ten times the maximum human dose in proportion to the body weight being used. The immediate mortality was such that the dosage was lowered and from three to five times the maximum human dose, *i.e.* from 0.04 to 0.06 gm. per kilo, was given. While healthy turkeys are not seriously affected by these doses, sick birds show a pronounced prostration which appears in from 10 to 20 minutes and may last 6 hours or longer.

The young turkey is evidently much more susceptible than the rat to neoarsphenamine, for Federal Government regulations<sup>9</sup> require

<sup>9</sup> Roth, G. B., *Pub. Health Rep., U. S. P. H.*, 1921, xxxvi, 2523.

that white rats weighing from 100 to 150 gm. shall survive 7 days after the intravenous injection of 0.2 gm. per kilo, or from three to five times the doses here employed in the treatment of spontaneous blackhead. The smaller turkeys often succumb within 24 hours, but nearly all those surviving the treatment show a pronounced improvement on the following day. In infected turkeys the toxic reaction of neoarsphenamine is almost immediate, and the delayed effects noted in rats<sup>10</sup> have not been observed. As in rats, there is a pronounced tendency to hemorrhage following injection, so that unless pressure is maintained for a considerable time over the punctured wing vein, large hematomas result. One turkey acted as though blind following the injection, but after a few days showed improvement in vision. On further observation the difficulty appeared to be one of accommodation rather than actual blindness, in that distant objects appeared near. Many months after recovery from the blackhead infection this turkey still showed slightly defective accommodation and usually pecked twice in order to strike a particle of food.

There are two features which occur quite constantly in spontaneous blackhead that of themselves militate against recovery. First, the infected turkeys do not rest normally by squatting on the ground, but keep constantly on their feet, either moving listlessly about or standing hunched up for long periods usually with head under wing. Secondly, there is loss of appetite, which, with the diarrhea usually present, results in weakness and a progressive loss of weight.

As the toxic effects of the neoarsphenamine disappear, the treated turkeys show at first a pronounced thirst, drinking large amounts of water, and then an improvement in appetite. They subsequently rest for long periods, squatting normally on the ground. Such improvement in function can scarcely fail to have a favorable influence on the course of the disease. The treated turkeys soon become more brisk in their movements, and it may be difficult after a few days to distinguish them from the healthy birds. The subsequent course of the disease varies in different cases; some show steady improvement and are fully recovered within a week, others are alternately active and depressed, either recovering after a prolonged convalescence or

<sup>10</sup> Roth, G. B., *Pub. Health Rep., U. S. P. H.*, 1921, xxxvi, 1990.

eventually succumbing to the disease. The most favorable response to treatment was obtained in the early stages of infection, and recovery was rarely obtained in advanced cases. One turkey (No. 15) had a relapse with characteristic symptoms following a period of marked improvement. The results of the injection of nearsphenamine in cases of blackhead are furnished in Table I. Although the reduction of the mortality to 41 per cent was encouraging, the high cost of this drug, its instability, the difficulty of obtaining ampules in amounts suitable for given occasions, and its high toxicity in doses sufficiently large to influence the disease constitute serious obstacles to its employment.

*Arsenious Acid Treatment.*—It was thought possible that small amounts of arsenious acid added to the daily ration might serve to prevent infection. On September 7, liquor acidi arsenosi, an acidulated 1 per cent aqueous solution of arsenic ( $\text{As}_2\text{O}_3$ ), was added to the daily ration of sour milk in an amount to furnish 1 cc. to 20 kilos of body weight of turkeys. Thus in proportion to the body weight about one-tenth of the maximum sublethal dose for a rabbit was given each day. On September 11, the above dose was doubled, on September 12, it was tripled, and again increased on September 16, when about one-half the sublethal rabbit dose was given. On account of the death at this time of a large turkey without any lesions of blackhead, no arsenic was given on or after September 17. For the 8 days preceding the administration of arsenic there had been on the average approximately one new case of blackhead a day. This rate of incidence was somewhat lower for a time after the treatment was established but showed a pronounced increase following the increase in dosage of arsenic. It was obvious that the administration of this drug failed to prevent blackhead, and it is even possible that the larger doses finally given were increasing the incidence of infection. It is of interest to note that one turkey weighing nearly 3 kilos evidently succumbed to arsenical poisoning. The symptoms were almost identical with those of blackhead infection; *i.e.*, weakness, loss of appetite, and chrome-yellow droppings. There was, however, no marked loss of weight. On postmortem examination the liver and spleen were engorged and of a dark purplish color. There were no blackhead lesions either in the ceca or in the liver.

TABLE I.

Group.	Turkey No.	Date of onset of disease.	Symptoms.	Injections.	Result.	Remarks.
		1922				
	1	June 8	Typical.		June 13. D.	Typical blackhead lesions.
	2	" 15	Mild.		Recovered.	Recovery from a mild case of blackhead.
	3	" 17	Typical.		June 25. D.	Typical blackhead lesions.
	4	" 26	"		" 28. "	"
	5	July 6	"		July 10. "	"
	6	" 10	"		" 21. "	"
	7	" 12	"		" 24. "	"
	8	" 14	"		" 21. "	"
	9	" 20	Severe.		" 20. "	Cecal blackhead; no liver lesions.
	10	" 25	Typical.		" 25. K.	Blackhead. Recovery precluded by the severity of the infection.
	11	Aug. 1	"		Aug. 8. D.	Typical blackhead lesions.
	12	" 2	"		" 8. "	"

1  
Untreated.

1 Untreated.	31	Sept. 7	Mild.		Recovered.	A spontaneous recovery from probable blackhead.
	33	" 13	Typical.		Sept. 16. D.	Typical blackhead lesions.
	36	" 18	"		" 23.	"
	63	Oct. 26	"		Oct. 28. K.	Blackhead. Recovery unlikely with such extensive liver involvement.
2 Treated with neo-arsphenamine.	13	Aug. 7	Typical.	Aug. 9, 0.10 gm. in vein. " 12. 0.10 " " "	Recovered.	First injection followed by impairment of vision which gradually improved but remained defective.
	14	" 10	"	" 10. 0.10 " " " " 12. 0.15 " " "	"	Normal 14 days after onset.
	15	" 10	"	" 10. 0.05 " " " " 12. 0.05 " " " " 22. 0.09 " " "	"	Marked improvement after second injection followed by relapse on Aug. 21. Apparent recovery followed the third injection. On Sept. 10, again very ill; symptoms atypical; photophobia; apparently in pain. Recovery without further treatment.
	16	" 10	"	" 10. 0.08 " " " " 12. 0.010 " " "	"	Normal 14 days after onset.

TABLE I—Continued.

Group.	Turkey No.	Date of onset of disease.	Symptoms.	Injections.	Result.	Remarks.
		1922				
	17	Aug. 10	Typical.	Aug. 10. 0.07 gm. in vein and breast.	Aug. 11. D.	Dose of neoarsphenamine undoubtedly lethal. Hemorrhage into peritoneum and caeca. Typical blackhead lesions.
	18	" 11	"	" 12. 0.05 " " "	" 13. "	Lethal dose of neoarsphenamine. Early blackhead lesions.
	19	" 11	"	" 12. 0.05 " " "	" 12. "	Lethal dose of neoarsphenamine. Early blackhead lesions.
	20	" 11	Severe.	" 12. 0.05 " " " " 22. 0.05 " " " " 25. 0.05 " " "	Recovered.	Slow recovery.
	21	" 14	Typical.	" 18. 0.10 " " " " 22. 0.15 " " "	"	Improvement gradual.
	22	" 15	Severe.	" 18. 0.05 " " "	Aug. 19. D.	Typical blackhead lesions.
	23	" 23	Typical.	" 25. 0.075 " " " " 30. 0.06 " " "	" 31. "	" " "
	24	" 30	"	" 30. 0.06 " " " Sept. 2. 0.075 " " " " 6. 0.06 " " "	Recovered.	Rapid improvement after last injection.

2  
Treated with neoarsphenamine.



25	Aug. 30	Typical.	Aug. 30. 0.02 gm. in vein. Sept. 2. 0.025 " " " " 6. 0.03 " " "	Sept. 7. D.	Typical blackhead lesions.
26	Sept. 1	Severe, acute.	" 2. 0.20 " " "	" 2. "	An excess of neoarsphenamine. Early blackhead lesions.
27	" 1	Typical.	" 2. 0.15 " " " " 6. 0.12 " " " " 13. 0.075 " " "	" 15. "	Blackhead lesions; healing.
28	" 5	"	" 6. 0.13 " " " " 13. 0.075 " " "	Recovered.	Severe infection in a large turkey. Recovery slow; 2 1/4 wks.
29	" 6	"	" 6. 0.07 " " " " 8. 0.05 " " "	"	Steady improvement following injection.
30	" 6	"	" 6. 0.04 " " " " 8. 0.05 " " "	Sept. 8. D.	Lethal dose of neoarsphenamine. Typical blackhead lesions.
32	" 10	Mild.	" 21. 0.10 " " "	Recovered.	At no time very ill.
37	" 21	Typical.	" 21. 0.07 " " "	"	Rapid improvement.
38	" 21	"	" 21. 0.05 " " "	"	Gradual "
39	" 21	"	" 21. 0.08 " " "	"	" "

2  
Treated with neoarsphenamine.

TABLE I—Continued.

Group.	Turkey No.	Date of onset of disease.	Symptoms.	Injections.	Result.	Remarks.
	40	1922 Sept. 21	Typical.	Sept. 21. 0.5 cc. of 1 per cent solution in breast.	Sept. 24. D.	Typical blackhead lesions.
	41	" 21	"	Sept. 21. 0.5 cc. of 1 per cent solution in breast. Sept. 24. 2 cc. of 1 per cent solution in breast. Sept. 29. 1.5 cc. of 1 per cent solution in vein.	" 29.	" "
	42	" 21	"	Sept. 21. 0.5 cc. of 1 per cent solution in breast. Sept. 24. 2 cc. of 1 per cent solution in vein.	Recovered.	Appears well 8 days after onset.
	48	" 24	"	Sept. 24. 5 cc. of 1 per cent solution in vein. Sept. 29. 3.5 cc. of 1 per cent solution in vein.	Sept. 30. D.	Typical blackhead lesions.
3 Treated with atoxyl.	49	" 27	"	Sept. 27. 5 cc. of 1 per cent solution in vein. Sept. 29. 5.5 cc. of 1 per cent solution in vein.	Recovered.	Extreme emaciation; weakness after many weeks.
	50	" 27	"	Sept. 27. 2 cc. of 1 per cent solution in vein.	"	Recovery slow.
	51	" 29	"	Sept. 29. 5 cc. of 1 per cent solution in vein.	Sept. 29. D.	Typical blackhead lesions.

34	Sept. 13	Typical	Sept. 13. 0.10 gm. of neoarsphenamine in vein. Sept. 29. 5 cc. of 1 per cent solution of atoxyl in vein.	Oct. 1. D.	Typical blackhead lesions. Largest male.
35	" 13	"	Sept. 13. 0.10 gm. of neoarsphenamine in vein. Sept. 21. 0.14 gm. of neoarsphenamine in vein. Sept. 29. 4 cc. of 1 per cent solution of atoxyl in vein.	Recovered.	Disease ran a chronic course. Large hen turkey.
44	" 21	"	Sept. 21. 0.05 gm. of neoarsphenamine in vein. Sept. 29. 3 cc. of 1 per cent solution of atoxyl in vein.	Sept. 30. D.	Typical blackhead lesions.
45	" 21	Severe.	Sept. 21. 0.05 gm. of neoarsphenamine in vein. Sept. 29. 3 cc. of 1 per cent solution of atoxyl in vein.	" 30. "	" " "
46	" 21	"	Sept. 21. 0.13 gm. of neoarsphenamine in vein. Sept. 29. 4 cc. of 1 per cent solution of atoxyl in vein.	Recovered.	Disease ran chronic course.
47	" 21	"	Sept. 21. 0.15 gm. of neoarsphenamine in vein. Sept. 29. 3 cc. of 1 per cent solution of atoxyl in vein.	"	" " "

4  
Treated with atoxyl  
and neoarsphenamine.

TABLE I—Continued.

Group.	Turkey No.	Date of onset of disease.	Symptoms.	Injections.	Result.	Remarks.
		1922				
	52	Oct. 7	Typical.	Oct. 9. 7 cc. of 25 per cent solution in vein.	Recovered.	Prompt improvement. Well 7 days after onset.
	53	"	"	Oct. 9. 8 cc. of 25 per cent solution in vein. Oct. 15. 12 cc. of 25 per cent solution in vein.	"	Improvement after second injection.
	54	"	"	Oct. 9. 6 cc. of 25 per cent solution in vein. Oct. 15. 6 cc. of 25 per cent solution in vein.	"	Improvement gradual.
5 Treated with try- parsamide.	55	"	10 Severe.	Oct. 10. 4 cc. of 25 per cent solution in vein. Oct. 12. 4 cc. of 25 per cent solution in vein. Oct. 15. 5 cc. of 25 per cent solution in vein.	Oct. 16. D.	Liver extensively involved. Difficulty in finding food on account of pock lesions on eyelids.
	56	"	15 Typical.	Oct. 15. 6 cc. of 25 per cent solution in vein. Oct. 20. 4 cc. of 25 per cent solution in vein. Oct. 26. 6 cc. of 25 per cent solution in vein.	Recovered.	Improvement gradual.

57	Oct. 15	Typical.	Oct. 16. 15 cc. of 25 per cent solution in vein.	Recovered.	Improvement prompt.
58	" 16	"	Oct. 16. 5 cc. of 25 per cent solution in vein and 22 cc. subcutaneously. Oct. 26. 14 cc. of 25 per cent solution in vein.	"	" gradual.
59	" 17	"	Oct. 17. 26 cc. of 25 per cent solution in vein. Oct. 18. 11 cc. of 25 per cent solution subcutaneously.	"	" prompt.
60	" 18	"	Oct. 18. 9 cc. of 25 per cent solution in vein.	"	"
61	" 18	"	Oct. 18. 7 cc. of 25 per cent solution in vein.	"	"
62	" 18	Severe.	Oct. 20. 8 cc. of 25 per cent solution in vein.	Oct. 26. D.	Typical blackhead lesions.
64	" 29	Typical.	Oct. 29. 22 cc. of 25 per cent solution in vein.	Recovered.	Improved steadily.
65	" 29	"	Oct. 31. 14 cc. of 25 per cent solution in vein.	"	"

5  
Treated with try-  
parsamide.

TABLE I—*Concluded.*

Group.	Turkey No.	Date of onset of disease.	Symptoms.	Injections.	Result.	Remarks.
		1922				
	66	Oct. 30	Typical.	Oct. 31. 7 cc. of 25 per cent solution in vein and 8 cc. subcutaneously.	Recovered.	Improved steadily.
	67	Nov. 24	"	Nov. 25. 14 cc. of 25 per cent solution in vein.	"	"
	68	" 25	"	Nov. 25. 8 cc. of 25 per cent solution in vein and 8 cc. subcutaneously.	"	"
	69	Dec. 17	"	Dec. 17. 14 cc. of 25 per cent solution in vein.	"	"
5 Treated with try- parsamide.						
	43	Sept. 21	Typical.	Sept. 21. 0.5 cc. of 1 per cent solution of atoxyl in vein. Oct. 14. 4 cc. of 25 per cent solution of tryparsamide in vein.	Recovered.	Course of infection chronic.
6 Treated with atoxyl and tryparsamide.						

While it is possible that the above estimated doses of arsenic may not be especially harmful to turkeys, bad results may follow the administration of so powerful a poison in the general ration for a flock. One especially greedy bird may take much more than his share of the food, and if the arsenic is given in milk or water the thirst produced by the poison may result in its being taken in undue amount.

To obtain an idea of the amount of arsenic that may be administered in a single dose, two turkeys, one weighing  $1\frac{1}{2}$  and the other 2 kilos, were given 0.7 and 1 cc. respectively of liquor acidi arsenosi by mouth. The first had 2 days previously received 0.05 gm. of neoarsphenamine and the second 11 days later received 0.08 gm. of the same, in both cases injected intravenously. These two turkeys were markedly prostrated after taking the arsenic, one not eating for several days. There was eventually some improvement, but both finally died without recovery from the blackhead infection. Since both birds were weakened by the disease and received additional arsenic in the neoarsphenamine injected on another occasion, it appears probable that the turkey may withstand as large doses of arsenic as the rabbit. The results obtained in these two instances also show that arsenic when given in large doses to infected turkeys fails to control blackhead.

*Atoxyl Treatment.*—Atoxyl was employed in a small number of cases, but as the turkeys had previously been receiving arsenic in their food, it was not considered advisable to employ more than very moderate doses. Consequently the results are not especially conclusive. There were, however, several recoveries among the treated birds.

*Tryparsamide Treatment.*—Another arsenic compound, tryparsamide, or *N*-phenylglycineamide-*p*-arsonic acid, prepared by Jacobs and Heidelberg<sup>11</sup> and introduced by Brown and Pearce<sup>12-17</sup> in the treatment of trypanosome and spirochete infections was finally utilized

<sup>11</sup> Jacobs, W. A., and Heidelberg, M., *J. Exp. Med.*, 1919, xxx, 411.

<sup>12</sup> Brown, W. H., and Pearce, L., *J. Exp. Med.*, 1919, xxx, 417.

<sup>13</sup> Pearce, L., and Brown, W. H., *J. Exp. Med.*, 1919, xxx, 437.

<sup>14</sup> Pearce, L., and Brown, W. H., *J. Exp. Med.*, 1919, xxx, 455.

<sup>15</sup> Brown, W. H., and Pearce, L., *J. Exp. Med.*, 1919, xxx, 483.

<sup>16</sup> Pearce, L., and Brown, W. H., *J. Exp. Med.*, 1921, xxxiii, 193.

<sup>17</sup> Pearce, L., *J. Exp. Med.*, 1921, xxxiv, No. 6, Suppl. 1.

in the treatment of turkeys infected with blackhead. A 25 per cent solution of tryparsamide in distilled water was injected, usually into the wing vein but occasionally subcutaneously. Since this solution on boiling became yellowish in the course of a few days, the salt was as a matter of routine dissolved in sterile distilled water and used without further sterilization. It was thought inadvisable to use the dregs of the solution, which showed a fine sediment, for intravenous injection, and this portion was therefore injected subcutaneously. In contrast with neoarsphenamine, tryparsamide produced no evidence of pain when injected subcutaneously, and no more than a transient local edema followed the injection of large doses beneath the skin on the back of the head and neck. Little or no bleeding resulted from the puncture of the vein. No blindness or other untoward result followed the tryparsamide treatment of cases of natural infection. After the intravenous injection of the drug there was a variable amount of weakness which lasted only a few hours at most, and this was usually followed by pronounced improvement. The birds at first showed a marked thirst, after which the appetite returned. Another favorable effect was that they would now squat down and rest.

From a study of the pathology of this disease, it is readily understood why recovery occurs so infrequently in young turkeys if the disease is allowed to run its natural course. Not only is the function of the cecum more or less completely interfered with, but often the destruction of liver parenchyma is so extensive as to preclude recovery. In a series of seventeen cases of spontaneous blackhead treated with tryparsamide there were two deaths, or a mortality of less than 12 per cent.

One of the two tryparsamide-treated birds that failed to recover had both eyes closed with lesions of bird-pox so that it was unable to feed without assistance, the other was extremely ill before receiving treatment. Stained sections of the diseased liver of the first of these showed a total disappearance of liver parenchyma in the involved areas, leaving a reticulum transversed by blood vessels and in certain areas extensively infiltrated with cells, chiefly of lymphoid type. There were early regenerative changes present but repair was far from complete.



The dosage employed was based on that determined by Brown and Pearce<sup>12</sup> for laboratory animals. From 1 to 1.5 gm. per kilo may be safely injected intravenously into infected turkeys, but somewhat smaller doses should be employed with very young turkeys. From the favorable results here obtained, it should not be assumed that tryparsamide is a certain cure for all cases of blackhead. On account of the extensive destruction of the liver parenchyma, it would appear futile to undertake the cure of blackhead in an advanced stage. The treatment should begin as soon as the diagnosis can be established. Experience has shown that delay in beginning treatment both reduces the probability of recovery and prolongs the period of convalescence if the bird survives. Since blackhead may produce death within a relatively short period after the lesions are sufficiently developed to produce symptoms, prompt action is more essential than in the treatment of a mild or more chronic disease. After a time the following course was adopted for the detection of cases of blackhead at the earliest possible moment. The ground beneath the night shelter was examined each morning for sulfur droppings and, if they were found, a search was made for a moping turkey, which was then placed under observation until the diagnosis could be established by the discharge of sulfur droppings, lack of appetite, etc., and by the elimination of other possible ailments such as obstruction of the crop or other portions of the alimentary tract. If the symptoms were characteristic of blackhead, the turkey was placed in the hospital yard and treatment at once instituted.

From the results obtained in the treatment of spontaneous blackhead, it appeared that tryparsamide was the most effective of the compounds employed. The percentage of recoveries following the treatments outlined above should not, however, be taken as a true index of the therapeutic value of the drugs. It is probable that the mortality in cases of blackhead would have diminished as the season advanced, and it seems fairly well established that the chances of recovery increase as the turkey matures. The turkeys of the group treated with tryparsamide were on the average somewhat larger than those of the other groups, but they were of various ages and sizes. Furthermore, recovery followed tryparsamide treatment in a number of relatively small turkeys weighing from 1 to 2 kilos (roughly 8 to 15 weeks old).

The clinical action of the drugs with respect to the amelioration of serious symptoms should also be considered. Neoarsphenamine in adequate doses for the treatment of blackhead shows a pronounced toxicity and may give an immediate high mortality in young birds. Improvement of appetite, increase of vigor, and ability to rest usually succeed the period of prostration that follows the injection of this drug. Tryparsamide, however, may be given in large doses without producing marked prostration and, judging from clinical symptoms, acts fully as beneficially on the course of the disease.

On account of the lack of adequate controls with respect to age, season, and severity of infection in the treatment of spontaneous cases of blackhead, it seemed desirable to subject these drugs to experimental test.

#### *Inoculated Blackhead.*

The following experiment was planned to compare the effects of treatment of inoculated blackhead in turkeys of approximate age and weight with these drugs. Inoculated blackhead has been found to run a more uniform course than the natural infection, and since it is possible to make direct observations on the development of the local lesion, all question of error in diagnosis is eliminated.

*Experiment.*—October 9, 1922. Twelve young turkeys weighing from 555 to 890 gm. were inoculated in the left breast with bits of liver lesions from a case of spontaneous blackhead. Eight of these turkeys were 60 days old and four were 68 days old. October 14 (5 days after inoculation). All but one (No. 71) showed well marked swellings on the breast at the site of inoculation. In this single exception an indurated mass situated deep in the muscle was found on the 11th day after inoculation. These turkeys were distributed in four groups of three each, the first group serving as controls, the second receiving neoarsphenamine, the third atoxyl, and the fourth tryparsamide treatment. All three drugs were administered intravenously in three successive doses given at 48 hour intervals, beginning 5 days after inoculation. The neoarsphenamine was dissolved in a small amount of sterile distilled water and injected immediately into the wing vein. A 1 per cent solution of atoxyl and a 25 per cent solution of tryparsamide were prepared by dissolving the required amounts of the salts in sterile distilled water. These solutions were used without further sterilization. The details of the injections together with other data are furnished in Table II.

The dosage employed was based on that previously determined for spontaneous blackhead. Since treatment was commenced in this

experiment before the turkeys showed any impairment of health due to the infection, larger initial doses than those employed might have been more effective. Furthermore, as the turkeys were later on distinctly weaker on account of the extensive lung involvement, the third and last dose was unreasonably large and evidently hastened death in a number of cases.

Nearsphenamine treatment instituted 5 days after inoculation of blackhead failed to check the development of the local lesion in the breast or to prevent the dissemination of the disease to the lungs. The injection of three doses of 0.04, 0.06, and 0.11 gm. into an 890 gm. turkey was followed by extensive paralysis. The two other nearsphenamine-treated turkeys also showed pronounced toxic symptoms after the third injection. None recovered.

Atoxyl in the amount administered failed to arrest the development of inoculated blackhead, but possibly larger doses might have been employed. There were no recoveries.

Tryparsamide treatment was attended in one instance by improvement and recovery after two doses, in another by recovery after the injection of sufficient drug to cause partial paralysis, and in the third by general collapse terminating fatally.

There were thus only two recoveries in this series of twelve turkeys inoculated with blackhead, both being turkeys that were treated with tryparsamide.

Late in the experiment, when it was seen that there was no prospect of cure in any except the tryparsamide-treated cases, large doses (2 to 2.5 gm. per kilo of body weight) of this drug were injected into the three controls, and smaller doses (1 gm. per kilo of body weight) into the nearsphenamine- and atoxyl-treated turkeys. In no instance did the administration of tryparsamide late in the inoculated disease result in a recovery. A period of unfavorable weather undoubtedly hastened the death of the treated turkeys.

The histological study of the lesions from this series indicates that healing of liver lesions may occur under treatment while the disease is still active and extending in the lung and possibly in the local breast lesions. Thus a control turkey (No. 70) which received tryparsamide treatment late in the infection, showed advanced healing of the liver lesions with no demonstrable organisms, whereas in the lungs there

TABLE II.

Group.	Turkey No.	Inoculation.	Weight.	Injections into wing vein.	Result.	Remarks.
1 Controls.	70		Oct. 14. 720 gm. " 18. 780 "		Oct. 29. D.	Inoculated blackhead, involving breast, lungs, and liver.
	71		" 14. 725 " " 18. 780 "		" 24. "	Inoculated blackhead, involving breast and lungs, also cecal and liver lesions due to spontaneous blackhead.
	72		" 14. 780 " " 18. 835 "		" 24. "	Inoculated blackhead, involving breast, lungs, and liver. A core without any microscopic evidence of blackhead in cecum.
2 Treated with neoarsphenamine.	73		Oct. 14. 780 gm. " 16. 805 "	Oct. 14. 0.04 gm. " 16. 0.05 " " 18. 0.08 "	Oct. 21. D.	Inoculated blackhead; lesions of breast, lungs, liver, and pancreas.
	74		" 14. 890 " " 16. 975 "	" 14. 0.04 " " 16. 0.06 " " 18. 0.11 "	" 23. "	Inoculated blackhead; lesions of breast and lungs. Paralysis following last injection suggestive of overdosage.
	75		" 14. 780 " " 16. 835 "	" 14. 0.03 " " 16. 0.04 " " 18. 0.07 "	" 24. "	Inoculated blackhead; lesions of breast and lungs. Spontaneous blackhead with lesions in cecum and liver.
3 Treated with atoxyl.	76	Oct. 9, 1922. Each inoculated in left breast with blackhead liver lesions Oct. 14. A well defined lesion	Oct. 14. 780 gm. " 16. 835 "	Oct. 14. 2 cc. of 1 per cent solution. Oct. 16. 3 cc. of 1 per cent solution. Oct. 18. 6 cc. of 1 per cent solution.	Oct. 25. D.	Inoculated blackhead; lesions of breast and lungs.

3 Treated with atoxyl.	77	developing locally in each, except No. 71, in which the swelling was first noted on Oct. 20.	Oct. 14. 555 gm. " 16. 615 "  " 14. 835 " " 16. 945 "	Oct. 14. 1.5 cc. of 1 per cent solution. Oct. 16. 2 cc. of 1 per cent solution. Oct. 18. 4 cc. of 1 per cent solution.  Oct. 14. 2 cc. of 1 per cent solution. Oct. 16. 4 cc. of 1 per cent solution. Oct. 18. 8 cc. of 1 per cent solution.	Oct. 24. D.  " 21. "	Inoculated blackhead; lesions of breast, lungs, and liver.  Inoculated blackhead; lesions of breast and lungs.
4 Treated with trypanarsamide.	79		Oct. 14. 890 gm. " 16. 975 "	Oct. 14. 3 cc. of 25 per cent solution. Oct. 16. 6 cc. of 25 per cent solution.	Recovered.	Breast lesion 2 by 3 by 1 cm.; regressed following second injection. General condition gradually improved although growth somewhat checked.
	80		" 14. 835 " " 16. 865 "	Oct. 14. 4 cc. of 25 per cent solution. Oct. 16. 6 cc. of 25 per cent solution. Oct. 18. 6 cc. of 25 per cent solution.	Oct. 24. D.	Inoculated blackhead; lesions of breast and lungs. Impaired vision and weakness of legs following last injection.
	81		" 14. 610 " " 16. 670 "	Oct. 14. 4 cc. of 25 per cent solution. Oct. 16. 8 cc. of 25 per cent solution. Oct. 18. 9 cc. of 25 per cent solution.	Recovered.	Appetite unimpaired throughout infection. Growth unchecked. Partial paralysis following last injection; mincing gait with hyperextension of toes persists.

were active lesions and moderately numerous apparently healthy organisms. In one of the neoarsphenamine-treated turkeys which received an injection of tryparsamide late in the disease, there were an active breast lesion and extensive disease in the lungs but the liver lesions showed an unusual picture; the affected areas presented a connective tissue reticulum through which extended the dilated sinusoids but in which there was only a trace of necrotic parenchyma. The prompt absorption of the necrotic tissue and the maintenance of circulation in the infected portions are undoubtedly favorable to repair. The histological evidence that blackhead infection is more readily checked in the liver than elsewhere in the body by chemotherapeutic agents indicates the reason that recovery occurs on treatment so much more frequently in the spontaneous than in the inoculated disease.

#### CONCLUSIONS.

The natural form of blackhead, although very fatal to young turkeys, is favorably influenced by treatment with certain of the newer arsenical compounds.

Neoarsphenamine injected intravenously in toxic doses has a somewhat favorable effect on the course of the spontaneous disease and evidently lowers the mortality. Its instability, its pronounced toxicity for young turkeys, as well as the difficulty of obtaining amounts suitable for the cases to be treated on any one occasion, serve to make its employment impracticable.

Less favorable results were obtained with atoxyl.

Arsenious acid fed daily in small doses failed to prevent blackhead and possibly increased the incidence of infection. In larger doses it failed to cure spontaneous blackhead.

Tryparsamide may be injected in cases of spontaneous blackhead in doses as high as 1 gm. per kilo of body weight either intravenously or subcutaneously without serious toxic effect. Prompt clinical improvement usually follows and the mortality is undoubtedly greatly lowered.

With inoculated blackhead recovery is more difficult to obtain. Birds treated by injections of either neoarsphenamine or atoxyl failed

to recover. There were two recoveries among three tryparsamide-treated cases. An attempt to cure inoculated blackhead at an advanced stage by the injection of tryparsamide failed.

The study of the lesions of treated turkeys shows that tryparsamide, and to some extent nearsphenamine, have a more pronounced effect on the liver lesions than on the disease in the lung, which accounts for the discrepancy in the results of the treatment of spontaneous and inoculated blackhead. There is histological evidence of prompt absorption of necrotic liver parenchyma; moreover, the protozoa are destroyed more rapidly in the liver than in the lung.

Tryparsamide has the most pronounced curative action on blackhead infection of any of the drugs thus far tested. Either intravenous or subcutaneous injection of appropriate doses of this drug at the first appearance of symptoms should serve to cure the majority of cases of blackhead.