COMPARATIVE RESISTANCE AMONG DIFFERENT BREEDS OF CHICKENS TO THE ROUNDWORM ASCARIDIA LINEATA (SCHNEIDER)

by

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INTRODUCTION

The resistance of a host to invading organisms is an important factor of disease prevention. Often times bacterial diseases are combated by inoculating into a host antigens that cause the body to produce specific antibodies, thereby protecting the host. Helminthiasis apparently is not readily controlled in this manner. Antigens from metazoan parasites either do not stimulate the host to production of antibodies, or the antibodies are incapable

of establishing immunity. However, some workers (McCoy, 1931 and others) in this field are certain of having demonstrated acquired immunity to helminths, and it is probable that more evidence will be secured. But at present, there is little proof of acquired resistance to metazoan parasites, particularly the intestinal forms. On the other hand, natural resistance which commonly consists of species, racial and individual resistance is more readily demonstrated. Certain species will not have diseases of others, for example, man is not susceptible to hog cholera. Black races are more resistant to malaria and yellow fever than are white races, but they are more susceptible to typhoid. An individual may seem more resistant to a disease than does another member of his family. Less frequently associated with natural immunity are breed and age resistance. Breeds have diseases of other breeds but are not affected to the same extent, and it is known that older hosts are more resistant than the younger ones.

The resistance of one breed of chickens, the Single Comb White Leghorns, to the intestinal nematode Ascaridia lineata (Schneider) was studied through a period of several years by Ackert and his associates. They found, for example, that White Leghorn chicks two months or less of age were very susceptible to the A. lineata, but by the time the

chickens were three months old they were very resistant to the viability and growth of the nematodes (Ackert, 1930). Whether or not all breeds of chickens react to this parasite in the same manner is unknown. To obtain information on this question five experiments were conducted in which Rhode Island Reds, White Plymouth Rocks and White Leghorns were compared for breed and age resistance to the intestinal nematode of chickens, Ascaridia lineata (Schneider).

ACKNOWLEDGMENTS

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MATERIALS AND METHODS

During the two years that these five experiments were in progress 727 chickens were received from an accredited hatchery. The chicks were placed in screened, steam heated, well lighted and ventilated pens. That these pens were suitable for normal growth and development of the chickens was shown by Herrick, Ackert and Danheim (1923) who were the first to raise in confinement a considerable number (300) of chickens to maturity. Some of these fowls remained

in the pens over three years and laid eggs which yielded normal hatches.

The chickens were given a carefully planned diet consisting of yellow cornmeal, 40 gms.; oatena, 17 gms.; alfalfa leaf meal, 4 gms.; meat meal, 10.4 gms.; powdered milk, 6.4 gms.; cracked wheat, 15 gms.; and cod liver oil, 1.69 gms. To retain the vitamin A potency of the cod liver oil, a barrel of feed sufficient for 300 chickens was mixed each week.

When the chickens were between one and two weeks old each was banded and the numbers recorded in a ledger. Weekly weights were kept for each chicken, and at the termination of the experiments these weights were averaged for each breed to provide data on growth rates. From these figures comparative growth curves were constructed.

The parasite used in these studies is the fowl nematode Ascaridia lineata (Schneider), which belongs to the Family Ascaridae, Superfamily Ascaroidea, Order Eunematoda, and Phylum Nemathelminthes.

A week after obtaining the chickens mature A. lineata were received from a poultry house in Topeka, Kansas, where they had been taken from chickens. These worms were for the purpose of procuring egg cultures to be used later in infesting experimental chickens. To secure the eggs, the

posterior end of a worm was excised and the internal organs pressed into a sterile petri dish; the uteri were isolated and transferred to another sterile dish. After spreading out the uteri ribbon-like, they were punctured at various points thereby liberating the eggs. At such places the eggs were inspected under the microscope to determine their fertility by the presence of a light area in the center (Ackert, 1931). The parts of uteri containing fertilized eggs were isolated, the eggs teased out, drawn up with a pipette and transferred to the final sterile petri dish. In this way pure cultures were obtained without any tissue remaining to contaminate and induce fungous growth. Distilled water plus three or four drops of two per cent formalin were added to prevent bacterial or fungous growth. The eggs were incubated at 30° C. for three weeks which allows ample time for them to become fully infective, known as the coiled embryo stage. Each day the dish covers were removed to insure an adequate supply of oxygen.

When the chickens were two weeks of age each breed was divided into small lots. Group A was parasitized and Groups B, C, D and E, unparasitized, were placed in other pens. This precaution was taken to prevent infection from the parasitized group.

To parasitize the chickens a drop of water containing the eggs of A. lineata was placed under a microscope and 50 ± 5 eggs were counted out by means of a mechanical stage. The work of Ackert, Graham, Nolf, and Porter (1931) showed that 50 eggs were best suited for this type of work. The eggs were wiped off the slide with a small piece of filter paper and then loosely rolled with the eggs on the outside; this was placed in the chick's esophagus with forceps.

As these chickens grew older, they were spread out to prevent crowding and cannibalism. For three weeks after parasitizing each group was permitted to grow and then was killed. The evening before autopsy the chickens were taken off feed and placed on clean cement floors where food was unobtainable. In this way the intestines were rather free from debris which hindered the examination for parasites. The chickens were decapitated and the small intestine from the gizzard to the caeca removed. This was broken into three or four pieces after the total length was measured, and by means of warm water under pressure (Ackert and Nolf, 1929), the contents including worms were washed out into jars, and preserved in 10 per cent formalin until examined.

Examination was made by means of a wide angle binocular microscope. The worms from each chicken were preserved in vials with 4 per cent formalin. In measuring, the shadow

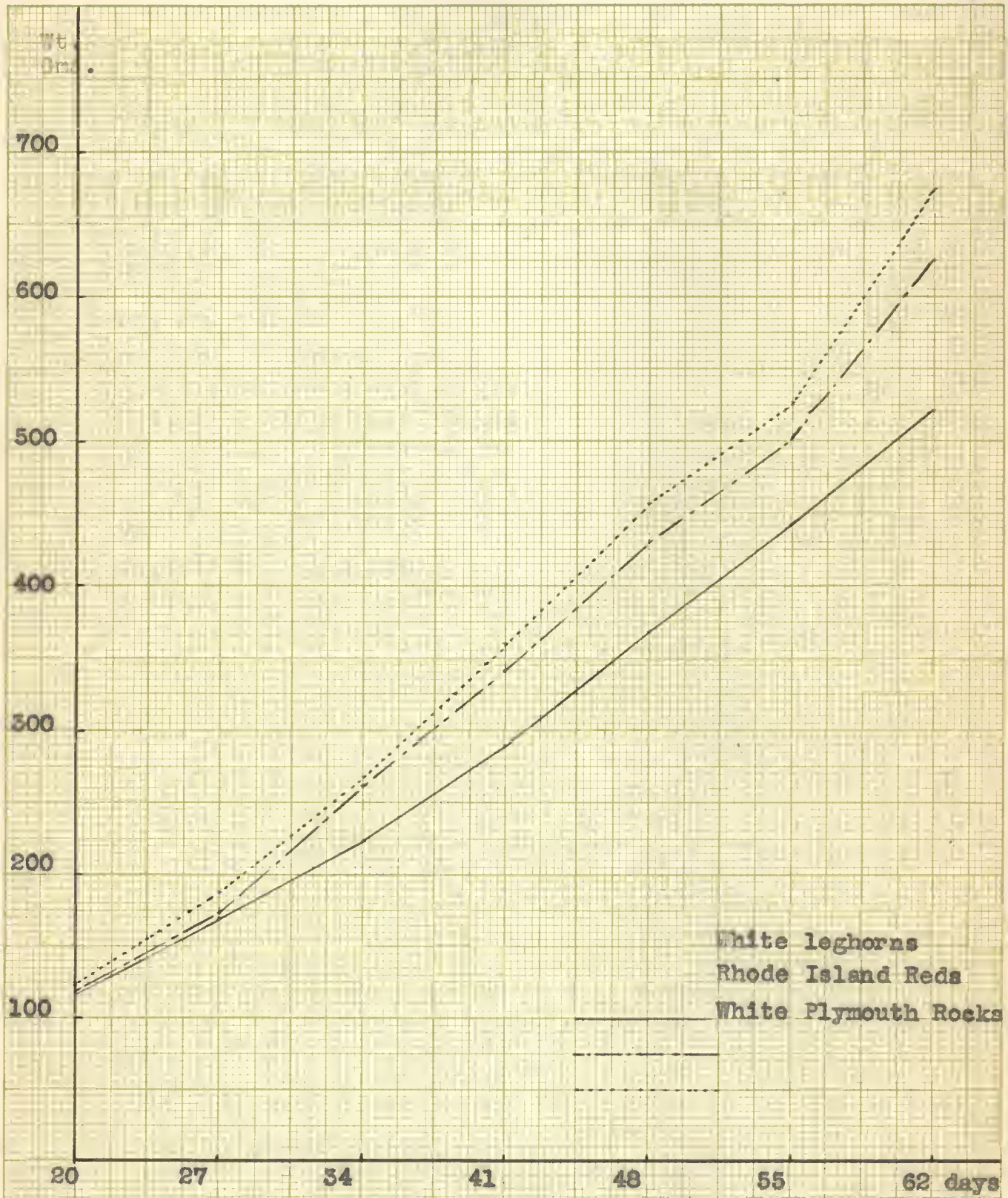

Fig. 1. Growth curves of chickens in Experiment I; includes Group C parasitized when 30 days old, and group E parasitized when 44 days of age.

of each worm magnified six times was thrown on the ground glass of a photographic bellows and traced on onion skin paper; from these tracings measurements were made with a milled wheel having divisions enlarged six times. Later, the worm lengths and numbers were recorded in Record Book XIII, pages 152-243, along with the weekly weights of the chickens. The data were treated statistically thereby facilitating the interpretation of the results.

EXPERIMENTAL DATA

Experiment I

On October 3, 1932, day-old chicks from an accredited hatchery were received as follows: 50 White Plymouth Rocks, 50 White Leghorns, and 50 Rhode Island Reds. When these chickens had grown to 30 days of age, they were divided into two equal groups: C and E. Group C was then parasitized, and Group E was parasitized two weeks later. Ackert (1923) found that at about the tenth day the young A. lineata begin to bore between the villi and penetrate the intestinal mucosa destroying Lieberkühn's glands; after the eighteenth day the worms free themselves and withdraw into the lumen of the duodenum. For this reason each group was kept three weeks after parasitizing. The birds were

then killed and the intestinal contents examined. The results were studied under the respective Groups C and E.

Group C. Group C contained 24 White Leghorns, 25 Rhode Island Reds and 27 White Plymouth Rocks, a total of 76 chickens. Counting the A. lineata from the Leghorns disclosed that 19 of the 24 fowls were infested. The range of infestations was from one to 22 worms; the average for the Leghorns was 5.33 worms. The examination of the White Plymouth Rocks showed six of the 27 infested. The range was one to 22 worms, which gave a mean number of 3.85, or 1.48 less worms than those from the Leghorns. Of the 25 Rhode Island Reds all were infested but two. The range was from one to 16 worms, the average being 2.92 worms or 2.41 fewer worms than from the Leghorns. The differences in numbers of A. lineata from these breeds were slight and not significant (Table 1).

In this work a ratio of 3.0 is considered indicative of a significant difference between two groups. If the difference is three or more times its probable error it is regarded as being beyond the limits of experimental error. A ratio of 3.0 means that in one hundred cases there is only one chance of the difference being due to chance variation. If a ratio of 4.5 is obtained there is one chance out of 417 for an error; a ratio of 5.0 indicates one

Table 1. Comparison of Ascaridia lineata found in chickens in Experiment I, Group C, hatched October 2, 1932, parasitized November 1, 1932 and killed three weeks later when seven weeks old.

	Breed	Number of hosts	Mean	Stand. Dev.	Error of mean	Dif.	Probable error of difference	Ratio D / P.E.D.
Numbers of Worms	W. Leghorns	24	5.33	5.50	.764			
						2.41	.876	2.75
	R. I. Reds	25	2.92	3.19	.430			
						0.93	.822	1.13
	W. P. Rocks	27	3.85	5.40	.701			
						1.48	1.03	1.44
	W. Leghorns	24	5.33	5.50	.764			
Lengths of Worms (mm.)	W. Leghorns	24	22.29	6.86	.409			
						5.37	.662	8.11
	R. I. Reds	25	16.92	6.55	.520			
						2.78	.693	4.01
	W. P. Rocks	27	19.70	6.04	.430			
						2.59	.614	4.21
	W. Leghorns	24	22.29	6.86	.409			

chance out of 1351 trials for an error; and a ratio of 8.0 suggests one chance in 14,705,832 of the difference being due to chance variation.

As a further means of judging resistance, the lengths of the worms were determined. The White Leghorns harbored the longest parasites, averaging 22.29 mm. The A. lineata in the White Plymouth Rocks averaged 19.70 mm., and the Rhode Island Reds had the shortest worms, an average of 16.92 mm. The difference between the mean lengths of the Reds and Leghorns was 5.37 mm., which was significant (8.11 times its probable error). Between the Reds and Rocks the difference in mean lengths of A. lineata was 2.78 mm., and between the Leghorns and the Rocks, 2.59 mm. Both of these differences were significant (Table 1). Summarizing it is seen that the Leghorns had the largest number of worms and the longest ones, the Reds had the least worms and the shortest ones, and the Rocks had intervening numbers and lengths of nematodes. These results, then, point to the tentative conclusion that the Rhode Island Reds are the most resistant, the White Rocks next, and the White Leghorns the least resistant to the viability and growth of A. lineata when the chickens are parasitized at 30 days of age.

Group E. At 44 days of age Group E was parasitized and at 65 days was killed. Twenty-four White Leghorns, 19 Rhode Island Reds, and 24 White Plymouth Rocks were involved. Of the 24 Leghorns six had no worms, and one to seven was the range of infestation; the mean number was 1.66 worms. The Rocks had the same average number; but 10 negatives were found in the 24 chickens. The infestation ranged from one to six worms in a chicken. Out of 19 Reds 12 proved negative. The range of infestation was one to three worms per chicken with 0.89 worm as an average. None of the differences in numbers of worms from these groups of chickens was significant (Table 2).

As to the rate of growth of the A. lineata, those in the White Leghorns averaged 17.31 mm. long and the worms from the White Rocks, 18.72 mm. The difference between the two was 1.41 mm. which was not significant. The Rhode Island Reds had worms 14.89 mm. in length, which was 2.42 mm. shorter than those in the Leghorns. This difference likewise was not significant. The difference in length of the worms between the Rocks and the Reds was 3.83 mm.; this was 3.96 times the probable error and, hence, probably significant (Table 2). As in Group C the Rhode Island Reds had the fewest and the shortest worms. The A. lineata from the White Leghorns and those from the White Plymouth

Table 2. Comparison of Ascaridia lineata found in chickens in Experiment I, Group E, hatched October 2, 1932, parasitized November 15, 1932, and killed three weeks later when nine weeks old.

	Breed	Number of hosts	Mean	Stand. Dev.	Error of mean	Dif.	Probable error of difference	Ratio D / P.E.D.
Numbers of Worms	W. Leghorns	24	1.66	1.60	.220	0.77	.953	0.81
	R. I. Reds	19	0.89	1.37	.207	0.77	.341	2.26
	W. P. Rocks	24	1.66	1.97	.271	0.00	.350	0.00
	W. Leghorns	24	1.66	1.60	.220			
Lengths of Worms (mm.)	W. Leghorns	24	17.31	5.59	.596	2.42	1.01	2.40
	R. I. Reds	19	14.89	4.97	.813	3.83	.966	3.96
	W. P. Rocks	24	18.72	4.88	.458	1.41	.791	1.78
	W. Leghorns	24	17.31	5.59	.596			

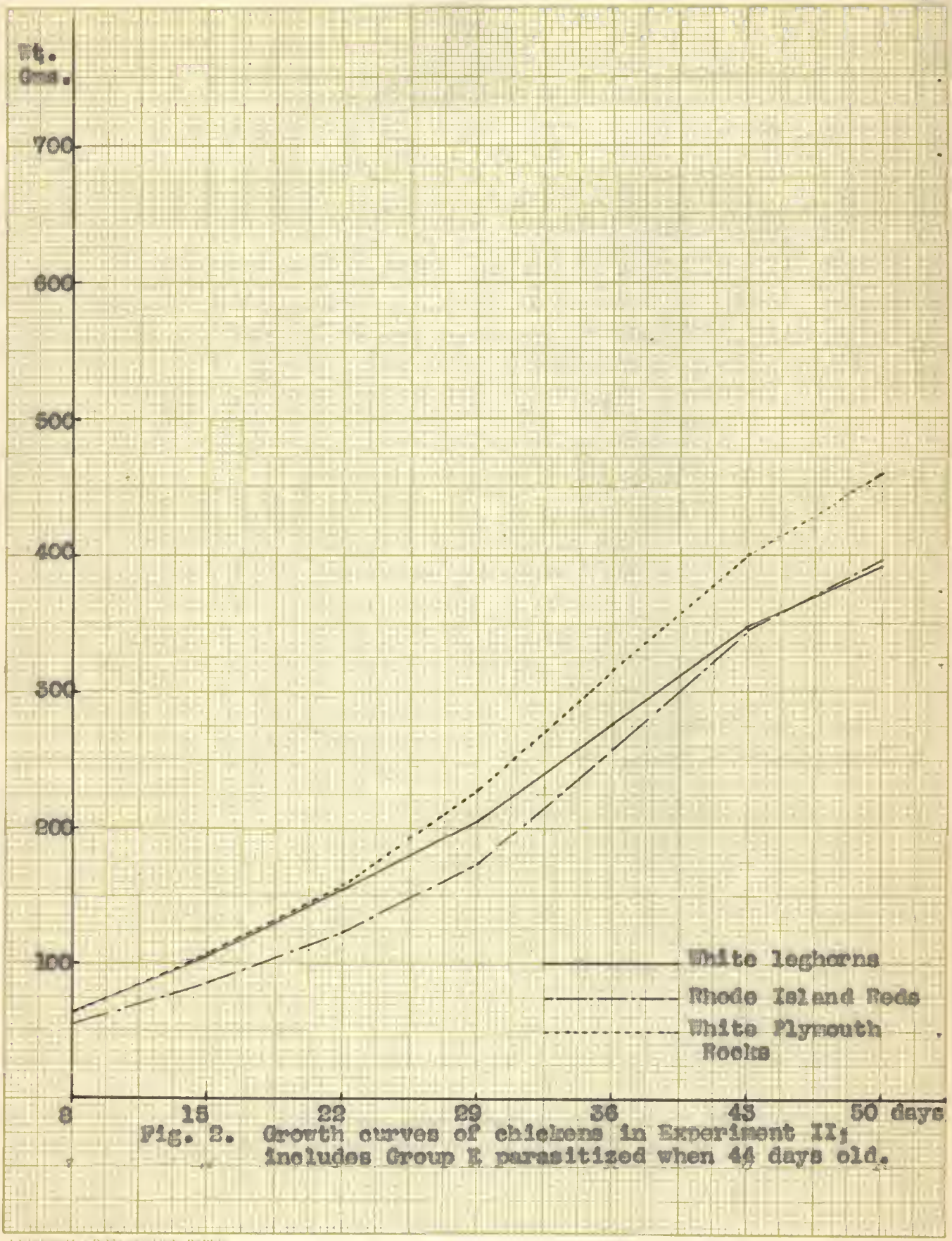

Fig. 2. Growth curves of chickens in Experiment II; includes Group E parasitized when 44 days old.

Rocks did not vary much.

Experiment II

The chickens for this experiment were hatched February 6, 1933. The same numbers were received as in Experiment I, namely, 50 White Leghorns, 50 White Plymouth Rocks, and 50 Rhode Island Reds. These were fed, banded, weighed and kept under the same conditions as those in Experiment I. At 30 days of age the chickens were divided into Group C and Group E. The former was given embryonated eggs at that time and Group E when 44 days of age.

Group C. Due to faulty egg cultures this portion of the experiment failed; only negative results were obtained. The weights were recorded as in Group E.

Group E. After having been parasitized for 21 days the chickens of Group E were killed and examined. Seventeen of the 22 Leghorns were infested with from one to six *A. lineata*, an average of 2.36 worms. Eleven of the 17 Rhode Island Reds had a range of one to seven nematodes, which averaged 1.88 worms. Of the 13 White Rocks, six harbored from one to eight worms, resulting in an average of 1.76 *A. lineata*. While none of the differences in average numbers of worms among these groups was significant, the Leghorns had the most.

Table 3. Comparison of Ascaridia lineata found in chickens in Experiment II, Group C, hatched February 6, 1933, parasitized March 21, 1933, and killed three weeks later when nine weeks old.

	Breed	Number of hosts	Mean	Stand. Dev.	Error of mean	Dif.	Probable error of difference	Ratio D / P.E.D.
Numbers of Worms	W. Leghorns	22	2.36	1.84	.265			
						.48	1.30	0.37
	R. I. Reds	17	1.88	1.91	.313			
						.12	1.10	0.12
	W. P. Rocks	13	1.76	2.62	.472			
						.60	.961	0.62
	W. Leghorns	22	2.36	1.84	.265			
Lengths of Worms (mm.)	W. Leghorns	22	24.63	6.40	.599			
						5.53	1.29	4.28
	R. I. Reds	17	19.10	9.23	1.14			
						1.76	1.51	1.16
	W. P. Rocks	13	20.86	7.04	.991			
						3.77	1.16	3.25
	W. Leghorns	22	24.63	6.40	.599			

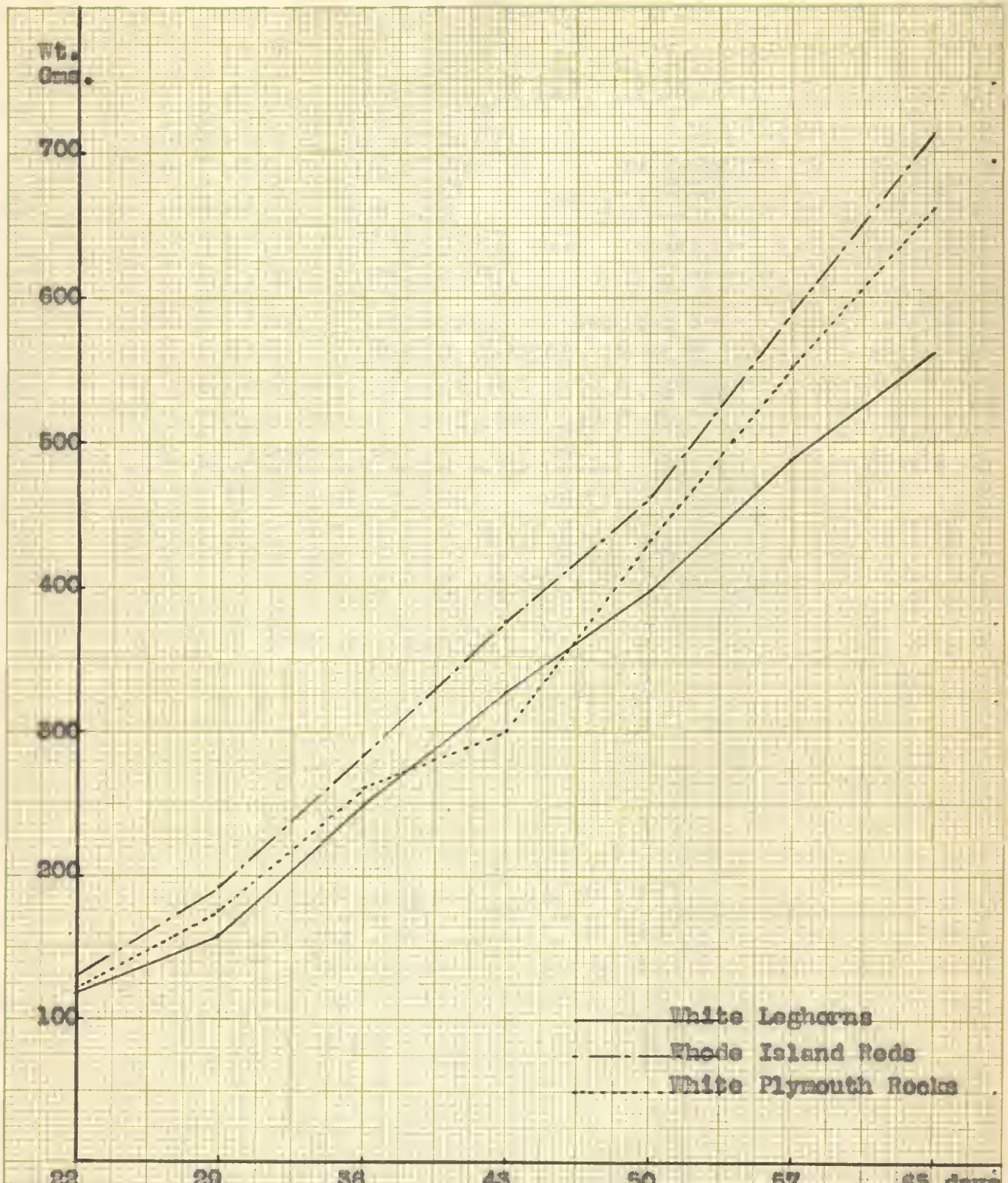

Fig. 3. Growth curves of chickens in Experiment III; includes Group C parasitized when 30 days old.

Concerning the growth of worms in the chickens, the A. lineata in the Leghorns had a mean length of 24.63 mm.; while the Reds had worms 19.10 mm. long. The difference between these two mean lengths was 5.33 mm., which was 4.28 times its probable error. The worms in the Rocks averaged 20.86 mm. in length, being 3.77 mm. shorter than those in the Leghorns. This difference was significant (Table 3). As in Experiment I, the Leghorns had the most and the longest worms. The Reds had the shortest A. lineata, but the Rocks had slightly less worms.

Experiment III

This experiment was conducted during the early summer of 1933 to replace Group C of Experiment II which failed from a faulty egg culture. The chicks were parasitized when 30 days of age and killed when 51 days old. Of the 25 Leghorns parasitized, 17 were infested, the range being from one to 37 worms. The average per chicken was 6.12 A. lineata. The difference, which was not significant, was 3.40 worms. Ten of the 18 Reds harbored A. lineata; the range was from one to 15 worms, which averaged 2.72 worms. Eighteen cases of infestation were found in the 34 White Rocks. They had worms ranging from one to 16 in number, or an average of 3.03 worms per chicken; this difference of 0.31 worm more than in

the Reds was negligible. The Leghorns and Rocks had a difference of 3.09 worms, but this likewise was not significant. As in Group C of Experiment I, the Leghorns of this group had the most nematodes and the Reds the least.

The lengths of worms in the Leghorns averaged 26.07 mm. and in Reds 24.96 mm. or 1.11 mm. shorter, which was not significant. The A. lineata from the Rocks averaged 21.24 mm. in length, or a significant difference of 4.83 mm. between the worms in the Rocks and Leghorns. Also, between the worms from the Rocks and those from the Reds there was a significant difference of 3.72 mm. in length.

As in Group C of the first experiment, the Leghorns had the longest and largest number of worms. The Reds averaged fewer worms per chicken, but they were not the shortest. In this experiment the Rocks had the shortest worms. The results are summarized in Table 4.

Experiment IV

This experiment was begun September 24, 1933, when a large number of one day old chicks were received from the same hatchery as those in previous experiments. The procedure was the same as before, except that different ages were used. Instead of using Groups C and E, Groups A, B and D were used. Group A contained 34 White Leghorns, 11

Table 4. Comparison of *Ascaridia lineata* found in chickens in Experiment III, Group C, hatched May 6, 1933, parasitized June 5, 1933, and killed three weeks later when seven weeks old.

	Breed	Number of hosts	Mean	Stand. Dev.	Error of mean	Dif.	Probable error of difference	Ratio D / P.E.D.
Numbers of Worms	W. Leghorns	25	6.12	7.94	1.07			
						3.40	1.27	2.68
	R. I. Reds	18	2.72	4.27	.673			
						.31	.835	0.37
	W. P. Rocks	34	3.03	4.26	.490			
						3.09	1.18	2.62
	W. Leghorns	25	6.12	7.94	1.07			
Lengths of Worms (mm.)	W. Leghorns	25	26.07	6.27	.316	1.11	.5	
						1.11	.564	1.97
	R. I. Reds	18	24.96	4.86	.468			
						3.72	.620	6.00
	W. P. Rocks	34	21.24	6.11	.408			
						4.83	.516	9.36
	W. Leghorns	25	26.07	6.27	.316			

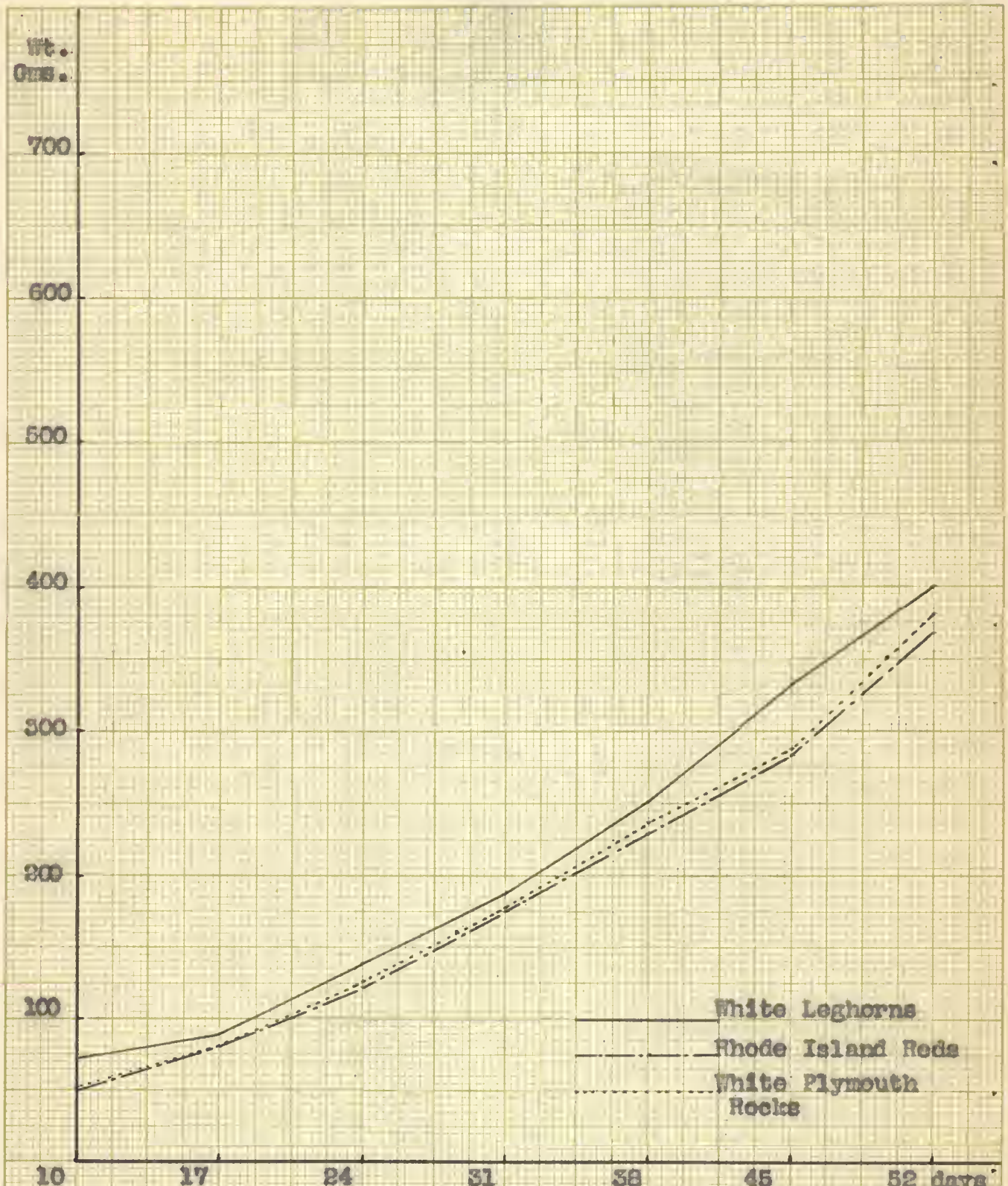

Fig. 4. Growth curves for Experiment IV; includes:
Group A, parasitized when 12 days old
Group B, parasitized when 19 days old
Group D, parasitized when 33 days old.

White Rocks. This group was killed at 54 days of age after infecting them when 33 days old.

Group A. The Leghorns, which were parasitized at 12 days of age, harbored from one to 43 worms or an average of 10.00 worms per chick. All chickens were infected. The Reds varied from one to 10 worms and averaged 6.09 A. lineata. Only one chick was free of the nematode. Two negatives were found in the White Rocks and 34 worms in one chicken. An average of 9.33 worms was found. The actual difference between these and the Reds was 3.24. The differences among the breeds were not significant (Table 5).

Apparently the Leghorns were less resistant because they had worms averaging 22.10 mm. in length as compared to 16.99 mm. in the Reds. This resulted in a difference of 5.11 mm. and a ratio of 10.23. The average lengths of worms in the Rocks was 16.38 mm. The actual difference of worms in the Reds and Rocks was not significant. A significant difference of 5.72 was found between the Rocks and Leghorns (Table 5). Again the Leghorns had the most and longest worms, the Reds the fewest worms. The Rocks had slightly shorter worms than did the Reds.

Group B. Table 6 shows the results from this group of chickens which were parasitized at 19 days of age. Forty-seven of 49 Leghorns were infested, the range being from

Table 5. Comparison of Ascaridia lineata found in chickens in Experiment IV, Group A, hatched September 24, 1933, parasitized October 6, 1933, and killed three weeks later when five weeks old.

	Breed	Number of hosts	Mean	Stand. Dev.	Error of mean	Dif.	Probable error of difference	Ratio D / P.E.D.
Numbers of Worms	W. Leghorns	34	10.00	10.67	1.24			
						3.91	1.87	2.09
	R. I. Reds	11	6.09	6.84	1.38			
						3.24	2.99	1.08
	W. P. Rocks	12	9.33	11.80	2.50			
						0.67	2.80	0.24
	W. Leghorns	34	10.00	10.67	1.24			
Lengths of Worms (mm.)	W. Leghorns	34	22.10	4.55	.167			
						5.11	.497	10.28
	R. I. Reds	11	16.99	5.69	.469			
						0.61	1.05	0.58
	W. P. Rocks	12	16.38	14.72	.938			
						5.72	0.952	6.08
	W. Leghorns	34	22.10	4.55	.167			

Table 6 . Comparison of Ascaridia lineata found in chickens in Experiment IV, Group B, hatched September 24, 1933, parasitized November 3, 1933, and killed three weeks later when six weeks old.

	Breed	Number of hosts	Mean	Stand. Dev.	Error of mean	Dif.	Probable error of difference	Ratio D / P.E.D.
Numbers of Worms	W. Leghorns	49	8.18	7.05	.680			
	R. I. Reds	25	5.84	6.84	.788	2.34	1.04	2.25
	W. P. Rocks	21	4.38	4.67	.688	1.46	1.05	1.40
	W. Leghorns	49	8.18	7.05	.680	3.80	.966	3.93
Lengths of Worms (mm.)	W. Leghorns	49	17.87	9.81	.331			
	R. I. Reds	25	13.59	5.26	.294	4.28	.482	8.88
	W. P. Rocks	21	11.35	6.54	.459	2.24	.545	4.11
	W. Leghorns	49	17.87	9.81	.331	6.47	.566	11.43

one to 19; the average, 8.18 worms. Twenty-four of the 25 Reds harbored parasites, a maximum of 24 worms in one chicken. The average was 5.84 worms. All but two Rocks were infested; the range was from one to 18 worms; an average of 4.38 worms per chicken. The Rocks had 3.80 worms per chicken less than the Leghorns, which gave a significant difference. There was no constant difference between the Leghorns and the Reds nor between the Reds and the Rocks.

In worm lengths a marked difference occurred among the breeds. The Leghorns had A. lineata 17.87 mm. long, and the Reds, 13.59 mm., or a difference of 4.28 mm. (a ratio of 8.88). The Rocks possessed worms 11.35 mm. long. This was 2.24 mm. shorter than in the Reds and resulted in a significant difference. The difference between the Rocks and the Leghorns was 6.47 mm., which was significant (a ratio of 11.43). In this group the Rocks had the fewest and shortest worms rather than the Reds, while the Leghorns remained the least resistant because they harbored the longest and the largest number of worms.

Group D. These chickens were parasitized at 33 days of age. Whereas, 15 of the 25 Leghorns were infested, only five of 14 Reds, and seven of 11 Rocks had worms. The range of infestation in the Leghorns was one to 10; in

the Reds, one to six; and in the Rocks, one to five. The Leghorns had an average of 1.44, the Rocks 1.82 and the Reds 0.86 worms per chicken, about the same number in all three breeds of chickens.

The longest worms were found in the Leghorns (19.94 mm.). The worms in the Reds averaged 18.71 mm., or a difference of 1.23 which was not significant. The Rocks had worms measuring 15.80 mm. or 2.91 mm. shorter than in the Reds. This difference was significant. A difference of 4.14 mm. between the worms in the Rocks and the Leghorns gave a ratio of 4.16. As in the previous experiments, the Leghorns had the longest and the Reds had the fewest A. lineata. The largest number of worms, however, and the shortest ones were in the White Rocks (Table 7).

Experiment V

The final experiment was completed in March, 1934. Groups A, B and D were used in order to support the previous experiment. The ages, conditions and periods of time were identical.

Group A. Nine of the 12 White Leghorns were infested; range, one to 11 worms; average, 5.42 worms per chicken. Twenty of the 24 Reds were parasitized, the largest infestation being 13 worms; the average, 4.33 A. lineata per

Table 7. Comparison of Ascaridia lineata found in chickens in Experiment IV, Group D, hatched September 24, 1933, parasitized October 27, 1933, and killed three weeks later when eight weeks old.

	Breed	Number of hosts	Mean	Stand. Dev.	Error of mean	Dif.	Probable error of difference	Ratio D / P.E.D.
Numbers of Worms	W. Leghorns	25	1.44	2.14	.246			
						0.58	.380	1.53
	R. I. Reds	14	0.86	1.60	.239			
						0.96	.459	2.09
	W. P. Rocks	11	1.82	1.75	.356			
						0.38	.433	0.88
	W. Leghorns	25	1.44	2.14	.246			
	W. Leghorns	25	19.94	7.11	.798			
						1.23	.804	1.53
	R. I. Reds	14	18.71	8.21	.160			
						2.91	.618	4.71
	W. P. Rocks	11	15.80	3.98	.597			
						4.14	.996	4.16
	W. Leghorns	25	19.94	7.11	.798			

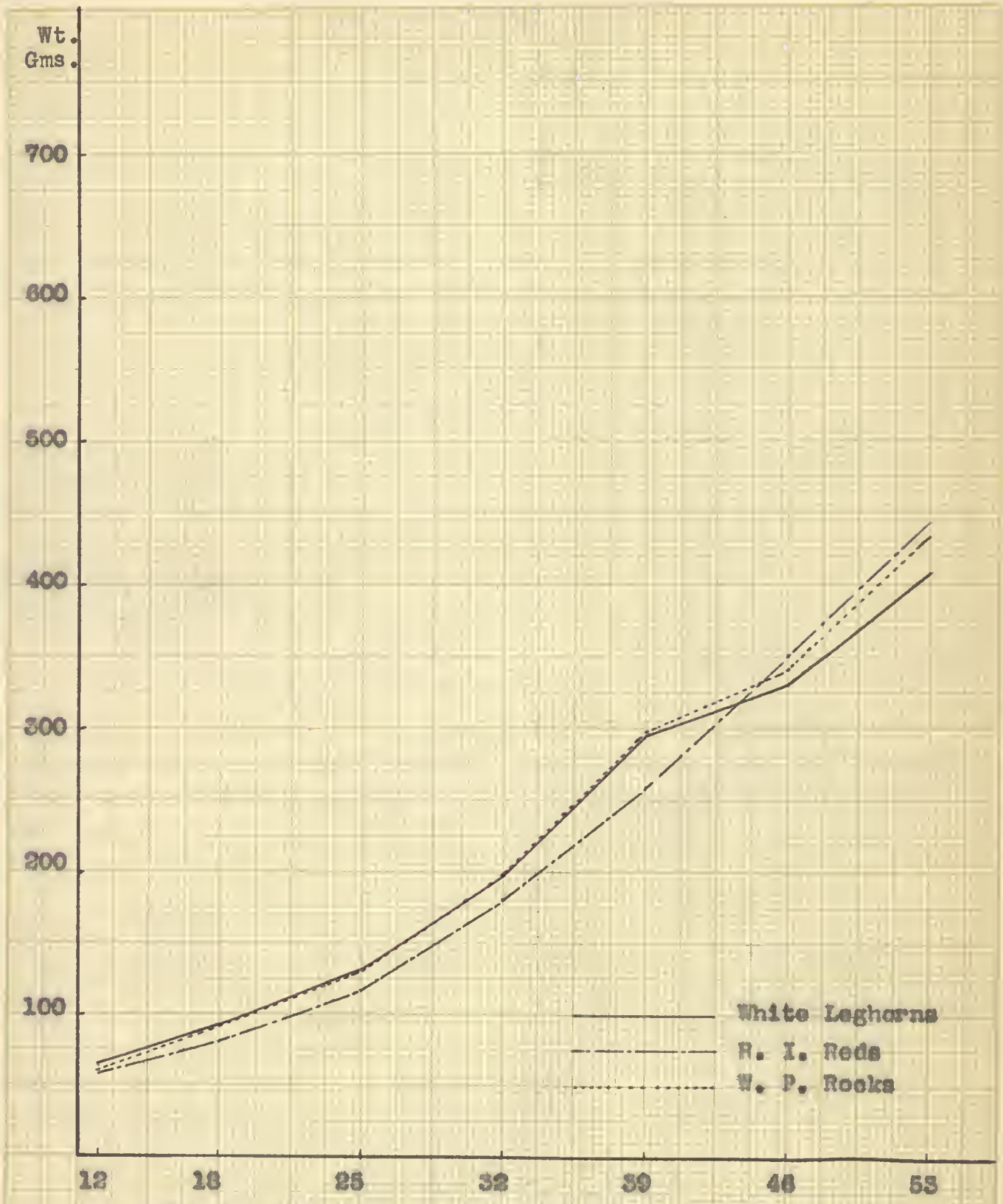

Fig. 5. Growth curves for Experiment V; includes:
Group A, parasitized when 12 days old
Group B, parasitized when 19 days old
Group D, parasitized when 33 days old.

bird. All but two of the 24 White Rocks were infested, the range being from one to 18 worms per chicken, average, 5.96. In this test the differences in numbers of worms lay within the range of experimental error (Table 8).

On measuring the worms, it was found that those from the leghorns averaged 21.95 mm. in length and that the worms from the Reds had a mean length of 18.10 mm. or a difference of 3.85 mm. which was significant. The White Rocks in this group were quite susceptible as their A. lineata averaged 23.45 mm. or 1.5 mm. longer than the Leghorns. This difference was not significant. Between the Rhode Island Reds and the White Plymouth Rocks there was a difference of 5.35 mm., which was significant. From this test it may be seen that, as in previous experiments, the Rhode Island Reds were the most resistant to the growth of the A. lineata, the White Leghorns and the White Plymouth Rocks being about equally susceptible.

Group B. Twelve Leghorns, 26 Reds and 34 Rocks were parasitised at 19 days of age and killed 21 days later. Nine of the 12 Leghorns were infested with from one to ten worms; an average of 3.91 worms. Of the 26 Reds, 12 harbored A. lineata with a range of one to nine worms; average, 1.46 worms. This was 2.45 less parasites than in the Leghorns which was a significant difference. Only 14 of 34

Table 8. Comparison of Ascaridia lineata found in chickens in Experiment V, Group A, hatched January 14, 1934, parasitized January 26, 1934, and killed three weeks later when five weeks old.

	Breed	Number of hosts	Mean	Stand. Dev.	Error of mean	Dif.	Probable error of difference	Ratio D / P.E.D.
Numbers of Worms	W. Leghorns	12	3.42	3.22	.627			
						0.91	.810	1.12
	R. I. Reds	24	4.33	3.74	.514			
						1.63	.844	1.93
	W. P. Rocks	24	5.96	4.87	.669			
						2.54	.916	2.77
	W. Leghorns	12	3.42	3.22	.627			
Lengths of Worms (mm.)	W. Leghorns	12	21.95	8.15	.859			
						3.85	.986	3.90
	R. I. Reds	24	18.10	7.32	.484			
						5.35	.635	8.43
	W. P. Rocks	24	23.45	7.33	.412			
						1.50	.953	1.57
	W. Leghorns	12	21.95	8.15	.859			

Table 9. Comparison of Ascaridia lineata found in chickens in Experiment V, Group B, hatched January 14, 1934, parasitized February 2, 1934, and killed three weeks later when six weeks old.

	Breed	Number of hosts	Mean	Stand. Dev.	Error of mean	Dif.	Probable error of difference	Ratio D P.E.D.
Numbers of Worms	W. Leghorns	12	3.91	3.04	.592			
						2.45	.676	3.62
	R. I. Reds	26	1.46	2.48	.327			
						0.22	.415	0.53
	W. P. Rocks	34	1.24	2.21	.256			
						2.67	.645	4.14
	W. Leghorns	12	3.91	3.04	.592			
Lengths of Worms (mm.)	W. Leghorns	12	20.98	6.88	.676			
						3.05	1.01	3.02
	R. I. Reds	26	17.93	6.82	.747			
						0.14	1.17	0.12
	W. P. Rocks	34	17.79	8.69	.904			
						3.19	1.13	2.82
	W. Leghorns	12	20.98	6.88	.676			

Rocks were infested with a range of one to 11 worms. The average was 1.24 or 2.67 worms less than from the Leghorns which gave a ratio of 4.14. The lengths of worms in the Leghorns averaged 20.98 mm. and those in the Reds, 17.93 mm. The difference of 3.05 mm. gave a ratio of 3.02. The worms from the Rocks averaged 17.79 mm. long, which was nearly the same as the Reds and gave no significant difference. The difference of 3.19 mm. between the worms of the Rocks and those of the Leghorns was not significant. In Table 9 it is noted that the Leghorns had the most and the longest worms, indicating that they were the least resistant. The Reds and Rocks had about the same amount of resistance to the A. lineata.

Group D. When 54 days old, this group of chickens was killed and the worms counted and measured. Four worms were the most found in one Leghorn and 15 of the 26 harbored A. lineata. Twenty-two of 43 Reds were infested and 16 worms were in one chicken of this breed. Twelve worms were found in one Rock and only 17 of 50 Rocks were infested. The Leghorns and White Rocks each averaged one worm per chicken while the Reds averaged 1.80 worms. No significant differences in numbers of worms were found among these groups of chickens.

Concerning resistance to the growth of the A. lineata, the worms in the Leghorns averaged 17.81 mm., in the Reds, 16.28 mm. and in the Rocks 20.04 mm. While the Leghorns had longer worms than those of the Reds, they were not significantly so. Here, the Rocks were the most susceptible, the difference of 3.76 mm. between the Rocks and the Reds being significant (Table 10).

Combined Experiments

As the number of individuals was small in some of the groups, the results of the five experiments described were combined, and chickens of the same age added together giving approximately 50 chickens of each breed in every group.

Group A. The White Leghorns in Group A, which were parasitized at 12 days of age, harbored an average of 8.28 worms per chicken, the Rocks, 7.03 and the Reds, 4.89 worms. The Reds had a significant difference of 3.39 less worms per chicken than the Leghorns. Differences in numbers of worms between the Reds and the Leghorns and between the Rocks and the Leghorns were not significant.

The longest worms were in the White Leghorns which had A. lineata averaging 22.03 mm. The Rocks had worms 20.34 mm. long, and the Reds, 17.66 mm. The differences in lengths of worms in all three breeds of chickens were

Table 10. Comparison of Ascaridia lineata found in chickens in Experiment V, Group D, hatched January 15, 1934, parasitized February 16, 1934, and killed three weeks later when eight weeks old.

	Breed	Number of hosts	Mean	Stand. Dev.	Error of mean	Dif.	Probable error of difference	Ratio D / P.E.D.
Numbers of Worms	W. Leghorns	26	1.00	1.11	.147			
						0.80	.368	2.17
	R. I. Reds	45	1.80	3.37	.338			
						0.80	.392	2.04
	W. P. Rocks	50	1.00	2.06	.197			
						0.00	.246	0.00
	W. Leghorns	26	1.00	1.11	.147			
Lengths of Worms (mm.)	W. Leghorns	26	17.81	6.67	.882			
						1.53	.983	1.56
	R. I. Reds	45	16.28	5.79	.434			
						3.76	.866	4.34
	W. P. Rocks	50	20.04	7.78	.750			
						2.23	1.16	1.92
	W. Leghorns	26	17.81	6.67	.882			

significant. This combined group shows that the Leghorns are the most susceptible and the Reds the least, while the Rocks are not as resistant as the Reds nor as susceptible as the Leghorns to the growth of the A. lineata.

Group B. These chickens were parasitized one week later or when 19 days old and killed when 40 days old. The worms in the Leghorns averaged 7.34 per chicken. The mean number of worms in the Reds was 3.91 and in Rocks, 2.44. The difference was not significant between the Rocks and Reds, but there was a significant difference of 4.90 worms (Table 11) between the Rocks and the Leghorns. The worm lengths in the Leghorns averaged 13.19 mm., those in the Reds 14.46 mm. and in the Rocks 13.37 mm. A significant difference of 3.73 mm. was found between the Leghorns and the Reds; a significant difference (4.82 mm.) was found, likewise, between the Leghorns and the Rocks. The difference in average worm lengths between the Rocks and the Reds was negligible. In this group the Leghorns were the most susceptible to the viability (numbers) and growth (lengths) of the A. lineata (Table 11).

Group C. This group, which was parasitized at 30 days of age and killed when 51 days old, is known as Group C. The Leghorns averaged 5.73 worms per chicken and the Reds 2.84 worms, or 2.89 less worms than the Leghorns. The

Table 11. Summary of the combined results of Experiments I, II, III, IV and V.

	Average number worms	Dif.	Ratio	Average length	Dif.	Ratio
Group A						
W. Leghorns	8.82			22.08		
		3.39	3.03		4.42	10.83
R. I. Reds	4.89			17.66		
		2.19	2.05		2.68	4.43
W. P. Rocks	7.08			20.34		
		1.20	0.68		1.74	3.24
W. Leghorns	8.28			22.09		
Group B						
W. Leghorns	7.34			18.19		
		3.70	4.75		3.73	7.80
R. I. Reds	3.61			14.46		
		1.17	1.76		1.09	1.20
W. P. Rocks	2.44			13.37		
		4.90	7.34		4.82	5.46
W. Leghorns	7.34			18.19		
Group C						
W. Leghorns	5.73			23.83		
		2.89	3.79		3.65	6.78
R. I. Reds	2.84			20.18		
		0.55	0.98		0.28	0.52
W. P. Rocks	3.39			20.46		
		2.34	2.99		3.37	7.61
W. Leghorns	5.73			23.83		
Group D						
W. Leghorns	1.22			20.00		
		0.36	1.18		3.41	4.61
R. I. Reds	1.58			16.59		
		0.43	1.38		0.34	0.47
W. P. Rocks	1.15			16.93		
		0.43	1.80		3.07	3.71
W. Leghorns	1.22			20.00		
Group E						
W. Leghorns	2.00			21.45		
		0.64	1.42		3.87	4.03
R. I. Reds	1.36			17.58		
		0.34	0.55		2.34	2.52
W. P. Rocks	1.70			19.92		
		0.30	0.49		1.53	2.45
W. Leghorns	2.00			21.45		

difference was significant. The Rocks had 3.39 worms, which was not significantly different from the Reds or Leghorns. For lengths of worms, the Leghorns averaged 23.83 mm. and the Reds, 20.18 mm. or a significant difference of 3.65 mm. and a ratio of 6.78. The Rocks had worms averaging 20.46 mm. in length or 3.37 mm. less than those from the Leghorns. The ratio was 7.61. Summarizing, the Leghorns had the longest and the most worms, whereas, the Reds had the shortest and fewest worms per chicken.

Group D. Group D was parasitized at 33 days of age and killed when 54 days old. The Leghorns had an average of 1.22 worms, the Reds, 1.59 and the Rocks, 1.15 worms per chicken. This was the only group in which the Leghorns did not average the most worms. However, no difference between any two of the groups was significant. In the worm lengths the Leghorns had the longest (20.00 mm.) and the Reds the shortest (16.59 mm., or 3.41 mm. less than the former). The ratio was 4.61. The worms in the Rocks averaged 16.93 mm. long, 3.07 mm. less than the Leghorns, which was significant (Table 11).

Group E. The oldest group was 44 days old when parasitized and 65 days of age when killed. The worms in the Leghorns numbered 2.00; in the Reds, 1.36; and in the Rocks, 1.70 A. lineata per chick. The differences were not

significant. As was generally the case, the Leghorns had the most and the Reds the fewest worms.

The longest worms were found in the Leghorns (21.45 mm.) and the shortest ones in the Reds (17.58 mm.). The difference of 3.87 mm. was significant, as it was 4.03 times the probable error of the difference. The worms in the Rocks averaged 19.92 mm. in length, but this was not significantly different from the other worm lengths of either of the other breeds.

The general conclusion for all groups of chickens is that the Rhode Island Reds are the most resistant to the growth and viability of A. lineata; the White Plymouth Rocks rank next, and the White Leghorns are the least resistant of the three breeds studied. Table 11 shows that during any of the ages studied this same principle holds true in general.

EVIDENCE OF AGE RESISTANCE

In order to ascertain whether or not the resistance of the chickens to A. lineata increased with age the individuals of each breed of chickens were grouped together and the differences in worms found between Groups A, B, C, D and E. When examined for A. lineata each successive group of chickens was about one week older than the previous group.

As shown in Table 12, the youngest White Leghorns (Group A) examined when five weeks old harbored an average of 8.23 worms, whereas the oldest group (E) had 2.00 worms per chicken. Intervening groups of White Leghorns showed a progressive decrease in worm numbers excepting Group D which was 1.22 or 0.88 worm less than in Group E; however, the difference was not significant. The average number of A. lineata found in the Rhode Island Reds decreased in constant amounts from 4.89 to 1.36 worms per chicken during the five weeks period. In the White Plymouth Rocks the worms decreased irregularly from 7.08 in the youngest group to 1.70 in the oldest group. Between Groups A and B there was a significant decrease in worm numbers, also, between Groups C and D. Those that were irregular, however, did not have significant differences. Summarizing, the White Leghorns examined at 33 days of age had 8.23 worms per chicken, and those at 65 days old, 2.00 worms; the Rhode Island Reds, of the same ages, averaged 4.89 and 1.36 A. lineata; and the White Plymouth Rocks, 7.08 and 1.70 worms per chicken.

These results point directly to age resistance. The resistance of Leghorns increased with greater rapidity than did that of the Reds, which at the beginning were more resistant than the Leghorns. Although the Reds were rela-

Table 12. Summary of results from the Groups arranged in successive order to determine the possibility of age resistance in each breed of chickens.

	Age when parasitized (days)	Mean number worms	Dif.	Ratio D / P.E.D.	Mean length worms	Dif.	Ratio D / P.E.D.
White Leghorns							
Group A	12	8.28			22.08		
			0.94	0.83		3.89	10.46
Group B	19	7.34			18.19		
			1.61	1.85		5.64	12.84
Group C	30	5.73			23.83		
			4.51	6.61		3.83	5.67
Group D	33	1.22			20.00		
			0.88	2.54		1.45	1.88
Group E	44	2.00			21.45		
Rhode Island Reds							
Group A	12	4.89			17.66		
			1.28	1.65		3.20	6.31
Group B	19	3.61			14.46		
			0.77	1.19		5.72	10.04
Group C	30	2.84			20.18		
			1.26	2.76		3.59	5.83
Group D	33	1.58			16.59		
			0.22	0.52		0.99	1.05
Group E	44	1.36			17.58		
White Plymouth Rocks							
Group A	12	7.08			20.34		
			4.64	4.80		6.97	7.24
Group B	19	2.24			13.37		
			0.95	1.78		7.09	8.02
Group C	30	3.39			20.46		
			2.24	4.98		3.53	5.41
Group D	33	1.15			16.93		
			0.55	0.99		2.99	4.22
Group E	44	1.70			19.92		

tively resistant when young, their resistance did not increase in the same proportion as did that of the Leghorns. The White Plymouth Rocks increased their resistance in about the same proportion as the Leghorns, but they were more resistant at the beginning. When nine weeks old (Group E) the resistance to the A. lineata was about equal in all three breeds of chickens.

Whereas, the numbers of worms clearly demonstrate age resistance in the chickens, it is obscured when the lengths of the worms are considered. The worm lengths in the Leghorns averaged 22.00 mm. in Group A and 18.19 mm. in Group B. Group C followed with an increased average length of 23.83 mm. Group D Leghorns had worms 20.00 mm. and Group E 21.45 mm. long. The A. lineata from the Reds and Rocks had this same sort of variation (Table 12).

There were significant decreases in the lengths of worms between Groups A and B and between Groups C and D in all three breeds of chickens. The average worm lengths in Groups A and E chickens were nearly the same despite the age. It is to be noted that the irregularity of worm lengths is constant. The worm lengths in the chickens of Groups C and E seem to be the cause. These two groups of chickens were used during the first year of the work, while Groups A, B and D were used this year. In this way differ-

ent strains of chickens or worms would become a factor. In fact, it was disclosed that the hatchery, from which these chickens had come, had used eggs from two different flocks of chickens.

It may be that strains of chickens or strains of A. lineata were sufficiently different to cause this variation. Sandground (1929) states that as a result of recent researches with Ancylostoma caninum it had been shown that a species is comprised of strains better adapted to one species of host than to another, and it has been postulated that a higher degree of age resistance is exhibited in the host parasitized with a foreign strain of nematodes. This theory might explain the situation under consideration. Those worms in chicken Groups B and D may have been normal strains while those strains of worms in Groups A, C and E were foreign. The idea is quite plausible. Perhaps if all foreign strains had been used there would have been a consistently graded decrease in the lengths of worms as the hosts grew older. Or if all were normal strains the worm lengths might have proved identical. The irregularity in the acquisition of age resistance to the worms may have been due to one of these factors or to others.

Figures 1 to 5 illustrate the comparative growth rates of the three breeds of chickens in each experiment. On the

whole the White Leghorns were the heaviest during the first one or two weeks of age. After that time the Reds and the White Rocks gained rapidly in weight until one or the other proved the heavier. The Leghorns usually lagged behind. No interesting changes occurred after parasitizing nor at any other period. Perhaps this was due to the light infestations of A. lineata.

DISCUSSION

The experiments described apparently prove that breeds of chickens have varying degrees of resistance to A. lineata. In the three breeds studied the Rhode Island Reds were the most resistant, the White Plymouth Rocks next and the White Leghorns were the least resistant to the viability and growth of the nematodes.

That variations in race resistance occur was shown by Cort and his coworkers (1929) who found that Indian-Negro mixtures were quite resistant to hookworm infestation, while Indian-White mixtures had greater susceptibility. In 1932 Chandler reported that an African suffers less from a given degree of hookworm infestation than does a white man.

In discussing resistance, Chandler (1932) stated that factors affecting resistance and susceptibility are age,

sex, pregnancy, chronic diseases, exposure, species and race of a host. He stated, also, that different species of animals in which a parasite can reach maturity are by no means equally affected by comparable grades of infection. Just as this is true of species, it, apparently is true of breeds. Variations in resistance occur in all three breeds of chickens studied. The younger the chickens, the greater the differences among the breeds. This work supports Herrick's finding (1926) that young chickens are exceedingly susceptible to parasitism. As the chickens grow older, worms are eliminated and finally the hosts are almost totally resistant. Each breed was found to have this same phenomenon in different degrees; as the chickens became older, the A. lineata had more difficulty in maintaining an existence. When about nine weeks of age, the amount of parasitism was nearly the same in all three breeds of chickens.

It is possible that the presence of A. lineata in the intestine causes the body to secrete a substance for combating the worms, thus producing an acquired resistance. If this is true, the antigenic power of the worm proteins must be found and the antibodies determined by intricate tests which are not in use at present. On the other hand, Taliaferro (1934) states that macrophages are prominent in

active immunity. The same cells are involved, in varying degrees, whether the animal be responding to a local or a general invasion by protozoa, helminths, bacteria or other infectious agents. Chandler (1930) contends that parasitic infections are quantitative in nature; that the severity of infection is not controlled almost entirely by the resistance of the host, but very largely by the actual number of worms present.

In 1931 McCoy, who studied the immunity reactions of dogs against A. caninum, inferred that the first dose causes an acquired resistance, because the second dose never affects the animal as badly as the first. This idea is in opposition to Taylor's (1928) who noted that infections from Starling gapeworms in chickens causes light infection the first time, but a second infection will bring about a heavy infestation. This may be due to the abnormal host, or it may be due to the refractibility of chickens to antibody formation. The dog is the normal host of A. caninum, also, the dog is not considered refractory and is able to produce antibodies.

As already noted, age resistance was demonstrated in worm numbers, but was poorly demonstrated with worm lengths in any of the three breeds of chickens. According to Sandground (1929) age resistance depends upon the presence

of foreign strains of host or parasite. He observed, also, that foreign species produce different results. For example, normal hosts of Ancylostoma duodenale and Syngamus trachea, respectively, man and turkey, exhibit no appreciable age resistance, while in the respective abnormal hosts, dog and chickens, age resistance was demonstrated, that is, the abnormal hosts were more resistant. In another paper Sandground (1928) stated that age resistance is seldom found without a background of partial abnormality between host species and parasitic species involved. Abnormality is due to foreign strains. As stated in the experimental data this is the only explanation offered now for the variation in the length of worms shown in Table 12. Groups C and E were studied in 1932-1933, while Groups A, B and D were observed during 1933 and 1934. This gave an excellent chance for foreign strains to appear.

In conjunction with Sandground's theory of foreign strains of parasites resulting in marked age resistance, while normal strains do not cause any age resistance, Ransom (1921) found that chickens, an abnormal host, were very susceptible to Syngamus trachea and produce marked age resistance; whereas turkeys, the normal host, are not affected and produce no age resistance. Ackert and Eisenbrandt (in press) when they used A. lineata rather than

S. trachea, found that turkeys, the abnormal host in this case, are much more resistant to A. lineata than are Leghorn chickens, the normal host.

Such an explanation might be made for breed resistance. Perhaps Leghorns are the original hosts of A. lineata and the other breeds are foreign breeds. Hence, the Rhode Island Reds would be more resistant than the White Leghorns. The more probable explanation is that the Reds have some factor that makes them resistant even at an early age. As the chickens grow older this substance increases to a certain extent thereby making existence of the worms more unfavorable. The Leghorns do not have as much of this substance present in their bodies thus providing a more favorable habitat for the worms. As the Leghorns age the factor intensifies more rapidly than in the Rocks or the Reds, so that by nine weeks of age they are almost as resistant as the other breeds of chickens.

It is interesting to note that while the young Leghorns are not very resistant to A. lineata, they are exceedingly resistant to general conditions. In all of the experiments it was rare for a Leghorn chick to die, while among the other breeds deaths were of common occurrence.

SUMMARY

1. Studies on the comparative resistance of White Leghorn, White Plymouth Rock and Rhode Island Red chickens to the nematode Ascaridia lineata (Schneider) were conducted during a period of two years. Five experiments involving 727 chickens were carried out in groups as follows: Group A, parasitized at 12 days of age; Group B, at 19 days; Group C, at 30 days; Group D, at 33 days; and Group E, at 44 days of age.

2. Twenty-one days after parasitizing the chickens were killed and the worms collected, counted and measured. The average numbers and lengths of the worms were used as criteria for comparing the resistance of the different breeds of chickens.

3. On analyzing the data biometrically, it was found that the Rhode Island Reds were the most resistant, the White Plymouth Rocks next and the White Leghorns the least resistant to the A. lineata.

4. Evidences of age resistance in each breed were demonstrated by the numbers of worms. The lengths of the worms, however, did not indicate a consistent acquisition of resistance although it was notable among some groups.

The variations may have been due to the use of foreign and normal strains of either chickens or nematodes.

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