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To the Graduate Council:

I am submitting herewith a thesis written by Denis Cecil De Loach entitled "Investigation of the poisoning of cattle consuming fescue grass." I have examined the final electronic copy of this thesis for form and content and recommend that it be accepted in partial fulfillment of the requirements for the degree of Master of Science, with a major in Animal Husbandry.

Charles S. Hobbs, Major Professor

We have read this thesis and recommend its acceptance:

M. C. Bell, George M. Merriman

Accepted for the Council: Carolyn R. Hodges

Vice Provost and Dean of the Graduate School

(Original signatures are on file with official student records.)

July 15, 1954

To the Graduate Council:

I am submitting herewith a thesis written by Denis Cecil DeLoach entitled "Investigation of the Poisoning of Cattle Consuming Fescue Grass." I recommend that it be accepted for nine quarter hours of credit in partial fulfillment of the requirements for the degree of Master of Science, with a major in Animal Husbandry.

Charles & Hoffy) Major Professor

We have read this thesis and recommend its acceptance:

M. C. Bell S. M. Merrinon

Accepted for the Council:

Dean of the Graduate School

INVESTIGATION OF THE FOISONING OF CATTLE CONSUMING FESCUE GRASS

230

A THESIS

Submitted to The Graduate Council of The University of Tennessee in Partial Fulfillment of the Requirements for the degree of Master of Science

by

Denis Cecil DeLoach

Sauth Thirty Attend

August 1954

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Denis Cecil DeLoach

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CHAPTER I

INTRODUCTION

Experimental results and field observations in the United States, New Zealand, and Australia indicate that fescue contains a toxic agent that is detrimental to cattle of both beef and dairy breeds.

Tall fescue grass (Festuca arundinacea) has been recommended and used extensively as permanent pasture in Tennessee, Kentucky, and other states for the past several years. The acreage of fescue grass, in spite of the possible detrimental effects, is still increasing.

Observations at the Middle Tennessee Experiment Station, at Alcoa, and possibly at Calderwood, as well as correspondence with other stations, indicate that the disease now known as fescue poisoning may be a serious problem to some Tennessee cattlemen and dairy farmers.

The fescue problem in livestock feeding is one of increasing importance. Fescue poisoning could develop into a problem of major importance.

It is significant that fescue lameness has been noted on farms where there is a heavy soil or a soil heavily fertilized.

Symptoms of fescue poisoning in cattle are: loss of condition, rough or unshed hair coat, humped back, loss of coordination of the hind limbs, and loss of hair on the hind limbs on an area extending from the Coronary band about one-half of the way to the hock joint. In this same area, inflammation of the skin and a general swelling of the tissues are sometimes noted. In advanced stages of the disease, there may or may not be severe lesions about the hoof, which makes it very difficult for the animal to stand. Symptoms of this condition are not consistent.

In view of these facts, an experiment was set up in an attempt to experimentally reproduce the disease and to study the clinical and hematological pictures in this disease.

CHAPTER II

REVIEW OF LITERATURE

Only limited information appears in the literature concerning the clinical, hematological, and post-mortem pictures in this disease. A few workers, however, have reported their experience and work with fescue as a pasture grass.

Although there are several species of fescue, they, of course, are fairly closely related. Fescue poisoning of cattle has been reported from pastures of the following species: Alta fescue, Kentucky 31 fescue, and Reed fescue. According to some workers, meadow fescue and Reed or Alta fescue integrate somewhat in some areas. Therefore, since most fescues have been shown to be closely related, and many of them have been shown to cause poisoning, hereafter, the word fescue will be used in place of a list of those fescues actually reported to have caused poisoning.

At the present time, no one knows the cause of this condition called "fescue lameness." Some workers believe it to be ergot; others believe fescue leaves contain some substances which act like ergot alkaloids. Fescue poisoning could be confused with foot rot, injury to feet or legs, a low plane of nutrition, chronic anaplasmosis and possibly other conditions. In all of the reports of fescue poisoning, there is a conspicuous absence of post-mortem findings.

Cunningham (1949) of New Zealand reported that some of the cattle pastured on tall fescue grass, where there was little other vegetation, developed a condition commonly known as "fescue foot." He described this condition as starting with extreme lameness followed by a progressive necrosis of one or both of the hind limbs. He further compared these symptoms with those of chronic ergot poisoning, but pointed out that ergot poisoning is usually seasonal. By feeding fescue which contained no seed heads, these workers were able to reproduce the disease in cattle. These workers stated that horses are not susceptible to this condition. There are probably species differences in the reaction to "fescue poisoning." These results suggest that there is present in fescue grass leaves or stems some substance which acts like ergot alkaloids in causing vasoconstriction and dry gangrene in the extremities of the limb. The difference between the reactions of different individuals may be due to differences in susceptibility. Cunningham further stated, similar differences have been noted in the field since it is rare for all in any group of cattle to be affected. No information has yet been obtained as to the nature of the toxic substances in tall fescue. Attempts to induce lesions in laboratory animals by feeding the dried fescue as part of the ration have so far proved unsuccessful and a ready test to control extraction and isolation of the toxic principle is therefore not yet available. It has been suggested that tall fescue grass should be classified as a toxic plant and not as a plant which acquired toxicity through infestation by Claviceps purpurea. Such a difference in classification entails a different approach to fescue, which, as a poison plant, should be regarded as an even less desirable fodder for cattle than it is at present. (Cunningham, 1949).

Brentzel (1947) reported that <u>Claviceps purpurea</u> (ergot fungus) is a parasite (Ascomycete) producing disease of grass, especially rye grass. This organism is of some importance in medicine since it produces in the infected grain a substance which produces contractions of involuntary muscles. It has produced poisoning both in man and in domestic animals and caused abortion by inducing contraction of the uterus and gangrene by causing constriction of the blood vessels.

The fungus grows in the seeds which, as they develop, become much larger than healthy grains. The tissue of the grain is gradually replaced by mycelia. As the grain ripens, the tissue of infected seeds is completely replaced by mycelia, which dries out to a dense, hard, black mass, the sclerotium.

The sclerotium retains the general form of the seed but is larger and projects considerably from the head of grain. It is these ergot grains which contain the poison.

Ergot grains fall to the ground and remain dormant over the winter months. In the spring the ergot sprouts a number of stalks which develop rounded masses of tissue containing asci when mature. These asci are forcibly discharged into the air by the wind. If they fall on grain, they germinate, forming mycelia which invade the grain, and start the cycle over again. Some of the most common grasses affected by this organism are red top, Kentucky bluegrass, and English rye grass.

Atansoff (1920) found 2700 sclerotia in a one-pound sample of fescue seed. There are approximately 227,000 fescue seeds in a onepound sample, so sclerotia composed 1.2 per cent of this sample.

Atansoff further stated that ergot was first used as a drug in 1582, to induce uterine contraction. There is reason to believe that sclerotia from all species of <u>Claviceps</u> are poisonous. There have been reports from many parts of the world on the poisonous effects of ergotized pastures and ergotized hay when consumed by domestic animals. The fungi involved are mainly <u>Claviceps purpures</u> and <u>Claviceps paspal</u>. Ergotism may appear in many forms which seemingly have no relationship to each other. An explanation for this might be that ergot is composed of many constituents which act differently. The two common types are the gangrenous and the nervous.

The gangrene affects those parts poorest in blood supply, the extremities, tail, and ears. The lesions may vary from simple sores around the top of the hoof, upon the teats, to loosening of the sole or wall or sloughing of an entire limb. If the entire limb sloughs, there is usually a circular line of demarcation between the healthy and gangrenous area. In man, the sloughing usually appears first upon the fingers and toes. The gangrene may be dry or moist and does not differ essentially from other gangrene. Horses, pigs, cattle, and poultry are susceptible to gangrene caused by ergot.

The nervous or spasmodic form is ushered in by anosthesia or hyperthesia of the skin. Both may exist in different parts of the body at the same time. These conditions begin at the extremities and spread toward the main part of the body. This indicates that they may be due to circulatory changes. There are also some disturbances of sensation in the digestive tract. There may be a craving for food and a lack of

appetite at the same time; digestion is impaired; vomiting and diarrhea are common. The involvement of the central sensory apparatus is shown by the loss of special senses. The motor area later begins to be affected, causing twitching, tremors, convulsions, and painful contractions of muscles. Animals frequently die from convulsions.

The similarity between fescue poisoning and ergot poisoning is obvious from previous descriptions, and this similarity has led to the assumption that poisoning by tall fescue grass is due to eating ergot sclerotia with the fescue. The New Zealand workers point out several reasons why they believe that poisoning by ergot cannot be associated with fescue poisoning as it appears in the field. First, ergot occurs seasonally while fescue poisoning exhibits no seasonal effect. In Australia. if only sclerotia are toxic, ergot poisoning can occur only from about January to March; if all stages of the life cycle of Claviceps purpures are toxic, the extreme period of ergot poisoning should be between November and March. Second, it is virtually impossible for grazing cattle to pick up from the ground enough ergot sclerotia to cause lameness and necrosis of the limbs. Cunningham (1949) has shown in feeding experiments with cattle that approximately 100 grams of sclerotia must be fed each day for about ten days in any ration to produce lameness and necrosis. He further stated that in these particular paddocks it would be practically impossible to pick from the ground the quantity necessary to produce ergotism. The most conclusive proof was exhibited when fescue hay containing no seed heads or sclerotia produced typical fescue lameness. (Cunningham, 1949).

The Australian workers state another reason why they believe it is not ergot that caused this condition. During the spring of 1946, the fescue was extensively damaged by insects so that very few seed heads were formed. This, however, had no apparent effect on the toxicity of the grass.

In the spring of 1953, cattle at the Middle Tennessee Experiment Station were showing definite symptoms of "fescue lameness" while grazing fescue grass and Ladino clover pasture. Associate Professor J. K. Underwood of the University of Tennessee Agronomy Department visited the station and observed these pastures very closely. He found no sclerotia on the fescue grass and took samples for microscopic study. Upon microscopic examination of the fescue, he found no sclerotia.

At the present, the only prevention seems to be to remove animals from fescue pastures before advanced symptoms appear. The New Zealand workers recommend that cattle be restricted to fescue grazing to periods of one week at a time.

Pulsford (1950) reported that observations in South Australia support the conditions as described by Cunningham (1949). The Australian workers observed the symptoms of lameness within 3 to 10 days after cattle were placed on fescue and the cattle recovered within 7 to 10 days if moved to a different pasture when they were showing the early symptoms. Additional symptoms observed were rapid loss of condition, puffiness around the eyes, the animals becoming hidebound with a clear line of demarcation noted in the area of the fetlock joint, which is usually the point of sloughing of the hoof. They further found no

abortion and no sloughing of extremities other than the hind limbs. They had also noted that sheep scoured on these affected pastures. When this condition was first noticed, it was described by the local stockman as being due to the ergot (<u>Claviceps purpurea</u>) which is present in all stands of <u>Festuce arundinacea</u> examined in this district. The assumption is readily made from the similarity of the symptoms in each case. Unlike the condition as reported by Cunningham, there is in South Australia a very marked seasonal incidence, the condition being prevalent from May to October, but rarely seen during the summer. Most cases occur and symptoms appear more quickly during the period when this grass is making its most rapid vegetative growth. Cattle which have not had previous access to the fescue are affected more rapidly than those which are accustomed to it.

Goodman (1952) reported that similar conditions have been noted by cattlemen in Colorado for the past thirty years. The lameness noted by these cattlemen had not been associated with fescue pastures until after the report of New Zealand work by Cunningham (1949). The Colorado report by Goodman indicated that the worst outbreak of the fescue poisoning occurred during the winter of 1948-49 and in 1951-52 when they found sloughing of the hoof of one or both hind feet and occasionally the loss of the tail and tip of the ears in addition to the lameness. He further stated, "They have endeavored to associate ergot with this trouble since it was first reported some 50 years ago, but we have never been able to find any ergot in the flowering portion of the plant in the two areas in Colorado where this disease has appeared." These workers stated that

their experience with fescue lameness in Colorado is very similar to that reported by Dr. N. F. Pulsford in Australia, in that the trouble seems to be reasonable lameness as a rule, not developing until cold weather sets in. Goodman further stated, "Certainly we have the conditions which have been called 'fescue foot', and in some pastures it is extremely severe." Cattle affected with fescue poisoning appear to be highly nervous; some of them develop diarrhea; and many of the cows following calving develop a prolapse of the uterus. "Our present questions are: Is the fescue which was supposed to be Alta fescue and Kentucky 31 actually these varieties? or Is contamination with another fescue responsible for the difficulty?"

Garrigus (1953) at Kentucky has stated through personal communication that poisoning of cattle pastured on fescue has been encountered in Kentucky, Florida, Mississippi, South Dakota, Missouri, and Tennessee in addition to those reported in Colorado, New Zealand, and Australia. There has been considerable variation in the symptoms reported by the various workers who have noted the condition of "fescue poisoning." "Some of this information is contradictory. Apparently more than a normal amount of lameness has occurred with cattle grazed on fescue. Some of this resembles ergotism in that a dry gangrene and later sloughing off of the feet, tail, ears, or other extremities often occurs." He further stated, "This condition is more apparent in older stands of fescue than in newer stands. This may be due to an accumulation of infection or to a reduction in the percentage of legumes or other grasses in the stand." Early symptoms which he reported were incoordi-

nation and stiffness, rather than swelling or inflammation of the feet. In later stages, a ring-like scab will develop above the dewolaws and completely around the cannon bone. Dry gangrene and complete sloughing off of the foot will soon follow.

Davis (1953) stated that the two conditions, fescue foot and ergotism, certainly have a great deal in common. In the case of development on peat soils, there was no chance of its being ergot; first there were no seed heads, and second, the time of the year was such as to rule against the development of ergot in any form. Davis further stated, "The condition as developed in Florida, developed rapidly within three weeks of the time the animals were placed on the pasture. The first symptom was lameness. Upon close examination, the hoofs were noticed to have cracked and the skin above the hoofs was roughened, sore, and gave the appearance of something like photosensitization. Animals that were left for any length of time after lameness developed showed a sloughing of the hoofs and in some severe cases the entire leg below the knee was raw, sore, and practically down to the bone. "These animals stopped eating. and there was a tremendous loss of weight, condition, and, in the case of severely affected animals, death apparently resulted from the overall anorexia." It was noticed that apparently most of the keratinized tissues were affected because the hair of the switch fell out, and, in some severe cases, it was noticed that hair on other parts of the body also fell out. They further stated that if cattle were removed from the pasture immediately and placed on another feed at the first sign of lameness, they would usually recover in a period of from a week

to a month. Animals that did not develop the condition within the first three weeks apparently never did.

Stearns (1953) reported that Kentucky 31 fescue seemed toxic during the drought while most of the other pasture forages such as orchard grass and ladino clover had died out. He further stated, "It does not grow as tall as the Reed variety and is one of the species that is not supposed to cause trouble. Where there has been mixed grasses for grazing, no trouble has been reported."

CHAPTER III

EXPERIMENTAL PROCEDURE

Beef Cattle

The cattle used in this experiment were three pregnant grade Hereford cows purchased locally in February, 1954. These animals tested were negative for tuberculosis and brucellosis. Prophylactic Brucella vaccine was administered to all animals after arrival. These cattle were not on fescue pasture prior to their purchase for this experiment.

The experiment was conducted from February 22, 1954 to June 6, 1954 at the Tennessee Agricultural Experiment Station at Knoxville. Upon arrival, these cattle were placed in individual box stalls and preliminary work was done on them from February 5, 1954 to February 20, 1954. Good quality mixed hay containing at least fifty per cent alfalfa was fed <u>ad lib</u>. The hay was chopped to promote uniform consumption and to facilitate easier weighing and feeding. These animals were fed for approximately two weeks pre-experimentally in order to get them on feed and to accustom them to individual feeding facilities. During this pre-experimental period, the animals were fed a mineral mixture containing 50 per cent salt, 25 per cent steamed bone meal, and 25 per cent dicalcium phosphate and had free access to water. Chopped mixed hay was fed and gradually increased in amount until they were eating approximately twenty-two pounds of hay at the time the experiment began.

During the pre-experimental period, the cattle were sprayed with DDT to rid them of lice. Fecal samples were taken and found to be negative for worm eggs. The initial pre-experimental average weight was 889 pounds. Pregnancy tests were run on these cattle and all three were heavy with calves. These cows were in fair condition at the beginning of the pre-experimental work. Analysis of the mixed hay fed during the pre-experimental period and the fescue hay fed during the experimental period is shown in Table I. Blood samples were taken three times during

TABLE I

PERCENTAGE COMPOSITION OF HAY FED CATTLE DURING PRE-EXPERIMENTAL PERIOD AND EXPERIMENTAL PERIOD

Noteturo	Witnessen	Protoin	Ach	Ether	Crude	Nitrogen Free
Dan Dan and	manitel Denie	4			12001	LAUGUV
tre-expert	mental rerio	r G				
10.86	1.14	7.12	6.63	1.61	39.94	36.84
Experiment	al Period					
10.09	0.72	4.50	5.37	1.15	39.26	39.63

the pre-experimental period.

The cattle were taken off feed completely for a 48-hour period between the pre-experimental period and the experimental study to encourage consumption of the fescue hay which was of very poor quality.

At the beginning of the experimental period, the average weight of the cows was 908 pounds. The animals were fed ad lib baled fescue

hay obtained from the Middle Tennessee Experiment Station, Columbia. This hay came from a field on which fescue poisoning had been encountered during the season which it was cut. These cattle had free access to water and to a mineral mixture containing 50 per cent salt, 25 per cent steamed bone meal, and 25 per cent dicalcium phosphate. They were fed individually in compartmented feed racks so as to feed the mineral mix and the hay separately and to avoid waste. The animals were fed once daily. The hay for each animal was weighed out into a burlap bag or tub and placed in the feed rack in the evening and weighbacks were taken the following evening. The cattle were confined in the individual box stables at all times. The amount of hay offered was adjusted when necessary to provide <u>ad lib</u> consumption. Hay samples were taken as the hay was fed to make a composite sample for analysis at the end of the experiment, as shown in Table I.

Wood shavings were used for bedding to prevent cows from eating any feed other than fescue hay.

These cattle were weighed every 28 days. Blood samples were taken weekly for the determination of hemoglobin, hematocrit, sedimentation, red and white cell counts, and differentials. The Wright's Stain method, as described by Parker (1948), was used for the determination of the differential count. The red cell count was determined by the Haymes solution method, as described by Farker (1948), and the sodium eitrate method, as described by Gradwohl (1948). Glacial acetic acid method was used in the determination of the white cell count, as described by Parker (1948). The hemoglobin was determined by the Evelyn's

Colorimeter method, (1948). The hematocrit was determined by the Wintrobe method as described by Kolmer (1943).

The cattle were observed regularly for visible symptoms of "fescue poisoning."

These cattle were weighed at the end of this phase of the experiment and then they were moved to the Middle Tennessee Experiment Station on June 9, 1954, ending this phase of the experiment.

An analysis of the fescue stems taken from the Middle Tennessee Experiment Station, in the spring of 1953, from different areas within the field, where cattle had reportedly suffered from fescue lameness, was obtained and is shown in Table II. This analysis was determined by K. B. Sanders, Associate General Chemist, University of Tennessee.

TABLE II

Sample	N	itrogen					
Description	Total	a	Ъ	SiO2	P	Ca	K
Dry Area	0.86	0.66	0.20	1.46	0.21	0.21	1.92
High Area	1.00	0.79	0.21	1.93	0.26	0.44	1.29
High Area	1.27	1.00	0.27	1.43	0.17	0.41	1.84
Low Area	1.50	1.08	0.42	2.45	0.44	0.45	2.48
Low Area	1.41	1.10	0.31	2.11	0.37	0.46	1.97

PERCENTAGE COMPOSITION OF SAMPLES* OF FESCUE STEMS FROM THE MIDDLE TENNESSEE EXPERIMENT STATION WHERE BEEF CATTLE REPORTEDLY SUFFERED FROM "FESCUE FOOT"

These data are reported on moisture free basis. *Samples were taken from areas within the field with different elevation and fertility. ^aProtein nitrogen.

bNon-protein nitrogen.

The Cock's Comb Test

The second phase of this experiment was with six young White Leghorn cockerels, with an average weight of 3.8 pounds.

Samples of fescue pasture taken as a representative sample from pasture where difficulties had been encountered at Columbia Experiment Station and fescue hay that was fed at the University of Tennessee Experiment Station were dried and finely ground in a Wiley mill. A sample of 454 grams of this material was extracted with a combination of chemicals following a modification of extraction procedure as outlined in the Merck Index (1953). One-pound samples were extracted using four different combinations of chemicals as listed below:

Number	One	700 ml. of methanol 1500 ml. of acetone
		3000 ml. of alcohol (ethyl) 15 ml. of NaOH
Number	Two	700 ml. of methanol 1500 ml. of acetone 3000 ml. of alcohol (ethyl) 20 ml. of HCl
Number	Three	5000 ml. of H20 15 ml. of HC1
Number	Four	The extract from the Number Two sa

This extraction progressed for eighteen hours in a round bottom six liter flask. Mixing was accomplished by frequent agitation. Separation of the extract was obtained by first pouring the mixture through a cheese cloth, then by filtering it through a filter paper with suction. Final concentration was obtained by evaporation on a water bath at 80° C. The final extract was a greenish-black, viscous, oily mass of about 25

sample

grams. The extract was defatted using a continuous extraction with petroleum ether. This final extract was dissolved in 1 part H₂O, 1 part 50 per cent alcohol, and 2 ml. of HC1.

Different levels of this solution, from the final extract, using the four different chemical combinations, was injected into the cockerels, following the cock's comb test, using a negative control group and a positive control with a solution of an ergot compound. Following the injections, the cockerels were closely observed for reactions and visible symptoms of cyanosis. The level of the extract injected in cockerels ranged from 2 cc. to 8 cc.

The cockerels later were injected with different levels of a 50 per cent alcohol to determine the level required to produce symptoms of drunkenness or to get the same symptoms noted from the injection of fescue extract.

Albino Rats

The third phase of this experiment was with albino rats following exactly the same procedure and using the same samples of fescue extract, except that the level of injection was much lower.

These investigations with rats and cockerels were of an exploratory nature in an attempt to determine the effect of fescue extracts on laboratory animals and to compare these with those obtained for biological tests for ergot alkaloids.

CHAPTER IV

RESULTS AND DISCUSSION

Beef Cattle Phase

At the time the experiment started, each animal was consuming approximately fourteen pounds of fescue hay per day. Three months and twelve days later, their feed consumption had increased to approximately twenty-five pounds. Table III shows the average daily feed consumption and gains during the pre-experimental period.

TABLE III

PRE-EXPERIMENTAL GAINS AND AVERAGE DAILY FEED CONSUMPTION BY COWS FED MIXED HAY

			Pounds	
Cow	Number	Hay	Average Daily Gain	Total Gain
	1a	20.43	1.31	21
	1b	20.37	1.13	18
	10	19.50	1.06	17

February 5, 1954 - February 20, 1954

Table IV shows the average daily gain, the average daily feed consumption, and total gain during the experimental period by 28-day periods.

Table IV shows that average daily feed consumption increased by

AVERAGE DAILY FEED CONSUMPTION, AVERAGE DAILY GAIN AND TOTAL GAIN DURING EXPERIMENTAL PERIOD

February 22, 1954 - June 6, 1954

	445	
Pounds		
in		
Gain		0
and		
Period		
28-Day	1	Carrow
Per		
Consumed	2	
Feed	1	
	1	
30	1999	

**

#1

		•											
COW	Hay	Gain*	Gain**	Hay	Gain*	Gain**	Нау	Gain*	Gain**	Hay	Gain*	Gain**	Gain**
le	16.05	-0.79	-22	19.84	-2.14	-60	24.66	-2.18	19-	25.39	-2.17	-39	-182
Tb	18.13	-0.46	-13	21.56	-0.11	60	21.61	-0.11	1	21.50	-0.22	4	- 23
Ic	13.20	-0.46	-13	14.55	וויו-	-31	16.95	10.07	61 1	17.22	0.17	89	- 39

*Average deily gain. **Total gain by 28-day periods. ***Total gain for all four periods. ****Period Four was only 18 days.

28 day periods while the average daily gain was a minus figure. These cattle began to show a loss of weight in a short time after the experiment started. The total weight loss for animal la, lb, and le were 182 pounds, 23 pounds, and 44 pounds, respectively.

On March 2, 1954, scouring occurred in all three cows and became very severe for six or seven days. This scouring condition cleared up completely in two weeks.

On March 6, 1954, cow la gave birth to a 65 pound calf and this calf appeared to be healthy and strong at birth. This calf seemed to be nursing almost continuously, which indicated that the cow was giving very little milk. When this phase of the experiment was finished and the cattle were moved to the Middle Tennessee Experiment Station, the calf had gained only 19 pounds. On April 2, 1954, cow la seemed to have a slight limp in the left rear foot and the cow lb appeared to be pale about the eye lids and udder and had a very slight limp of the right rear foot. These symptoms were confirmed by Dr. G. M. Merriman.

As the experiment progressed, this lameness became more pronounced in cows la and lb and slight in lc. These cattle showed a slight but definite loss of coordination of the hind limbs, loss of weight, and a rough hair coat. The anemic appearance was visible only in cow lb. The symptome listed above were apparent throughout the experiment after April 2, 1954. The analysis of cattle blocd during the pre-experimental and the experimental periods is shown in Table V. Normal ranges of blood cellular elements, as described by Coffin (1953), and the average blood cellular elements of cattle used in this experiment during the

ANALYSIS OF CATTLE BLOOD DURING PRE-EXPERIMENTAL AND EXPERIMENTAL PERIODS

Cow la

			Mean			Diff (per	eren r ce	tials nt)			
Sample Date	Mean Corpus- cular Volume	Mean Corpus- cular Hemo- globin	Corpus- cular Hemo- globin Concen- tration	Mature Neutrophils	Inmature Weutrophils	wmphocytes	Myelocytes	<i>lionocytes</i>	Sosinophils	Basophi ls	Basket cells
Pre-Exp	erimental	Period									
2-10-54	46.71	15.30	32.76	28	0	52	0	13	7	0	0
2-15-54	52.74	17.63	33.43	11	0	62	0	17	10	0	0
2-22-54	51.99	16.53	31.80	25	0	59	0	11	5	0	0
Average	50,48	16.49	32.66	21.3	0	577	0	13.7	7.3	0	0
Experim	ental Per	iod									
3- 2-54	55.36	18.12	32.73	23	0	55	0	18	4	0	0
3- 6-54	(calf born	1)	1. 1. 1.				1999			
3- 8-54	56.47	18.26	32.34	-	-	-		-	-	-	-
3-15-54	51.09	17.47	34.19	-	-		-	-	-	-	-
3-22-54	46.07	14.73	31.98	22	0	51	0	11	15	0	0
3-30-54	53.18	17.24	32.41	14	0	70	0	11	5	0	0
4- 6-54	53.22	17.24	32.39	9	0	42	0	36	13	0	0
4-13-54	53.15	17.37	32.67	22	0	67	0	7	3	1	0
4-20-54	44.53	14.88	33.41	22	0	64	0	9	5	0	0
4-27-54	53.30	16.24	30.47	28	0	53	0	12	5	0	2
5- 4-54	50.54	15.57	30.80	22	0	62	0	10	6	0	0
5-10-54	44.84	14.45	32.23	20	0	68	0	10	2	0	0
6- 3-54	50.09	17.23	34.40	19	0	70	0	9	2	0	0
Average	51.03	16.57	32.50	20,.1	0	60.2	0	13.3	6	.1	.2
Total A	verage*	1		oper-							
	50.88	16.55	32.51	20.4	0	59.6	0	13.4	6.3	.08	.15

ANALYSIS OF CATTLE BLOOD DURING PRE-EXPERIMENTAL AND EXPERIMENTAL PERIODS (continued)

Sample Date	Hemo- globin (gms/ 100 ml.)	Hemato- crit (% cells)	Sedimen- tation	White Blood Count	Red Blood Count (million)	Description of Red Cells
Pre-Exp	erimental	Period				
2-10-54	12.70	38.77	2.50	8,400	8.30	appear normal
2-15-54	12.87	38.50	2.25	6.800	7.30	slight anisocytosis
2-22-54	12.35	38.84	2.50	14,000	7.47	slight anisocytosis
Average	12.64	38.70	2,42	9,730	7.69	
Experim	ental Per	iod				
3- 2-54	12.54	38.31	2.25	8.200	6.92	R.B.C. crenated
3- 6-54	(calf hor	n)			
3- 8-54	13.26	41.00	2.00	9.900	7.26	
3-15-54	14.03	41.03	2.00	7.950	8.05	
8-22-54	13.26	41.46	2.00	8.750	9.00	elight anisocytosis
8-80-54	12.70	30.46	2.25	7.000	7.42	elight anisoavtosis
4- 6-54	12.79	39.49	2.50	8,150	7.42	slight anisocytosis,
	PROBANCE TO	10.12	and the second			slight microcytic
4-13-54	12.61	38.59	3.25	8,650	7.26	rouleau formation, slight microcytic
4-20-54	13.45	40.26	2.50	9,900	9.04	appear normal
4-27-54	11.92	39.12	3.00	8,250	7.34	slight microcytic
5- 4-54	11.74	38.11	3.00	7,850	7.54	slight microcytic
5-10-54	11.59	35.96	3.00	7,100	8.02	slight microcytic, target cells seen
6- 8-54	11.51	33.46	3.25	8,950	6.68	appear normal
Average	12.62	38.85	2.58	8,309	7.66	
Total A	verage*				n d same	
	12.63	38.82	2.55	8,717	7.67	

Cow la

ANALYSIS OF CATTLE BLOOD DURING PRE-EXPERIMENTAL AND EXPERIMENTAL PERIODS (continued)

			Mean	ŝ		Diffe (per	eren r ce	tials nt)			
Sample Date	Mean Corpus- cular Volume	Mean Corpus- cular Hemo- globin	Corpus- cular Hemo- globin Concen- tration	Mature Neutrophils	Immature Neutrophils	Lymphocytes	Myelooytes	Monocytes	Rosinophils	Basophils	Basket cells
Pre-Exp	erimental	Period									
2-10-54 2-15-54 2-22-54	63.39 60.71 51.08	20.06 20.39 16.00	31.65 33.59 31.32	39 24 14	010	35 48 56	000	15 20 21	11 7 9	000	000
Average	58.39	18.82	32.19	25.7	.3	46.3	0	18.7	9	0	0
Experim	ental Per	iod									
3- 2-54 3- 8-54 3-15-54 8-22-54	56.13 56.78 58.68	17.99 17.04 18.84 18.00	32.04 30.02 32.10 31.55	10	0 0	57	0 0	17	16	0110	0110
3-30-54 4- 6-54 4-13-54	48.74 43.96 52.27	15.59 14.01 15.34	31.99 31.87 29.35	22 20 17	000	51 58 56	000	20 15 16	7 6 10	011	000
4-20-54 4-27-54 5- 4-54 5-10-54	51.75 50.70 51.12 48.97	16.74 15.64 15.31 15.32	32.34 30.85 29.95 31.29	17 29 17 30	0000	61 60 71 56	0000	16 5 5	6265	0010	0400
6- 3-54	53.04	18.95	35.70	25	0	64	0 0	5	5	1	0
rverage	22.45 Toro cost	10.00	01.09	2019	0	01	0	7410	1.4.1	•**	***
TO CAT N	53.62	17.01	31.71	22	.08	54.5	0	15.2	7.9	.3	.3

Cow 1b

ANALYSIS OF CATTLE BLOOD DURING PRE-EXPERIMENTAL. AND EXPERIMENTAL PERIODS (continued)

Sample Date	Hemo- globin (gms/ 100 ml.)	Hemato- crit (% cells)	Sedimen- tation	White Blood Count	Red Blood Count (million)	Description of Red Cells
Pre-Exp	erimental	Period				
2-10-54	12.36	39.05	2.00	9,700	6.16	slight anisocytosis, slight achromia
2-15-54 2-22-54	12.87	38.31 38.31	2.00	6,650 5,000	6.31 7.50	slight anisocytosis slight anisocytosis
Average	12.41	38.56	1.92	7,117	6.66	
Experim	ental Per	lod				
3- 2-54	13.94	43.50	2.25	9,650	7.75	R.B.C. crenated*
3- 8-54	14.13	47.07	2.00	9,800	8.29	
3-15-54	12.96	40.37	2.00	8,750	6.88	
3-22-54	12.44	39.43	3.00	8,800	6.91	appear normal
3-30-54	12.10	37.82	1.50	14,100	7.76	slight anisocytosis
4- 6-54	10.76	33.76	5.00	7,400	768	moderate microcytic, bordering on moderate achromia
4-13-54	10.60	36.12	3.25	9,250	6.91	slight microcytic
4-20-54	12.52	38.61	3.00	5,700	7.48	appear normal
4-27-54	11.09	35.95	2.25	8,900	7.09	slight microcytic, slight anisocytosis
5- 4-54	11.33	37.83	3.00	7,000	7.40	appear normal (very bad smear - dust)
5-10-54	10.85	34.67	3.00	7,700	7.08	slight achromia, slight microcytic
6- 3-54	10.85	30.39	3.25	7,250	5.78	moderate achromia, slight microcytic
Average	11.96	37.97	2.61	8,692	7.25	
Total A	verage**		S. J			
	12.05	38.09	2.48	8,377	7.13	

-	1.00		-	
	23.1	10.00	10.0	50.
2.00	233	- N	100.00	63
-	- M			

*Smears made in cold, damp barn. **Total average includes pre-experimental and experimental data.

ANALYSIS OF CATTLE BLOOD DURING FRE-EXPERIMENTAL AND EXPERIMENTAL PERIODS (continued)

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00	164	10	ł

Protesso Profil Drawn	and and an approximate an inter		Mean	1997 - 1997 - 1997 - 1997 - 1997 - 1997 - 1997 - 1997 - 1997 - 1997 - 1997 - 1997 - 1997 - 1997 - 1997 - 1997 -		Diff (per	eren r ce:	tia ls nt)			
Sample Date	Mean Corpus- cular Volume	Mean Corpus- cular Hemo- globin	Corpus- cular Hemo- globin Concen- tration	Mature Neutrophils	Inmature Neutrophils	Lymphooytes	Kyelocytes	Monocytes	Eosinophils	Basophi 1s	Basket cells
Pre-Exp	erimental	Period									
2-10-54	60.51	17.96	29.68	18	0	46	0	25	11	0	0
2-15-54	52.61	17.23	32.76	15	0	45	0	30	10	0	0
2-22-54	49.48	16.19	32.71	11	0	67	0	13	9	0	0
Average	54.20	17.13	31.72	14.7	0	52.7	0	22.7	10	0	0
Experim	ental Per	iod	and and a								
8- 2-54	47.16	15.13	32.09	4	-	-	-	-	-	-	-
3- 8-54	49.92	16.22	32.48	-	-	-	-	-	-	-	-
3-15-54	41.46	13.61	32.82	7	0	67	0	15	11	0	0
3-22-54	43.70	14.04	32.12	17	0	55	0	17	11	0	0
3-30-54	50.52	16.46	32.57	7	0	74	0	7	12	0	0
4- 6-54	45.51	14.77	32.46	10	0	70	0	10	10	0	0
4-13-54	43.58	14.09	32.34	6	0	80	0	8	5	1	0
4-20-54	52.60	16.39	31.17	18	0	63	0	9	9	1	0
4-27-54	47.03	14.59	31.03	10	0	73	0	15	2	0	0
5- 4-54	45.13	14.33	\$1.75	4	0	68	0	10	17	1	0
5-10-54	46.35	16.25	35.06	10	0	64	0	10	16	0	0
6- 3-54	50.48	16.98	33.64	24	0	55	0	9	10	2	0
Average	46.95	15.24	32.46	11.5	0	66.9	0	11	10.3	.5	0
Total A	verage*										
	48.40	15.62	32.31	12.1	0	63.6	0	13.7	10.2	•4	0
							1000				

ANALYSIS OF CATTLE BLOOD DURING PRE-EXPERIMENTAL AND EXPERIMENTAL PERIODS (continued)

Sample Date	Hemo- globin (gms/ 100 ml.)	Hemato- crit (% cells)	Sedimen- tation	White Blood Count	Red Blood Count (million)	Description of Red Cells
Pre-Exp	erimental	Period				and the second second
2-10-54	13.74	46.29	1.50	7,500	7.65	moderate anisocytosis
2-15-54	14.03	42.83	1.50	10,100	8.14	moderate anisocytosis
2-22-54	13.84	42.31	1.75	12,000	8.55	slight anisocytosis
Average	13.87	43.81	1.58	9,867	8.11	
Experim	ental Per	iod				
3- 2-54	14.03	43.72	1.75	13,000	9.27	
3- 8-54	15.00	46.18	2.00	11.950	9.25	
3-15-54	15.31	46.64	2.00	10.400	11.25	moderate anisocytosis
3-22-54	12.97	40.38	2.00	11.350	9.24	moderate anisocytosis
3-30-54	12.26	87.64	2,50	9,000	7.45	moderate anisocytosis, moderate achromia
4- 6-54	11.42	35.18	3.00	10,100	7.73	moderate achromia, moderate microcytic
4-13-54	11.74	36.30	3.50	9,250	8.33	moderate microcytic, slight achromia
4-20-54	12.87	41.29	2.50	7,000	7.85	slight microcytic
4-27-54	12.96	41.76	2.25	9,900	8.88	slight microcytic, slight achromia
5- 4-54	13.54	42.65	2.00	9,900	9.45	slight microcytic, slight achromia
5-10-54	13.26	37.82	2.00	10,450	8.16	slight microcytic
6- 3-54	13.06	38.83	2.50	9,400	7.69	slight microcytic, slight achromia
Average	12.32	40.70	2.33	10,141	8.71	
Total A	verage*	Gel?				M. G. S. Marker
	13.33	41.32	2.18	10,087	8.59	
		N. C. S.				

Cow le

pre-experimental and experimental periods are shown in Table VI.

The experimental data in Table VI are within the normal range for cattle blood as listed by Coffin (1953). There were some variations in the data for individual blood samples which were out of the normal ranges. However, these variations were not consistent with the paleness about the eyes and udder which was noted in cow 1b, a drop in the cell concentration would be expected. The results in Table V show that there was only a slight drop in hemoglobin and red blood cell concentration. The values were still within the normal range for hemoglobin and red blood cell concentration.

White Leghorn Cockerel Phase

The second phase of this experiment was conducted with cockerels. The cock's comb test was first run on six White Leghorn cockerels injecting different levels of extract from the fescue hay that had been fed the cattle in this experiment and fescue grass clipped from pastures at the Middle Tennessee Experiment Station. No visible symptoms were noted below the 6 cc. level; however, above the 6 cc. level, the cockerels showed lack of coordination, vomiting, and a possible frosty appearance of the comb. These cockerels were observed very closely following injection of the extract and symptoms were noticed within a few minutes after injection. Using different levels of the extract on all six of the cockerels, the same condition was produced in every case above the 6 cc. level.

Several days elapsed between injections using extracts from different chemical mixtures and different samples. The injection levels

NORMAL RANGES OF BLOOD CELIULAR ELEMENTS IN CATTLE AND AVERAGE BLOOD CELIULAR ELEMENTS OF CATTLE USED IN THIS EXPERIMENT DURING THE PRE-EXPERIMENTAL AND EXPERIMENTAL PERIODS

Cow la

Blood Cellular Elements	Range	Pre-Experimental Period	Experimental Peri
Hemoglobin	8.0-14.5	12.64	12.62
Hematoorit	30-40	38.70	38-85
Mean Corpuscular Hemoglobin	14.4-18.6	16.49	16.57
Mean Corpuscular Volume	49.5-60.7	50.48	51.03
Mean Corpuscular Hemoglobin Concentration	52-34	32.66	32.50
Red Blood Cells (million)	5.4-9.0	7.69	7.66
White Blood Cells	4,500-13,000	9,730.0	8, 309.0
Neutrophils	15-55	21.3	20.1
Eosinophils	1-15	7.5	6.0
Basophils	5	0*0	1.0
Lymphocytes	40-70	57.7	60.2
Monocytes	3-15	13.7	13.5

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NORMAL RANGES OF BLOOD CELLULAR ELEMENTS IN CATTLE AND AVERAGE BLOOD CELLULAR ELEMENTS OF CATTLE USED IN THIS EXFERIMENT DURING THE PRE-EXPERIMENTAL AND EXPERIMENTAL PERIODS (continued)

Cow 1b

Blood Cellular Elements	Range	Pre-Experimental Period	Experimental Period
Hemoglobin	8.0-14.5	12.41	11.96
Hematoorit	30-40	38.56	37.97
Mean Corpuseular Hemoglobin	14.4-18.6	16.82	16.56
Mean Corpuscular Volume	49.5-60.7	58.39	52.44
Mean Corpuseular Hemoglobin Concentration	32-34	52.19	31.59
Red Blood Cells (million)	5.4-9.0	6.66	7.15
White Blood Cells	4,500-13,000	0-711-7	8,692.0
Neutrophils	15-55	25.7	20.9
Eosinophils	1-15	0*6	1.7
Basophils	61	0.0	0.4
Lymphocytes	40-70	46.3	57.0
Monocytes	3-15	18.7	14.2

NORMAL RANGES OF BLOOD CELLULAR ELEMENTS IN CATTLE AND AVERAGE BLOOD CELLULAR ELEMENTS OF CATTLE USED IN THIS EXPERIMENT DURING THE PRE-EXPERIMENTAL AND EXPERIMENTAL PERIODS (continued)

Cow le

Blood Cellular Elements	Range	Pre-Experimental Period	Experimental Period
Kemoglobin	8.0-14.5	13.87	12.32
Hematoorit	30-40	45.81	40.70
Mean Corpuseular Hemoglobin	14.4-18.6	17.15	15.24
Mean Corpuseular Volume	49.5-60.7	54.20	46.95
Mean Corpuscular Hemoglobin Concentration	32-54	81.72	32.46
Red Blood Cells (million)	5.4-9.0	8.11	7.87
White Blood Cells	4,500-13,000	9,867.0	10,141.0
Neutrophils	15-55	14.7	11.5
Eosinophils	1-16	10.0	10.3
Basophils	5	0*0	0.5
Lymphocytes	40-70	52.7	66*9
Monocytes	3-16	22.7	0.11

of the extract taken from pasture clippings at the Middle Tennessee Experiment Station and using all four chemical mixtures are shown in Table VII.

TABLE VII

LEVEL OF PESCUE EXTRACT INJECTED IN COCKERELS

	Level (1	n ccs.)	
lst Injection ^a	2nd Injectionb	3rd Injection©	4th Injectiond
2	8	6	4
4	6	6	2
4	6	2	6
0	0	0	0
6	4	4	8
8	2	4	8
	lst Injection ^a 2 4 4 4 0 6 8	Level (ilst2ndInjection®Injectionb284646466482	Level (in ccs.)lst2nd3rdInjection®Injection®286466462000644824

Fescue Taken from Pasture Clippings from the Middle Tennessee Experiment Station

*Extract using chemical mixture Number One. bExtract using chemical mixture Number Two. CExtract using chemical mixture Number Three. dExtract using chemical mixture Number Four. *Control lot.

The level of the extract injected taken from fescue hay that was fed cattle in the first phase of this experiment is shown in Table VIII.

The symptoms were exactly the same using both samples and all chemical mixtures except the Number Three mixture. There were no visible symptoms noted following this injection of the water extract.

TABLE VIII

LEVEL OF FESCUE EXTRACT INJECTED IN COCKERELS

		Level (i	n ces.)	
Cockerel Number	lst Injection ^a	2nd Injection ^b	3rd Injection®	4th Injectiond
1	2	8	6	4
2	4	6	6	2
3	4	6.31	2	6
46	0	0	0	0
5	6	4	4	8
6	8	2	4	8

Fescue Taken from Hay Fed Cattle in the First Phase of This Experiment

^SExtract using chemical mixture Number One. ^DExtract using chemical mixture Number Two. ^CExtract using chemical mixture Number Three. ^dExtract using chemical mixture Number Four. ^SControl lot.

This phase of the experiment was to determine the nature of the toxic factor. Some workers believe that "fescue lameness" is the same thing as ergotism, while other workers have experimentally eliminated that possibility.

In this work with cockerels, an ergot compound was injected at one and two cc. levels in two cockerels, getting a greater response from the 2 cc. level. There was a slight change in the color of the comb at the low level with a definite cyanosis at the high level, but the ergot injection failed to get the lack of coordination and the vomiting effect. Levels of 50 per cent alcohol were injected in these cockerels to determine if the vomiting and lack of coordination was due to the alcohol content of the extract, but these symptoms were not found.

Albino Rat Phase

The third phase of this experiment was conducted with the use of fifty-five albino rats with an average weight of fifty grams. These rats were set up in groups of one rat each and injected following the same procedure and the same extract using all four chemical compositions as in the second phase of this experiment. Levels of the extract injected from both samples of fescue clippings and fescue hay ranged from 0.5 cc. to 1.5 ccs.

These animals were observed very closely following injection of the fescue extract. Injections below 0.5 cc. had no effect on any of the rats. However, above 0.5 cc. the rats became less active immediately after the injection, showed an increased heart beat, lack of coordination of the hind limbs and death followed after two or three hours. After death, the skin on these rats was a very dark blue color.

The different levels of injections, by groups, of fescue extract using all four chemical mixtures taken from pasture clippings at the Middle Tennessee Experiment Station are shown in Table IX. Table X shows the level of fescue extract injected by groups using all four

chemical mixtures from the fescue hay samples used in the cattle work phase of this experiment. The symptoms were the same from all four chemical mixtures.

TABLE IX

LEVEL OF FESCUE EXTRACT INJECTED IN RATS

	Level (in ccs.)						
Lot Numbera	lst Injection ^b	2nd Injection©	3rd Injectiond	4th Injectione			
lf	0.0	0.0	0.0	0.0			
2	0.5	0.5	0.5	0.5			
5	0.75	0.75	0.75	0.75			
4	1.0	1.0	1.0	1.0			
5	1.5	1.5	1.5	1.5			

Fescue Taken from Pasture Clippings from the Middle Tennessee Experiment Station

^aEach lot contained one rat. ^bExtract using chemical mixture Number One. ^cExtract using chemical mixture Number Two. ^dExtract using chemical mixture Number Three. ^eExtract using chemical mixture Number Four. ^fControl lot.

TABLE X

LEVEL OF FESCUE EXTRACT INJECTED IN RATS

		Level (in ccs.)							
Lot Numbera	lst Injection ^b	2nd Injection ^c	3rd Injectiond	4th Injection ^e					
lf	0.0	0.0	0.0	0.0					
2	0.5	0.5	0.5	0.5					
3	0.75	0.75	0.75	0.75					
4	1.0	1.0	1.0	1.0					
5	1.5	1.5	1.5	1.5					

Fescue Taken from Hay Fed Cattle in the First Phase of This Experiment

^aEach lot contained one rat. ^bExtract using chemical mixture Number One. ^cExtract using chemical mixture Number Two. ^dExtract using chemical mixture Number Three. ^eExtract using chemical mixture Number Four. ^fControl lot.

CHAPTER V

SUMMARY

Three cows were used in a 102-day test to study the effects of feeding fescue pasture clippings from a pasture that had caused fescue poisoning to cattle grazed on the fescue. Fescue extract from both pasture clippings and hay samples injected in six White Leghorn cockerels and fifty-five albino rats was also studied.

The cattle in the experiment were observed very closely for visible symptoms of "fescue lameness," and a complete blood analysis on them was run weekly and studied. There was no apparent change in the blood picture of these cattle as compared with normal cattle. There were slight visible symptoms of "fescue lameness" noted in these cattle. The weight losses in these cattle could be attributed in part to poor quality hay.

The extracts of both the hay and pasture samples, when injected in cockerels, above the 6 cc. level caused a slight cyanosis in the comb color and caused vomiting and lack of coordination. When these extracts were injected in rats, above the 0.5 cc. level they caused death.

More research needs to be conducted on the causes and effects of fescue poisoning. These experiments are being conducted under pasture conditions.

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