Case Report – Myonecrosis in Feedlot Cattle

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Michael Apley, DVM PhD DACVCP[1]; Brent Meyer, DVM MS[2]; Annette O'Connor, BVSc MVSc DVSc[2]; David Villar, DVM PhD[2]; Bruce Janke, DVM PhD[2]; Kent Schwartz, DVM MS[2]; Karl Kersting, DVM MS[2]
[1]*Department of Clinical Sciences, College of Veterinary Medicine, Kansas State University*
[2]*Department of Veterinary Diagnostic and Production Animal Medicine, College of Veterinary Medicine, Iowa State University*

Abstract

This report describes an outbreak of disease in a Northwestern Iowa feedlot from January to March of 2001. The cattle had been received in the feedlot in July and August, 2000. Clinical signs included severe lameness, recumbency and death. Lameness was not apparent early in the outbreak and the initial diagnosis was central nervous disease. No infectious or toxic cause could be demonstrated. Due to poor performance, approximately a third of the heifers were held back after the main group was sold. Half of these poor performing heifers displayed visible stiffness. Myonecrosis was demonstrated by significantly elevated serum creatine kinase concentrations in visibly affected cattle as compared to visibly unaffected cattle. Histological lesions were confirmed in cardiac muscle but skeletal muscle was not examined. The cattle had been fed a predominantly corn diet with a liquid supplement containing vitamin E calculated at 12.5 IU/head per day until late in the feeding period, when they were switched to a dry supplement delivering 40 IU/head per day. Serum and liver vitamin E concentrations in sampled animals were below the normal range.

Common limitations in field investigations include a failure to test un-affected animals to enable comparisons between groups, testing of animals after disease onset resulting in an inability to demonstrate a temporal relationship between the cause and effect, and small sample sizes. Our case-report suffers to some extent from all these factors; however we suspect that the myonecrosis likely occurred due to Vitamin E deficiency. This presumptive diagnosis is based on the combination of knowledge of vitamin E, creatine kinase, (CK) and Aspartate Amino Transferase (AST) values in the sampled cattle, clinical signs observed, elimination of other possible etiologies and supportive statistical analyses. Investigation of unexplained debilitation in feedlot cattle, especially when accompanied by lameness, should include evaluations of serum and/or liver vitamin E concentrations, serum (AST) and (CK) concentration, muscle histology, and ration vitamin E concentration.

Introduction

This case report involves high death loss encountered by a Northwestern Iowa feedlot from January to March of 2001. The winter of 2000-2001 was characterized in Iowa by muddy pen conditions due to multiple precipitation events combined with freeze-thaw cycles. A weather summary for this area during this time period is presented in Table 1.

Case History

Between January 23 and March 3, 2001, 27 (6.2%) of 438 yearling feeder heifers in a northern Iowa feedlot died. The heifers were predominantly medium framed, British and Continental breeds. The heifers arrived as separate shipments on July 6th, August 12th and August 16th of 2000 from 3 origins in the North Central US.

In the same feedlot from February 12 to February 27, 2001, 6 of 159 yearling steers (3.7%) died. The steers were medium framed, apparently of Angus breeding and arrived August 17, 2000 from Nebraska. All of the cattle would have been classified as high quality, northern calves on arrival. Although they initially had good winter hair coats, muddy pen conditions led to extensive hair loss on the abdomen, chest, and legs.

In addition to the mortalities, four heifers and a steer with limited recovery were sold for salvage value during January and February, 2001. Three pens of unaffected cattle were received as calves on October 20th, November 5th, and November 6th, 2000.

Presenting clinical signs were an initial period of ambulatory difficulty followed by recumbency, then death within a few days. Rectal temperatures in affected animals were subnormal. Early in the period of death loss, a high prevalence of epistaxis was reported in the heifers. Necropsies of several initial cases by the referring veterinarian showed no visible lesions. These findings, combined with apparent altered mental status and seizuring of some of the affected animals, led to an initial diagnosis of Central Nervous System (CNS) disease. Response to antimicrobial, anti-inflammatory, and thiamine therapy was poor, as all cases either died or were sold for salvage value after partial recovery.

The heifers were housed in two, open, dirt floor feeding pens with shaded concrete bunks and aprons. A shed was available to one heifer pen during the entire feeding period and through January for the other pen. The steers were housed in one pen similar to the heifer pens, but with no available shed.

Wet corn gluten feed had been part of the diet until it was removed on January 29 due to the possibility of excess sulfur

in the ration contributing to polioencephalomalacia. The ration varied over the period reported, but after exclusion of the wet corn gluten feed consisted of 83% corn, 11% ground corn stalks, 4% liquid protein supplement, and 2% soybean meal on an as-fed basis. The liquid supplement was added to the ration at a rate calculated to achieve an intake of 1.25 lbs/day per animal. According to the manufacturer's label, this supplement included a minimum of 50% crude protein (with not more than 48% equivalent crude protein units from non-protein nitrogen), a minimum of 40,000 IU vitamin A per pound and a minimum of 10 IU vitamin E per pound. The liquid supplement was discontinued on February 17, 2001 due to initial consideration of possible monensin or urea toxicity, and was replaced by a dry supplement containing 40 IU vitamin E per pound. The dry supplement was included in the feed at 1 lb/day per animal, which was maintained for the remainder of the feeding period.

Harvest shipment dates for the first two groups of heifers (unaffected heifers with the best performance) were February 27, 2001 (176 head) and March 4, 2001 (86 head). Approximately 50% of the remaining heifers displayed visible lameness. The steers had not been shipped as of a site visit on March 9, 2001, but were nearing finish. Approximately 30% of the steers were estimated to be lame at this time.

Diagnostic Findings

A live heifer in a terminal stage was submitted to the Iowa State University Diagnostic Lab on February 12, 2001. At this time, 9 heifers (including this animal) and 1 steer had died after displaying typical clinical signs. Pertinent diagnostic laboratory findings were:

Histology: No significant lesions in brain, spinal cord, liver, kidney, heart

Bacteriology: No bacterial growth from brain, liver, or kidney

Two additional heifers were submitted on February 15, 2001 with a history of rapid deterioration and death. Laboratory findings were as follows.

Gross findings: Heifer 1 - Fibrin clots in enlarged right hock, "old" rumen ulcer, rumen pH 5.4, Heifer 2 - Catarrhal exudate and ulcers in abomasum, clumped rumen papillae, rumen pH 5.6

Histology: Brains: Mild hyalinization of scattered vessels, mild endothelial swelling, Hearts: Mild, subtle, multifocal acute myofibrillar necrosis and hyalinization, Rumens: Edema in papillae, multifocal suppurative inflammation, Abomasums: Multifocal hemorrhages,

Bacteriology: Arcanobacterium pyogenes from the hock of heifer 1.

Immunohistochemistry: Negative for BVD.

Toxicology: Testing ruled out urea toxicosis (by feed and rumen content analysis), excess sulfur (by analysis of feed and water submissions), nitrate poisoning (ocular fluid), hypomagnesemia (ocular fluid). Copper and selenium concentrations in the livers were within normal limits. Analysis of two supplement samples confirmed the monensin concentration was not above specifications.

Serum from a heifer in the early stages of disease (ambulatory difficulty but still standing) was also submitted on February 15. The serum creatine kinase (CK) concentration was 23,960 IU/L. The expected upper range of normal is 350 IU/L.

The case fatality rate in the affected pens continued to be reported as almost 100% although clinical success with thiamine on similar appearing cases in an adjacent pen of calves was reported. At this time, the owners were unwilling to pursue further diagnostic tests, so a combination of research funds from the authors and support from the Diagnostic Laboratory was used to continue the investigation.

On March 1, 2001, one live-affected and three dead animals were submitted to the diagnostic lab. In addition, whole blood and serum from an animal that was still alive and standing at the feedlot were submitted. Including these animals, 26 heifers and 6 steers had been lost to date. Results were as follows.

The live heifer was conscious but unable to rise from lateral recumbency. It received treatment for CNS disease but died on the afternoon of the next day. Nystagmus was noted on physical examination. Analysis of a cerebral spinal fluid tap reported cellularity within the normal range, high protein content and negative bacterial culture. The entire spinal cord was extracted and no gross or histological lesions were noted. Brain sodium levels were slightly elevated but not enough to suggest salt toxicity. The slightly elevated brain sodium levels were considered due to dehydration.

Clinical Pathology: The serum creatine kinase value of the animal at the feedlot was 48,952 IU/L.

Histology: Hearts from two animals showed a few scattered foci of myofiber loss and replacement with fibrous connective tissue.

Chemistry/toxicology: Vitamin E and selenium analysis results from two livers submitted March 1 and one liver submitted February 15 are reported in Table 2.

A site visit was conducted on March 5, 2001 prior to laboratory results from the March 1 submissions. The first two harvest shipments of heifers had been completed by this time and only the poorest performing heifers remained. The disease history was clarified at this time. Approximately 50% of the heifers and 30% of the steers in the affected groups appeared to be lame. The typical stance of the severely affected animals is represented in Figure 1.

Affected animals walked with a stilted gate, with stiffness especially notable in the hindquarters. No history of excessive fluctuations in feed intake was apparent. No history of a sudden occurrence of drunk and/or down animals or loose, bubbly stools was reported to support a presumptive diagnosis of acidosis-related founder. The managers had a history of good feed consumption management and no previous history of excessive founder rates. Although a few "long toes" were observed, they were not common among the affected, lame individuals.

A follow-up site visit was conducted on March 19, 2001. During this visit, 20 serum samples were collected; 9 from clinically normal animals (visibly unaffected) and 11 from animals that were visibly lame and having difficulty moving (visibly affected). Serum Aspartate Amino Transferase (AST), Creatine Kinase (CK), and vitamin E levels for these samples are reported in Table 3.

Statistical Analysis of Serum Sample Results

All analyses were conducted in SAS V 8.1 (SAS Inc. Cary. NC). Comparison of means for serum AST, CK and Vitamin E between visibly affected and unaffected animals were conducted after testing for normality of distribution using the Wilk-Shapiro test. Vitamin E was normally distributed; therefore, a t-test for two samples was used to determine if a group effect existed. The two sample Wilcoxon (Mann-Whitney) tests were used to determine if group differences existed between groups for AST or CK, which were not normally distributed. Exact p values were calculated.

The animals were also categorized into two groups; those with values within the normal range and those with values above the normal range. Stratifications are illustrated in Table 4. As the expected value of some cells was less than five, Fishers exact tests (right sided) were used to determine the probability of being visibly affected in the above normal range AST and CK groups vs. being affected in the normal groups. Odds ratios and 95% confidence intervals were also calculated.

Statistical Analysis Results

The t-test for differences in mean serum vitamin E concentrations between groups found no significant difference ($p = 0.92$). The two sample Wilcoxon (Mann-Whitney) tests for the AST values resulted in exact p value=0.06 for the one-sided test and exact p value =0.13 for the two-sided test. The corresponding results for CK were exact p value =0.04 and exact p value =0.08 respectively.

The point estimate of odds ratio for AST values was 6.6 (95% confidence interval 0.6-73). The p value for the Fishers exact test for differences in the proportions was 0.11. The point estimate of odds ratio for CK values was 9.0 (95% confidence interval: 1.1-71) and the Fishers exact test p value was 0.04. These results suggest that visibly affected animals were associated with above normal range AST or CK values, however due to small sample size the degree of uncertainty about the exact association is unclear, as shown by wide confidence intervals.

Discussion

It is often difficult to reach conclusions about the cause of disease outbreaks because of a lack of resources for proper investigation of the disease. Common limitations in field investigations include a failure to test un-affected animals to enable comparisons between groups, testing of animals after disease onset resulting in an inability to demonstrate a temporal relationship between the cause and effect, and small sample sizes. Our case-report suffers to some extent from all these factors; however we suspect that the myonecrosis likely occurred due to Vitamin E deficiency. This presumptive diagnosis is based on the combination of knowledge of vitamin E, creatine kinase, and AST values in the sampled cattle, clinical signs observed, elimination of other possible etiologies and supportive statistical analyses.

Creatine kinase is one of the most specific diagnostic enzymes in the blood, indicating active muscle necrosis. The half-life in cattle is reported to be approximately 4 hours, with return to normal serum concentrations 2-3 days after the necrosis stops.[2] Elevated concentrations are common in cattle unable to rise due to conditions such as hypocalcemia. However, in this investigation, we found markedly elevated CK serum concentrations in ambulatory animals. Although elevated CK values could be attributed to lame animals spending more time in recumbency, the finding of myocardial lesions along with diagnostic elimination of other possible etiologies supports the hypothesis of a primary myonecrosis.

Serum vitamin E concentrations were below normal and not significantly different between visibly affected and visibly unaffected poor-doing heifers. However, the sample population consisted of the poorest performing heifers retained after the first two groups had already been shipped so these groups may not represent the most powerful comparison. Ideally we should have compared the Vitamin E levels of the poor doers to the "normal doers" but this was not possible.

The OR and Fishers exact test results for CK and AST weakly indicate that the visibly affected poor-doers were more likely to have elevated values as compared to the visibly unaffected poor-doers as the confidence intervals either barely include or exclude the OR of 1 which would suggest no association. However, given the limitation of small sample sizes, we believe that as the point estimates the ORs are skewed to the right (away from OR=1) they support the

hypothesis that high CK and AST, and by extension vitamin E, was associated with the outbreak.

A primary question in this investigation was whether the vitamin E supplementation rate had contributed to the incidence of disease. The liquid protein supplement had been included in the ration through February 16, 2001 at a rate calculated to provide 12.5 IU of vitamin E per head per day. Beginning February 17, 2001 a dry supplement providing vitamin E at the rate of 40 IU per head per day was included in the ration until harvest. The liquid supplement was also used at a nearby feedlot experiencing similar cases. However, reports were received of other feedlots in the area using this supplement that were not experiencing high morbidity and mortality. These reports were not confirmed and differences in the feedlots were not investigated.

Vitamin E requirements in feedlot cattle have been reported by Hutcheson as 200-800 IU/head per day during the receiving period, 100-200 IU/head per day during the growing period, and 50-100 IU/head per day during the finishing period.[3] The vitamin E requirement for young calves has also been estimated as between 15–60 IU/kg of dry feed, with optimal growth in finishing cattle achieved at supplementation of vitamin E at 50–100 IU of vitamin E/head per day.[4]

A conventional practice has been to supplement vitamin E to achieve requirements due to unpredictability of feedstuff vitamin E contents. Corn has been reported as averaging 18.7 IU of vitamin E/kg with a range of 10.8 to 36.1 IU/kg.[3] If the cattle in this report consumed 11 kg of corn per day, they should have received a minimum of 119 IU of vitamin E per day. However, feedstuffs are extremely variable in their vitamin E content and extensive decreases may occur due to storage conditions such as natural drying, heat, and moisture.[4] A deficiency in this investigation was the lack of feed analysis for actual vitamin E content.

It is also possible that the stress of the winter contributed to increased vitamin E requirements. Sconberg, et al, found that the stress of shipping and handling, or stress simulation by injection of ACTH and epinephrine, decreased plasma and red blood cell α-tocopherol content while raising serum creatine kinase concentrations.[5] This reference pointed out that "stress and vitamin E status affect the ability of muscle, and possibly organ cell membranes to withstand oxidative insult". By completely removing vitamin E supplementation from a corn silage-based ration, the authors were able to decrease mean plasma α-tocopherol concentrations to 1.2 ± 0.2 ppm, which is consistent with the serum concentrations reported in Table 2. In contrast, on arrival these same cattle had plasma α-tocopherol concentrations of 4.33 ± 0.37 ppm.

Njeru, et al., concluded that "Serum and liver tocopherol reflected Vitamin E intake and could be used reliably to estimate vitamin E status in young cattle".[6] By placing heifers on a corn-based concentrate and poor quality Bermuda grass hay the authors were able to "deplete vitamin E stores" and lower serum α-tocopherol concentrations to a mean of 1.8 ppm after 28 days. Supplementation with 500 IU vitamin E per day resulted in a rapid rise in serum α-tocopherol concentrations, achieving a mean of approximately 3.5 ppm by day 20 of supplementation. The authors of this paper used additional literature sources to suggest that serum α-tocopherol concentrations below 1.0 to 1.5 μg/ml could be considered deficient in ruminants since concentrations lower than these are often found in cattle with nutritional muscle degeneration.

Nockels, et al, found that feeding heifers a control diet assayed at 12.1 IU/Kg dry diet resulted in mean plasma α-tocopherol concentrations of 1.67 ppm while heifers supplemented with an additional 1000 IU/head per day had mean plasma α-tocopherol concentrations of 3.28 ppm.[7] In stressed cattle, the authors noted that this paper supported the findings of Sconberg, et al, in that stress may raise the plasma α-tocopherol concentration while in fact the tissues (liver, red blood cells) are experiencing decreased concentrations. Therefore, plasma concentrations of α-tocopherol in stressed cattle may be overly optimistic concerning total body vitamin E status.

Conclusion

The affected cattle in this report became recumbent and died, or became severely lame, following an extended feeding period on a predominantly corn diet with vitamin E supplementation calculated at 12.5 IU/head per day throughout the feeding period. No infectious or toxic cause could be demonstrated through laboratory efforts. Myonecrosis was demonstrated by significantly elevated creatine kinase in visibly affected cattle as compared to visibly unaffected cattle and by cardiac muscle histology. Serum vitamin E concentrations were interpreted as inadequate while liver vitamin E concentrations were well below the normal range.

Unexplained debilitation in feedlot cattle, especially when accompanied by lameness, should be investigated by including evaluations of serum and/or liver vitamin E concentrations, serum AST and CK values, muscle histology, and ration vitamin E status. Veterinarians should be aware of vitamin E supplementation recommendations and work with their clients to assure adequate vitamin E concentrations are present in the feed for both health and performance.

Figure 1. Affected animal on March 5, 2001.

Table 1. Weather summary data for a city close to the feedlot described in this report for the period January – March, 2001.

	Average High Temperature C(F)	Average Low Temperature C(F)	Snowfall cm(in)	Precipitation cm(in)
January	-3.6 (25.6)	-12.6 (9.3)	21.8 (8.6)	3.0 (1.2)
February	-3.9 (25.0)	-16.5 (2.3)	16.8 (6.6)	3.3 (1.3)
March	0.5 (32.9)	-8.5 (16.7)	10.3 (4.1)	2.6 (1.0)

Table 2. Selenium and Vitamin E results from liver analysis

Submission date	Selenium (ppb)	Vitamin E (ppm)
2/15/01	680	1.4
3/1/01	590	1.0
3/1/01	450	0.6

Normal selenium range: 250 - 500 ppb
Normal vitamin E range: 2.6 – 5.0 ppm

Table 3. Serum Aspartate Amino Transferase (AST), Creatine Kinase (CK), and Vitamin E results from samples collected 3/19/01.

	AST (IU/L)	CK (IU/L)	Vitamin E (ppm)
Normal upper	125	350	4.5
Normal lower	55	100	2.5
Visibly normal 1	99	203	1.4
Visibly normal 2	87	388	1.8
Visibly normal 3	92	325	0.8
Visibly normal 4	87	285	1.1
Visibly normal 5	109	797	1.7
Visibly normal 6	100	296	1.1
Visibly normal 7	114	232	1.1
Visibly normal 8	91	176	1.6
Visibly normal 9	198	5831	1.5
Visibly normal mean	**109**	**948**	**1.34**
Visibly affected 1	170	354	1.2
Visibly affected 2	116	245	1.5
Visibly affected 3	86	180	1.6
Visibly affected 4	478	7399	0.8
Visibly affected 5	155	1095	2.2
Visibly affected 6	99	470	1.6
Visibly affected 7	165	1960	0.8
Visibly affected 8	133	903	0.6
Visibly affected 9	106	664	1.2
Visibly affected 10	96	1508	1.2
Visibly affected 11	101	2341	2.3
Visibly affected mean	**155**	**1556**	**1.36**

Table 4. Stratification of AST and CK values by animal group.

AST	Visibly Affected Animals	Visibly Unaffected Animals
Above normal	5	1
Normal	6	8
CK		
Above normal	9	3
Normal	2	6