

## COMPOSITION OF LIPIDS OF CEREAL FORAGES AS RELATED TO TETANY IN CATTLE

V.R. Bohman and H.F. Mayland and D.L. Grunes  
University of Nevada-Reno 89557 and  
USDA-ARS Kimberly, Idaho 83341  
and Ithaca, New York 14853

## Summary

Forage samples were collected from mixed winter wheat and rye (*Triticum aestivum* L. and *Secale cereale* L.) pastures, from December through April at El Reno, Oklahoma, while they were grazed by 32 mature cows to determine if the concentrations and speciation of naturally occurring higher fatty acids (HFA) in forage were related to the incidence of tetany. Forage samples were analyzed for N, K, aconitic acid, total lipids and total and individual C11 to C18:3 HFA. Total HFA and lipid values were high in the very immature forage, but these values decreased as the winter season progressed. In early March, these values increased sharply coinciding with rapid forage growth and were near maximum on the day (19 March) when tetany occurred in five cows. Forage N concentrations were also maximum on that day and then declined rapidly with time. A positive correlation ( $P < .05$ ) occurred between forage N, total lipids, HFA, K, aconitic acid, and C18:3. A negative correlation ( $P < .05$ ) occurred between total lipids and C18:2 and C16; HFA and C18:2, and between C18:3 and C18:2 and C16. Aconitic acid and K were also negatively correlated with C16 and C18:2. Linolenic (C18:3) was the predominant fatty acid (62 percent), followed by palmitic (C16 at 15 percent) and linoleic (C18:2 at 10 percent). Other fatty acids (C12, 14, 16:1, 18 and 18:1) constituted the remaining 13 percent. The fatty acid composition was similar to that reported in other gramineae forages. It was concluded that the naturally occurring HFA could be a factor in tetany of grazing animals.

Keywords: Higher fatty acids, linolenic acid, palmitic acid, linoleic acid, nitrogen, aconitic acid, potassium, cereal forages, tetany, lipids

## Introduction

Tetany in the mature grazing ruminant is the clinical manifestation of a variety of interacting events of both plant and animal origin that may result in the death of the animal. Hypomagnesemia is the most consistent symptom of this anomaly (Fontenot, 1979; Littledike et al., 1981). However, both plasma Mg and Ca may be low at times and with "wheat pasture poisoning" severe hypocalcemia is

the primary cause of tetany (Bohman et al., 1983a). These conditions hypomagnesemia, and hypocalcemia, do not always reflect low dietary intakes of these cations, but may be induced by high intakes of various metabolites including lipids and their degradation products which limit the absorption of Ca and Mg from the digestive tract.

The purpose of this study was to identify the lipid composition of cereal forages sampled during the grazing period and to related lipid fractions and other metabolites to the occurrence of tetany in cattle.

## Experimental Procedure

Thirty-two pregnant Angus or Hereford beef cows (average age 10.2 yr, range 8 to 15 yr) were placed on three 8-ha pastures (replicates) in December 1979, at the Southwestern Livestock and Forage Research Station, El Reno, Oklahoma. These pastures were seeded to hard-red winter wheat and rye the preceding September. The forage was approximately 10 cm high when sampling was initiated in December. Two forage samples weighing approximately 750 g (wet weight) were collected from each pasture by clipping random plants to a 1.5 cm stubble height at intervals of 10 days in December, 14 days in January and February and 7 days in March and April. One sample was freeze-dried while the second sample was oven-dried at 65°C. Dried samples were ground through a 1-mm stainless steel screen and stored for analysis. Oven-dried samples were used for N, K and aconitic acid analysis while the freeze-dried samples were used for lipid determinations.

The lipids were extracted from the freeze-dried plant samples by the method of Folch et al. (1957) and aliquot was dried for total lipids. Another aliquot was used for the preparation of methyl-esters of the fatty acids (Mason and Waller, 1964) and these were determined on a Perkin-Elmer 990 gas chromatograph. Total HFA were determined by the method of Molloy et al. (1974).

The analysis of variance, regression and correlation analysis were calculated on a Cyber 730 computer system according to the SPSS computer package of Nie et al. (1975).

## Results and Discussion

The temperature, precipitation, accumulative plant growth and percent forage dry matter is shown in figure 1. Five cows developed tetany on day 105 of the study. The lipid content of the forage was high initially, decreased during the winter period (Dec. to March) and rose to a maximum near the time of tetany (Mar. 19) then decreased to the end of the study. Since total lipids represent a composite of various entities and their level in plant tissue is related to other metabolites, the interrelations of total lipids, lipid fractions, nitrogen and related components were evaluated (fig. 2). The pattern of HFA and total lipids was similar. The correlation

<sup>1</sup>Contribution from the Nevada Agri. Exp. Sta. Reno Nevada in co-operation with USDA-ARS, Kimberly, Id and Ithaca, N.Y.

<sup>2</sup>Appreciation is extended to A.C. Mathers for N and K analysis.

<sup>3</sup>Dept. Animal Science, Univ. of Nevada Reno 89557.

<sup>4</sup>USDA-ARS Snake River Conservation Research Center, Kimberly, ID 83341.

<sup>5</sup>USDA-ARS Soil, Plant and Nutrition Laboratory, Ithaca, N.Y. 14853

<sup>6</sup>Mention of a trade name does not constitute a recommendation for use by the USDA or State Agri. Exp. Sta.

was .82 between these components (table 1). Although N, total lipids and HFA all peaked at the time of tetany, the N content of the forage plants remained relatively constant, but high, until February (day 63) when it was appreciably lower. Thereafter it increased until the time of tetany and then decreased rapidly thereafter.

The interrelation of total lipids, lipid fractions and nitrogen is shown in table 1. The correlation of the N content of the forage and total lipids and HFA was .63 and .59, respectively. Mayland et al. (1976) regressed HFA concentration against N concentration of several different forages and found that the slope varied from 8.4 to 24.5 Meq HFA/kg for each percentage of N. In the current study this rate was 50 meq/kg which was similar to the value of 46 meq/kg reported for mixed forages in the Netherlands (Kemp et al. 1966). The plants in the study of Mayland et al. (1976) were grown under growth chamber conditions while the other studies involved field-grown forages. Mayland et al. (1976) suggested that the difference in rate of increase with N may have reflected the degree of light saturation of the plants under growing conditions. Day length, temperature or other factors may also have attributed to the reduced response under growth chamber conditions. Other studies (Barta, 1955; Molloy et al., 1974) reported intermediate values for field-grown forage. However, when sequential samples were taken from the same forage in this study the relationship was not as high ( $r=.59$ ; Mayland et al., 1976; .63 to .96). High forage N has been consistently associated with hypomagnesemia and tetany in grazing ruminants but when exogenous N is fed or administered to experimental animals, it has no effect on magnesium absorption (Sell and Fontenot, 1980). Highly related components such as lipids or lipid fractions may have a greater effect.

The patterns of the individual fatty acids in relation to the time of tetany in cattle differ (fig. 3). Linolenic acid (C18:3) is the predominant fatty acid present in the wheat-rye forage. Its concentration ranges from 52% to 72% of the total HFA, being high during December and at the time of tetany. These values are similar to those of 59 to 75% reported for C18:3 by Mayland et al. (1976). It is positively correlated with percent N, HFA, aconitic acid, K and total lipids ( $r=.33$  to .61 table 1 and 3) but negatively related to palmitic acid ( $r=-.75$ ). Linoleic acid (C18:2) is negatively correlated to total lipids ( $r=-.48$ ), aconitic acid ( $-.45$ ), K ( $-.38$ ) and linolenic acid ( $-.58$ ). Aconitic acid and K concentrations of the plant are related to tetany as well as these lipids (Bohman et al., 1983b). Linolenic acid is the predominant acid in cereal forage lipids (fig. 2). It constitutes about 62% of the total fatty acids (table 3). Palmitic acid (14.7%) and linolenic acid (10.4%) are the next dominant acids. These 3 acids account for 87 percent of the fatty acids in the immature cereal forage. The other fatty acids, C12, 14, 16:1, 18 and 18:1, collectively account for 13% of the fatty acids (fig. 3).

Since lipids are rapidly hydrolyzed in the rumen to their constituent fatty acids and glycerol (Maynard et al., 1979), the fatty acids are free to form soaps within the digestive tract of the host animals and thus complex Mg and Ca from the diet. Since fatty acids are not absorbed in the rumen, a maximum opportunity exists for this phenomena to occur.

#### Literature Cited

- Barta, A.L. 1975. Higher fatty acid content of forages as affected by species and by nitrogen and potassium fertilization. *Crop Sci.* 15:169-171.
- Bohman, V.R., F.P. Horn, E.T. Littledike, J.G. Hurst and D. Griffin. 1983a. Wheat pasture poisoning II. Tissue composition of cattle grazing cereal forages as related to tetany. *J. Anim. Sci.* 57:1364-1373.
- Bohman, V.R., F.P. Horn, B.A. Stewart, A.C. Mathers and D.L. Grunes. 1983b. Wheat pasture poisoning I. An evaluation of cereal pastures as related to tetany in beef cows. *J. Anim. Sci.* 57:1352-1363.
- Folch, J., M. Lees and G.H. Sloane Stanley. 1957. A simple method for the isolation and purification of total lipids from animal tissues. *J. Biol. Chem.* 226:497-509.
- Fontenot, J.P. 1979. Animal nutrition aspects of grass tetany. P.51-52. In V.V. Rendig and D.L. Grunes (ed). *Grass Tetany*. Amer. Soc. Agron. Spec. Publ., No. 35. Madison, Wisc.
- Garton, G.A. 1960. Fatty acid composition of the lipids of pasture grasses. *Nature* 187: 511-512.
- Kemp, A., W.B. Diejs and E. Kluvers. 1966. Influence of higher fatty acids on the availability of magnesium in milking cows. *Neth. J. Agri. Sci.* 14:290-295.
- Littledike, E.T., J.W. Young and D.C. Beitz. 1981. Common metabolic diseases of cattle: ketosis, milk fever, grass tetany and downer cow complex. *J. Dairy Sci.* 64:1465-1482.
- Mason, M.E., and G.R. Waller. 1964. Dimethoxypropane induced transesterification of fats and oils in preparation of methyl esters for gas chromatographic analysis. *Anal. Chem.* 36:583-586.
- Mayland, H.F., L.F. Molloy and T.W. Collie. 1976. Higher fatty acid composition of immature forages as affected by N fertilization. *Agron. J.* 68:979-982.
- Maynard, L.A., J.K. Loosli, F. Hintz and R.G. Warner. 1979. *Animal Nutrition* (7th ED.) McGraw-Hill Book Co., New York.
- Molloy, L.F., D.J. Giltrap, T.W. Coolie and A.J. Metson. 1974. Degradation of higher fatty acids in perennial rye-grass and white clover following drying and storage. *J. Sci. Food Agri.* 25:595-606.
- Nie, N.H., H.C. Hill, J.G. Jenkins, K. Steinbrenner and D.H. Brent. 1975. *Statistical package for the social sciences* (2nd ED.) McGraw-Hill Book Co., New York.
- Sell, J.F. and J.P. Fontenot. 1980. Magnesium in Animal Nutrition. Nat'l. l. Feed Ingredients Assn. West Des Moines, Iowa 50265.

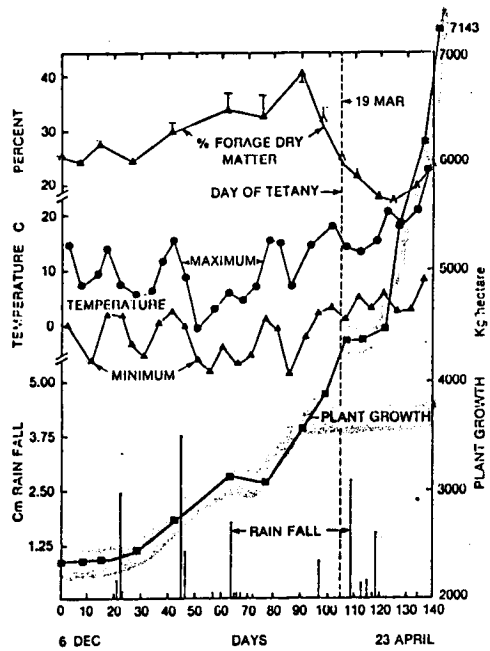


Fig. 1. The temperature, precipitation, accumulation plant growth and percent forage dry matter of experimental pastures.

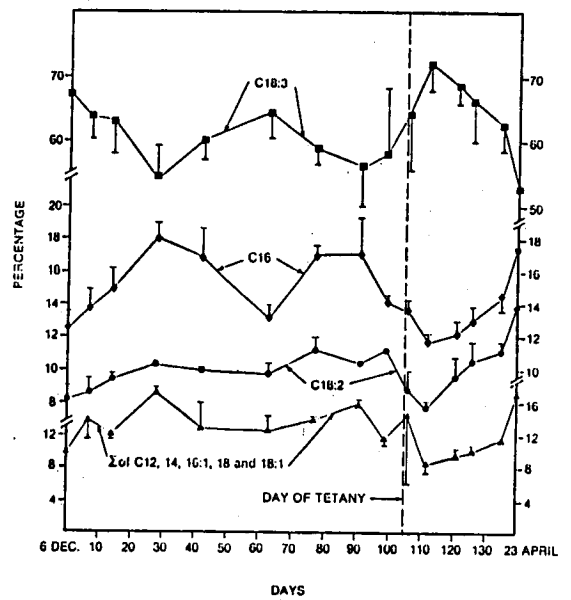


Fig. 3. The fatty acid composition of the lipids of immature cereal forages as related to time and the occurrence of tetany in cattle

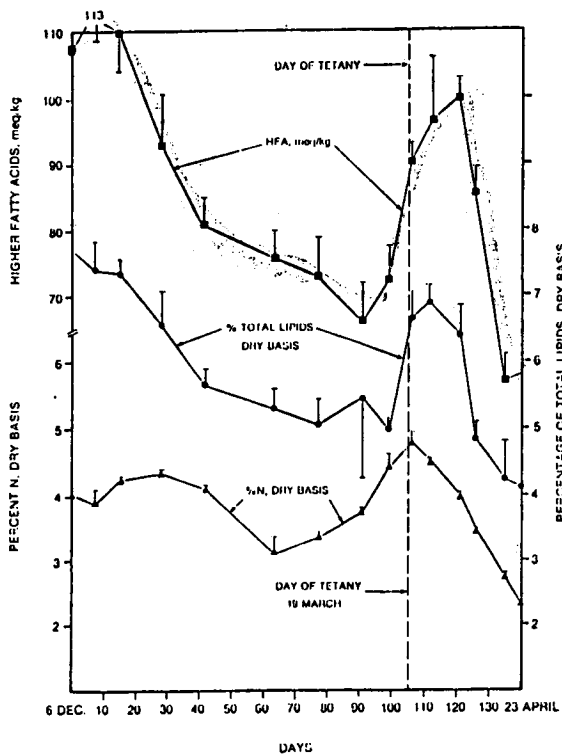


Fig. 2. The higher fatty acids, total lipids and nitrogen content of immature cereal forages as related to time and the occurrence of tetany in cattle.

Table 1. The interrelations of lipids, lipid fractions and nitrogen in cereal forages (n=45)

X and Y Parameters	Regression equation	r	x		y	
			$\bar{x}$	SE	$\bar{y}$	SE
Percent N and HFA <sup>a</sup>	y = 50 x - 97.5	.59**	3.8	.10	85.3	2.83
Percent total lipids (PTL) and percent N	y = .35 x + 1.72	.63**	5.91	.19	3.8	.10
C16 <sup>b</sup> and percent N	y = -.059 x + 4.67	-.23	14.7	.40	3.8	.10
C18:3 <sup>b</sup> and percent N	y = .035 x + 1.58	.33*	62.2	.96	3.8	.10
HFA and PTL	y = .05 x + 1.27	.82***	85.3	2.83	5.91	.10
HFA and C16	y = -.04 x + 18.3	-.30*	85.3	2.83	14.7	.40
HFA and C18:3	y = .16 x + 48.6	.47**	85.3	2.83	62.2	.96
PTL <sup>c</sup> and C16	y = .61 x + 18.3	-.28	5.91	.19	14.7	.40
PTL <sup>c</sup> and C18:3	y = 2.06 x + 50.1	.40**	5.91	.19	62.2	.96
PTL <sup>c</sup> and C18:2	y = -1.17 x + 17.4	-.48**	5.91	.19	10.4	.46
C16 and C18:3	y = 1.08 x + 88.7	-.75***	14.7	.40	62.2	.96
C18:3 and C18:2	y = -.28 x + 27.6	-.58**	62.2	.96	10.4	.46

\*, \*\*, \*\*\*, statistically significant at P.05, P.01 and P.001, respectively

a Higher fatty acids, milliequivalents/kg.

b Length of fatty acid chain and degree of unsaturation. The fatty acids C12 to C18:3 are in the units of percentage of total.

*Percent Total Lipids*

Table 2. The correlation coefficients of potassium and aconitic acid with plant lipids, HFA and HFA species.

Lipids	Aconitic acid meq/kg	K %
Total lipids, % dry basis	.71**	.61**
HFA, meq/kg, <sup>a</sup>	.75**	.74**
Percent of acids		
C12	-.10	.05
C14	-.28	-.004
C16	-.49*	-.30*
C16:1	-.29	-.33*
C18	-.14	-.16
C18:1	-.26*	-.19
C18:2	-.45**	-.38**
C18:3	.61**	.41**

\*, \*\*, statistically significant at P.05 and P.01 respectively

Table 3. Fatty acid composition of various green forages, percent of total acids in each group.

Fatty acids	Forages				
	Cereal <sup>a</sup>		Wheat <sup>b</sup>	Grasses <sup>b</sup>	Grasses <sup>c</sup>
	mean	SD	mean	mean	mean
C12	2.8	1.2			
C14	2.9	.8	2.5	2.3	1.1
C16	14.7	2.7	13.4	16.4	15.9
C16:1	2.8	1.0	.8	.7	2.5
C18	1.8	.9	1.2 <sup>d</sup>	1.4 <sup>d</sup>	2.0
C18:1	2.7	1.3			3.4
C18:2	10.4	3.1	9.1	14.7	13.2
C18:3	62.2	6.4	73.1	79.1	61.3

<sup>a</sup>This study.

<sup>b</sup>Hayland, et al., 1976.

<sup>c</sup>Carton, G.A., 1960.

<sup>d</sup>Includes both C18 and C18:1.