

EPIDEMIOLOGICAL FEATURES OF THE SELENIUM STATUS
IN CATTLE OF NORTHERN CALIFORNIA

H.F. Mayland, USDA-ARS, Kimberly, Idaho 83341;

J.D. Williams, previously UC-Davis, NSW Dept. of Agr., P.O. Box 488, Lismore, NSW, Australia

B.B. Norman, W.H. Johnson and J.R. Dunbar, University of California, Davis, California.

Summary

Selenium (Se) is an essential mineral for animal health. Deficiencies result in white muscle disease (WMD), retained placenta, and ill thrift in cattle. These health problems are known to occur in some northern California herds. This survey was initiated to provide knowledge on the epidemiological features of Se deficiency in cattle in this region.

Blood samples were drawn from 10 cows on each of 10 ranches in each of the northern 22 counties. Whole blood Se and glutathione peroxidase (GSH-Px) activities were determined, and these values were statistically compared with each other and with data obtained by questionnaire. The latter included information on animal diseases, soils, forages, and general ranch descriptors like elevation and rainfall. The GSH-Px spot test produced either a positive (+), negative (-) or intermediate (+/-) enzyme activity classification for each cow. Herd classification was identified as + or - if >70% cow-values fell in that class, otherwise it was identified as +/-.

The overall geometric mean blood-Se value was 47.6 ng/ml. Regressions of individual and herd GSH-Px-class against blood-Se values produced $r = .75$ and $r = .82$, respectively. Significantly low blood-Se levels were recorded in herds with: 1) negative GSH-Px values, 2) past histories of WMD and ill thrift, 3) predominantly hay diet, 4) pregnant and early lactating cows, and 5) pure bred cows. The Se status of herds using Se-salt blocks was not different from non-supplemented herds. Descriptive ranch variables did not reliably assess the Se status of herds in this study.

Introduction

Selenium deficiency in grazing animals is known to occur in the eastern and western regions of the United States (Kubota and Alloway, 1972). Deficiencies are quite severe in western Oregon and Washington and in northern California. This paper reports the results of a survey undertaken to provide information on the epidemiological factors associated with selenium deficiency in northern California.

Materials and Methods

Blood samples were collected in a convenience sample of 1465 adult cows from 150 herds in 22 counties in northern California over a five month period from January to May 1980. A maximum of 10 cows in no more than 10 herds in each county were included in the survey. Samples were obtained from cows that had 1) had at least one calf, 2) been on the ranch for at least 60 days and 3) been grazing

the same pasture for at least 30 days prior to sampling. These stipulations ensured that all animals in the survey had blood selenium chemistry indicative of local management practices and environmental conditions. Nearly all cows sampled were beef cattle.

Blood samples were collected into purple-top collection tubes containing EDTA and forwarded on ice (not frozen) to Davis. At Davis, the glutathione peroxidase (GSH-Px) spot test was performed using the method of Segall et al. (1977). This procedure produced either a positive (+), negative (-) or intermediate (+/-) enzyme activity classification for each cow. These categories corresponded to values observed in selenium-supplemented calves which showed increased weight gains (+ group) and unsupplemented calves which showed poor weight gains (- group). All GSH-Px spot tests were performed within seven days of collection.

Individual GSH-Px classifications were used to assign a GSH-Px classification to the herd. A herd was arbitrarily assigned to group 1 if 70% or more cows in the sample returned negative (-) enzyme activity results. Similarly a herd was assigned to group 3 if 70% or more cows returned positive (+) enzyme activity results. Herds with GSH-Px values falling between these categories were classified as group 2.

The selenium concentration in the whole blood was determined at the USDA-ARS laboratory near Kimberly, Idaho using the fluorometric method of Olson et al (1975). The herd-selenium concentration was calculated as the mean blood selenium concentration for cows in the herd.

At the time of blood sampling, a questionnaire on various ranch and animal characteristics was completed. The ranch variables included topography, elevation, rainfall, fertilizer history, soil type, feed type, disease history, selenium supplementation history and herd size, as well as individual animal variables like production and breed data for the sample cows.

Questionnaire responses to all disease variables referred to the past disease history of the ranch and the disorders were not necessarily apparent at the time of sampling. Responses to the remaining variables referred to the status of the cows at the time of sampling and for at least one month prior to that. In many cases the animals in this survey had been under the same ranch conditions for the previous four months.

The injectable supplementation group referred to supplementation via either injections of vitamin E (vitamin ADE complex) alone or in combination with sodium selenite (vitamin E 68 IU/ml, Na selenite 10.95 mg/ml). Only one of these ranches was also

TABLE 1. SOME RESULTS OF THE ONE-WAY ANALYSIS OF VARIANCE COMPARING THE GEOMETRIC MEAN HERD SELENIUM CONCENTRATIONS BETWEEN GROUPS WITHIN GIVEN RANCH VARIABLES.

Variable	Group	No. herds in group	Group herd blood ng Se/ml ²	ANOVA level of significance (P)
Herd GSH-Px	1	52	25a	.000
	2	41	43b	
	3	49	94c	
White muscle disease	No	110	53a	.000
	Yes	27	31b	
	?	13	38ab	
Illthrift	No	107	52a	.006
	Yes	26	33b	
	?	17	40ab	
Weak calf syndrome	No	102	50a	.083
	Yes	39	38a	
	?	9	48a	
Retained placenta	No	71	45a	.76
	Yes	71	48a	
	?	8	52a	
Undiagnosed abortions	No	74	48a	.84
	Yes	61	46a	
	?	15	43a	
Goiter	No	125	48a	.16
	Yes	10	52a	
	?	15	34a	
Feed	Pasture	36	58a	.009
	Both	88	46ab	
	Hay	26	34b	
Supplementation	No	128	46a	.53
	Vit E ± Se injection	14	56a	
	Se Salt blk	8	42a	
Great soil groups	Alfisol	35	50a	.45
	Entisol	10	44a	
	Inceptisol	15	48a	
	Mollisol	25	38a	
	Vertisol	15	57a	
Elevation	0- 999'	87	46ab	.006
	1000-1999'	25	60b	
	2000-2999'	13	51ab	
	3000-3999'	8	21a	
	4000-4999'	13	44ab	
5000' +	4	58ab		

¹See text for description of GSH-Px, feed and supplement groups. Disease variables are: no history of the disease (No), disease has occurred in herd (Yes), or history not known (?).

²Variable-group means followed by a similar letter are not different (P = .05).

supplementing with selenium salt blocks.

Logarithmic (base 10) transformation of individual and herd selenium data allowed all variables used in the study to be normally distributed (BMDP 2D) for subsequent analyses using the BMDP medical Computer Programs (Dixon and Brown, 1978). One-way analysis of variance (BMDP 7D) was made on the transformed blood selenium data, as the response variable, and the different groups within each descriptor (Table 1). Differences between group means were evaluated at $P \leq .05$ (Kleinbaum and Kupper, 1978). Duncan's multiple range test was used for pairwise testing for differences between herd means at different elevations. The data were also analyzed (BMDP 1F) by the Chi Square frequency distribution test (Remington and Schork, 1970) to compare the results of the one-way analysis of variance (Table 2).

The variances of the selenium response data for individual animals were not homogenous. Thus Kruskal-Wallis non-parametric one-way analysis of variance (BMDP 3S) was used to statistically ascertain the effect of animal groupings (Table 3). Differences between groups or ranks were evaluated at $P = .05$ using Dunn's multiple comparison procedure (Hollander and Wolfe, 1973). Simple linear regression of GSH-Px on herd selenium values and animal GSH-Px on whole blood selenium concentrations was performed using BMDP 1R.

A non parametric discriminant analysis using the classification of Anderson (1966) was conducted using herd GSH-Px and all other ranch variables in a biased data base (BMDP 7M). The 50 lowest selenium value herds (range 6.4 to 31.7 ng Se/ml) and the highest value herds (range 62 to 219 ng Se/ml) were used to determine if the remaining ranch variables had any ability to discriminate herd selenium status.

Since there was no significant difference between supplemented and non-supplemented herds (Table 1) no further distinction was made between these subpopulations in any analyses of the data.

Results

The overall geometric mean blood selenium level for cows in this study was 47.6 ng/ml. Below average county values occurred in the coastal region above Sonoma, the northern mountain regions, north central Sierra Nevadas and eastern Sacramento Valley. Above average values occurred in the north central coastal, western Sacramento Valley and Trinity Lassen and Plumas counties in the mountain region. A significant correlation ($r = .75, P < .0001$) was measured between the individual animal GSH-Px spot test classifications and individual blood selenium concentrations. A significant correlation ($r = .75, P < .0001$) was also measured between the herd GSH-Px spot test classifications and herd blood selenium concentrations.

Tables 1 and 3 detail the geometric group means of blood selenium for some of the variables. The geometric means are the antilog_{10} values of the means of the initial logarithmic transformation of the blood selenium data. Table 2 lists the results of the Chi squared frequency distribution analysis. The following significant details can be extracted from the data summaries.

TABLE 2. RESULTS OF THE CHI SQUARE FREQUENCY DISTRIBUTION ANALYSIS OF VARIABLE GROUPS VERSUS LOW OR HIGH SELENIUM HERDS.

Variable	Degrees of freedom	X ² value	Level of significance
Ranch variables			
Herd GSH-Px	2	84.	.000
White muscle disease	2	7.55	.022
Illthrift	1	4.95	.027
Weak calf syndrome	1	2.84	.11
Retained placenta	2	2.61	.27
Undiagnosed abortions	2	.28	.87
Goiter	2	1.30	.52
Feed	2	7.86	.017
Supplementation	2	7.05	.025
Great soil groups	5	3.02	.70
Elevation	5	5.76	.33
Herd size	4	4.17	.38
Rainfall	8	8.23	.41
Topography	4	6.86	.14
Time on ranch	4	7.97	.093
Time on pasture	5	3.99	.55
Fertilization	1	.03	.86
Animal variables			
Production	4	54.	.000
Breed			
Purebred vs crossbred	1	14.3	.001
Hereford vs Angus vs crossbred	2	6.4	.041
Animal GSH-Px	2	834.	.000

Herds reporting the occurrence of white muscle disease had a mean blood selenium level of 31 ng/ml whereas those not reporting the disorder had a mean value of 53 ng/ml. An arbitrary level of 40 ng Se/ml was selected as a critical level to categorize herds into selenium deficient (<40 ng Se/ml) or selenium adequate (>40 ng Se/ml). Herds in which illthrift occurred also had lower blood selenium (33 vs 52 ng/ml) than did herds free of illthrift. Weak calf syndrome occurred in herds with low selenium levels (38 vs 50 ng/ml), but this relationship was significant only at P = .083. Retained placenta and undiagnosed abortions were not related to herd blood selenium (P = .76 and .84, respectively) in this study.

Significantly higher (P = .009) blood selenium levels were associated with herds grazing pasture (58 ng/ml) than those fed hay (34 ng/ml). Blood selenium values were intermediate (46 ng/ml) for herds grazing pasture and receiving hay. Low blood selenium (21 ng/ml) was associated with herds on ranches lying at 3000 to 3999 feet elevation (900 to 1200m) and this was less (P ≤ .05) than the high value (60 ng/ml) for herds maintained on ranches at 1000

to 1900 feet (300 to 600m). All other comparisons of elevational groupings were non-significant (P = >.05).

No significant difference in mean blood selenium levels was observed between herds that were 1) injected with vitamin E and or selenium, 2) given access to selenium salt block or 3) not given any supplement (Table 1). However, under Chi squared analysis, significantly (P = .025) more ranches using injectable vitamin E alone or in combination with selenium were observed than expected with blood levels above 40 ng Se/ml (not shown). A similar difference was not observed for the ranches using selenium salt block supplementation or no supplementation.

Dry pregnant and early (0 - 3 months) lactation cows had lower blood selenium levels than non-pregnant or late (over 3 months) lactation cows (Table 3). This relationship was also supported by the Chi squared analysis as many more than expected dry pregnant and early lactation cows were recorded in the low selenium group.

Both statistical procedures (Tables 2 and 3) reveal purebreds to have lower blood selenium concentration than crossbreds. No difference (P ≤ .05) was observed in blood selenium values in Hereford and Angus, but Angus cows had significantly (P ≤ .05) lower selenium values than the Hereford-Angus crossbred cows.

The 3 GSH-Px groups (-, +, ±) were each associated with significantly (P < .000) different blood selenium concentrations when based on either mean herd values (Tables 1 and 2) or individual animal data (Table 3).

Using non parametric techniques and a biased data base of the 50 lowest and 50 highest blood selenium herds, good discrimination was obtained using three variables; herd GSH-Px, herd size and ranch history of retained placenta to correctly classify 80% of the herds into the low or high selenium categories. Similar techniques correctly classified 72% of the low and high selenium categories when the discriminating variables included feed, white muscle disease, retained placenta and time on the ranch. Parametric techniques indicated that the last four variables correctly accounted for 67% of the herd selenium when using the biased data base. Good discrimination of herd selenium status was only achieved in this study by use of the GSH-Px spot test data in combination with some of the simple ranch variables.

Discussion

Biasing of this survey was unavoidable, but nevertheless imposed some statistical limitations and results can be extrapolated only cautiously. Sampling criteria minimized a source of error in that blood selenium levels respond rapidly to changes in selenium intake while GSH-Px would lag by 10 to 30 days (Hoffman et al, 1978). The lower correlations between blood selenium levels and GSH-Px values, when compared with other studies (Scholz and Hutchinson, 1979), may have resulted from limiting the GSH-Px classification to three categories.

The selenium uptake in forage plants is determined by the availability of soil selenium. Availability is a function of soil type, form and level of selenium and sulfur and soil pH. Soils of volcanic origin generally have low selenium levels while sedimentary soils have higher levels (Muth and Allaway, 1963). Many soils in northern California are of volcanic origin. For purposes of the survey,

TABLE 3. SIGNIFICANT RESULTS OF KRUSKAL-WALLIS ONE WAY ANALYSIS OF VARIANCE (X^2) COMPARING MEAN BLOOD SELENIUM CONCENTRATIONS BETWEEN DIFFERENT ANIMAL VARIABLE GROUPS AND THE LEVEL OF SIGNIFICANCE (P).

Variable Group	# Cows in group	Blood Se ng/ml ^{1/}	df	X^2	P
Production					
Dry, nonpregnant	71	63a	4	90	.000
Dry, pregnant	318	40b			
Lactating, < 3 mo.	517	42b			
Lactating, > 3 mo.	444	51a			
Dry	15	45ab			
Breed					
Purebred	976	42a	1	25	.000
Crossbred	390	50b			
Hereford	747	42ab	2	2/	.025 ^{2/}
Angus	195	40a			
Hereford X Angus	178	49b			
GSH-Px					
GSH-Px 1	587	25a	2	866	.000
GSH-Px 2	210	39b			
GSH-Px 3	578	82c			

¹Variable-group means followed by a similar letter are not different (P = .05).

²These data were analyzed by the common one way analysis of variance.

the predominate great soil group occurring on each ranch was identified even though other soils were often present.

Contrary to the well established relationship between selenium deficiency* and retained placenta (Julien et al., 1976) it appears that the low incidence (< 10% in reporting herds) here may involve some other factor. The occurrence of illthrift in this area, however might respond to selenium supplementation (Andrews et al., 1976).

Blood selenium in herds provided the FDA approved selenium levels of 20 ppm were not different from those values in non selenium herds (P = .53). Substantially higher selenium concentrations (300 ppm) are undoubtedly required in northern California (Hathaway et al., 1980).

Crossbred animals had higher selenium than purebreds in agreement with the findings of Langlands et al. (1980). They reported that genotype was a significant source of variation in blood selenium and GSH-Px in cattle and sheep. It is postulated that the physiological demands for selenium are higher during pregnancy and early lactation, thus depressing blood selenium values below those measured in other groups.

The results of this survey have confirmed the good relationship of GSH-Px with blood selenium concentrations in cattle and verified that GSH-Px is a good indicator of animal and herd selenium status. It is also apparent that many cattle sampled would respond to increased selenium supplementation.

Anderson, T.W. 1966. Some non-parametric multivariate procedures bases on statistically equivalent blocks. P.R. Krishnaiah (ed.) In: Proc. Int'l. Symp. Multivariate Anal. Dayton, Ohio. 1965. Academic Press, NY.

Andrews, E.D., W.J. Hartley and A.B. Grant. 1968. Selenium responsive diseases of animals in New Zealand. N.Z. Vet. J. 16:3.

Dixon, W.J. and M.B. Brown (eds.) 1979. BMDP Biomedical Computer Programs, P-series. Univ. of California Press. Berkeley. 880p.

Hathaway, R.L., J.E. Oldfield, M.R. Buettner, M. Hansen and G.E. Carter. 1980. Effective levels of selenium in mineral-salt mix for supplementing grazing heifers. Proc. Western Section Am. Soc. Anim. Sci. 31:223.

Hoffman, C., B. Rivinus and L. Swanson. 1978. Effect of intramuscular administration of selenium and vitamin E in dairy heifers on erythrocyte glutathione peroxidase activity and blood selenium levels. J. Anim. Sci. 47:192.

Hollander, M. and D.R. Wolfe. 1973. Non parametric statistical methods. John Wiley and Sons. NY. p 115-132.

Julien, W.E., H.R. Conrad and A.L. Moxon. 1976. Selenium and vitamin E and incidence of retained placenta in parturient dairy cows. J. Dairy Sci. 59:1954.

Kleinbaum, D.G. and L.L. Kupper. 1978. Applied regression analysis and other multivariate methods. Duxbury Press. North Scituate, Mass. p 263-265.

Kubota, J., W.H. Allaway, D.L. Carter, E.E. Cary and V.A. Lazar. 1967. Selenium in crops in the United States in relation to selenium responsive diseases of animals. J. Agric. Food Chem. 15:448.

Langlands, J.P., J.E. Bowles, G.E. Donald, T.S. Chang, R. Evans, H. Hearnshaw and T.B. Post. 1980. Genotype as a source of variation in selenium and glutathione peroxidase activity of whole blood from grazing sheep and cattle. Aust. J. Agric. Res. 31:839.

Muth, O.H. and W.H. Allaway. 1963. The relationship of white muscle disease to the distribution of naturally occurring selenium. J. Am. Vet. Med. Assoc. 142:1379.

Olson, O.E., I.S. Palmer and E.E. Cary. 1975. Modification of the fluorimetric method of selenium in plants. J. Assoc. Official Anal. Chemists. 58:117.

Remington, R.D. and M.A. Schork. 1970. Statistics with application to the biological and health sciences. Prentice-Hall. New Jersey. p 229-244.

Scholz, R.W. and L.J. Hutchinson. 1979. Distribution of glutathione peroxidase activity and selenium in the blood of dairy cows. Am. J. Vet. Res. 40:245.

Segall, H.J., D.M. Siegel, B.B. Norman and M.N. Oliver. 1977. A rapid screening blood spot test for selenium-responsive disease in cattle. Calif. Vet. 31:10.