



Project Summary

Effects on Cattle from Exposure to Sewage Sludge

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This research was initiated to examine the effects of sewage sludge disposal on agricultural land and the potential for transmitting sludge contaminants into the food chain. Soils, forages, and cattle grazing on a sludge disposal site were examined for trace metals and persistent organics. Cattle were also fed diets containing digested sludge to simulate worst-case conditions of cattle grazing on sludge-fertilized pastures.

Soils at the sludge disposal site had increased concentrations on Zn, Cu, Ni, Cd, and Pb. Forages from these soils treated with sludge had higher levels of Zn, Cd, Cu, and Ni and lower Pb concentrations than forages from soils that had not received sludge.

Cattle fed digested sewage sludge as a percentage of their diet remained healthy. The sludge had no positive or negative effects on cattle performance other than to act as a diet diluent. The direct ingestion of sewage sludge led to increased levels of Cd and Pb in kidney and liver tissues. The amount of Cd increase was related to: (1) the concentration of Cd in the diet; (2) the Cd source (Fort Collins sludge versus Metro Denver sludge), and (3) the time period of sludge ingestion. Concentrations of Cu, Pb, and Cd in liver tissues were shown to increase in a linear fashion during sludge ingestion. Cadmium did not decrease in liver or kidney tissues when sludge was removed from the diet; however, Cu and Pb in liver decreased significantly.

Fat tissue from all sludge ingestion studies showed significant increases in PCB's DDE, dieldrin, and oxychlor-dane. DDE and dieldrin decreased significantly when sludge was removed from the diet, but PCB's did not.

The effects to cattle from exposure or ingestion of sewage sludge appear benign. Thus, the principal health hazards associated with grazing cattle on sludge-fertilized pastures would be an increased level of heavy metals entering the human food chain through kidney and liver consumption.

Additional studies were conducted measuring the die-off rate of pathogens in liquid sludge as the sludge dried in earthen basins. Analysis showed that as the sludge dried, the number of fecal coliforms, total coliforms, salmonellae, f2 and T7 bacteriophage, and *Ascaris* ova decreased to low levels within periods ranging from weeks to months.

This Project Summary was developed by EPA's Municipal Environmental Research Laboratory, Cincinnati, OH, to announce key findings of the research project that is fully documented in a separate report of the same title (see Project Report ordering information at back).

Introduction

Although sewage sludge contains a number of valuable nutrients and humus materials that can be used to increase soil fertility, problems are associated with the recycling of sewage sludge. Much of our sewage is not only derived from garbage or fecal material but is the product of an industrial society; thus, heavy metals and persistent organics, many of which are products or byproducts of industry, turn up in sludge. In addition to these elements and compounds, there is a disease potential. The problems associated with many of these elements, compounds, and biological agents become particularly acute when they are potentially harmful to crops, animals, or the human food chain. The principal objective of this research was to

examine cattle exposed to varying amounts of sewage sludge to determine whether and to what extent contamination of cattle tissue had occurred as a result of sludge exposure. To accomplish this objective several separate, but interrelated, studies were conducted.

The Metropolitan Denver Sewage Disposal District No. 1 (Metro Denver) had been recycling sewage sludge to land since 1969. The sludge had been applied to the soil surface, plowed under, and planted to forages such as winter wheat, oats, sorghum, or sudan grass. A herd of beef cattle that had been on the sludge recycling site since the inception of sludge disposal there subsequently grazed these forages. These cattle had been exposed to sewage sludge for 6 years and had ample opportunity to ingest contaminants by consuming sludge enriched forage or by direct ingestion of soil and sludge. This situation represented a unique opportunity to examine some long-term effects of cattle exposure to sludge from a somewhat typical ongoing practice.

Soils, forages, and cattle from the sludge disposal site (the abandoned Lowry Bombing Range or LBR) were examined for heavy metals and persistent organics. Disease incidence, tissue heavy metal concentrations, and persistent organics in fat tissues from these cattle exposed to sludge were compared with range cattle from a nearby control ranch (no sludge exposure) and with a larger group of cattle of known age and no exposure to sludge.

Kidney and liver tissues (tissues known to accumulate heavy metals) from cattle of known age and no exposure to sewage sludge were also examined so that information concerning normal background concentrations of these tissues could be developed. This information was then compared with metal concentrations found in tissues of cattle exposed to sludge.

Additional feeding studies were designed around the premise that the application of sludge to soil or pasture land will result in the direct ingestion of sludge constituents. To assess the magnitude of uptake of heavy metals and persistent organics into edible and other tissues of cattle, the effects on cattle directly ingesting sludge were examined.

Three sludge feeding studies were conducted: (1) young feed-lot steers were fed 0%, 4%, or 12% Metro Denver sludge (20 ppm Cd) for 3 months; (2) young feed-lot steers were fed 0% and 12% Ft. Collins sludge (100 ppm Cd) for 3 months; (3) older cows and young steers were maintained on 0% and 12% Ft. Collins sludge

diets for 9 months, which was followed by a 4-month withdrawal period in which no sludge was fed. The quantities of sludge were chosen to bracket likely ingestion ranges including a worst-case situation. Although cattle might ingest 12% soil, the likelihood that they would ingest 12% sludge is indeed remote. Special pains were taken to feed a digested sludge relatively high in cadmium concentration. The objective was to have enough sludge with enough cadmium to produce a measurable uptake and then attempt to relate uptake to health effects.

This report also describes work that examined the time required for the destruction of human pathogens. Pathogens such as enteric viruses, bacteria, protozoan cysts, and helminth ova have been shown to survive anaerobic digestion of sewage sludges, although the numbers of these organisms are usually greatly decreased if not eliminated by this process. Air drying in beds and storage in stockpiles has the potential to completely eliminate these organisms. Anaerobically digested liquid sewage sludge was placed in earthen drying basins. The survival of endemic salmonellae, fecal coliform, inoculated *Ascaris* ova, and inoculated f2 and T7 bacteriophages were followed until these indicator organisms could no longer be detected.

Results

Heavy Metal Levels in Cattle Tissue

Certain nonessential trace metals such as Cd and Pb have been shown to increase

in specific tissues (kidney, liver, and bone) during the life span of animals. Concentrations of Fe, Zn, Cu, Cd, and Pb in kidney and liver tissues of cattle not exposed to sewage sludge were examined to augment existing information concerning normal tissue levels of these trace metals in cattle exposed to different conditions. These cattle served as control animals for the LBR tests.

The selected cattle had known birth dates and no previous exposure to sewage sludge; the selection also provided a range of dietary regimen, sex, and age. Animals chosen for the study were: 4 beef cows from the University of Wyoming; 25 range beef cows from the same ranch in eastern Colorado; 15 dairy cows from Colorado State University; and 12 young feed-lot steers used as controls in sludge feeding trials. The 29 range cows varied in age from 5 to 15 years, the steers were approximately 18 months old, and the dairy cows ranged in age from 2 to 8 years at the time of slaughter.

Table 1 shows the mean concentrations of Zn, Cu, Cd, Pb, and Fe in kidney and liver tissues from the dairy and range cattle. The Cu concentrations in liver tissues from the dairy cattle were for the most part an order of magnitude greater than those in liver tissues from the range cattle. The large difference in liver Cu concentrations was probably because the dairy cattle received dietary trace salt supplements added directly to their feed, whereas the range cattle would not have received similar supplements. The significant difference in kidney Cd concentrations between the range cattle and dairy cattle was caused by

Table 1. Mean Concentration of Trace Metals Determined in Kidney and Liver Tissues from Control Cattle of Known Age

Tissue and Element	$\mu\text{g/g}$ (dry weight)	
	Range Cattle of Known Age	Dairy Cattle of Known Age
Kidney:		
Zn	88.4 \pm 17.6*	96 \pm 46
Cu	15.8 \pm 3.2a†	21.7 \pm 2.6b
Cd	13.4 \pm 8.0a	2.8 \pm 1.7b
Pb	3.6 \pm 1.7	1.9 \pm 1.3
Fe	225 \pm 73	293 \pm 64
Liver:		
Zn	132 \pm 25	118 \pm 26
Cu	21.3 \pm 32.9a†	216 \pm 96b
Cd	1.06 \pm 0.6	0.74 \pm 0.9
Pb	1.8 \pm 1.1	1.0 \pm 1.0
Fe	256 \pm 99	250 \pm 72

* Mean \pm standard deviation.

† Values followed by different letters are significantly different at the 1% level using Student's *t*-test.

‡ If the 4 range cows from Wyoming are excluded from this analysis, the mean of the 25 Colorado range cows becomes $9.5 \pm 3.18 \mu\text{g/g}$ Cu. The difference between range and dairy cattle is still significant.

a difference in age; the range-cattle age averaged 10 years, whereas the dairy-cattle age averaged 4 years (Figure 1). The range of Cd concentration was from 0.83 ppm in a 2-year-old dairy cow to 31.2 ppm in a 13-year-old range cow. Levels of other metals did not increase or decrease in liver or kidney tissues with the age of the animals.

Heavy Metals and Persistent Organics at the LBR Sludge Disposal Site

The heavy metal status of soils at the LBR sludge disposal site showed that concentrations of Zn, Cu, Ni, Cd, Pb, and Hg were increased (Table 2), and concentrations of As, Se, Mo, and 22 organics were unaffected by sludge amendment. Forage crops grown on these sludge-amended soils showed increased tissue concentrations of Zn, Cu, Cd, and Ni (Table 3); however, none of the metal concentrations were elevated to a phytotoxic level or to a range that would be considered abnormal for forage tissues. Concentrations of As, Se, and Hg in forage tissues were unchanged, and Pb concentrations decreased as a result of sludge applications. None of the forage tissues examined contained detectable levels of 22 persistent organics.

Heavy Metal and Persistent Organics Content of Cattle Tissues from the LBR Sludge Disposal Site

Twelve older range cows, which had been on the LBR sludge disposal site since the beginning of disposal operations, were slaughtered and various tissues sampled for heavy metals and persistent organics analysis. Heavy metal concentrations in

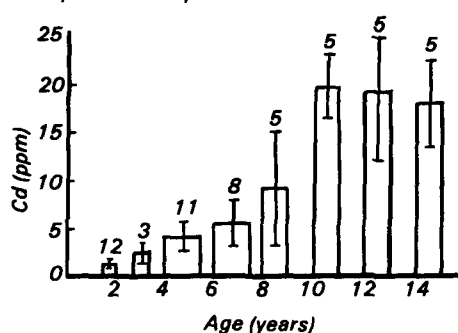


Figure 1. Mean concentration of Cd in kidney tissues (dry weight) as a function of cattle age. Numbers above bars represent number of animals for each group. Tick marks represent standard deviation for each group.

kidney, liver, and muscle tissues were, for the most part, within a range that is considered normal for older beef-type cows (Table 4). Muscle tissues, the primary food tissue, contained levels of Cd and Pb that were below detection limits. The Cu concentrations of liver tissues were unexpectedly low. These low liver Cu levels indicated that dietary Cu levels must have been low; thus, little sludge could have been consumed by the cattle.

Fat tissues were the only tissues to show any detectable levels of organic residues. Detectable levels of alpha-BHC, HCB, DDE, dieldrin, and PCB's were found in both the sludge-exposed cattle and cattle from a nearby control ranch; however, there were no differences between the two groups that appear to be attributable to sludge.

Cattle grazing the LBR sludge disposal site were healthy and showed no greater incidence of disease or death loss than other herds in the area not exposed to sludge. Postmortem pathological examination of internal organs showed no significant differences between the LBR-site cattle and a nearby control herd.

Effects of Sludge Ingestion by Cattle

Three separate experiments were carried out in which dried sewage sludge was fed to cattle (as a percentage of their diet) to simulate worst-case conditions of cattle grazing on sludge-fertilized pastures. All of the cattle in these experiments remained healthy throughout the duration of the experiments, and the sludge was shown to have no negative or positive effects on

Table 2. Mean, Median, and Range of Heavy Metals in Surface Soil Samples from Areas of Sludge Application and Control Areas Within LBR Site

Element	$\mu\text{g/g}$ (dry weight)			
	No Sludge Applications		Areas of Sludge Application	
	Mean	Range	Median	Range
Zn	50.8	39.5 - 62.5	93	40 - 252
Cd	0.43	0.18 - 0.72	1.45	0.43 - 4.40
Cu	10.6	6.75 - 18.8	11.0	8.6 - 50.0
Ni	11.8	6.0 - 43	21.8	6.00 - 43.0
Pb	18.2	14.0 - 22.0	36.2	14.2 - 101
As	<3	<3 - 3	3	<3 - 5
Se	<1	<1	1	<1 - 2
Hg	<0.027	0.010 - 0.090	0.150	<0.010 - 0.580

Table 3. Heavy Metal Concentrations of Forages from the LBR Site

Element	$\mu\text{g/g}$ (dry weight)		
	No Sludge Applications		Sludge Applied
	Mean	Median	Range
Zn	11.7	75.8	37.2 - 153
Cd	0.11	1.08	0.25 - 3.70
Cu	2.28	12.5	3.80 - 22.0
Ni	<0.57	2.80	0.25 - 15.5
Pb	2.3	0.75	<0.65 - 4.0
As	<0.25	<0.25	<0.25
Se	<0.35	<0.25	0.25
Hg	0.024	0.020	0.010 - 0.100

Table 4. Mean Tissue Concentration of Control Range Cattle (Known Age) and Sludge Exposed Cattle at LBR Site

Tissue	Animals Sampled	$\mu\text{g/g}$ (dry weight)				
		Cd	Cu	Zn	Pb	Fe
Kidney:						
Control (range)	29	13.4a*	15.8a	88.4a	3.6a	225
LBR Site	12	16a	16.1a	93a	0.8a	ND†
Liver:						
Control (range)	29	1.1a	9.5a	132a	1.8a	256
LBR Site	12	0.8a	4.6b	129a	0.3a	ND

* Values followed by different letters within a column are significantly different at the 1% level (Student's t-test).

† Not determined.

cattle performance other than to act as a diet diluent.

The first feeding study was conducted with the use of young feed-lot steers that were fed feed-lot rations containing 0%, 4%, or 12% (dry weight) Metro Denver sludge. In the second feeding study, similar steers were fed feed-lot rations containing 0% or 12% (dry weight) Ft. Collins sludge-sludge with Cd and Cu levels five times and two times, respectively, that of the Metro Denver sludge. Other metal concentrations were similar for the two sludges. The duration of both feeding studies was approximately 3 months. Analysis of kidney and liver tissues revealed significant increases of Cd and Pb because of the increased dietary levels of these metals (Table 5). The estimated availability or absorption and retention of Cd from the sludges into the cattle tissues measured 0.07% for the Ft. Collins sludge and 0.02% for the Metro Denver sludge. Not all of the cattle tissues were analyzed for uptake of heavy metals; thus, total uptake by all cattle tissues could be higher, but probably less than 1% at best. Even a 1% uptake through consumption of sewage sludge is significantly less than the 2% to 9% reported by other researchers utilizing Cd salts and a variety of other animal species; thus indicating that Cd uptake from both sources of sewage sludges were less available for tissue uptake and/or retention. Muscle tissues showed no heavy increases due to the sludge ingestion after this 3-month period.

It is known that young animals of most species absorb greater percentages of dietary heavy metals (particularly Pb) than older animals, but the extent to which this might occur in cattle had not been investigated. The feeding trials described herein used young feed-lot steers; thus, the metal uptake levels could have been higher in select animal tissues than would have occurred with older animals. Another aspect not examined in these studies was the possibility that some portion of the sludge-borne tissue contaminants could be eliminated once the sludge was removed from the cattle diet.

A third sludge-feeding study was then conducted. An older group of 16 Hereford cows (3 to 7 years old) and a group of 16 Hereford steers (6 months old) were each divided into two groups of eight each and fed maintenance diets consisting of 0% (controls) or 12% (dry weight) Ft. Collins sewage sludge. Thus, the 12% sludge diet was fed to eight cows and eight steers (two of the four treatment groups) for 9 months. At the end of the 9-month period,

four cows and four steers from each treatment group were slaughtered. The remaining animals were kept on test for another 4 months, but sludge was removed from the diet of those that had been fed sludge. All of the remaining animals were then slaughtered at the conclusion of the 4-month sludge withdrawal period. Periodic bone and liver biopsies taken over the 13-month test period permitted a more detailed investigation of uptake and depletion rates of heavy metals in these tissues.

Analyses of liver biopsies from the sludge fed cattle showed that Cd increased in liver tissues with time in a linear fashion (Figure

2). Cu and Pb also increased significantly, but decreased when sludge was removed from the diet (Figures 3 and 4).

The ingested sludge resulted in significant increases of Cd, Pb, and Zn in kidney tissues (Table 6). Cadmium concentrations did not decrease after the sludge withdrawal period, but Pb concentrations appeared to have decreased slightly during the withdrawal period.

Fat tissues were analyzed for 22 persistent organics. DDE and TNC increased significantly in fat tissues due to the sludge ingestion, but decreased significantly during the 4-month withdrawal period,

Table 5. Mean Cd, Zn, Cu, and Pb of Ft. Collins and Metro Denver Sludges and Tissue Concentration from Cattle Fed These Sludges

Tissue Types and % Sludge in Diet	$\mu\text{g/g(dry weight)}$			
	Cd	Zn	Cu	Pb
Ft. Collins Sludge:	98	1700	1700	470
Kidney - 0%	1.2 a*	93a	23a	0.95a
Kidney - 12%	14 b	96a	21b	11 b
Liver - 0%	0.19a	142a	127a	0.31a
Liver - 12%	4.9 b	132a	113a	4.3 b
Metro Denver Sludge:	21	1500	710	780
Kidney - 0%	1.1a	84a	17a	0.9a
Kidney - 12%	2.4b	82a	15b	15.8b
Liver - 0%	0.2a	87a	124a	0.2a
Liver - 12%	0.4b	101b	240b	4.6b

* Means followed by different letters within a column and tissue type are significantly different at the 5% level (Student's t-test).

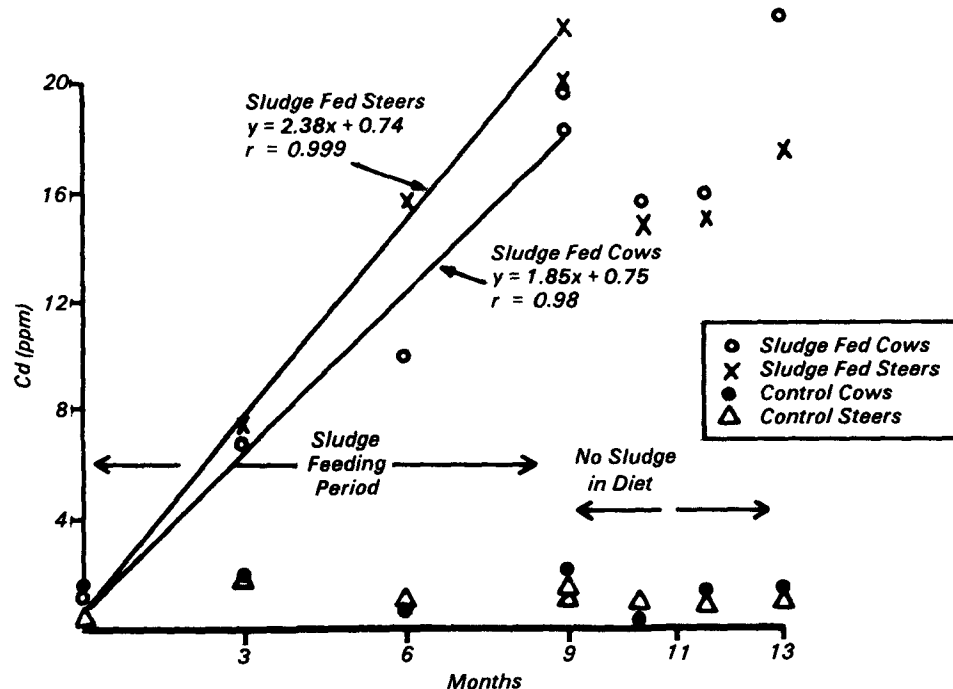


Figure 2. Cadmium concentrations (dry weight) of liver biopsies during the 9-month sludge ingestion period and 4-month withdrawal period.

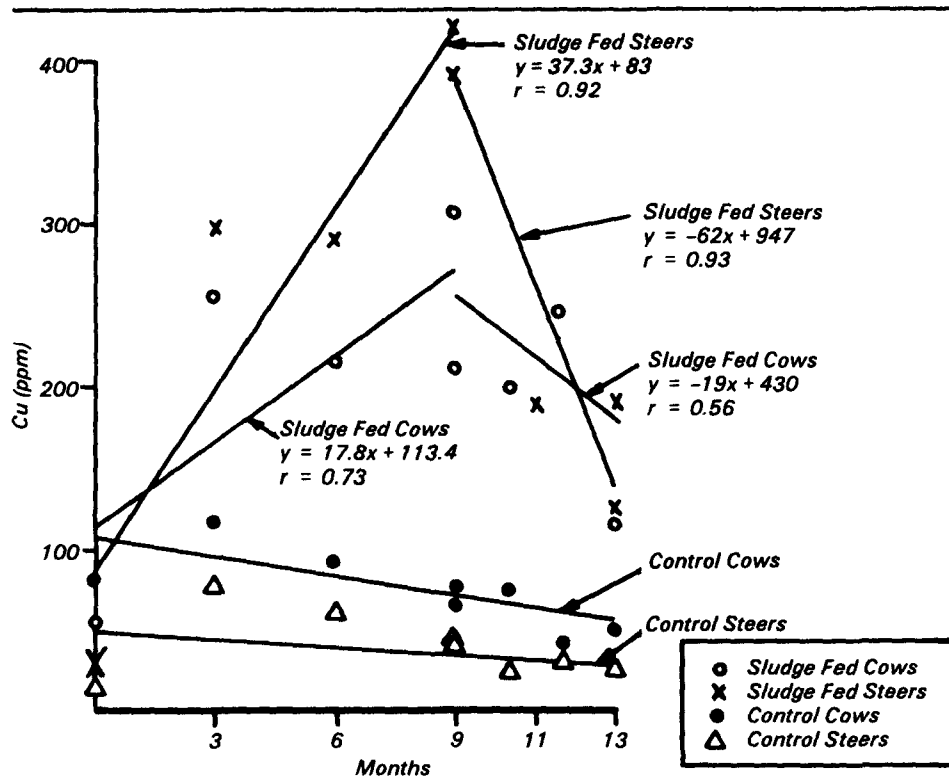


Figure 3. Copper concentrations (dry weight) of liver biopsies during the 9-month sludge ingestion period and 4-month withdrawal period.

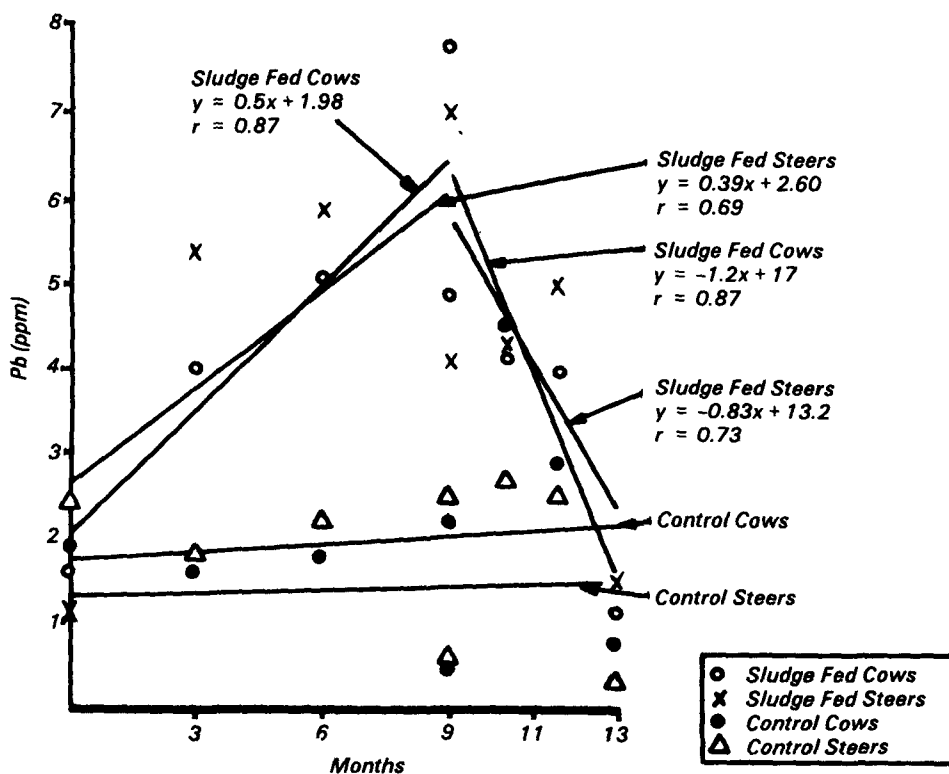


Figure 4. Lead concentrations (dry weight) of liver biopsies during the 9-month sludge ingestion period and 4-month withdrawal period.

which indicates that both compounds were either metabolized or eliminated from fat tissues. Dieldrins, PCB's, HCB, and oxy-chlordane all showed significant increases because of sludge ingestion and decreased after sludge withdrawal. These decreases were not statistically significant, however. Trace levels of DDT, heptachlor epoxide, and chlordane were found in most fat samples analyzed but were not elevated because of sludge ingestion.

Microbiological Studies of Air-Dried Sludge

The microbiological studies showed that with adequate drying and residence time in earthen drying beds, any enteric pathogens remaining in the sludge after anaerobic digestion should be destroyed. The virus indicator organism f2 had a maximum survival time of 19 weeks even though initial concentrations were orders of magnitude in excess of any enteroviruses. Salmonellae and fecal coliform bacteria persisted for 70 and 73 weeks, respectively, because of their ability for regrowth. Sludge desiccation may have been the most important factor in bringing about the disappearance of these persistent organisms. Salmonellae were not detectable after the sludge had dropped to about 30% moisture. Fecal coliforms were detectable at less than 5% moisture, but at very low concentrations. *Ascaris* ova were also sensitive to moisture. Only a small portion of the inoculated ova were viable when the sludge moisture content had dropped to 7.7%.

Storage of sludge for a year with a resulting decrease in sludge moisture to 10% or less should reduce any pathogenic organisms left after anaerobic digestion to undetectable levels. The likelihood of pathogenic microorganisms moving from the sludge into the groundwater before being inactivated is negligible.

Conclusions

The effects on cattle from exposure or ingestion of sewage sludge appear to be benign. The ingestion of high levels of sewage sludge by cattle will, however, raise heavy metal concentrations of a number of tissues (mainly kidney and liver) and will also raise the persistent organics content of fat tissues. The degree of elevation, of course, is a function of the amount of ingestion and concentration of these substances in the sludge. The data collected permit estimates of the elevation to be made as a function of the extent of ingestion.

Muscle tissue, the principal food tissue for man, was effectively screened from

excessive accumulations of the contaminants examined in this study. The entry of Cd and other heavy metals into the diet of man would be largely through liver consumption. Although high levels of Cd were reached in liver and kidney tissues of cattle consuming Ft. Collins sludge for 9 months, this was an extreme case and is not likely to occur under normal agricultural practices. It appears that cattle would act as an effective screen against these metals if sludges are properly used for the fertilization of agricultural land. Thus, if low metals sludges, such as the Metro Denver sludge, are applied to calcareous soils as a fertilizer and subsequently tilled into the plow layer, there would appear to be little potential hazard to the food chain.

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Table 6. Heavy Metals Concentrations in Kidney Tissues of Cattle After 9 Months of Sludge Ingestion and a 4-Month Withdrawal Period

Treatment	$\mu\text{g/g}$ (dry weight)				
	Cd	Pb	Zn	Fe	Cu
Cows:					
Control *	7.4a†	1.4a	77a	320a	17a
Sludge Fed‡	54.0b	4.3b	88b	281a	15a
Sludge Withdrawn §	69.0b	3.4b	131c	343a	17a
Steers:					
Control*	3.5a	1.1a	82a	255a	19a
Sludge Fed‡	57.0b	5.2b	98b	290a	16a
Sludge Withdrawn §	64.0b	3.4b	116c	247a	16a

* Average of all control animals, 9-month and 4-month withdrawal period.

† Values within a group followed by different letters are significantly different at the 5% level using Student's t-test.

‡ Cattle fed diet containing 12% sludge and slaughtered at 9 months.

§ Cattle fed diet containing 12% sludge and slaughtered after 4-month withdrawal period.

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Gerald Stern is the EPA Project Officer (see below).

The complete report, entitled "Effects on Cattle from Exposure to Sewage Sludge," (Order No. PB 83-170 589; Cost: \$16.00, subject to change) will be available only from:

National Technical Information Service
5285 Port Royal Road
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