LONG-TERM EFFECTS OF COPPER RICH SWINE MANURE APPLICATION ON CONTINUOUS CORN PRODUCTION

by

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(ABSTRACT)

Three long-term field experiments were established in the spring of 1978 and continued through 1988 to evaluate corn (*Zea mays* L.) response to high Cu levels from Cu rich swine manure and $CuSO_4$ applications. The field research was conducted on soils with diverse properties, i.e., on a Bertie fine sandy loam (Aquic Hapludults), a Guernsey silt loam (Aquic Hapludalfs), and a Starr-Dyke clay loam (Fluventic Dystrochrepts-Typic Rhodudults). Three treatments in the long-term field experiments on the three soils were a control, annual applications of Cu as Cu rich swine manure, and annual applications of Cu as $CuSO_4$ equivalent to that in the manure. After the 11 years, 1109 mt ha^{-1} of wet Cu rich swine manure were applied for the manure treatment. The Cu rich manure contained an average of 1316 mg Cu kg^{-1}. An average of 325 kg Cu ha^{-1} added to the soils from the manure application over the 11 years exceeded U.S.E.P.A. guidelines for safe copper loading levels for cropland by 45 kg ha^{-1}. Copper concentrations in corn ear leaves were within the normal range of 3 to 20 mg kg^{-1} where the 325 kg Cu ha^{-1} were applied to the soil as either Cu rich manure or $CuSO_4$. Concentrations of Cu in the grain were also in the normal range of 1 to 5 mg kg^{-1} where the high level of the two Cu sources was applied to soils. There was no decrease in corn yield on the three soils from application of either Cu source.

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Chapter 1

Introduction

At the close of 1988, there were 400,000 swine (*Sus scrofa domesticus*) on Virginia farms (Virginia Agricultural Statistics Service, 1989). At that time, 7,500 Virginia farmers were faced with the disposal of manure produced by these swine. Depending on the feeding method, the amount of wet manure produced by swine ranges from 4 to 14 liters pig^{-1} day^{-1} (Simpson, 1986). Multiplying the number of swine by the lowest production level of 4 liters manure pig^{-1} day^{-1}, the manure production of Virginia swine was at least 1,600,000 liters day^{-1}.

The use of cropland as a land base for manure disposal has been both convenient and beneficial to the farmer. Swine manure is a valuable source of nutrients and also increases soil organic matter. Because of the addition of large amounts of copper to swine feed, swine manure contains considerable copper. The application of copper rich swine manure to cropland has been questioned because of potential toxicity to crops and possible detrimental effects on the environment (Chaney, 1974; Council of Agricultural

Science and Technology, 1976; de Haan et al., 1976; Lexmond and de Haan, 1977; Lexmond, 1981).

There is a need for long-term research on the effects of copper rich swine manure application on various crops and soils. The research herein discusses the effect of copper rich swine manure and $CuSO_4$ application on continuous corn production. Grain corn, which was harvested from 119,475 hectares in Virginia in 1988 (Virginia Agricultural Statistics Service, 1989), responds well to nutrients from manure application (Brady, 1984). Due to the diversity of physiographic regions in Virginia, soil properties vary across the state. Field research for this study was conducted on diverse soils in three of the physiographic regions of Virginia to ensure wide applicability of the experimental results.

This research was initiated in 1978 and continued through 1988. Objectives of this research were as follows:

1. To determine corn response to long-term applications of copper rich swine manure and $CuSO_4$,

2. To determine environmental effects of applying copper rich swine manure and $CuSO_4$ to soils with diverse chemical and physical properties, and

3. To establish safe guidelines for the application of copper rich swine manure and $CuSO_4$ to soils for corn production.

Chapter 2

Literature Review

Use of Copper Rich Swine Feed

Copper is essential for normal metabolism in swine. The dietary requirement for Cu is approximately 6 mg kg^{-1} in the total diet (Cunha, 1977; National Research Council, 1979) and can be met by supplementing swine diets with copper sulfate, copper carbonate, or copper oxide.

The potential of Cu as a growth promoter for swine was first conceived by Braude in the 1940s, upon observing swine licking Cu pipes within their pens (Braude, 1981). This apparent craving of swine for Cu instigated research (Barber et al., 1955a,b) to ascertain if there would be a growth response to Cu supplements in swine diets. In 1955, they reported that the addition of 250 mg Cu kg^{-1} to swine diets improved the performance of growing pigs via improved feed conversion efficiency (Barber et al., 1955b). Research on Cu supplementation of swine diets was continued by others (Wallace, 1967; Hedges and Kornegay, 1973; Meyer and Kroger, 1973; David, 1974; United Kingdom

Agricultural Supply Trade Association, 1978; Aviotti et al., 1980; Braude, 1981; Cromwell et al., 1982) and was shown to stimulate growth and to increase feed efficiency at levels of 125 to 250 mg Cu kg^{-1}.

Supplementation of Cu at levels above those required for swine physiological needs improves performance, but the exact mode of action of Cu is not understood. The growth response with Cu supplementation is similar to that obtained by antibiotics in some cases (Cunha, 1977). Ludvigsen (1981) suggested that Cu may have an anthelmintic effect resulting from interaction of Cu on microflora of the intestinal tract. High levels of Cu in swine may have a metabolic effect, reducing maintenance requirements, which in turn increases growth rate and improves feed efficiency (Ludvigsen, 1981).

Copper toxicity in swine may occur at 250 mg Cu kg^{-1} in the feed and is probably due to a lack of Zn and Fe (Cunha, 1977). Harmful effects from using 250 mg Cu kg^{-1} have been prevented by feed supplementation of 130 mg Zn and 150 mg Fe kg^{-1}; however, 270 mg Fe kg^{-1} is required to counteract these effects in baby pigs (Cunha, 1977). Swine can tolerate 450 mg Cu kg^{-1} if the intake of Fe and Zn is adjusted (Ludvigsen, 1981). Copper toxicity is more likely to occur when Ca is in excess (National Research Council, 1979) and may be affected by the source and level of protein in the diet (Agricultural Research Council, 1981). The Agricultural Research Council recommends that Cu should not exceed 250 mg kg^{-1} in swine diets containing recommended levels of Fe and Zn (Agricultural Research Council, 1981).

Only about 5 percent of dietary Cu is absorbed by swine (Bowland et al., 1961; Miller et al., 1979); subsequently, the Cu content of excreta is high. Copper concentrations in manure produced by swine fed Cu-enriched diets commonly range from 600 to 2370 mg kg^{-1} of dry matter (Batey et al., 1972; Kornegay et al., 1976; Mullins et al., 1982).

Soil Copper

The concentration of Cu in U.S. soils ranges from almost 1 to over 40 mg Cu kg^{-1}, averaging 9 mg Cu kg^{-1} (Tisdale et al., 1985). Copper deficiency has been encountered in organic soils containing peat and muck (Histosols) and in highly leached acid sandy soils. Copper toxicity has occurred in soils where Cu levels have been increased through applications of manure, sewage sludge, pesticides, and mine wastes containing Cu (Tisdale et al., 1985).

Copper exists in many forms in soil. Labile forms of Cu include (Lindsay, 1979):

1. Cations in soil solution: free and complexed,

2. Nonspecifically adsorbed cations: electrostatically held on normal cation exchange sites, and

3. Specifically adsorbed cations: covalently held on surfaces of layer silicate clays, oxides of either Fe, Al, or Mn , carbonates, and organic matter.

It has been suggested that only labile forms of Cu are available to plants (Lindsay, 1979). Viets (1962) and McLaren and Crawford (1973) concluded that other Cu fractions, which include Cu in the lattice structure of primary and secondary minerals and Cu occluded in oxides and hydroxides, are not major sources of Cu for plants. Copper is absorbed by plant roots as the cupric ion, Cu^{2+}, and may also be absorbed as a component of organic complexes. Organically complexed Cu is particularly important since a very high percent of Cu in soil solution is in this form (Hodgson et al., 1966)

The concentration of Cu in soil solution is usually in the range of 0.6 to 63 ppb (Tisdale et al., 1985). The species of Cu in soil solution is influenced by soil pH, with Cu^{2+} dominant at pH values below 6.9, $CuOH^{+}$ important near pH 7.0, and $Cu(OH)_2^0$ dominant at pH values above 6.9. There is a 100-fold increase in the solubility of Cu^{2+} for each unit decrease in pH (Tisdale et al., 1985). Other important forms of Cu in solution are the complexes $CuSO_4^0$ and $CuCO_3^0$.

The plant availability of applied Cu differs with soils, because adsorption of Cu varies with pH, amount and kind of clay, and amount and properties of humus (Baker, 1974). Research has shown that with time, a substantial portion of Cu applied as Cu rich swine manure, $CuSO_4$, and sewage sludge reverts to plant-unavailable forms (Mullins et al., 1982a,b; Logan and Chaney, 1983; Payne et al., 1988). The applied copper probably converts to insoluble oxyhydroxide and organic forms (Payne et al., 1988). Baker (1974) attributes the wide range in available Cu found in literature, 0.1 to 10 mg kg^{-1}, to the numerous extraction methods used to measure soil Cu.

Research indicates limited downward movement of Cu from surface application of Cu rich manure, sewage sludge, or soluble Cu salts (Batey et al., 1972; Emmerich et al., 1982; Mullins et al., 1982a,b). During the eighth year of this study Payne et al. (1988) analysed subsoil samples of the three soils and detected only slight increases in Cu concentrations in subsoils of the Bertie fine sandy loam and the Starr-Dyke clay loam. Leaching of applied Cu was greater in the Bertie soil because it has less adsorption sites than the Starr-Dyke soil. Relatively large increases in subsoil Cu concentrations of the Guernsey silt loam were attributed to downward movement from plowing rather than from leaching. Subsoil Cu concentrations were equivalent from the manure and $CuSO_4$ sources.

Concerns exist about Cu contamination of groundwater from applying Cu to cropland. According to Public Health Service drinking water standards (Gates, 1966), a Cu concentration of 1.0 ppm in drinking water is safe for human consumption. Baker (1974) concluded it is not possible to grow crops at high enough Cu levels to render groundwater unsafe. Applied Cu could contaminate streams and lakes from erosion of topsoil containing Cu.

High levels of soil Cu have inhibited plant uptake of Zn (Schmid et al., 1965). It has been reported that Cu and Zn compete for the same absorption sites (Bowen, 1969). Indications are that high Cu levels also reduce the uptake of Fe and P (Reuther and Smith, 1952; Spencer, 1966; Adriano et al., 1971). High levels of Cu in soil have also reduced the rate of N mineralization (Broadbent and Nakashima, 1971; Quaraishi and Cornfield, 1971). This has been attributed to the toxic effect of Cu on soil microorganisms.

The distribution of Cu in soil may not be uniform since little movement of Cu occurs in soil (Jones and Belling, 1967). This might allow root proliferation to occur in soil areas containing lower Cu levels. Plants may be able to tolerate soils with high Cu levels, if their roots avoid soil areas with high Cu concentrations.

Plant Copper

Copper has various functional roles in plants. It is present in oxidase enzymes, important in photosynthesis, and involved in nitrogen fixation (Tisdale et al., 1985).

Copper deficiencies are likely to occur when Cu levels are below 4 mg kg^{-1} in mature leaves (Jones, 1972). Copper deficiency symptoms vary with crops. When deficiency begins in corn, the youngest leaves become yellow and stunted. As deficiencies become more severe, the younger leaves become pale and the older leaves die back. In advanced stages, Cu deficiency in corn resembles K deficiency, with necrosis occuring along leaf tips and edges. Copper, like most of the micronutrients, is toxic in large amounts. Jones (1972) states that Cu levels greater than 20 mg kg^{-1} in leaves are excessive. Copper toxicity symptoms include a reduction in shoot vigor, poorly developed root systems, and leaf chlorosis.

The normal range in Cu concentration for various parts of corn plants (Jones and Eck, 1973) are as follows:

1. Whole plants, at the 3-to 4-leaf stage: 7 to 20 mg kg^{-1},

2. Ear leaf at silk: 3 to 15 mg kg^{-1},

3. Stalk at silk, above ear node: 3 to 15 mg kg^{-1},

4. Stalk at silk, below ear node: 3 to 10 mg kg^{-1}, and

5. Grain at maturity: 1 to 5 mg kg^{-1}.

Copper is supplied to plant roots mainly by root interception. Differences in copper nutrition of plants can be attributed to the following genotypic differences in roots: rate of Cu absorption, root length per plant, length of root hairs, and root exudation (Tisdale et al., 1985). Another genotypic difference amongst plants that influences Cu nutrition is tissue requirement for Cu (Tisdale et al., 1985). Crops such as soybeans, pasture grasses, and rye have lower requirements for Cu than wheat, alfalfa, corn and other crops which are highly responsive to Cu (Tisdale et al., 1985). There are also genetic differences in the efficiency of plants to transport Cu from roots to shoots. This efficiency can differ within species as well as amongst species (Thomas et al., 1963; Dragun et al., 1976). Research on the effect of Cu on element accumulation by two single-cross corn hybrids revealed differences in Cu concentrations in hybrid shoots (Dragun et al., 1976). Two single-cross corn hybrids grown on the same soil, had a range of 13 to 31 mg Cu kg^{-1} in ear leaves (Thomas et al., 1963). Genetic control of element accumulation in plants has also been reported by Gorsline et al.(1964), Baker et al.(1967) and Epstein (1972). A potential exists for reducing Cu toxicity in crops by crop breeding techniques.

The uptake of Cu appears to be greatest at the end of the growing season (Fleming, 1973). Once Cu is absorbed by plants, it tends to accumulate in roots, and little translocation to shoots occurs (Dragun et al., 1976; Jarvis and Whitehead, 1981; Minnich et al., 1987). Root Cu levels were 10 times higher than shoot levels in snapbeans (*Phaseolus vulgaris* L.) that received Cu rich sewage sludge application (Minnich, 1987). Mean top concentrations of Cu in corn grown in solutions containing various amounts of Cu were well below mean root concentrations (Dragun et al., 1976). Root concentrations of Cu were much higher than leaf and grain concentrations of Cu in corn grown on soil that received long-term application of Cu rich sewage sludge (Kirkham, 1975). Copper concentrations in tobacco (*Nicotiana tobacum* L.) and orange (*Citrus* sp.) tops increased only slightly with an increase in concentration of Cu in solution, however, the increase in Cu concentration in roots was substantial (Smith and Specht, 1953; Struckmeyer et al., 1969).

Response of Corn To Copper Application

For most crops the effective concentraion of Cu at the root surface should be 0.02 to 0.04 mg kg^{-1}, although toxicity has occurred in some crops at 0.1 to 0.3 mg kg^{-1} ppm (Smith and Specht, 1953; Struckmeyer et al., 1969). Solution Cu concentrations above 0.16 mg kg^{-1} were toxic to two single-cross corn hybrids (Dragun et al., 1976). Plants grown in those solutions exhibited stunted tops, reduced root growth, and P and K deficiencies.

In a Cu toxicity study by Lexmond and de Haan (1980), the main effects of increased Cu concentration in various soils on growth and composition of corn were as follows:

1. Decreased dry matter production,

2. Increased Cu and Mn levels in plants,

3. Decreased phosphate content, and

4. Increased organic-N, free NO_3-N, and K contents.

The decrease in phosphate content was attributed to decreased uptake of P as a result of Cu injury to the root system.

Kornegay et al. (1976) determined the effect of three annual applications of 24 kg Cu ha^{-1} as Cu rich swine manure on corn production on a Groseclose silt loam (clayey, mixed, mesic, Typic Hapludults). A small increase in the Cu content of the corn ear leaf occurred from the Cu rich manure application. There were no differences in corn grain Cu content or yield from the Cu rich manure applications. Studies by Sutton et al. (1983) showed that four annual applications of 15.3 kg Cu ha^{-1} from Cu rich swine manure neither increased Cu concentrations in corn plants nor decreased corn grain yield on a Crosby silt loam (fine, mixed, mesic Aeric Ochraqualfs). Payne et al. (1988) reported that application of $CuSO_4$ supplying 415 kg Cu ha^{-1} over a 20 year period did not affect corn grain or silage yield.

Effect of Manure on Soil Organic Matter

Benefits of soil organic matter include contribution to soil carbon and plant nutrients and increased infiltration rate and retention of water in soil. Long term benefits of manure on soil organic matter levels have been reported by Jenkinson and Rayner (1977). Twenty annual applications of farmyard manure were followed by 80 years of no manure application on a plot at Rothamstead. After the 100 years, the manured plot still retained more organic C and nitrogen than the control plot that did not receive manure. Nine applications of manure on a Holtville silty clay (Typic Torrifluvents) resulted in increased water infiltration rates during the growing season (Meek et al., 1982).

High application rates of manure are necessary for significant long-term effects on soil organic matter. Many manures have a high water content. Slurries contain more than 90% water; subsequently, 1 t of slurry will add only 100 kg of organic matter to soil (Simpson, 1986). Much of the organic matter supplied by manures is lost during decomposition in soil. The final contribution of 40 t of farmyard manure to soil organic matter would be 2.5 t ha^{-1} after humification is complete (Simpson, 1986). Slurries would contribute less. Simpson (1986) suggests that annual applications of 40 t ha^{-1} of farmyard manure would be needed to increase soil organic matter. Annual application of 44 t ha^{-1} was required to maintain soil organic matter on a Typic Ochraqualf used for continuous corn production (McIntosh and Varney, 1973).

Effect of Manure on Soil Phosphorus

Fresh swine manure contains 0.6 to 2.5% P (Sommers and Sutton, 1980), which usually remains in the manure as it is not subject to significant volatilization. Swine excrete significant amounts of P in both feces (83.1%) and urine (16.9%) (Sommers and Sutton, 1980). Soluble P in swine manure averaged 43% of the total P in studies by Townshend et al. (1969). Losses of some of the water soluble P can occur through seepage and runoff.

Seventy-five to 80% of the P in manure is available to plants (Olsen and Barber, 1977). Plants can absorb P as either the $H_2PO_4^-$ ion or the HPO_4^{2-} ion. Plant uptake of $H_2PO_4^-$ is faster than it is with HPO_4^{2-}. At pH levels below 7.22, $H_2PO_4^-$ is dominant and, at pH levels above 7.22, HPO_4^{2-} is more important. The phosphate anions do not all remain in exchangeable form; they can undergo reversion due to Ca, Al, or Fe or be reincorporated into organic matter. Only part of the P supplied by pig slurry is in mineral form, which includes compounds in which P is combined with Ca, Mg, Fe, F, and Al. Phosphates can also form insoluble complexes with clay.

Organic P can comprise as much as 85% of total soil P (White, 1981), with levels ranging from almost 0 to over 0.2% (Tisdale, 1985). It is usually higher in the surface horizon of soil than in the subsoil (Tisdale, 1985). The organic portion includes organic compounds in plant and animal residues, microbial synthesis products, and living cells. Plants also can absorb some soluble organic phosphate compounds. Most of the P added to soils by manure is incorporated into the soil microbial biomass (Birch, 1961) - in a process called immobilization. The P in microbial tissue is eventually released in inorganic form in a process called mineralization, the rate of which is dependent upon both growth and decay rate of the microbial population and on how easily the microbial metabolites decompose. The residual effects of manure P in cropland are important because the amount of P applied in manure often exceeds the amount needed for crops.

Phosphorus concentrations in most plants is between 0.1 to 0.45% (Tisdale, 1985). The normal P concentrations in various corn parts according to Jones and Eck (1983) are as follows:

1. Whole plants, the 3- to 4-leaf stage - 0.4 to 0.8%,

2. Ear leaf at silk - 0.2 to 0.4%,

3. Stalk at silk, above ear node - 0.1 to 0.2%,

4. Stalk at silk, below ear node - 0.1 to 0.2%, and

5. Grain at maturity - 0.2 to 0.4%.

High P levels in soils affect uptake of Cu in plants, because phosphate adsorbs Cu in soil and high P levels reduce mycorhizal absorption of Cu (Wijesundara, 1988). Spencer (1966) reported that phosphate lessened Cu toxicity in citrus, by reducing uptake of Cu, and thereby reducing Cu concentrations in leaves and roots.

A concern with high rate of P addition to soil is the possibility of water pollution from leaching of P to ground water. Research has shown that leaching of P from land application of swine manure is insignificant (Duthion et al., 1980; Furrer and Stauffer, 1986). In lysimeter experimentation conducted by Furrer and Stauffer (1986), application of 12 t ha^{-1} of pig slurry supplying 270 kg P ha^{-1} did not increase the P content in drainage water. After application of 100 t ha^{-1} of cattle slurry on a clay soil and a sand soil, leaching of P was insignificant - less than 0.02 g m^2 (Kofoed and Klaussen, 1986). Vetter and Steffens (1981) determined that movement of P supplied by pig slurry is greater than that supplied by fertilizer. They also observed that more downward movement of P occurs in soils with already high P levels and in acid soils with low humus contents. After their long-term high applications of slurry, they found that concentrations of P in shallow groundwater were at levels acceptable for drinking water.

Runoff of soil containing high P concentrations poses a pollution hazard to surface water. Results of a study by Sherwood (1980) suggest that phosphate may be present in runoff water for 6 to 8 weeks after application of pig slurry, and that spreading of the slurry during the growing season or when a soil moisture deficit exists does not prevent phosphate presence in runoff water. Addition of P to bodies of freshwater can result in eutrophication, whereby algal productivity and decomposition of organic matter is increased with a subsequent depletion of oxygen. It has been suggested that the lower limit of advanced eutrophication occurs at a concentration of 20 $\mu g\ l^{-1}$ total P in water (Webber, 1981).

Summary

The use of growth promoting levels of Cu in swine diets, which improves feed efficiency and rate of weight gain in swine, results in manure containing high levels of Cu. Application of Cu rich swine manure has increased both available and non-available forms of Cu in soil, implying that some of the applied Cu reverts to insoluble forms. Little translocation of the absorbed Cu from roots to shoots occurs. Various studies have shown that Cu concentrations in leaf and grain are within the normal range for corn after Cu rich swine manure and high $CuSO_4$ applications. Copper phytotoxicity does not appear to occur in corn from such applications , as grain yields are not reduced. High concentrations of P and Cu in soil from Cu rich swine manure application do not appear to pose pollution risks to groundwater, as they tend to accumulate in topsoil and little downward leaching occurs. More mobile elements, such as NO_3^-, are easily leached and have polluted groundwater after slurry application (Duthion et al., 1980; Smilde, 1980; Vetter and Steffens, 1980; Unwin, 1986).

Chapter 3

Materials and Methods

Soil and Location

Long-term field experiments were initiated in 1978 on three diverse soils (Table 1) and continued through 1988 to determine the effects of Cu rich swine manure application or $CuSO_4$ on corn production. The field plots were located on a Bertie fine sandy loam (fine-loamy, mixed, thermic Aquic Hapludults), a Guernsey silt loam (fine, mixed, mesic Aquic Hapludalfs), and a Starr-Dyke clay loam (fine-loamy, mixed thermic Fluventic Dystrochrepts-clayey, mixed, mesic Typic Rhodudults). These soils are located in the Atlantic Coastal Plain, Ridge and Valley, and Piedmont regions of Virginia, respectively.

Table 1. Properties of soils in 1978 prior to initiation of long-term field experiments.

Field Experiment Location	Soil Series	Soil Texture	Soil pH	Organic Matter	Soil CEC†
				%	$cmol_c\ kg^{-1}$
Ridge and Valley Region	Guernsey	silt loam	5.7	1.8	10.4
Coastal Plain Region	Bertie	fine sandy loam	6.2	1.7	5.0
Piedmont Region	Starr-Dyke Complex	clay loam	5.9	1.4	12.3

†Soil Cation Exchange Capacity.

Field Procedures

Three treatments applied in quadruplicate in a randomized complete block design at each location were as follows: 1) a control, 2) cumulative total of 1109 mt ha^{-1} Cu rich swine manure from 1978 through 1988, and 3) Cu as $CuSO_4$ applied annually at levels equivalent to that in the wet Cu rich swine manure. Annual rates of manure application varied over the 11 years. A rate of 67.2 mt ha^{-1} was applied from 1978 through 1980. This rate was then increased to exceed the maximum Cu loading levels in the soils during a shorter period of time. The maximum Cu loading levels were based on U.S. Environmental Protection Agency guidelines for sewage sludge application in agricultural soils (U.S. Environmental Protection Agency, 1983). The increased rates ranged from 134.4 mt ha^{-1} in 1981 and 1982, to 168 mt ha^{-1} in 1983 and 1984, and to 84 mt ha^{-1} in 1985 and 1986. In 1986 the maximum Cu loading rate of 280 kg ha^{-1} was exceeded and the rate of application was decreased to 67.2 mt ha^{-1} in 1987 and 1988.

Manure for field experimentation was collected from swine fed diets supplemented with Cu as $CuSO_4$. The Cu concentration averaged 251 mg kg^{-1} in the feed and 1316 mg kg^{-1} in the manure over the 11 years (Table 2). Swine were housed on sloped solid concrete floors and manure was collected daily and placed in plastic lined containers until land application. Average solid content in the manure over this period was 21 percent. The relatively high amount of solids in the manure reflect drainage of urine from the sloped floors and drying during collection. The 1109 tons of wet Cu rich swine manure supplied a cumulative total of 321 to 329 kg Cu ha^{-1} to the soils. Variation in the amount of applied Cu reflects differences in Cu concentration and percent solids in batches of manure.

Table 2. Nutrient concentrations in swine feed and manure for 1988 and for the 11-year average from 1978 through 1988.

Element	Swine Feed		Swine Manure	
	1988	11-Year Ave.	1988	11-Year Ave.
	%			
Ca	0.86†	0.94	3.8	4.0
K	0.61	0.81	1.9	1.7
Mg	0.13	0.17	0.75	0.80
N	2.5	2.7	4.9	4.5
Na	0.15	0.14	0.40	0.45
P	0.65	0.58	2.0	2.2
	$mg\ kg^{-1}$			
B	10.7	9.3	22	20
Cu	249.5	251.0	1281	1316
Fe	157.5	165.4	920	1137
Mn	29.3	39.0	194	246
Zn	77.3	67.7	440	345

†Each value is the average of triplicate analyses for two subsamples.

The pH levels of the experimental areas on the three soils were maintained above 6.5 over the 11 years through dolomitic limestone applications. Corn nutrient requirements were met by fertilizer applications based on the results of annual soil and plant tissue analyses (Tables 3 and 4). The amounts of N, P, and K applied to the plots that received swine manure were based on the estimated supply of these elements by the manure (Donohue and McCart, 1977).

"Pioneer 3140" corn was grown at a population of 57,400 plants ha^{-1} on the Bertie fine sandy loam and Starr-Dyke clay loam and at a population of 47,380 plants ha^{-1} on the Guernsey silt loam. Corn grain yields measured at plant maturity were adjusted to 155 g moisture kg^{-1}.

Soil Analyses

Soil samples were taken from the 0-to 15-cm surface layer of each plot in the spring prior to application of treatments. Soil pH was determined in water with a 1:1 soil/solution ratio after a 1 hour equilibration period, soil organic matter by a chromic acid oxidation procedure (Walkley and Black, 1934), and extractable P by the dilute HCl-H_2SO_4 method (Nelson et al., 1953). Soil CEC was measured by atomic absorption spectrometry after saturation with 0.5M Ca acetate (pH 7.0) followed by displacement with neutral 0.5 M Mg acetate (pH 7.0) (Rich, 1961). Extractable soil Cu and Zn were determined by the DTPA (diethylenetriamenepentaacetic acid) method (Baker and Amacher, 1982).

Table 3. Cumulative amounts of macronutrients as inorganic salts and of swine manure applied from 1978 through 1988 for treatments.

Treatment No.	Treatment Description	Macronutrients† K	Macronutrients† N	Macronutrients† P	Swine Manure
		----------kg ha^{-1}----------			mt ha^{-1}
		Guernsey silt loam			
1	Control	1755	2380	400	-------
2	Swine Manure	115	56	159	1109
3	$CuSO_4$	1755	2380	400	-------
		Bertie fine sandy loam			
1	Control	1742	2212	355	-------
2	Swine Manure	22	90	----	1109
3	$CuSO_4$	1742	2212	355	-------
		Starr-Dyke clay loam			
1	Control	2020	2408	562	-------
2	Swine Manure	22	349	----	1109
3	$CuSO_4$	2020	2408	562	-------

†Inorganic nutrient sources were KCl, NH_4NO_3, and triple superphosphate.

Table 4. Cumulative amounts of micronutrients applied as inorganic salts from 1978 through 1988 for treatments.

Treatment		Micronutrients†			
No.	Description	B	Cu	Mn	Zn
		--------kg ha^{-1}--------			
		Guernsey silt loam			
1	Control	8.8	------	56	124
2	Swine Manure	8.8	------	56	124
3	$CuSO_4$	8.8	329	56	124
		Bertie fine sandy loam			
1	Control	7.7	------	224	124
2	Swine Manure	7.7	------	224	124
5	$CuSO_4$	7.7	326	224	124
		Starr-Dyke clay loam			
1	Control	7.7	------	56	107
2	Swine Manure	7.7	------	56	107
3	$CuSO_4$	7.7	321	56	107

†Inorganic nutrient sources were sodium borate and copper, manganese, and zinc sulfates.

Swine Feed And Manure Analyses

Nutrient concentrations in swine feed and swine manure were determined by the same procedures. Samples were dried at 105°C for 16 hours and ground to pass a 20-mesh screen. One gram subsamples were dry ashed at 450°C for 3 hours and leached with acidic solution. The amounts of Ca, Cu, Fe, K, Mg, Mn, Na, and Zn in solution were then determined by atomic absorption spectrometry. Nitrogen was determined by a micro-Kjeldahl technique (McKenzie and Wallace, 1954) and P by a molybdivanadophosphoric acid method (Kitson and Mellon, 1944). Boron in the samples was determined by an Azomethine-H procedure (Parker and Gardner, 1981). Manure samples were dried at 105°C for 16 hours before percent solids were determined gravimetrically.

Plant Tissue Analyses

Ear leaves were sampled at the early silk growth stage and grain at plant maturity. The plant tissue was dried at 70°C for 16 hours and ground to pass a 20-mesh screen. B, Ca, Cu, K, Mg, Mn, N, P, and Zn concentrations in the ear leaf tissue were obtained by the procedures used for these determinations in swine feed. One-half gram subsamples of grain were digested in a HNO_3-$HClO_4$ acid mixture, and the Cu and Zn in the digests were determined by atomic absorption spectrometry.

Statistical Analyses

Plant and soil data were evaluated by LSD mean separation procedures at the 0.05 level of significance (Steele and Torrie, 1980).

Chapter 4

Results and Discussion

Long-term field experiments were initiated in 1978 and continued through 1988 to determine the effects of high Cu levels from Cu rich swine manure and $CuSO_4$ applications on continuous corn production. A cumulative total of 1109 mt ha^{-1} of wet Cu rich swine manure applied over the 11 years supplied 321 to 329 kg Cu ha^{-1} (Table 5). Applications of Cu as $CuSO_4$ were equivalent to that in the manure. The effects of applied Cu on corn production were evaluated by measurement of corn grain yields, nutrient concentration in corn tissue, soil properties, and soil levels of Cu and P.

Corn Grain Yields

Corn grain yields were not decreased on the three soils under study by the high rate of Cu appplication over the 11 years as either manure or $CuSO_4$ (Table 5). Similar findings on the effect of Cu rich swine manure application on corn grain yields have been

Table 5. Effect of 11 annual applications of copper on copper concentration in corn grain and on corn grain yields.

Copper Treatment		Grain Copper	Grain
Amount	Source	Concentration	Yield
kg Cu ha^{-1}		mg kg^{-1}	kg ha^{-1}
		Guernsey silt loam	
0	Control	2.1ab†	11,830a
329	Swine Manure	2.0b	12,590a
329	$CuSO_4$	2.2a	11,170a
		Bertie fine sandy loam	
0	Control	2.2a	8,080a
326	Swine Manure	2.0a	8,180a
326	$CuSO_4$	2.3a	8,270a
		Starr-Dyke clay loam	
0	Control	2.1a	7,830a
321	Swine Manure	1.9a	9,660a
321	$CuSO_4$	2.2a	8,200a

†Column means for each experiment followed by different letters are significantly different at the 0.05 probability level.

reported by others (Kornegay et al., 1976; Sutton et al., 1983). Three annual applications of 24 kg Cu ha^{-1} as Cu rich swine manure did not decrease corn grain yields on a Groseclose silt loam (clayey, mixed, mesic Typic Hapludults) (Kornegay et al., 1976). A study by Sutton et al. (1983) showed that four annual applications of 15.3 kg Cu ha^{-1} from Cu rich swine manure did not depress corn grain yield on a Crosby silt loam (fine, mixed, mesic Aeric Ochraqualfs). Much higher levels of Cu were used in this study (a cumulative average of 325 kg Cu ha^{-1}) than in the research conducted by Kornegay et al. (1976) or Sutton et al. (1983). Wijesundra (1988) reported that 22 annual applications of $CuSO_4$, supplying up to 469 kg Cu ha^{-1}, did not decrease corn grain yields on a Davidson silty clay (clayey, kaolinitic, thermic Rhodic Paleudult). Higher levels of Cu application than used by Wijesundra as sewage sludge did not depress corn grain yields (Page, 1974).

In 1988 corn grain yields averaged 8,180, 11,860, and 8,560 kg ha^{-1} on the Bertie, Guernsey, and Starr-Dyke soils, respectively (Table 5). These yields are relatively high for non-irrigated corn and reflect a suitable amount and distribution of rainfall during the 1988 growing season. In Virginia the average corn grain yield in 1988 was 4955 kg ha^{-1} (Virginia Agricultural Statistics Service, 1989). The higher yields obtained in these field experiments can be attributed to appropriate fertilization of the corn, as evidenced by plant tissue analysis. That is, concentrations of B, Ca, K, Mg, Mn, N, P, and Zn (Tables 6 and 7) were within the normals for ear leaves at the early silk growth stage (Jones and Eck, 1973).

The higher yield on the Guernsey silt loam can not be attributed to soil productivity, because this soil has been placed in Soil Productivity Group IV - below average in yield (Donohue and Hawkins, 1979). The Bertie fine sandy loam is also placed in Soil

Table 6. Effect of 11 annual applications of copper as either copper sulfate or swine manure on macronutrients in corn ear leaves on three soils.

Copper Treatment		Macronutrients				
Amount	Source	Ca	K	Mg	N	P
kg Cu ha^{-1}		%				
		Guernsey silt loam				
0	Control	0.35a†	2.0a	0.21a	3.0a	0.26a
329	Swine Manure	0.37a	2.1a	0.19a	3.2a	0.25a
329	$CuSO_4$	0.34a	2.0a	0.21a	3.1a	0.25a
		Bertie fine sandy loam				
0	Control	0.37a	2.0a	0.26a	2.6b	0.28b
326	Swine Manure	0.37a	2.0a	0.24a	2.9a	0.30a
325	$CuSO_4$	0.39a	2.0a	0.25a	2.9a	0.26c
		Starr-Dyke clay loam				
0	Control	0.48a	2.1a	0.21a	3.0a	0.22b
321	Swine Manure	0.44b	2.0b	0.21a	3.0a	0.25a
321	$CuSO_4$	0.44b	2.1a	0.22a	3.1a	0.24ab

†Column means for each experiment followed by different letters are significantly different at the 0.05 probability level.

Table 7. Effect of 11 annual applications of copper as either copper sulfate or swine manure on micronutrients in corn ear leaves on three soils.

Copper Treatment		Micronutrients			
Amount	Source	B	Cu	Mn	Zn
kg Cu ha^{-1}		-------------mg kg^{-1}--------------			
		Guernsey silt loam			
0	Control	21.7a†	8.6a	54b	41a
329	Swine Manure	21.9a	9.6a	60a	37b
329	$CuSO_4$	21.4a	8.0a	55b	37b
		Bertie fine sandy loam			
0	Control	21.2a	8.8a	26a	45a
326	Swine Manure	20.6a	8.7a	26a	41b
326	$CuSO_4$	20.6a	8.9a	26a	45a
		Starr-Dyke clay loam			
0	Control	16.0b	9.2a	64b	30a
321	Swine Manure	16.2b	9.5a	71a	28b
321	$CuSO_4$	16.8a	9.5a	64b	31a

†Column means for each experiment followed by different letters are significantly different at the 0.05 probability level.

Productivity Group IV, whereas the Starr-Dyke clay loam falls in Soil Productivity Groups I (most productive) and II (above average). The higher yield on the Guernsey silt loam can be explained by the adequate rainfall during silking stage at that field experiment location, which was in Blacksburg, Virginia (Virginia Agricultural Statistics Service, 1989). The average corn grain yield at that location in 1988 was 5,802 kg ha^{-1}, which was considerably higher than the state average of 4,955 kg ha^{-1} (Virginia Agricultural Statistics Service, 1989).

Soil Copper

Copper concentrations in Cu rich swine manure are similar to those found in sewage sludge (Valdares et al., 1983; Williams et al., 1984). Guidelines for maximum Cu loading of cropland from sewage sludge application are based on soil cation exchange capacity and a soil pH $\geq$ 6.5 (U.S. Environmental Protection Protection Agency, 1983). For the soils under study with cation exchange capacities of 5.0 cmol kg^{-1} (Bertie fine sandy loam), 10.4 cmol kg^{-1} (Guernsey silt loam), and 12.3 cmol kg^{-1} (Starr-Dyke clay loam) and a pH $\geq$ 6.5 (Table 6), the safe Cu loading rate is 280 kg ha^{-1} based on guidelines for sewage sludge application (U.S. Environmental Protection Agency, 1983). A higher amount of Cu was applied to the three soils from either the Cu rich manure or the $CuSO_4$ application over the 11 year period from 1978 through 1988. Overall, from 321 to 329 kg ha^{-1} were added to the soil from the cumulative total application of 1109 mt ha^{-1} of wet swine manure (Table 5).

Applications of the high levels of Cu as either Cu rich manure or $CuSO_4$ increased the DTPA extractable Cu in the three soils (Table 8). Equivalent increases occurred from application of the two Cu sources. Increases in DTPA extractable Cu were not accompanied by increases in ear leaf Cu (Table 7). Plant uptake of Cu was restricted by decreased availability of Cu at the nearly neutral pH of the soils (Table 8). Copper uptake in the manure treatments may have been depressed by the high amounts of soil P and organic matter (Bingham et al., 1958; Zhu et al., 1990). Studies have shown that after plant roots absorb Cu, little translocation from roots to shoots occurs (Andrew et al., 1962; Fiskell and Leonard, 1967; Dragun et al, 1976). The combination of restricted plant uptake of Cu and root accumulation of Cu were responsible for normal Cu levels in corn ear leaves and grain.

Levels of DTPA extractable Cu in the Ap horizon were highest in the Starr-Dyke clay loam and lowest in the Bertie fine sandy loam (Table 6). Earlier research on these three soils by Mullins et al. (1982) shows that lower amounts of clay and free Fe and Mn were present in the Ap horizon of the Bertie fine sandy loam than in the Ap horizons of the Guernsey silt loam and the Starr-Dyke clay loam. This would result in less Cu sorption (Jenne, 1977) and therefore more leaching of Cu in the Bertie fine sandy loam.

Nutrient Concentration in Corn Tissue

The high level of Cu application from either source did not increase Cu concentrations in corn ear leaves as compared with the control (Table 7). Copper concentrations in ear leaf tissue from the three treatments were in the normal range of 3 to 15 mg kg^{-1} (Jones and Eck, 1973). Likewise, Cu applications from either source did not

Table 8. Effect of 10 annual applications of copper on pH, extractable copper, extractable phosphorus, and organic matter in soil.

Copper Treatment		Soil	DTPA	Ext.	Organic
Amount	Source	pH	Ext.Cu	P	Matter
kg Cu ha^{-1}			mg kg^{-1}	mg kg^{-1}	%
		Guernsey silt loam			
0	Control	7.0a†	1.6b	100b	2.3a
311	Swine Manure	6.8a	38.6a	614a	2.8a
311	$CuSO_4$	6.9a	35.4a	109b	2.5a
		Bertie fine sandy loam			
0	Control	6.8a	1.7c	149b	2.7b
308	Swine Manure	6.9a	21.2b	522a	3.1a
308	$CuSO_4$	6.7a	22.6a	152b	2.9ab
		Starr-Dyke clay loam			
0	Control	7.1a	8.2b	30b	2.5b
303	Swine Manure	7.1a	51.4a	403a	3.6a
303	$CuSO_4$	7.2a	46.4a	41b	2.5b

†Column means for each experiment followed by different letters are significantly different at the 0.05 probability level.

increase Cu concentrations in corn grain (Table 5). Levels of Cu in the grain from all treatments were in the normal range of 1 to 5 mg kg^{-1} (Jones and Eck, 1973). Normal concentrations of Cu in corn tissue reflect slight translocation of Cu from corn roots to shoots as reported by Dragun et al. (1976).

The effect of treatments on nutrient concentrations in corn ear leaves are shown in Tables 6 and 7. Concentrations of B, Ca, K, Mg, Mn, N, P, and Zn were within the normal range for ear leaves at the early silk stage (Jones and Eck, 1973). Soil levels of Zn in the manure treatments were higher than in the other treatments due to the presence of high amounts of Zn in Cu rich swine manure. Zinc is used to counteract Cu toxicity in swine (Cunha, 1977; Ludvigsen, 1981) and is excreted in swine manure. Levels of Zn in corn ear leaves were lower in the manure treatments than in the other treatments, probably due to P induced Zn-deficiency from the high levels of P in the manure (Olsen, 1972; Marschner and Schropp, 1977; Murphy et al., 1981).

Soil pH and Organic Matter

The pH of the three soils were ≥ 6.7 in the spring of 1988 (Table 6). These pH levels are in the recommended range to be used for sewage application based on U.S. Environmental Protection Agency (1983) guidelines. At high pH levels availability of soil Cu declines (Brady, 1984), resulting in less uptake of applied Cu by plants.

Organic matter content in the three soils is reported in Table 8. The percentage of organic matter was higher in the manure treatment on the Bertie fine sandy loam and the Starr-Dyke clay loam. Large increases in organic matter did not occur from application of the cumulative total of 1109 mt ha^{-1} wet swine manure. Research has shown that levels of organic matter are not easily increased with manure applications (Simpson et al., 1986). Six annual applications of swine lagoon effluent at an average rate of 53.6 cm ha^{-1} on a loamy, siliceous, thermic Arenic Paleudult and a fine-loamy, siliceous thermic Typic Paleudult did not affect soil organic matter concentrations (King et al., 1985). Simpson (1986) indicated that annual applications of 40 t ha^{-1} of farmyard manure are needed to increase soil organic matter. Since farmyard manure contains 2.5 times more organic matter than slurry (Simpson, 1986), around 100 t ha^{-1} of slurry would need to be applied annually to increase soil organic matter.

Levels of organic matter in all treatments were high to very high for Virginia soils (Donohue and Hawkins, 1979). The warm and humid climate in Virginia results in an average organic matter level of less than 2% (Donohue and Hawkins, 1979). Appropriate management procedures over the 11 years of this study may be responsible for the high to very high organic matter levels in the three soils.

The differences between the initial organic matter levels in 1978 and the levels in 1988 reflect changes in sampling depth during the study and should not be interpreted as large increases in organic matter. Organic matter levels may have been increased in all of the treatments from the addition of corn root residues and in some instances the incorporation of cornstalk residues. In a corn residue management study, Barber (1979) found that at least 18% of the carbon in corn roots and about 11% of the carbon in cornstalk residues enters the soil organic matter pool. Larsen et al. (1972) conducted an 11 year study on the effect of corn on soil organic matter levels on a Marshall silty clay loam (Typic Hapludoll) and concluded that, to prevent the loss of organic carbon through the return of cornstalk residues, the addition of approximately 6 t of carbon

ha^{-1} yr^{-1} would be required. Corn residues prevent a decline in organic matter, but large quanities are needed to increase organic matter levels. A 12 year study comparing the effect of of incorporporation of total crop residues vs. incorporation of farmyard manure on soil humus content revealed that contribution from crop residues to soil organic matter were not as great as those from manure (Hoffman and van Ruymbeke, 1980).

The beneficial effects of soil organic matter on soil properties may be partially responsible for the relatively high yields obtained on the three soils (Table 5). Soil organic matter improves water retention in the surface of soils (Brady, 1984). Improved water retention in the soil surface could prevent crust formation on the soil surface and hence increase corn germination.

Soil Phosphorus

Application of the 1109 mt of Cu rich swine manure ha^{-1} supplied 5017 kg P ha^{-1} over the 11 years and, as expected, increased the dilute extractable P in the three soils (Table 6). Levels of extractable P ranged from 403 to 614 mg kg^{-1} on manure treatments in the three soils. Phosphorus levels $\geq$ 55 mg kg^{-1} are considered to be very high based on calibration data for the soil test (Olsen and Sommers, 1982).

Amounts of P in the plant tissue were not excessively high at these levels of extractable P (Jones and Eck, 1973). After the 11 years, ear leaf P ranged from 0.22 to 0.30 percent on the three soils (Table 7). The average range of P concentration in ear leaves at silk for optimum plant growth is between 0.2 and 0.4 percent (Jones and Eck, 1973). The largest increase in ear leaf P over the control was 0.03% with the manure treatment on the Starr-Dyke clay loam.

High levels of P have been shown to interfere with plant availability of Cu (Bingham and Martin, 1956; Bingham et al., 1958; Bingham and Garber, 1960; Spencer, 1960, 1963, 1966). The high amounts of soil P may have decreased Cu uptake in the manure treatments on the three soils under study. Potentially the high levels of P in the manure treatment also could induce Zn deficiency in corn. Liberal applications of Zn in these field experiments (Table 4) probably prevented P induced Zn deficiency on the manure treatments.

In summary, applications of Cu as either Cu rich swine manure or $CuSO_4$, which exceeded U.S. Environmental Protection Agency guidelines for sewage sludge application from 41 to 49 kg Cu ha^{-1}, were not toxic to corn plants on three soils with diverse properties. Proposed explanations for the lack of toxicity are as follows:

1. Low uptake of applied Cu at the nearly neutral pH of the three soils,

2. Little translocation of absorbed Cu from roots to shoots, and

3. Decreased uptake of Cu in the manure treatments from high amounts of soil P and/or organic matter.

Chapter 5

Summary and Conclusions

Copper supplementation in swine diets improves feed efficiency and rate of weight gain. The manure produced by swine fed growth promoting levels of Cu contains high concentrations of Cu. The application of Cu rich manure on cropland has raised concerns about possible toxicity to crops and detrimental effects on the environment. The purpose of this investigation was to evaluate detrimental effects from long-term application of Cu rich swine manure.

Field experimentaion was conducted on three diverse soils for 11 years to evaluate the long-term effects of Cu rich swine manure application on continuous corn production. Objectives of this research were to determine the response of corn to high Cu levels and to establish guidelines for safe application of Cu rich swine manure on the three soils. Manure was produced by swine fed diets supplemented with an average of 251 mg Cu kg^{-1} and contained an average of 1316 mg Cu kg^{-1}. Application of 1109 mt ha^{-1} of wet swine manure over the 11 years supplied an average of 325 kg Cu ha^{-1} to the soils. The soils used in field experimentation were a Bertie fine sandy loam (fine-loamy, mixed, thermic, Aquic Hapludults), a Guernsey silt loam (fine, mixed, mesic, Aquic Hapludalfs), and a Starr-Dyke clay loam (fine-loamy, mixed, thermic Fluventic Dystrochrepts-clayey, mixed, mesic, Typic Rhodudults).

U.S. Environmental Protection Agency guidelines for safe Cu loading levels for cropland were exceeded by 45 kg ha^{-1} in the three soils. Concentrations of Cu in corn leaves and grain were within the normal range for corn and no decrease in grain yields occurred over the 11 years on the three soils. These results suggest that Cu phytotoxicity did not occur in corn at the rates of Cu rich swine manure application used in this study on these soils.

Application of the 1109 mt of swine manure supplied 5,020 kg P ha^{-1}. Levels of P in ear leaf tissue were within the normal range for corn. Since little leaching of P occurs in soil, pollution of groundwater with P is not likely. The same holds true for Cu, which is fairly immobile in soil. Since high levels of both Cu and P accumulated in the surface soil, risk of water pollution to surface waters exists if erosion of topsoil occurs.

Beneficial effects of swine manure application at these rates were increased organic matter levels and increases in available P. These factors may have contributed to the relatively high yields obtained in this study. The high amounts of phosphorus and/or organic matter on the manure treatments also may have prevented Cu phytotoxicity on the three soils.

Annual rates of manure application in this study averaged 110 mt ha^{-1} over the 11 years. Swine manure is commonly applied at a rate of 22.4 to 33.6 mt ha^{-1} to soils for corn production (Donohue and McCart, 1977). At the higher rate of 33.6 mt ha^{-1}, it would take 33 years to reach the cumulative total of 1109 mt wet manure ha^{-1} applied in this study.

Application of Cu rich swine manure on cropland should be managed so as to maximize the benefits of manure on crop production and to minimimize the detrimental effects on the environment. Several recommendations concerning application of Cu rich swine manure on cropland for corn production can be based on this research. The recommendations are as follows:

1. Soil pH levels should be maintained at levels ≥ 6.5 to reduce availability of Cu,

2. Rates of manure application should not exceed those meeting the N requirements for corn to avoid leaching of NO_3^- into groundwater,

3. Manure should not be applied under conditions and in areas conducive for soil erosion to avoid runoff of high levels of P and Cu into surface waters,

4. Plant tissue should be regularly analysed for concentrations of Cu to prevent Cu phytotoxicity, and

5. Plant tissue analysis should also be used to determine if any nutrient imbalances are occuring from manure application.

Chapter 6

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