Final draft

Report to the State of Iowa Department of Public Health on the Investigation of the Chemical and Microbial Constituents of Ground and Surface Water Proximal to Large-Scale Swine Operations

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EXECUTIVE SUMMARY

As a direct follow-up to a Centers for Disease Control and Prevention (CDC) workshop on public health issues related to concentrated animal feeding operations (CAFOs), an investigation into the chemical and microbial constituents of ground and surface water proximal to large-scale swine operations was conducted in Iowa. The goal of this study was to obtain a broad profile of the chemical and microbial constituents of both ground and surface water that were potentially hazardous to human health.

The highest levels of chemical pollutants and zoonotic pathogens were generally found in samples collected from earthen manure lagoons, which amass and store waste from swine barns until it is applied to agricultural fields as crop fertilizer. The contaminants included nutrients, common ions, trace elements, antibiotics, parasitic oocysts of the protozoan *Cryptosporidium parvum*, and bacteria that demonstrated particular resistance to several antibiotics commonly used in swine management practice as feed supplements and therapeutics.

The results of this study also demonstrated the presence of trace metals, common ions, nutrients, pesticides, antibiotics, bacteria, and parasitic oocysts in samples obtained from collection points other than earthen manure lagoons. These sites include agricultural drainage ditches, agricultural drainage wells, tile line inlets, tile line outlets, earthen lagoon monitoring wells, and a river. These findings suggest the possibility of the movement of both chemical pollutants and microbial pathogens through soil and away from their point of highest concentration, the animal manure lagoons, and by overland flow away from the site of manure application.

Although chemical pollutants and zoonotic pathogens were identified in the environment on and proximal to large-scale swine feeding operations, the sample collection sites did not appear to be in locations that could pose a direct threat to human health. However, more research is recommended to accurately determine the potential level of risk, possible pathways of exposure, and critical control points to avoid any potential exposure to humans.

BACKGROUND

Continued expansion and intensification of large-scale swine feeding operations in the United States has given rise to some important environmental, agricultural, and public health issues. Waste management practices for these operations commonly involve using open lagoons, ponds, or slurry tanks for the temporary storage of manure in a liquefied form, which is subsequently applied as fertilizer on agricultural fields. This practice, under certain conditions, may contaminate the ground and surface water in the surrounding area. Although some quantifiable contaminants originating from these intensified animal production systems have been identified in water, research on the direct and indirect human health effects of this contamination is very limited.

A Centers for Disease Control and Prevention (CDC) workshop on public health issues related to concentrated animal feeding operations (CAFOs), held June 23-24, 1998, in Washington, D.C., concluded that more work needs to be done to quantify human exposure and evaluate the health impact of CAFOs. Public health concerns regarding the potential for human exposure by way of ground and surface water to chemical pollutants and microbial pathogens contained in the liquefied manure have lead to an increase in research into the environmental impact of large-scale swine operations.

As a direct follow-up to this workshop, we conducted an investigation of the chemical and microbial constituents of ground and surface water proximal to large-scale swine operations. We measured potential chemical (pesticides, antibiotics, trace metals, common ions, and nutrients) and microbial (*Escherichia coli*, *Salmonella sp.*, *Enterococcus sp.*, *Yersinia sp.*, *Campylobacter sp.*, *Cryptosporidium parvum*) contaminants that may be hazardous to human health.

METHODS

Manure, surface water, and ground water samples were collected from farm sites in Iowa counties with a high density of swine productions. The samples collected were tested for the presence of microorganisms, antibiotics, pesticides, organic and inorganic constituents, and sterols. Because of the extensive list of analytes, three separate laboratories were used.

Farm site selection

The Iowa Department of Health (IDPH), United States Geological Survey (USGS), University of Iowa (UI), and CDC participated in farm site selection. To be included in the study, the swine operations had to (a) have more than 1,000 confined animals, as defined for "concentrated animal feeding operations" in 40 *CFR* part 122, appendix B, section 23a (10); (b) have access to agricultural fields where manure could be applied; and (c) have swine producers willing to participate in the study.

Sampling and collection points

Sampling was conducted at nine participating large-scale Iowa swine operations between October and December 1998. Samples of surface and ground water were collected with the assistance of IDPH and USGS. The goal was to obtain a composite of samples from the following collection points:

1) Liquefied manure directly from earthen waste lagoons.

2) Surface water from rivers.

3) Surface water from agricultural drainage ditches, and related water bodies adjacent to swine operations or agricultural fields where swine manure was recently applied, and 24 to 48 hours following a substantial rain.

4) Ground water from lagoon monitoring wells and private wells.

5) Ground water from agricultural drainage wells, tile line inlets, and tile line outlets adjacent to large-scale swine operations or agricultural fields where manure was recently applied, and 24 to 48 hours following a substantial rain.

The USGS provided equipment for collecting samples and containers for collecting and shipping the samples to be analyzed for antibiotics. The University of Iowa Hygienic Laboratory (UHL) provided the materials for collecting and shipping all other analytes. All samples were packaged with ice and shipped to the corresponding laboratories for analysis.

Microbiology

The UHL performed testing for five organisms: *E. coli, Enterococcus sp., Salmonella sp., Campylobacter sp.*, and *Yersinia sp*. The bacterial isolates were cultured and identified using standard protocols.[1-4] CDC's National Center for Infectious Diseases (NCID) laboratory performed antibiotic resistance tests on microbial colonies grown and isolated from the environmental samples using both disk and dilution methods.[5,6]

Parasitology

NCID's parasitology laboratory tested 13 samples for the protozoan *Cryptosporidium parvum* by identifying, and quantifying, whole oocysts using an NCID method that involves solubilizing oocyst wall antigens, and analyzing for their presence by an immunoassay.[a]

[a]NCID's parasitology laboratory developed the methods used for this analysis

Antibiotic residue analysis

The USGS laboratory in Raleigh, North Carolina analyzed samples for β-lactams, aminoglycosides, tetracyclines, sulfonamides, macrolides, and fluroquinolones using an immunoassay.[b] The results were confirmed using liquid chromatography/mass spectrophotometry.[b]

Organic and inorganic constituent analysis

UHL analyzed all 23 samples for select organic and inorganic constituents. The limit of detection and methods used in the analysis are reported in Table 1.

Table 1. Environmental study proximal to concentrated animal feeding operations (CAFOs), chemical analysis of organic and inorganic constituents, Iowa, October-December, 1998.

Constituent	Limit of Detection (LOD)	Method
Trace Metals and Common Ions		
Total Arsenic	0.01 mg/L	SM 3113B[10]
Total Barium	0.05 mg/L	EPA 200.7[9]
Total Bromide	0.5 mg/L	EPA 300.0[9]
Total Cadmium	0.02 mg/L	EPA 200.7[9]
Total Chloride	0.5 mg/L	EPA 300.0[9]
Total Chromium	0.02 mg/L	EPA 200.7[9]
Total Copper	0.05 mg/L	EPA 200.7[9]
Total Fluoride	0.05 mg/L	EPA 300.0[9]
Total Lithium	0.05 mg/L	EPA 200.7[9]
Total Mercury	0.001 mg/L	EPA 245.1[11]
Total Selenium	0.01 mg/L	SM 3113B[10]
Total Sulfate	0.5 mg/L	EPA 300.0[10]
Total Zinc	0.02 mg/L	EPA 200.7[9]
Nutrients		
Total Nitrite Nitrogen as N	0.02 mg/L	EPA 300.0[10]
Total Nitrate Nitrogen as N	0.1 mg/L	EPA 300.0[10]
Total Ammonia Nitrogen as N	0.1 mg/L	TIM 780-86T[8]
Total Kjedahl Nitrogen as N	0.1 mg/L	TIM 786-86T[7]
Total Phosphate as P	0.1 mg/L	TIM 787-86T[9]
Organic Content		
Biochemical Oxygen Demand	1 mg/L	SM 5210A[11]
Chemical Oxygen Demand	1 mg/L	EPA 410.1[9]
Total Organic Carbon	0.5 mg/L	EPA 415.1[9]
Dissolved Organic Carbon	1 mg/L	EPA 415.1[9]

[b]USGS's laboratory in Raleigh, North Carolina developed the CHARM method used for this analysis

Sterol analysis

To determine whether sterol compounds could serve as an indicator of fecal pollution, UHL analyzed six water samples collected in the vicinity of the large-scale swine operations for total sterol concentration determined as cholesterol[c].

Pesticide analysis

UHL analyzed the samples for select pesticides. The limit of detection and methods used in the analysis are listed in Table 2.

Table 2. Environmental study proximal to concentrated animal feeding operations (CAFOs), chemical analysis of select pesticides, Iowa, October-December, 1998.

Pesticide	Limit of Detection (LOD)	Method
Aldicarb	1 μg/L	EPA 531.1[13]
Aldicarb sulfone	1 μg/L	EPA 531.1[13]
Aldicarb sulfoxide	1 μg/L	EPA 531.1[13]
Carbofuran	1 μg/L	EPA 531.1[13]
Oxymyl	1 μg/L	EPA 531.1[13]
Carbaryl in methyl	1 μg/L	EPA 531.1[13]
Methomyl	1 μg/L	EPA 531.1[13]
3-hydroxy-carbofuran	1 μg/L	EPA 531.1[13]
Methiocarb	1 μg/L	EPA 531.1[13]
Propoxur	1 μg/L	EPA 531.1[13]
Atrazine	0.1 μg/L	EPA 8141[14]
Carbaryl	0.1 μg/L	EPA 8141[14]
Terbufos	0.1 μg/L	EPA 8141[14]
Fonofos	0.1 μg/L	EPA 8141[14]
Chlorpyrifos	0.1 μg/L	EPA 8141[14]
Ethoprop	0.1 μg/L	EPA 8141[14]
Phorate	0.1 μg/L	EPA 8141[14]
Carbofuran pendimethalin	0.1 μg/L	EPA 8141[14]
Permethrin	0.1 μg/L	EPA 8081A[14]
Pyrethrins	0.5 μg/L	EPA 8081A[14]
Captan	0.05 μg/L	EPA 8081A[14]
Cypermethrin	0.1 μg/L	EPA 8081A[14]

[c]UHL developed the method used for this analysis based on an adaptation of EPA SW846 Method 8270.

RESULTS

A total of 23 samples were collected. The characteristics of the nine farm sites and specific collection points are reported in Table 3.

Table 3. Environmental study proximal to concentrated animal feeding operations (CAFOs), farm characteristics and samples collected, Iowa, October-December, 1998.

Operation type	Number of Farms	Number of Swine	Number of Samples	Collection point*
Farrowing	2	8,000	4	2 lg, 2 tli
Nursery	3	42,000	7	3 lg, 1 pw, 1 tlo, 1 mw, 1 add
Finishing	4	46,300	12	3 tlo, 3 mw, 2 lg, 2 adw, 1 add, 1 rv
Totals	2 Farrowing 3 Nurseries 4 Finishing	96,300	23	7 lg, 4 tlo, 4 mw, 2 tli, 2 add, 2 adw, 1 pw, 1 rv

*****lg = lagoon; pw = private well; tli = tile line inlet; tlo = tile line outlet; mw = monitoring well; add = agricultural drainage ditch; rv = river; adw = agriculture drainage well.

Sterol analysis

The samples collected, location, and total sterol concentration determined as cholesterol are reported in Table 4. Of the six samples tested for total sterol concentration determined as cholesterol, only one sample, collected from a tile line inlet, had detectable levels.

Table 4. Environmental study proximal to concentrated animal feeding operations (CAFOs), total sterol concentration determined as cholesterol, Iowa, October-December, 1998.

Farm	Collection site	Matrix	Total sterols
1	Private Well	Water	Not detected
2	Tile Line Inlet	Water	Not detected
2	Tile Line Inlet	Water	Trace detected
3	Ag. Drainage Ditch	Water	Not detected
4	River	Water	Not detected
8	Tile Line Outlet	Water	Not detected

Organic and inorganic constituent analysis

The ranges for organic and inorganic constituents detected in the 23 samples are reported in tables 5, 6A and 6B. With the exception of nitrate and sulfate, the highest levels of organic and inorganic constituents were detected, as expected, in samples obtained from earthen manure lagoons. One sample obtained from agricultural drainage well had a nitrate level of 26 mg/L and another sample obtained from a tile line inlet had a nitrate level of 21 mg/L. A sample obtained from an earthen lagoon monitoring well had a sulfate level of 120 mg/L, the highest value recorded among the 23 samples.

Table 5. Environmental study proximal to concentrated animal feeding operations (CAFOs), analytic ranges for trace metals, common ions, nutrients, organic content detected in ground and surface water samples (n=16) and lagoon samples (n=7), Iowa, October-December, 1998.

Constituent	Limit of Detection (LOD)	Ground and surface water samples over LOD (%) n=16	Range (mg/L)	Lagoon samples over LOD (%) n=7	Range (mg/L)
Trace Metals and Common Ions					
Total Arsenic	0.01 mg/L	1 (6)	0.07	3 (43)	0.01-0.07
Total Barium	0.05 mg/L	16 (100)	0.07-0.94	5 (71)	0.14-1.6
Total Bromide	0.5 mg/L	1 (6)	1.9	0	<0.5
Total Cadmium	0.02 mg/L	0	<0.02	2 (29)	≥0.02
Total Chloride	0.5 mg/L	16 (100)	11-390	7 (100)	240-760
Total Chromium	0.02 mg/L	0	<0.02	5 (71)	0.03-0.26
Total Copper	0.05 mg/L	1 (6)	0.16	6 (86)	0.22-38
Total Fluoride	0.05 mg/L	0	<0.05	7 (100)	4.4-220
Total Lithium	0.05 mg/L	0	<0.05	1 (14)	0.08
Total Mercury	0.001 mg/L	0	<0.001	0	<0.001
Total Selenium	0.01 mg/L	0	<0.01	4 (57)	0.03-0.04
Total Sulfate	0.5 mg/L	16 (100)	2.6-120	7 (100)	6.4-15
Total Zinc	0.02 mg/L	6 (24)	0.02-0.39	7 (100)	0.16-97
Nutrients					
Total Nitrite Nitrogen as N	0.02 mg/L	12 (75)	0.03-0.15	7 (100)	0.14-1.15
Total Nitrate Nitrogen as N	0.1 mg/L	13 (81)	0.1-35	2 (29)	0.26-9.3
Total Ammonia Nitrogen as N	0.1 mg/L	5 (31)	0.1-2.8	7 (100)	620-2,000
Total Kjedahl Nitrogen as N	0.1 mg/L	16 (100)	0.1-12	7 (100)	650-2,900
Total Phosphate as P	0.1 mg/L	16 (100)	0.1-130	7 (100)	45-510
Organic Content					
Biochemical Oxygen Demand (B.O.D.)	1 mg/L	16 (100)	1-38	7 (100)	270-21,000
Chemical Oxygen Demand (C.O.D.)	1 mg/L	16 (100)	2-250	7 (100)	1,100-38,000
Total Organic Carbon (T.O.C.)	0.5 mg/L	16 (100)	1.2-85	7 (100)	400-14,000
Dissolved Organic Carbon (D.O.C.)	1 mg/L	15 (94)	1.7-83	7 (100)	240-3,700

Table 6A. Environmental study proximal to concentrated animal feeding operations (CAFOs), analytic levels of trace metals and common ions of samples at each collection site, Iowa, October-December, 1998.

		Trace Metals and Common Ions												
Farm	**Collection site**	**Arsenic (mg/L)**	**Barium (mg/L)**	**Bromide (mg/L)**	**Cadmium (mg/L)**	**Chloride (mg/L)**	**Chromium (mg/L)**	**Copper (mg/L)**	**Fluoride (mg/L)**	**Lithium (mg/L)**	**Mercury (mg/L)**	**Selenium (mg/L)**	**Sulfate (mg/L)**	**Zinc (mg/L)**
6	Ag. Drainage Well	<0.01	0.16	<0.5	<0.02	13	<0.02	<0.05	<0.05	<0.05	<0.001	<0.01	32	<0.02
6	Ag. Drainage Well	<0.01	0.11	<0.5	<0.02	11	<0.02	<0.05	<0.05	<0.05	<0.001	<0.01	66	<0.02
4	River	<0.01	0.14	<0.5	<0.02	21	<0.02	<0.05	<0.05	<0.05	<0.001	<0.01	33	<0.02
3	Ag. Drainage Ditch	<0.01	0.1	<0.5	<0.02	18	<0.02	<0.05	<0.05	<0.05	<0.001	<0.01	25	0.06
4	Ag. Drainage Ditch	<0.01	0.12	<0.5	<0.02	14	<0.02	<0.05	<0.05	<0.05	<0.001	<0.01	22	<0.02
1	Lagoon	<0.01	0.81	<0.5	<0.02	380	0.21	38	19	<0.05	<0.001	0.03	13	97
2	Lagoon	<0.01	<0.05	<0.5	<0.02	300	<0.02	<0.05	4.4	<0.05	<0.001	<0.01	18	0.16
3	Lagoon	<0.01	0.14	<0.5	<0.02	240	0.03	3.2	55	<0.05	<0.001	<0.01	6.4	8.4
4	Lagoon	0.02	1.6	<0.5	0.02	680	0.26	16	220	0.08	<0.001	0.02	9.7	25
5	Lagoon	<0.01	<0.05	<0.5	<0.02	260	<0.02	0.22	15	<0.05	<0.001	<0.01	8.6	1.8
7	Lagoon	0.01	0.4	<0.5	<0.02	390	0.11	14	21	<0.05	<0.001	0.03	13	35
8	Lagoon	0.03	0.84	<0.5	0.02	760	0.26	3.3	170	<0.05	<0.001	0.04	15	29
3	Monitoring Well	<0.01	0.24	<0.5	<0.02	26	<0.02	<0.05	<0.05	<0.05	<0.001	<0.01	120	0.06
4	Monitoring Well	<0.01	0.28	<0.5	<0.02	13	<0.02	<0.05	<0.05	<0.05	<0.001	<0.01	56	0.04
8	Monitoring Well	0.07	0.94	1.9	<0.02	390	<0.02	<0.05	<0.05	<0.05	<0.001	<0.01	2.6	0.05
8	Monitoring Well	<0.01	0.12	<0.5	<0.02	140	<0.02	<0.05	<0.05	<0.05	<0.001	<0.01	10	<0.02
1	Private Well	<0.01	0.3	<0.5	<0.02	12	<0.02	<0.05	<0.05	<0.05	<0.001	<0.01	30	<0.02
2	Tile Line Inlet	<0.01	0.08	<0.5	<0.02	13	<0.02	<0.05	<0.05	<0.05	<0.001	<0.01	30	<0.02
2	Tile Line Inlet	<0.01	0.08	<0.5	<0.02	24	<0.02	<0.05	<0.05	<0.05	<0.001	<0.01	16	<0.02
3	Tile Line Outlet	<0.01	0.12	<0.5	<0.02	20	<0.02	<0.05	<0.05	<0.05	<0.001	<0.01	14	0.02
8	Tile Line Outlet	<0.01	0.07	<0.5	<0.02	15	<0.02	<0.05	<0.05	<0.05	<0.001	<0.01	11	0.39
9	Tile Line Outlet	<0.01	0.14	<0.5	<0.02	25	<0.02	<0.05	<0.05	<0.05	<0.001	<0.01	19	<0.02
9	Tile Line Outlet	<0.01	0.23	<0.5	<0.02	12	<0.02	<0.05	<0.05	<0.05	<0.001	<0.01	34	<0.02

Table 6B. Environmental study proximal to concentrated animal feeding operations (CAFOs), analytic levels of nutrients and organic content of samples at each collection site, Iowa, October-December, 1998.

		Nutrients					Organic Content			
Farm	**Collection site**	**Nitrite (mg/L)**	**Nitrate (mg/L)**	**Ammonia (mg/L)**	**Kjeldahl (mg/L)**	**Phosphate (mg/L)**	**B.O.D. (mg/L)**	**C.O.D. (mg/L)**	**T.O.C. (mg/L)**	**D.O.C. (mg/L)**
6	Ag. Drainage Well	0.15	26	0.1	1.6	0.6	1.0	26	9.1	9.0
6	Ag. Drainage Well	0.12	5.9	<0.1	1.3	0.3	3.0	26	8.7	7.9
4	River	0.04	9.4	<0.1	0.8	0.2	1.0	2	6.4	4.7
3	Ag. Drainage Ditch	<0.02	6.0	<0.1	0.4	0.2	<0.1	<0.1	3.3	2.9
4	Ag. Drainage Ditch	0.07	13	<0.1	<0.1	0.5	2.0	34	11	6.5
1	Lagoon	0.31	9.3	1,400	1,700	180	1,000	6,700	2,200	580
2	Lagoon	0.22	0.26	650	650	45	270	1,100	290	240
3	Lagoon	0.17	<0.1	860	980	78	1,200	3,300	1,000	720
4	Lagoon	0.37	<0.1	2,000	2,900	510	20,000	54,000	14,000	3,700
5	Lagoon	0.14	<0.1	620	720	55	620	1,600	400	440
7	Lagoon	0.54	<0.1	1,400	1,800	130	1,200	5,800	2,700	1,200
8	Lagoon	1.15	<0.1	1,800	2,900	500	21,000	38,000	2,900	2,800
3	Monitoring Well	<0.02	0.1	0.2	0.2	<0.1	<0.1	11	2.5	2.1
4	Monitoring Well	<0.02	<0.1	<0.1	0.4	0.3	2.0	38	3.5	1.7
8	Monitoring Well	0.12	<0.1	2.8	12	0.6	38	250	85	83
8	Monitoring Well	0.02	6.3	<0.1	0.6	<0.1	<0.1	11	3.6	2.7
1	Private Well	0.08	<0.1	0.3	0.5	<0.1	<0.1	6	1.2	1.7
2	Tile Line Inlet	0.05	21	<0.1	0.7	0.3	<0.1	18	5.5	4.1
2	Tile Line Inlet	0.03	5.8	0.2	0.8	0.1	<0.1	<0.1	4.9	5.8
3	Tile Line Outlet	<0.02	14	<0.1	0.2	<0.1	<0.1	<0.1	2.3	3.8
8	Tile Line Outlet	0.03	19	<0.1	0.1	<0.1	<0.1	3	1.4	-
9	Tile Line Outlet	0.13	12	<0.1	1.3	<0.1	4.0	18	6.6	5.1
9	Tile Line Outlet	0.04	35	0.4	1.0	<0.1	1.0	23	9.6	7.9

Table 7. Environmental study proximal to concentrated animal feeding operations (CAFOs), antibiotic levels and microbial isolates by site, Iowa, October-December, 1998.

		Antibiotic				Microbial Isolates[a]				
Farm	**Collection site**	**Tetracycline[b] (µg/L)**	**Sulfonamide[c] (µg/L)**	**β-Lactam[d] (µg/L)**	**Macrolide[e] (µg/L)**	***Salmonella*[f] *sp.* (MPN/100 mL)**	***E. coli*[g] (MPN/100 mL)**	***Enterococcus*[h] *sp.* (MPN/100 mL)**	***Yersinia*[i] *sp.* (MPN/100 mL)**	***C. parvum*[j]**
6	Ag. Drainage Well	<1	<5	<2	<10	<30	300	4,500	<30	na
6	Ag. Drainage Well	<1	<5	<2	<10	<30	740	4,500	<30	na
4	River	<1	<5	<2	<10	<30	340	490	<30	6
3	Ag. Drainage Ditch	<1	<5	<2	<10	<30	520	610	<30	0
4	Ag. Drainage Ditch	<1	<5	<2	<10	<30	3,700	13,000	<30	0
1	Lagoon	250	>20	<2	227	740	100,000	60,000	<300	2250
2	Lagoon	11	>20	<2	<10	<300	380,000	27,000	<300	na
3	Lagoon	150	>20	<2	60	<300	<100	1,800	<300	1560
4	Lagoon	68	>20	3.5	<10	<300	270,000	390,000	<300	na
5	Lagoon	66	>20	2.1	81	<300	69,000	12,000	<300	na
7	Lagoon	540	>20	2.1	275	9,300	310,000	1,900,000	<300	na
8	Lagoon	110	>20	2.9	15	<300	140,000	38,000	<300	1230
3	Monitoring Well	<1	<5	<2	<10	<30	<10	<10	<30	9
4	Monitoring Well	<1	<5	<2	<10	<30	<10	<10	<30	13
8	Monitoring Well	<1	<5	<2	<10	<30	10	80	<30	15
8	Monitoring Well	<1	7.6	<2	<10	<30	390	910	<30	0
1	Private Well	<1	<5	<2	<10	<30	<10	<10	<30	0
2	Tile Line Inlet	<1	<5	<2	<10	<30	45	1,400	<30	0
2	Tile Line Inlet	<1	<5	<2	<10	<30	520	1,500	<30	0
3	Tile Line Outlet	<1	<5	<2	<10	<30	10	30	<30	na
8	Tile Line Outlet	<1	<5	<2	<10	<3	2,900	2,400	<3	na
9	Tile Line Outlet	<1	<5	<2	<10	<3	<10	<10	<3	na
9	Tile Line Outlet	<1	<5	<2	<10	<3	230	1,500	<3	na

[a]all samples were culture negative for *Campylobacter sp.*, [b]Tetracycline concentrations as chlortetracycline; limit of detection is 1µg/L (ppb), [c]Sulfonamide concentrations as sulfamethiazine; limit of detection is 5 µg/L (ppb), [d]β-Lactam concentrations as penicillin G; limit of detection is 2 µg/L (ppb), [e]Macrolide concentrations as erythromycin: limit of detection is 10 µg/L (ppb), [f,i]Limit of detection for *Salmonella* and *Yersinia* is <3, <30, or <300 MPN/100 mL (based on dilution run), [g,h]Limit of detection for *E. coli* and *Enterococcus sp.* is <10, or <100 MPN/100 mL (based on dilution run), [j]Whole *Cryptosporidium parvum* oocysts/L, "na" = not analyzed

Pesticide analysis

Of the 23 samples tested for select pesticides, only 2 (9%) had detectable levels of any pesticide (0.14 μg/L atrazine was detected in a sample from a tile line inlet at farm #2 and 0.86 μg/L atrazine was detected in a sample from an agricultural drainage ditch at farm #4).

Antibiotic residue and microbial analysis

Table 7 shows which antibiotics and which microbes were detected at each site.

Antibiotic residue analysis

Detectable levels of four different classes of antibiotics were found in eight samples, one from each from all seven earthen lagoons and one from an earthen lagoon monitoring well. In seven of the samples, tetracyclines were detected at levels of 11, 66, 68, 110, 150, 250, and 540 μg/L, respectively. In one sample, sulfonamides were detected at a level of 7.6 μg/L. In four samples, β-lactams were detected at levels of 2.1, 2.1, 2.9, and 3.5 μg/L, respectively. In five samples, macrolides were detected at levels of 15, 60, 81, 227, and 275 μg/L, respectively. None of the 23 samples had detectable levels of fluorquinolones.

Bacterial cultures

Samples from the earthen manure lagoons contained the greatest number of bacterial isolates for *E. coli*, *Enterococcus sp.* and *Salmonella sp.* An isolate of one *Yersinia* species was obtained from an agricultural drainage ditch. *Enterococci* were isolated from 20 out of 23 (87%) samples and evaluated, *E. coli* were isolated from 18 out of 23 (78%) samples, and 3 species of *Salmonella* were isolated from 2 out of 23 (9%) samples. All samples were negative for *Campylobacter sp.* (Tables 7 and 8).

The antibiotic resistance patterns for the 41 bacterial isolates are reported in Table 8. Many of the 18 *E. coli* isolates demonstrated resistance to a particular antibiotic or combination of antibiotics. Eleven isolates demonstrated resistance to fluorfenicol (alone or in combination with other antibiotics), an antibiotic not approved for use in swine. All three *Salmonella* species isolated demonstrated resistance to particular combinations of antibiotics. Isolates of *Enterococcus* demonstrated resistance to particular combinations of antibiotics. Antibiotic susceptibility testing for the *Yersinia* isolate was not conducted.

Table 8. Environmental study proximal to concentrated animal feeding operations (CAFOs), antibiotic resistance patterns of microbial isolates from 23 water samples, Iowa, October-December, 1998

	Number of Resistant Microbial Isolates		
Antibiotic Resistance Pattern	***Escherichia coli* (%)**	***Salmonella* sp. (%)**	***Enterococcus* sp. (%)**
Pansusceptible[a]	2 (11.1)	0	na
Fluorfenicol alone	8 (44.4)	0	na
Tetracycline alone	1 (5.5)	0	na
Fluorfenicol + Tetracycline	1 (5.5)	0	na
Sulfamethoxazole + Tetracycline	1 (5.5)	1 (33.3)	na
Ampicillin + Florfenicol + Tetracycline	2 (11.1)	0	na
Streptomycin + Sulfamethoxazole + tetracycline	0	1 (33.3)	na
Apramycin + Bacitacin + Lincomycin	na	na	8 (40)[b]
Ampicillin + Apramycin + Bacitacin + Lincomycin	na	na	1 (5)[c]
Penicillin + Apramycin + Bacitacin + Lincomycin	na	na	2 (10)[d]
Synercid + Apramycin + Bacitacin + Lincomycin	na	na	9 (45)[e]
Fluorfenicol + Kanamycin + Streptomycin + Sulfamethoxazole + Tetracycline	1 (5.5)	0	na
Ampicillin + Cephalothin + Fluorfenicol + Sulfamethoxazole + Tetracycline	1 (5.5)	0	na
Amoxicillin-Clavulanic acid + Ampicillin + Ceftiofur + Cephalothin + Chloramphenicol + Fluorfenicol + Streptomycin + Sulfamethoxazole + Tetracycline	0	1 (33.3)	na
Amoxicillin-Clavulanic acid + Ampicillin + Ceftiofur + Cephalothin + Chloramphenicol + Fluorfenicol + Gentamycin + Kanamycin + Streptomycin + Sufamethoxazole + Tetracycline	1 (5.5)	0	na
Total	18 (100)	3 (100)	20 (100)

[a]Susceptibile to all antibiotics used in study, [b]Represents five isolates of *E. faecium*, two other species, and one isolate not speciated, [c]Represents one isolate of *E. faecium*, [d]Represents two isolates of *E. faecium*, [e]Represents eight isolates of *E. faecalis*, and one other species, "na" = not analyzed

Parasitology

Cryptosporidium parvum oocysts were isolated from 7 of the 13 (54%) samples analyzed (Table 7). The highest oocyst counts were detected in earthen manure lagoon samples; lower numbers of viable oocysts were found in three samples from three separate earthen lagoon monitoring wells and in one sample from a river. Oocysts were identified in samples obtained from both an earthen lagoon and an adjacent earthen lagoon monitoring well on one farm.

DISCUSSION

The study was successful in accomplishing the primary goal of obtaining a broad profile of the chemical and microbial constituents of both ground and surface water proximal to large-scale swine operations. Even though the results of some tests are still pending, this pilot project has generated some interesting findings. As anticipated, the highest levels of chemical pollutants and zoonotic pathogens were generally found in samples collected from earthen manure lagoons. The contaminants included trace metals, common ions, nutrients, antibiotics, parasitic oocysts of the protozoan *Cryptosporidium parvum*, and bacteria that demonstrated particular resistance to several antibiotics commonly used in swine management practice as feed supplements and therapeutics. Most bacterial isolates even showed resistance to fluorfenicol, an antibiotic not approved for use in swine. However, fluorfenicol is available for use in swine by veterinary prescription as an extra-label drug.

Our results also indicated the presence of trace metals, common ions, nutrients, pesticides, antibiotics, bacteria, and parasitic oocysts in samples obtained from collection points other than earthen manure lagoons. One sample obtained from an agricultural drainage and another sample obtained from a tile line inlet both had nitrate levels that exceed the drinking water standards. One sample obtained from an earthen lagoon monitoring well had the highest sulfate level found among the 23 samples. Trace levels of total sterols were also detected in a water sample from a tile line inlet suggesting possible fecal pollution.

Detectable levels of the pesticide atrazine were found in samples obtained from a tile line inlet and from an agricultural drainage ditch. Atrazine is a widely used agricultural herbicide, applied annually to cornfields. Compounds most likely used in swine barns for insect control were not found. Detectable levels of the antibiotic class Sulfonamides were also found in a sample from an earthen lagoon monitoring well.

Certain bacteria are indicators of fecal contamination. We isolated *E. coli* and *Enterococcus sp.* from samples collected from earthen lagoon monitoring wells, tile line inlets, tile line outlets, agricultural drainage ditches, and agricultural drainage wells.

Cryptosporidium parvum oocysts were also identified in a sample collected from an earthen lagoon and from an adjacent earthen lagoon monitoring well on the same farm site.

These discoveries suggest the possibility of the movement of both chemical pollutants and microbial pathogens through soil and away from their point of highest concentration, the animal manure lagoons, and by overland flow away from the site of manure application.

Sampling limitations

Samples were not obtained from all potential collection points on each participating farm. Furthermore, soil samples from agricultural crop fields where manure was applied were not obtained and analyzed for chemicals, microbes or parasites. The methods used for pesticides had very high limits of detection (low sensitivity), as did the newly developed methods for antibiotic detection; therefore, lower levels of pesticides and antibiotics present in the environment may have been missed. These limitations may restrict the overall picture of the environmental contamination existing on these farm sites.

Suggestions for future studies

In follow-up studies attempts should be made to obtain samples from all identified collection points, from every participating farm after manure application, and 24 to 48 hours after a substantial rainfall. This would help capture the movement of animal manure pollutants through runoff or soil filtration, providing more complete information on the movement of pollutants from animal manure through the soil and into the surrounding surface and ground waters.

Future studies should include information from farm management records on the therapeutic and prophylactic use of antibiotics on each specific farm, as well as any existing manure management plan; this information can be used to help correlate farm practices with findings obtained through the studies.

Testing for *Cryptosporidium parvum* oocysts proved to be a promising tool for both detecting the pathogen and its movements through the ground and surface water. In future studies, more samples should be tested for this protozoan, and DNA-fingerprinting techniques for *C. parvum* should be used to detect leakage from lagoons or crop fields where *C. parvum*-contaminated manure is applied.

Since microbial, parasitic, and chemical contaminants were identified in the earthen manure lagoons and manure is often applied as fertilizer on crop fields, future studies should look at the presence and survivability of microbial and parasitic pathogens, as well as chemicals, in the soil where the manure is applied. Studies should also be done to determine whether manure has been applied to an agricultural crop, and if so, whether the crop is destined for human consumption.

Finally, the occurrence of chemicals, microbes, and parasites in surface and ground water on non-CAFO farms (controls) should be determined for comparison.

CONCLUSIONS

We identified chemical pollutants and zoonotic pathogens in the environment on and proximal to large-scale swine feeding operations. However, the sample collection sites were not in locations that could pose a direct threat to human health. More research is recommended to accurately determine the level of risk, pathways of exposure, and critical control points to avoid any potential exposure.

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