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Association of farm management practices with risk of *Escherichia coli* contamination in pre-harvest produce grown in Minnesota and Wisconsin

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Abstract

Microbiological analyses of fruits and vegetables produced by farms in Minnesota and Wisconsin were conducted to determine the prevalence of *Escherichia coli* in pre-harvest fruits and vegetables. During the 2003 and 2004 harvest seasons, 14 organic (certified by accredited organic agencies), 30 semi-organic (used organic practices but not certified) and 19 conventional farms were sampled to analyze 2029 pre-harvest produce samples (473 organic, 911 semi-organic, 645 conventional). Before each harvest season, a farmer survey was conducted to collect relevant information on farm management practices that might affect the risk of *E. coli* contamination in fresh produce. The use of animal wastes for fertilization of produce plants increased the risk of *E. coli* contamination in organic (OR = 13.2, 95% CI = 2.2–61.2, *P*-value < 0.0001) and semi-organic (OR = 12.9, 95% CI = 2.9–56.3, *P*-value < 0.0001) produce significantly. Improper ageing of untreated animal manure significantly increased this risk in organic produce (OR = 4.2 95% CI = 1.7–12.3, *P*-value = 0.005) grown using such manure as a fertilizer. Organic growers who used cattle manure for fertilization of their crops showed significantly greater risk of contamination with the *E. coli* (OR = 7.4, 95% CI = 1.6–36.8, *P*-value = 0.003), compared to those who used other types of manure-based fertilizer. In Minnesota, organic and semi-organic produce collected from the southeastern (SE) part of the state were at a significantly greater risk of *E. coli* contamination (OR = 3.45, 95% CI = 1.8–35.2, *P* = 0.008), compared to those collected from farms located in the southern (S) regions of the state. In Wisconsin, organic and semi-organic produce collected from the southern (S) cluster of farms were at approximately 3-times greater risk of *E. coli* contamination (OR = 2.67, 95% CI = 1.3–9.4, *P* = 0.004), compared to those grown in the northern (N) cluster of farms.

Keywords: Organic foods; Produce; Farm management; Pre-harvest practices; Risk-factors; Prevalence; *Escherichia coli*

1. Introduction

Fresh fruits and vegetables are gaining popularity in our daily diet, as consumers are becoming more aware of the long-term health and nutritional benefits of sufficient quantities of these foods in our food habits. In this context, the U. S. Department of Agriculture (USDA) has emphasized the importance of sufficient number of servings of fresh produce in our daily diet (CNPP/USDA, 2005). Recently, the market for organic foods in general, and organic fresh produce in particular, has expanded at an annual rate of 20% (Dimitri and Greene, 2002). The USDA Organic Rule was implemented in 2002, and organic producers are required to conform to the farm management practices, including treatment and application of animal-manure-based fertilizers, that were specified in this set of regulations (NOP/USDA, 2002).

In recent years, outbreaks of foodborne infection from consumption of fresh fruits and vegetables have increased considerably (C.D.C., 2005; Sivapalasingam et al., 2004). Some people in the scientific community have expressed concerns about the microbial safety of organic produce, as organic growers primarily rely on animal wastes for fertilization of their produce plants (Stephenson, 1997). A few reports have

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documented that organic produce were not subjected to additional risks of contamination from foodborne pathogens, compared to their conventional counterparts (Mukherjee et al., 2004; Mukherjee et al., 2006; Pao et al., 2005; Phillips and Harrison, 2005). In a previous publication, Mukherjee et al. (2004) reported that certain farm management practices might result in significantly greater *Escherichia coli* prevalence in organic produce.

Numerous reports have documented that animals such as cattle, sheep, pig and chicken are major reservoirs of foodborne pathogens such as *E. coli* O157:H7 and *Salmonella* (Guan and Holley, 2003; Maule, 2000; Wang et al., 1996). When farmers use manure from these animals for fertilization of produce plants, these pathogens might get transmitted to fruits and vegetables harvested from those plants (Brackett, 1999). Such transmission was reported in lettuce when manure, inoculated with laboratory cultures of *E. coli* O157:H7, was used for fertilization (Solomon et al., 2002). Composting is an exothermic microbiological process, which generates high temperatures from 55 to 65 °C under proper conditions of aeration, moisture, particle size and carbon-to-nitrogen ratio for long enough duration that inactivates these foodborne pathogens (EPA, 2006). Published reports have documented the effectiveness of composting in destroying *E. coli* O157:H7 and *Salmonella* in cow manure (Lung et al., 2001). The USDA has specified composting techniques that organic growers are required to follow to minimize the risk of contamination from manure-based fertilizers (NOP/USDA, 2002). The USDA has also specified fertilization-to-harvest intervals that would reduce the chances of pathogen survival in non-composted manure. A few reports evaluated the effectiveness of such duration in minimizing the risk of contamination in vegetables (Ingham et al., 2005).

The reports available in the literature have documented the survival of contaminating bacteria in manure and in manure-amended soil (Cote and Quessy, 2005; Hutchison et al., 2004). Other laboratory and garden-scale studies have evaluated the effects of manure treatment and application to soil on the survival of pathogenic and fecal indicator bacteria in agricultural soil and vegetables grown in such soils (Ingham et al., 2004). The present study generated data on the microbiological quality of fresh produce at farms, and focused on finding associations of certain farm-level practices such as treatment and application of animal-manure-based fertilizers with increased risk of contamination of *E. coli* in fresh produce.

2. Materials and methods

2.1. Farmers' survey

A questionnaire was mailed to all the participating semi-organic, organic and conventional farmers at the beginning of each harvest season. General information such as names, contact information, years in business, market, certification status and produce types grown were collected. The major focus of the questionnaire, however, was use of animal waste and other pre-harvest production and handing practices used by produce growers. The relevant study questions are shown in Table 1. A stamped return-envelope, in which the farmer's responses were received, was attached with the questionnaire.

2.2. Classification of farm types

Farmers who grew fruits and vegetables were invited to participate in the study by telephone or by personal contacts. All the farmers who participated in the study during 2003 and 2004 were located in Minnesota and Wisconsin. The farms were classified into two categories: organic and semi-organic. Organic farms were those that were currently certified by an USDA-accredited organic certification agency. Semi-organic farms were those that reported using organic practices but were not certified.

2.3. Sampling of fresh fruits and vegetables

The participating farms were visited from 2 to 3 times during June, July, August and September, in the year of 2002, 2003 and 2004. Samples of fruits and vegetables were collected directly from the farm fields, and without any washing or rubbing-off of soil particles, the samples were transferred into sterile zip-lock bags. The gloves and knives were sanitized with alcohol swabs between collections in order to prevent cross-contamination. Produce types included the following: lettuce, other leafy greens such as spinach, cabbages, kale, Swiss chard, collard, and pepper, tomato, berries, broccoli, summer squash, cucumber, zucchini, and other types of produce in small numbers e.g. bok choi, cantaloupe, apple, kohlrabi, sprouts and peas. The sample amounts varied from one produce type to another. For leafy greens like spinach, kale, collard, about 500 g of leaves were collected from different plants in a portion of a field of cultivation, and those leaves constituted one sample. For

Table 1
Multiple-choice questions included in the farmers' survey on farm management practices before harvest of produce

Questions asked	Options for answers
Classification	Certified organic, non-certified organic, conventional
Years in production	<5 years, 5–10 years, 10–15 years,>15 years
Crops grown	Apples, bok choi, cabbage, cantaloupe, collard, cucumber, kale, lettuce, other leafy greens, pepper, raspberry, spinach, strawberry, summer squash, Swiss chard, tomato, zucchini
Use of animal waste	Yes, no
Type of animal waste	Cattle, beef, dairy, turkey, horse, sheep, goat, llama, chicken, duck, other
Use of compost:	Yes, no
If yes - the source of compost	Made in the farm, purchased
If made in the farm - procedure	Undisturbed pile, turned once, turned several times, monitored pile temperature
Application time from harvest	<30 days before, 30–90 days before, 90–120 days before,>120 days before
Duration of ageing	1/2–1 year, 1 year, 1–2 years, 2 years,>2 years
Application amount	Undetermined,<1 ton per acre, ≥1 ton per acre, please specify amount ______
Application season	Spring, fall, summer

Table 2
Response to the farmers' survey from semi-organic (SO), organic (O) and conventional (C) farmers in 2003 and 2004

Response to survey	Number (%) of respondents					
	2003			2004		
	SO	O	C	SO	O	C
Responders to the survey	19 (79)	11 (79)	12 (84)	19 (79)	7 (88)	19 (100)
Lettuce and leafy vegetable growers	11	8	4	15	7	5
Animal waste users	14	10	5	14	7	10
Among animal waste users:						
Compost users among	9	9	3	10	7	6
Ageing of manure for >1/2 year	6	4	5	7	2	9
Users of cattle manure	4	7	1	5	6	3
Manure application ≥90 days ahead of harvest	6	8	4	7	5	9

produce types such as cucumber, summer squash, pepper, tomato, from 2 to 5 of these vegetables were collected from different parts in a particular location of a field to constitute a sample. For produce types such as cantaloupe, cabbage, bok choi, and head lettuce, one head of lettuce or cabbage or bok choi or one cantaloupe constituted one sample. For fruits like raspberry and strawberry, about 500 g of fruits, ripe enough for harvest, from different plants in a particular location of the field were collected in sterile plastic boxes. For representative sampling from a field of cultivation, produce from different locations of a field were sampled. Most of the produce fields were sampled in triplicate, and for large fields of cultivation, as many as five samples were collected from different locations of the fields.

Sample bags and boxes were properly marked by recording produce type, farm identity, sample number, and date of collection on them. The sample bags were then placed in insulated coolers with ice-packs and were sent to the laboratory. Samples were received within 10 h of collection, and were stored at 4 °C until microbiological analyses began. The number of samples processed per run was variable depending on the number of farms visited per day, but it varied from 10 to approximately 50 samples. Most of the samples were analyzed in the following 24 h after delivery and all of them were processed within 48 h of sampling.

2.4. Microbiological analyses

Microbiological analyses of fruits and vegetables started with sample preparation, which depended on the type of fruits and vegetables. For leafy greens such as lettuce, spinach, kale, collards, cabbage and bok choi, leaves from the outside and inside sections of the produce were used to make up 25 g of sample. For produce types such as summer squash, zucchini, cucumber, tomato, pepper, one or two of these vegetables were cut into small pieces, and pieces from different portions of the vegetable were used to make up 25 g of sample. For cantaloupes small pieces of two fruits were cut from different parts including the peel to make up 25 g of sample. For fruits such as raspberry and strawberry, 2 to 6 (depending on size of each fruit) fruits were picked from different locations of the sample box to constitute 25 g of sample. Twenty five grams of produce sample were transferred into 225 ml of lauryl sulfate tryptose (LST, Neogen, Inc., Lansing, Michigan), in sterile stomacher bags. The sample was mixed with the enrichment broth in a stomacher (Tekmar Co., Cincinnati, Ohio) for 2 min.

Escherichia coli were determined by an initial enrichment in 9-ml tubes of LST (Neogen, Inc., Lansing, Michigan) broth. The LST tubes were incubated for 24 and 48 h at 37°C. LST tubes showing growth and gas production were transferred to 9-ml brilliant green bile (BGB; Neogen, Inc.) broth tubes each containing a Durham's tube. The BGB-broth tubes were incubated at 37 °C for 24 and 48 h for selective enrichment for coliforms. Broth cultures from the tubes showing both growth and gas production were streaked on eosin methylene blue (EMB; Neogen, Inc.) plates. The EMB plates were incubated for 24 h at 37 °C, and the plates were then examined for characteristic *E. coli* colonies, that had a dark center, with or without a greenish metallic sheen. The suspected *E. coli* colonies were confirmed using indole, methyl red, Voges Proskauer, and citrate fermentation tests.

Table 3
Number of samples collected from semi-organic, organic and conventional farms by produce types in 2003 and 2004

Produce types	Organic		Semi-organic		Conventional		Total	
	'03	'04	'03	'04	'03	'04	'03	'04
Berries	18	7	23	69	29	51	70	127
Bok-choi	3	3	9	8	0	1	12	12
Broccoli	15	9	21	40	6	14	42	63
Cabbages	21	28	43	41	27	41	91	110
Cantaloupe	0	3	9	6	12	6	21	15
Cucumber	12	25	26	34	41	44	79	103
Leafy greens	30	77	71	101	12	5	113	183
Lettuces	33	22	29	43	12	12	74	77
Others	0	12	8	12	6	14	38	38
Peppers	12	44	46	75	45	60	103	179
Summer squash	9	10	19	34	16	20	44	64
Tomatoes	13	39	39	58	46	45	98	142
Zucchini	12	16	29	18	45	35	86	69
Total	**178**	**295**	**372**	**539**	**297**	**348**	**847**	**1182**

2.5. Statistical analyses

Escherichia coli prevalence was calculated by using the number of samples tested positive for *E. coli*, and then dividing that by the total number of samples. The prevalence were compared at first individually using 2×2 Chi-square analyses (Moore and McCabe, 2003), and then *F*-test for multiple Chi-square analyses was performed using the PC SAS system for Windows operating system (SAS version 9.1, SAS Institute Inc., Cary, NC). Longitudinal data analyses were performed using the Generalized Estimating Equation, for determination of time-trend in the *E. coli* prevalence (Weiss, 2005). After performing an Exploratory Data Analyses (EDA), independent correlation structure was used in the GEE model fit to the *E. coli* prevalence data. The GEE model was chosen using backward elimination procedure. The chosen distribution was Binomial, as the outcome variable took only two values—0 for absence and 1 for presence of detectable *E. coli* in a produce sample. The odds ratios, 95% confidence intervals and the *P*-values were obtained from the PC SAS output. The Chi-square-values, *F*-values and the odds ratios (OR) were considered statistically significant at $P<0.05$.

3. Results

3.1. Farmers' response to the survey on their farm management practices

In 2004, approximately 88% of organic and 79% of semi-organic and all the conventional farms responded to the questionnaire on their farm management practices (Table 2). In 2003, these percentages ranged from approximately 79% among semi-organic and organic growers to 84% among conventional growers. Approximately 44 to 50% of conventional farms used animal waste as fertilizer, while 70 to 100% of the semi-organic and organic farms had animal manure as fertilizer. Among the organic farms, which used animal manure for fertilization of their produce plants, 90 to 100% used composting. Among semi-organic farms, this percentage ranged from 64 to 71 during 2003 and 2004. When farmers were asked about the duration of ageing of manure to be applied on produce plots as fertilizer, approximately 30 to 40% of organic and 40 to 50% of semi-organic farms used manure that was aged for more than 6 months. Among conventional growers that used animal waste for fertilization of their produce plants, as many as 90 to 98% used more than 6 months of ageing before application as fertilizer.

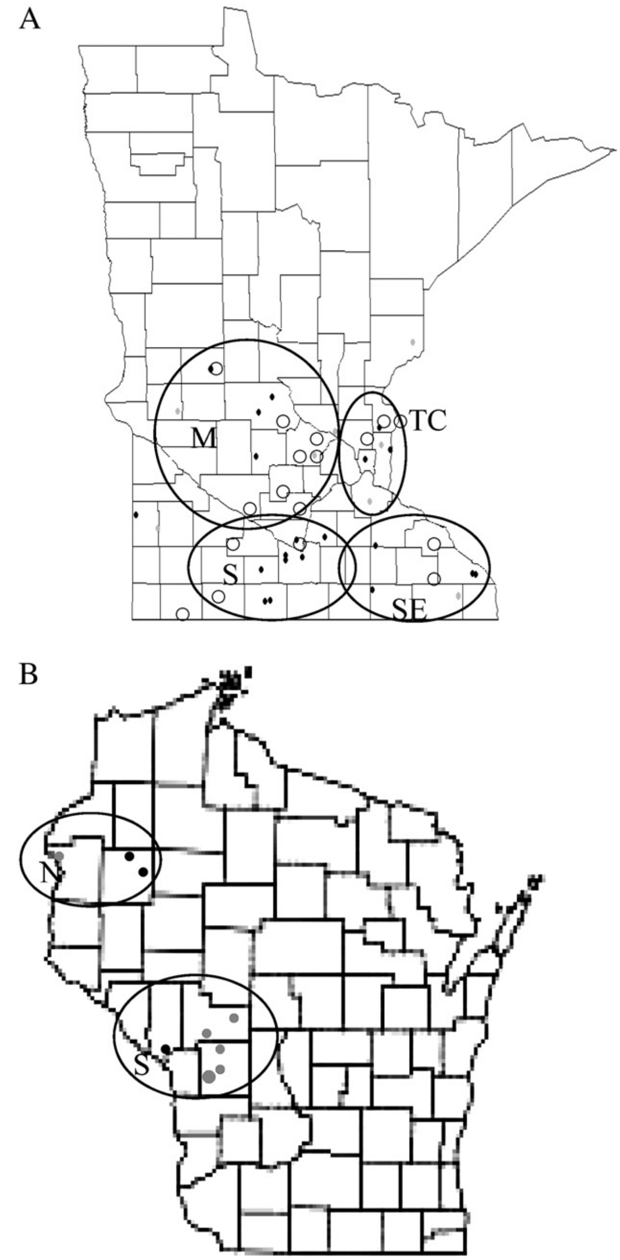

Fig. 1. Geographical distribution of semi-organic (●), organic (●) and conventional (○) farms among the middle (M), Twin Cities (TC), South (S) and South East (SE) locations in Minnesota (A) and in Wisconsin (B).

Table 4
Association of management practices and factors with risk of *E. coli* contamination in semi-organic and organic produce

Management practices and factors		Odds ratio (95% CI) for two farm types	
		Non-certified	Organic
Usage of animal waste: Users vs. non-users		12.9* (2.9–56.3)	13.2* (2.6–61.2)
Age of animal waste: <1/2 year vs. ≥1/2 year		1.3 (0.3–6.1)	4.2*(1.7–12.3)
Manure type: Cattle manure vs. others		2.2 (2.1)	7.4* (1.6–36.8)
Season of manure application:	Spring	1.07 (0.1–3.8)	1.8 (0.2–9.6)
	Both	1.3 (0.2–7.4)	1.04 (0.1–4.2)
	Fall^{ref}	–	–

Note: The odds ratios with * were statistically significant ($P<0.05$). The superscripted ref is the season of reference for calculation of OR.

3.2. Association of farm management practices with risk of *Escherichia coli* contamination

A total of 2029 samples were collected during the 2 years of the study (Table 3). In 2004, almost 40% more samples were collected than in 2003. The number of samples collected increased for each of the farm types from 2003 to 2004 approximately 66%, 44% and 17% for organic, semi-organic and conventional produce, respectively. Leafy greens, peppers and tomatoes contributed more than 230 samples over the two-year period. Leafy greens had the largest number of samples for organic and semi-organic farm types for both years combined, while peppers were the largest group of conventional produce.

Use of animal wastes in fertilization of fresh fruits and vegetable plots significantly increased the risk of *E. coli* contamination in fresh produce grown in semi-organic (OR = 12.9, 95% CI = 2.9–56.3, *P*-value < 0.0001) and organic (OR = 13.2, 95% CI = 2.2–61.2, *P*-value < 0.0001) farms (Table 4). Although composting did not affect the *E. coli* prevalence in semi-organic and organic produce, ageing of non-composted manure for more than 6 months contributed to a significant reduction of the risk (OR = 4.2 95% CI = 1.7–12.3, *P*-value = 0.005) among organically produced fruits and vegetables. Semi-organic and organic farms used various types of animal wastes such as chicken, hog, horse and cattle manure. Among these different manure types, use of cattle manure for fertilization of produce crops increased the risk of *E. coli* contamination by approximately 7-fold (OR = 7.4, 95% CI = 1.6–36.8, *P*-value = 0.003). Farmers who used animal wastes for fertilization applied manure either in the Spring or in the Fall or both in Spring and Fall. However, these different times of application of manure application did not have any significant effect on the *E. coli* prevalence in semi-organic and organic produce.

3.3. Variation of Escherichia coli prevalence in produce collected from farms in different geographical locations of Minnesota and Wisconsin

Farmers participating in this two-year study were grouped into four geographical locations such as Middle (M), Twin Cities (TC), South East (SE) and South (S) in Minnesota (Fig. 1A), and into two distinct groups such as North (N) and South (S1) in Wisconsin (Fig. 1B). The average percent prevalence of *E. coli* in produce from semi-organic and organic farms in the South East (SE), South (S), and Middle (M) portion of the state of Minnesota was significantly greater compared to that in farms located in the Twin Cities (TC) location (Table 5). In Wisconsin, organic and semi-organic produce from farms located in the South (S1) cluster had significantly greater average *E. coli* prevalence of 11% compared to 6% average prevalence in the farms located in the North (N) cluster. The distribution of leafy vegetables including lettuce and cabbage among these different locations in Minnesota and Wisconsin was quite consistent (Table 5).

Table 5
Percentage of *Escherichia coli* positives and percentage of leafy vegetables in farms, located at the four geographical regions in Minnesota and in the two regions in Wisconsin

States	Location	Percentage *E. coli* positives (Mean ± SE)	Percentage of leafy vegetables (Mean ± SE)
Minnesota	Southeast	10.8 ± 5.7[a]	48.1 ± 8.3
	South	10.3 ± 7.1[a]	20.7 ± 9.9
	Twin Cities	3.0 ± 1.7[b]	36.9 ± 8.3
	Middle	8.1 ± 1.8[a]	39.0 ± 5.5
Wisconsin	South	11.2 ± 3.7[a]	31.8 ± 4.7
	North	4.7 ± 2.9[b]	40.6 ± 3.8

Note: The average percentage positives were compared within the two states, between the geographical locations; percentages with different super-scripted letters were significantly different ($P < 0.05$).

Table 6
Variation in risk of *E. coli* contamination in fresh semi-organic and organic produce at different geographical locations in Minnesota and Wisconsin

Geographical locations	Odds ratio	95% Confidence Interval	*P*-value
Minnesota:			
Middle (M)	2.14	0.3–11.2	0.55
Twin Cities (TC)	1.15	0.1–15.6	0.74
South East (SE)	3.45	1.8–35.2	0.008
South (S)[ref]	–	–	–
Wisconsin:			
South	2.67	1.3–9.4	0.004
North[ref]	–	–	–

The superscripted ref is the geographic region of reference to calculate OR.

The analysis on the basis of these regions of Minnesota and Wisconsin determined that produce from the South East (SE) region of Minnesota had approximately 3.5-times (OR = 3.45, 95% CI = 1.8–35.2, *P* = 0.008) greater risk of *E. coli* contamination compared to those from the southern (S) region (Table 6). In Wisconsin, the organic and semi-organic produce from the farms located in the South (S1) region were at a 2.7-times (OR = 2.67, 95% CI = 1.3–9.4, *P* = 0.004) greater risk of such contamination compared to their counterparts cultivated in farms in the North (N) region. All the participating conventional produce growers were from Minnesota, and 6 of the 19 of these farmers had at least one *E. coli* contaminated samples from their farms. Prevalence of *E. coli* in conventional produce from the Middle (M), and South (S) regions of the state were 4.4 and 6.9%, respectively. None of the conventional produce grown in the Twin Cities (TC) and South Eastern (SE) region of Minnesota had detectable *E. coli* contamination.

4. Discussion

Several reports have found that some of the farm-level practices for handling and treating animal manure before their application as fertilizer might reduce the survival of contaminating bacteria in manure and in agricultural soil, in which manure had been amended as a fertilizer (Hutchison et al., 2004; Kudva et al., 1998; Lung et al., 2001). Thus a major portion of the survey's questionnaire was meant to collect information on manure-handling and treatment practiced by the semi-organic, organic and conventional produce growers who took part in this study.

During the two-year study, less than half of the conventional growers used animal manure for fertilization of their crops, while 70 to 90% of organic and semi-organic farmers applied animal manure for fertilization. This finding was expected as organic growers are not allowed to use most of the chemical fertilizers that conventional growers can use (AMS/USDA, 2000). Although the majority of the conventional farmers did not use animal waste as fertilizer, only 57% of the manure users composted animal manure before using it as fertilizer. As many as 70% of the semi-organic and almost all the organic farmers who used manure fertilizer applied composted manure.

Our findings, both during the present study and in a previous study, showed that only 1.5 to 2.5% of pre-harvest conventional produce had detectable *E. coli* contamination (Mukherjee et al., 2004; Mukherjee et al., 2006). This low number of positive samples prevented any risk-factor analyses involving conventional farms. However, among the 6 conventional farms which had at least one *E. coli* contaminated sample, 4 used manure-based fertilizers. In this study, only semi-organic and organic farms were included in the analyses of farm-level risk-factors associated with greater prevalence of *E. coli*. Among these two farm types, users of animal waste as fertilizer were at a significantly greater risk of *E. coli* contamination in their produce compared to those who did not use manure fertilizer (Table 4).

Ageing of animal wastes before their application as fertilizer was used by several of the participating farmers. Ageing of animal manure for less than 6 months increased the risk of *E. coli* contamination by more than four folds in organic produce (Table 4). However, this risk-factor did not show a significant effect on *E. coli* prevalence in semi-organic produce. Hutchison et al. (2004) reported that spreading livestock manure on the top of agricultural soil and let it age without mixing them into the soil significantly reduced counts of pathogenic bacteria such as *E. coli* O157:H7, *Salmonella enterica*, *Listeria monocytogenes*, and *Campylobacter jejuni*.

In another report, the persistence of *E. coli* was monitored in agricultural soils in Wisconsin, amended with non-composted bovine manure. No clear effect of fertilization-to-harvest intervals of 90, 100, 110 and 120 days was reported on *E. coli* contamination in radishes, carrots and lettuces produced in the manure-amended soils (Ingham et al., 2004). In the present study also, no strong association of fertilization-to-harvest interval of less than 90 days with increased risk of *E. coli* contamination in semi-organic and organic produce was evident. Among the semi-organic and organic produce, using cattle manure for fertilization increased the risk of *E. coli* contamination by 2-fold and 7-fold, respectively (Table 3). This finding was consistent with our previous report (Mukherjee et al., 2004). In that study, organic produce cultivated using cattle manure in fertilizer showed 16% *E. coli* prevalence, compared to 7% in those which were cultivated using other type of animal manure-based fertilizer.

Farmers located in Minnesota were spread all across the southern part of the state (Fig. 1A). They were classified into four groups, based on their geographical location at the mid-portion of the state, around the Twin Cities, southern and southeastern parts of the state. The greatest *E. coli* prevalence was in organic and semi-organic produce collected from the SE part of the state. The risk of such contamination in produce cultivated in this region of Minnesota was significantly greater than those cultivated in the S part of the state (Table 6). In Wisconsin, the farms were located in two distant and distinct areas—one in the northern portion one in the southern part of the state (Fig. 1B). Organic and semi-organic produce in the S location was 2.7-times the risk of such contamination in produce collected from the N location of the state, and this difference was statistically significant (Table 6).

The present study is one of the very few research works that have focused on farm management practices as they affect bacteriological contamination in fresh fruits and vegetables. In our previous report, we gathered information from the participating farmers, on the basis of which we used simple analytical comparisons between *E. coli* prevalence in pre-harvest produce, cultivated using different farm-level production practices (Mukherjee et al., 2004). In this research, we conducted formal farmers' surveys at the beginning of each of the 2003 and 2004 harvest seasons. Risk-factor analyses were conducted using an appropriate logistic regression model that has been used in other published reports on farm-level risk-factor analyses (Cho et al., 2006; Fossler et al., 2005; Kuhnert et al., 2005). Our findings suggested that a vast majority of organic and semi-organic produce growers used animal manure for fertilization of their produce plants, compared to less than half of the conventional growers who used such fertilization practices. To our knowledge, this study was the first report on the variation in microbiological quality of pre-harvest produce across different geographical locations in Minnesota and Wisconsin, where the participating farms were located. These results suggest a geographical effect, but because of the limited nature of this study they should not be extrapolated to other regions.

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