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UNITED STATES DISTRICT COURT

NORTHERN DISTRICT OF CALIFORNIA

IN RE: ROUNDUP PRODUCTS LIABILITY LITIGATION	MDL No. 2741 Case No. 16-md-02741-VC
This document relates to: ALL ACTIONS	

MONSANTO COMPANY'S NOTICE OF FILING

Please take notice that pursuant to Court's order during the March 14, 2018 hearing, defendant Monsanto Company submits the attached articles, each of which addresses an issue discussed at today's hearing:

1. Blair, A. & S. Zahm, Patterns of Pesticide Use among Farmers: Implications for Epidemiologic Research, 4 Epidemiology 55 (1993), Exhibit 303, admitted into evidence.

2. Bonner, M. et al., Occupational Exposure to Pesticides and the Incidence of Lung Cancer in the Agricultural Health Study, 125 Envtl. Health Perspective, 544 (2017), Exhibit 608 on Monsanto's Renumbered Exhibit List, ECF No. 1151, which Monsanto requests be entered into evidence.

3. De Roos, A. et al., An Application of Hierarchical Regression in the Investigation of

Multiple Paternal Occupational Exposures and Neuroblastoma in Offspring, 39 Am. J. of Indus. Med. 477 (2001), Exhibit 717 on Monsanto's Renumbered Exhibit List, ECF No. 1151, which Monsanto requests be entered into evidence.

4. Freeman, L. et al., Poultry and Livestock Exposure and Cancer Risk Among Farmers in the Agricultural Health Study, 23 Cancer Causes Control 663 (2012), Exhibit 910 on Monsanto's Renumbered Exhibit List, ECF. No. 1151, which Monsanto requests to be entered into evidence.

5. Hohenadel, K. et al., Exposure to Multiple Pesticides and Risk of Non-Hodgkin Lymphoma in Men from Six Canadian Provinces, 8 Int'l J. Envtl. Res. Public Health 2320 (2011), Exhibit 1011 on Monsanto's Renumbered Exhibit List, ECF No. 1151, which Monsanto requests be entered into evidence.

6. Koutros, S. et al., Risk of Total and Aggressive Prostate Cancer and Pesticide Use in the Agricultural Health Study, 177(1) American Journal of Epidemiology 59 (2013), Exhibit 1107 on Monsanto's Renumbered Exhibit List, ECF No. 1151, which Monsanto requests be entered into evidence.

7. Koutros, S. et al., Occupational Exposure to Pesticides and Bladder Cancer Risk, 45(3) International Journal of Epidemiology 792 (2016), Exhibit 1106 on Monsanto's Renumbered Exhibit List, ECF No. 1151, which Monsanto requests be entered into evidence.

DATED: March 14, 2018

Respectfully submitted,

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Patterns of Pesticide Use among Farmers: Implications for Epidemiologic Research

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Epidemiologic studies of farmers have linked pesticides with certain cancers. Information on exposures from many of these studies was obtained by interview of farmers or their next-of-kin. The reliability and validity of data on pesticide use obtained by recall, often years after the event, have been questioned. Pesticide use, however, is an integral component in most agricultural operations, and the farmers' knowledge and recall of chemicals used may be better than for many other occupations. Contrary to general belief, many farmers typically use only a few pesticides during their lifetimes and make only a few applications per year. Data from U.S. Department of Agriculture surveys indicate that herbicides are applied to wheat, corn, soybeans, and cotton and that application of insecticides to corn averages two or fewer times per year. In epidemiologic studies at the National Cancer Institute, the proportion of farmers ever reporting lifetime use of five or more different chemicals was 7% for insecticides and 20% for herbicides. Surrogate respondents have often been used in epidemiologic studies of cancer; they are able to recall pesticide use with less detail than the farmers themselves. The pesticides reported by surrogates were the same as reported by subjects themselves, but with less frequency. Comparison of reporting by cases and controls provided no evidence of case-response (differential) bias; thus, inaccurate recall of pesticide use by subjects or surrogates would tend to diminish risk estimates and dilute exposure-response gradients. (Epidemiology 1993;4:55–62)

Keywords: pesticides, farmers, interviews, proxy respondents, misclassification, herbicides, data collection.

Epidemiologic studies from a number of countries indicate that farmers tend to be at higher risk for selected cancers than the general population.[1-3] These excesses occur despite a lower mortality among farmers for all causes combined and for most major causes of death. The specific agents in the agricultural environment that might be involved have not been clearly identified, but pesticides have received the most attention. There is good reason to focus on pesticides because the carcinogenic potential of a number of these chemicals has been demonstrated in animal bioassays. For about 50% of the pesticides evaluated, the International Agency for Research on Cancer has concluded that there is limited or sufficient evidence for carcinogenicity in experimental studies.[4] Similar findings have been obtained by the National Cancer Institute/National Toxicology Program (NCI/NTP), in which, of the 41 pesticides tested, six were positive in both sexes of two species, 10 were positive in both sexes of one species, six were positive in one sex of one species, and 19 gave no evidence of carcinogenicity.[5] These summaries can be viewed optimistically or pessimistically depending on whether you consider a 50% positive rate as reassuring or alarming. It underscores, however, the need to identify which pesticides are likely to pose a cancer risk to humans.

The surest way to identify carcinogenic pesticides already on the market is by epidemiologic investigations. In epidemiologic studies, the need to extrapolate from artificially high exposures and from one species to another is not required, as it is in animal bioassays. Epidemiologic studies, however, have limitations. The concerns raised regarding studies of pesticides and cancer usually focus on the limitations of exposure assessment and arise from a belief held by many that farmers cannot reliably report their exposure history.[6,7] Assembling information on past pesticide use in epidemiologic studies is difficult, and the reliability and validity of exposures reported retrospectively by subjects should be assessed.

Questions raised regarding exposure assessment in epidemiologic studies of agricultural use of pesticides include: (1) Can farmers accurately recall the pesticides they used from the large number of formulations on the market? (2) Is there corroborative evidence regarding the accuracy of reported use of pesticides by farm-

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Submitted June 5, 1992; final version accepted September 14, 1992.

ers? (3) What is the quality of information obtained from surrogate respondents? and (4) Does the pesticide history-taking technique, that is, open-ended *vs* probe, differentially affect reporting by cases and controls? In this paper, we use data from National Cancer Institute studies and other resources to address these issues.

Methods

The data for this paper come from U.S. Department of Agriculture (USDA) surveys and National Cancer Institute case-control studies of cancer in Kansas,[8] Iowa and Minnesota,[9,10] and Nebraska.[11] These three case-control studies had similar designs, including a population-based series of adult cancer cases (lymphatic and hematopoietic system and soft tissue sarcoma) with controls selected by random-digit telephone dialing (for living cases under age 65), from the Health Care Finance Administration (for living cases 65 or older), and from death certificates (for deceased cases). The studies in Iowa/Minnesota[9] and Kansas[8] included only white men, whereas the study in Nebraska[11] included white men and white women. Interviews were conducted with subjects, or their next-of-kin (if the cases were deceased or incapacitated), and they followed a structured format. The interviews in Iowa and Minnesota were in person, whereas those in Kansas and Nebraska were by telephone. In each investigation, we sought detailed information on specific pesticides used.

Each of the National Cancer Institute studies included methodologic components to address issues in exposure assessment of pesticides. In the Iowa/Minnesota study, interviews with both farmers and their wives were obtained for a sample of subjects.[10] In Kansas,[8] we sought interviews with pesticide suppliers for 130 farmers to evaluate comparability of reporting. In Nebraska,[11] we obtained information on pesticides that the subjects reported in response to an open-ended question that did not name specific pesticides. In this study, we also collected information on pesticides recalled only after the interviewer provided a prompt by naming the specific chemical.

Results

Number of Pesticides Used

Table 1 provides information on agricultural use of pesticides in 1990 on different crops.[12] For some of these crops, many acres are not treated every year; for example, two-thirds of the acres of wheat were not treated with any herbicide. Another USDA survey found that the proportion of farmers reporting no pesticide use in 1982 by crop was 14% for corn, 3% for cotton, 37% for sorghum, 7% for soybeans, 76% for oats, 65% for wheat, 86% for alfalfa, and 90% for pasture.[13] Table 1 shows that the average number of applications per year exceeded two only for herbicides on cotton. Finally, for any crop/pest combination, there are several pesticides that may be used, but a few products tend to dominate the market. Three or fewer different pesticides account for 30–70% of the treated acres for crops listed in Table 1. Applications are sometimes mixtures of pesticides, but this is a recent technique, and even now, mixed applications seldom include more than three chemicals.

TABLE 1. Pesticide use by crop, 1990*

Crop	Pesticide	% Acres Treated	Average Times Applied	% of Acres Treated by 3 Pesticides
Wheat	Herbicides	34	1.07	57
Corn	Herbicides	95	1.41	44
Corn	Insecticides	32	1.09	70
Soybeans	Herbicides (north)	97	1.48	34
Soybeans	Herbicides (south)	93	1.60	32
Cotton	Herbicides	94	2.07	49

* Data from U.S. Department of Agriculture (12).

Table 2 lists the major herbicides and insecticides used in agriculture, according to USDA surveys in 1966, 1971, and 1976.[14,15] Although there are over 25 insecticides and 25 herbicides listed, according to poundage, only a few are widely used. For example, five insecticides account for 70% of all use by weight in 1966, 70% in 1971, and 73% in 1976. For herbicides, the top five by weight accounted for 68% in 1971 and 82% in 1976. The rank order of the pesticides by weight also has not changed radically over time. Four of the top five insecticides in 1966 were still in the top five in 1971, and three in 1976. For herbicides, four of the top five in 1971 remained in the top five in 1976.

From interviews in an epidemiologic study in Iowa and Minnesota, we found that farmers did not report using large numbers of pesticides during their lifetimes. Forty-six per cent reported that they used no herbicides, 17% no insecticides, and 91% no fungicides (Table 3). Seventeen per cent, 42%, and 9% of the farmers reported that they had only used one herbicide, insecticide, or fungicide, respectively. Only 20% reported ever using five or more herbicides, 7% five or more insecticides, and 0% five or more fungicides. We found similar results from a study in Nebraska (data not shown).[11] Nineteen per cent of Nebraska farmers reported that they had used five or more herbicides, and 33% reported use of five or more insecticides.

TABLE 2. Amount (× 1000 Pounds) and Relative Ranking of Major Pesticides Used in Agriculture in 1966, 1971, and 1976*

Chemical†	Pounds (Rank)		
	1966	1971	1976
Insecticides			
Toxaphene	34,605 (1)	37,464 (1)	30,700 (1)
DDT	27,004 (2)	14,324 (4)	0
Aldrin	14,761 (3)	7,928 (6)	900 (13)
Carbaryl	12,392 (4)	17,838 (3)	9,300 (4)
Parathion	8,452 (5)	9,481 (5)	6,600 (5)
Methyl parathion	8,002 (6)	27,563 (2)	22,800 (2)
Diazinon	5,605 (7)	3,167 (12)	1,600 (9)
Malathion	5,218 (8)	3,602 (10)	Not provided
TDE (DDD)	2,896 (9)	244 (25)	Not provided
Methoxychlor	2,578 (10)	3,012 (13)	1,400 (11)
Strobane	2,016 (11)	216 (26)	Not provided
Ethion	2,007 (12)	2,326 (15)	Not provided
Disulfoton	1,952 (13)	4,079 (8)	5,500 (7)
Bidrin	1,857 (14)	807 (20)	300 (17)
Heptachlor	1,536 (15)	1,211 (18)	1,600 (9)
Azinphos-methyl	1,474 (16)	2,654 (14)	300 (17)
Trichorfon	1,060 (17)	617 (22)	Not provided
Dichlorvos	912 (18)	3,176 (11)	864 (14)‡
Endosulfan	791 (19)	882 (19)	800 (15)
Dieldrin	724 (20)	332 (24)	Not provided
Lindane	704 (21)	650 (21)	Not provided
Endrin	571 (22)	1,427 (17)	600 (16)
Chlordane	526 (23)	1,890 (16)	1,400 (11)
Ronnel	391 (24)	479 (23)	Not provided
Phorate	326 (25)	4,178 (7)	6,300 (6)
Bux	39 (26)	3,606 (9)	Not provided
Methomyl	0 (27)	0 (27)	2,500 (8)
Carbofuran	Not provided	Not provided	11,600 (3)
Herbicides (no data for 1966)			
Atrazine		57,216 (1)	90,300 (1)
2,4-D		33,252 (2)	38,400 (3)
Propachlor		23,730 (3)	11,000 (6)
Alachlor		14,754 (4)	88,500 (2)
Trifluralin		11,427 (5)	28,300 (4)
Amiben		9,555 (6)	4,400 (11)
Arsenicals		7,837 (7)	3,500 (15)
Propanil		6,656 (8)	6,900 (9)
Butylate		5,915 (9)	24,400 (5)
EPTC		4,409 (10)	8,600 (7)
Vernolate		3,736 (11)	Not provided
Fluometuron		3,334 (12)	5,300 (10)
Alanap		3,332 (13)	4,300 (12)
MCPA		3,284 (14)	Not provided
Propazine		3,171 (15)	3,900 (13)
Nitralin		2,706 (16)	Not provided
Linuron		1,803 (17)	8,400 (8)
Simazine		1,723 (18)	2,500 (16)
2,4,5-T		1,339 (19)	Not provided
Fluorodifer		1,330 (20)	Not provided
Norea		1,323 (21)	Not provided
Diuron		1,229 (22)	900 (17)
Pebulate		1,062 (23)	300 (18)
Dalapon		1,032 (24)	Not provided
Dicamba		420 (25)	3,600 (14)

* 1966 and 1971 data from Ref 14; 1976 data from Ref 15.
† Acronyms include: DDT, dichlorodiphenyltrichloroethane; TDE, tetrachlorodiphenylethane; DDD, dichlorodiphenyldichloroethane; 2,4-D, (2,4-dichlorophenoxy)acetic acid; EPTC, S-ethyl dipropylthiocarbamate; MCPA, 2-methyl-4-chlorophenoxyacetic acid; 2,4,5-T, (2,4,5-trichlorophenoxy)acetic acid.
‡ Based on use on livestock only.

TABLE 3. Number (and Proportion) of Different Pesticides Reported by Farmers (Controls Who Farmed at Age 25 or Older) in Iowa and Minnesota

No. of Pesticides Reported	Number (%)		
	Herbicides	Insecticides	Fungicides
0	280 (46)	104 (17)	553 (91)
1	102 (17)	254 (42)	55 (9)
2–4	104 (17)	206 (34)	0 (0)
5+	122 (20)	44 (7)	0 (0)

Reliability of Recall of Specific Pesticides Used

In the study in Kansas,[8] pesticide suppliers were interviewed to see whether they could corroborate information on pesticide use reported by farmers. We sought information from the major suppliers for 130 farmers. Interviews were completed with 110 suppliers (17 suppliers could not be located, and three said our subjects were not farmers, even though we had considerable information that they were). The agreement between suppliers and farmers regarding herbicide and insecticide use is shown in Table 4. Agreement was approximately 60% for cases and controls for use of both herbicides and insecticides. Agreement for years of insecticide use on major crops was also approximately 60%, whereas agreement for years of herbicide use was slightly lower, particularly for sorghum.

TABLE 4. Agreement between Farmers and Suppliers Regarding the Kansas Farmer's Use of Pesticides on Specific Crops

Pesticide/Crop	All Subjects		Controls		Cases	
	Number*	%	Number	%	Number	%
Ever used						
Herbicides	65/45	59	40/29	58	25/16	61
Insecticides	65/45	59	42/27	61	23/18	56
Years† of herbicide use on:						
Wheat	51/59	46	34/35	49	17/24	41
Corn	65/45	59	41/28	59	24/17	59
Sorghum	42/68	38	25/44	36	17/24	41
Pasture	58/52	53	37/32	54	21/20	51
Years† of insecticide use on:						
Wheat	67/33	61	42/17	61	25/16	61
Corn	69/41	63	41/28	59	28/13	68
Sorghum	61/49	55	39/30	57	22/19	54
Pasture‡						

* Number of agreements/number of disagreements.
† Years in categories of 0, 1–5, 6 or more, and unknown.
‡ Fewer than 5 users.

Accuracy of Reports from Surrogate Respondents

In each of the National Cancer Institute case-control studies,[8,9,11] we interviewed next-of-kin of deceased farmers to obtain information on the decedent's use of pesticides. Data from Iowa/Minnesota[9] in Table 5 show that surrogate respondents were approximately twice as likely as the farmers to respond "I don't know" to at least one pesticide from the list. Surrogates also reported use of fewer pesticides than did the farmers (Table 6). A larger proportion of surrogates than farmers reported no use of pesticides and three to five times as many farmers as surrogates reported using five or more herbicides or insecticides.

TABLE 5. Comparison of Farmer and Surrogate Respondents from Iowa and Minnesota Providing at Least One "Don't Know" Response Regarding Use of Specific Pesticides (Controls over Age 25)

Type of Pesticide*	% (Number) Giving at Least One "Don't Know" Response	
	Farmer Interview	Surrogate Interview
Herbicides	35 (150)	65 (140)
Crop insecticides	45 (180)	65 (145)
Animal insecticides	14 (55)	30 (65)

* Subjects were asked about 38 herbicides, 34 crop insecticides, and 30 animal insecticides.

TABLE 6. Comparison of the Number of Pesticides Used Reported by Farmers and Surrogate Respondents (Controls over Age 25 from Iowa and Minnesota)

Number Used	% (Number)	
	Farmer Interviews	Surrogate Interviews
Herbicides		
0	38 (148)	62 (132)
1	16 (62)	19 (40)
2–4	19 (75)	13 (29)
5+	27 (108)	7 (14)
Crop insecticides		
0	46 (182)	63 (136)
1	18 (72)	22 (48)
2–4	20 (80)	11 (24)
5+	15 (59)	3 (7)
Animal insecticides		
0	13 (52)	24 (152)
1	37 (147)	50 (107)
2–4	39 (155)	24 (51)
5+	10 (39)	2 (5)

Table 7 compares the relative ranking of specific pesticides from subjects and surrogates by reporting frequency. The proportion of farmers reporting use of any specific chemical is typically two to five times larger than the proportion of surrogates. The rank order by number of times a specific chemical was reported by subjects and by surrogates, however, is quite similar, with Spearman correlation coefficients of 0.87 for herbicides, 0.71 for crop insecticides, and 0.80 for animal insecticides. When ranked by the number of persons reporting that the pesticide was used, the four most commonly reported herbicides [(2,4-dichlorophenoxy)acetic acid (2,4-D), atrazine, alachlor, and trifluralin] were reported in the same relative order for subjects and surrogates. The top four crop insecticides reported among farmers were in the top seven reported by surrogates, and four of the top five animal insecticides were the same for subjects and surrogates.

TABLE 7. Comparison of Reporting Frequency for Specific Pesticides from Farmers and Surrogate Respondents (Controls Who Farmed after the Age of 25 from Iowa/Minnesota)

	Farmer Interviews			Surrogate Interviews		
Pesticide*	No.	%	Rank	No.	%	Rank
Herbicides						
2,4-D	194	49	1	59	27	1
Atrazine	118	19	2	25	12	2
Alachlor	116	19	3	23	10	3
Trifluralin	103	26	4	17	8	4
Cyanazine	81	13	5	12	6	6
Dicamba	71	18	6	6	3	9
Chloramben	68	17	7	15	7	5
Bentazon	55	14	8	7	3	8
Glyphosate	53	13	9	2	1	11
Butylate	49	12	10	8	4	7
2,4,5-T	42	11	11	5	2	10
Crop insecticides						
Aldrin	95	24	1	13	6	3
DDT	64	16	2	26	12	1
Carbofuran	60	15	3	9	4	4
Phorate	48	12	4	6	2	7
Diazinon	40	10	5	6	3	7
Terbufos	39	10	6	4	2	12
Heptachlor	38	10	7	7	3	5
Copper arsenite	34	8	8	22	10	2
Fonofos	31	8	9	3	1	13
Carbaryl	30	8	10	6	3	7
Malathion	30	8	10	5	3	10
Dieldrin	26	7	12	2	1	15
Lindane	24	6	13	3	1	13
Chlordane	22	6	14	7	3	5
Lead arsenate	17	4	15	5	2	10
Toxaphene	15	4	16	0	0	17
Bufencarb	15	4	16	2	1	15
Animal insecticides						
Flyspray, NOS	236	60	1	129	60	1
DDT	106	27	2	45	21	2
Lindane	81	21	3	12	6	4
Malathion	59	15	4	9	4	5
Chlordane	37	9	5	4	2	6
Nicotine	36	9	6	13	6	3
Dichlorvos	35	9	7	3	1	7
Rotenone	21	5	8	1	<1	13
Famphur	17	4	9	3	1	7
Coumaphos	16	4	10	3	1	7
Toxaphene	16	4	10	3	1	7
Methoxychlor	15	4	12	2	1	12
Carbaryl	11	3	13	3	1	7
Ronnel	7	2	14	0	0	15
Dieldrin	6	2	15	1	<1	13
Trichlorfon	6	2	15	0	0	15

* See Table 2 for definitions of acronyms. NOS, not otherwise specified.

To compare directly responses from farmers with their next-of-kin surrogate respondents, we conducted interviews with wives and their farmer husbands (Table 8).[10] Surrogates tended to report fewer days per year of use of specific pesticides than the farmers. Correlations ranged from 0.23 to 0.80 for the different pesticides. Subjects and surrogate respondents agreed as to the category of frequency of use approximately 50–60% of the time, but it was better for more recent use, that is, after 1960, than for use before 1960.

Interview Technique: Open-Ended vs Probing with a List

In the Nebraska study,[11] subjects were first asked to respond to an open-ended question on their pesticide use. After they had volunteered all of the pesticides they could, the interviewer asked about the remaining

TABLE 8. Comparison of Farmers' and Their Spouses' Responses for Frequency of Pesticide Use*

Pesticide†	Used Before/After 1960	No. of Pairs	Correlation Coefficient	% Exact Agreement in Categories‡
Alachlor	After	25	0.80	52.0
Aldrin	After	30	0.63	66.7
Atrazine	After	30	0.78	60.0
Cyanazine	After	21	0.66	57.1
DDT	Before	23	0.23	30.4
Trifluralin	After	27	0.84	63.0
2,4-D	Before	26	0.30	48.4
	After	45	0.78	55.6
All herbicides		21	0.31	52.4
All insecticides		25	0.58	68.0

* Modified from Brown *et al.*[10]
† See Table 2 for definitions of acronyms.
‡ Categories for specific pesticides and all herbicides were 1–4, 5–9, and ≥10 days per year. Categories for all insecticides were 1–15, 16–60, and ≥61 days per year.

chemicals on a list previously developed by the investigators of commonly used pesticides to see whether this prompt could spark recall of having ever used the specific chemicals. A comparison of the number of pesticides mentioned in the open-ended questions with the number obtained from open-ended questions plus the prompts is shown in Table 9. This table includes farmers over the age of 25 who reported living on farms where pesticides were used. The number of pesticides volunteered by farmers was similar among cases and controls. Probing dramatically increased the number of pesticides reported. About 40% of the farmers who reported no use of insecticides or herbicides to the open-ended question responded positively to at least one of these chemicals when prompted with specific names. Among those who had volunteered no insecticides, 47% of the cases and 27% of the controls responded positively to at least one insecticide when prompted. Among farmers volunteering no use of herbicides, 49% of the cases and 38% of the controls were able to name at least one herbicide when prompted. The proportion of subjects reporting use of five or more pesticides also increased dramatically with probing. The distribution of the number of herbicides and insecticides reported from the open-ended question, however, was similar among cases and controls.

Table 10 displays volunteered *vs* probed information among cases and controls for selected pesticides. The proportion of farmers who volunteered that these pesticides were used was approximately equal among cases and controls, except for trifluralin, for which it was higher among controls. The proportions reporting use in response to prompts for specific pesticides were greater than the proportions from volunteered pesti-

TABLE 9. Number of Pesticides Reported from Open-Ended Questions (Volunteered) and Open-Ended plus Probed among Cases and Controls (White Male Farmers from Nebraska Age 26 or Older)

Pesticide and Number Used	Cases				Controls			
	Volunteered		Volunteered plus Probed		Volunteered		Volunteered plus Probed	
	Number	%	Number	%	Number	%	Number	%
Insecticides								
0	64	57	34	30	132	58	96	42
1	25	22	198	17	60	26	36	16
2–4	21	19	15	13	35	15	41	18
5+	2	2	45	40	1	<1	55	24
Total	113		113		228		228	
Herbicides								
0	35	38	18	20	84	49	52	30
1	30	33	19	21	41	24	35	20
2–4	22	24	27	30	42	24	41	24
5+	4	4	27	30	5	3	44	26
Total	91		91		172		172	

TABLE 10. Farmers' Reported Use of Selected Pesticides When Volunteered *vs* Probed by Case and Control Status (White Men from Nebraska)

Pesticide*	Cases				Controls			
	Volunteered		Probed		Volunteered		Probed	
	Number	%	Number	%	Number	%	Number	%
Insecticides								
DDT	16	33	33	67	38	41	54	59
Terbufos	7	33	14	67	8	30	19	70
Herbicides								
Alachlor	12	36	21	64	22	41	32	59
Cyanazine	3	19	13	81	9	26	25	74
2,4-D	47	64	26	36	74	64	42	36
Trifluralin	4	24	13	76	15	44	19	56

* See Table 2 for definitions of acronyms.

cides alone, except for 2,4-D. Sixty-four per cent of the cases and controls volunteered use of 2,4-D *vs* only 36% of those requiring a probe.

Discussion

The Federal Insecticide, Fungicide, and Rodenticide Act Amendment of 1988 mandated the review of the approximately 24,000 registered pesticide products on the market.[16] This large number may contribute to the general perception that each farmer uses many different pesticides each year. This impression, coupled with a belief that the specific pesticides used change from year to year, raises doubts regarding the validity of information on pesticide use obtained by interview. Obviously, if individual farmers used even a fraction of the pesticides available, it would be doubtful whether they could accurately recall the majority of them.

The number of different pesticides used on many agricultural commodities, however, is small. Data from USDA surveys and our epidemiologic studies indicate that, despite the availability of thousands of chemicals, the number of pesticides used by farmers is typically 10 or fewer rather than hundreds. Data from USDA also indicate that the specific pesticides used did not change radically between 1966 and 1976, at least for some types of agricultural commodities. Even for commodities for which pesticides are heavily used, such as vegetables, three or fewer chemicals typically account for 50% or more of the total amount of herbicides, insecticides, or fungicides used by weight.[17] The time period 1966–1976 was a time when rapid change might have been expected because of the shift from use of organochlorine to organophosphate insecticides. From 1966 to 1976, the share of the market for organochlorine insecticides on major crops dropped from 70% to 29%, whereas organophosphates rose from 22% to 49% and carbamates from 7% to 19%.[18] Thus, even during this period of relative instability, the problem of sorting out pesticide exposures in agriculture is probably no more difficult than for other exposures in many industrial situations. Studies in agriculture may possess a distinct advantage because farmers, who function as both owner and operator, may be able to provide more information on exposure than could usually be obtained from either workers or supervisors in industrial facilities. Farmers' use of pesticides is based on operational needs, and, consequently, they make reasoned decisions regarding pesticide use. Farmers must decide whether there is a pest problem, select the pesticide most likely to be effective, purchase the pesticide, record the purchase (costs are tax deductible), mix and apply the pesticide, and evaluate the success of the treatment. These activities tend to reinforce memory.

Methodologic efforts are needed, however, to assess the actual reliability and accuracy of farmers' reported use of pesticides. Our comparison of information from farmers with information from their pesticide suppliers indicates a moderate level of correspondence. It is important to remember, however, that the information from suppliers does not constitute a "gold standard." Thus, the overall accuracy of reports from farmers is probably better than suggested by this comparison, because some of the disagreement between farmers and suppliers must be due to errors from the suppliers.

Because of the rapidly fatal nature of many cancers, epidemiologic studies often must include interviews with surrogate respondents. For some factors of epidemiologic interest (for example, tobacco use), surrogates can provide reliable information. The accuracy of information on agricultural use of pesticides obtained from surrogates, however, is unknown. In one of our studies, we found that surrogate respondents were a poorer source of information than farmers themselves.[9] They reported a smaller number of pesticides ever used and a smaller proportion of farmers who used any specific pesticide, and they had a greater propensity to give an "I don't know" response. Studies including surrogate respondents, therefore, would have lower study power because fewer subjects would be classified as exposed. Interviews with the farmers themselves is obviously preferred. Interestingly, however, the rank orders of specific pesticides by the number of surrogates and subjects reporting the chemicals used were quite similar. This finding indicates that the chemicals reported by surrogates may essentially be the same as reported by farmers, but with lower absolute frequency. In the absence of evidence of case-response bias, it appears that errors associated with the reported use of pesticides would tend to bias risk estimates toward the null.[19]

Differential information bias is a concern in case-control studies.[19] Publicity about pesticides and disease and the tendency of individuals with cancer to try to identify events in their life that may have caused their disease could result in case-response bias. This bias moves risk estimates away from the null and could create false-positive findings. If case-response bias were a problem, we might anticipate that cases would be better prepared than controls to volunteer pesticides which they believed were associated with their disease and to recall more pesticides on open-ended questions. The number of insecticides and herbicides volunteered

by cases and controls, however, was quite similar, providing no support for this contention.

Data presented here indicate that the major problems in assessing agricultural pesticide exposure based on information obtained from interviews would result in nondifferential misclassification. This error tends to bias risk estimates toward the null and to dilute exposure response gradients. It may cause false-negative results, but it is unlikely, although not impossible,[20] to result in false-positive findings. The approach that one should take to minimize effects of misclassification errors on risk estimates depends upon the prevalence of the exposure of interest in the population.[21] When the prevalence of exposure is low, the critical concern is to avoid classifying unexposed subjects as exposed. If the exposure prevalence is high, the reverse is true. Since some agricultural pesticides may be used quite commonly and others infrequently, it may not be possible to have a single classification system across all pesticides.

Prospective studies of farmers would provide one solution to the misclassification problem, since periodic assessment of exposures would reduce the problem of long-term recall. Prospective studies could also be used to determine the magnitude of exposure misclassification from retrospective assessments, which would be invaluable in evaluating results from case-control studies. The National Cancer Institute, in collaboration with the Environmental Protection Agency and the National Institute of Environmental Health Sciences, is initiating a long-term prospective study of farmers, plus their spouses and children, to assess agricultural and life-style factors that may present hazards to farm families.

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A section 508–conformant HTML version of this article is available at http://dx.doi.org/10.1289/EHP456.

Occupational Exposure to Pesticides and the Incidence of Lung Cancer in the Agricultural Health Study

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Background: Occupational pesticide use is associated with lung cancer in some, but not all, epidemiologic studies. In the Agricultural Health Study (AHS), we previously reported positive associations between several pesticides and lung cancer incidence.

Objective: We evaluated use of 43 pesticides and 654 lung cancer cases after 10 years of additional follow-up in the AHS, a prospective cohort study comprising 57,310 pesticide applicators from Iowa and North Carolina.

Methods: Information about lifetime pesticide use and other factors was ascertained at enrollment (1993–1997) and updated with a follow-up questionnaire (1999–2005). Cox proportional hazards models were used to calculate hazard ratios (HRs) and 95% confidence intervals (CIs), adjusting for smoking (smoking status and pack-years), sex, and lifetime days of use of any pesticides.

Results: Hazard ratios were elevated in the highest exposure category of lifetime days of use for pendimethalin (1.50; 95% CI: 0.98, 2.31), dieldrin (1.93; 95% CI: 0.70, 5.30), and chlorimuron ethyl (1.74; 95% CI: 1.02, 2.96), although monotonic exposure–response gradients were not evident. The HRs for intensity-weighted lifetime days of use of these pesticides were similar. For parathion, the trend was statistically significant for intensity-weighted lifetime days ($p = 0.049$) and borderline for lifetime days ($p = 0.073$). None of the remaining pesticides evaluated was associated with lung cancer incidence.

Conclusions: These analyses provide additional evidence for an association between pendimethalin, dieldrin, and parathion use and lung cancer risk. We found an association between chlorimuron ethyl, a herbicide introduced in 1986, and lung cancer that has not been previously reported. Continued follow-up is warranted.

Citation: Bonner MR, Beane Freeman LE, Hoppin JA, Koutros S, Sandler DP, Lynch CF, Hines CJ, Thomas K, Blair A, Alavanja MCR. 2017. Occupational exposure to pesticides and the incidence of lung cancer in the Agricultural Health Study. Environ Health Perspect 125:544–551; http://dx.doi.org/10.1289/EHP456

Introduction

Lung cancer is the leading cause of cancer-related death in the United States (American Cancer Society 2017) and in the world (Torre et al. 2015). Lung cancer mortality and incidence is lower among farmers in the United States than among the general population (Blair et al. 1993; Blair and Freeman 2009) potentially because of the low prevalence of smoking among U.S. farmers (Alavanja et al. 2004; Blair et al. 1992). Nonetheless, increased lung cancer mortality among licensed pesticide applicators has been reported (Barthel 1981; Becher et al. 1996; Blair et al. 1983; MacMahon et al. 1988; Pesatori et al. 1994), raising the possibility that exposure to certain pesticides may increase the risk of lung cancer among farmers. Only a few epidemiologic studies have assessed exposure to specific pesticides (Austin et al. 1989; MacMahon et al. 1988; Pesatori et al. 1994). MacMahon et al. (1988) reported a slight increase in the lung cancer standardized mortality ratio (SMR) [SMR = 135; 90% confidence interval (CI): 114, 158] among pesticide applicators and termite control operators exposed to chlordane and heptachlor. Blair et al. (1983) also observed an excess of lung cancer among termite and other structural pest–control applicators. Using banked serum samples from 919 residents of Charleston, South Carolina, Austin et al. (1989) did not find an association between serum DDT levels and respiratory cancer mortality among 19 cases. In a small, nested case–control study of structural pesticide workers in Florida, Pesatori et al. (1994) observed suggestive positive associations for diazinon [odds ratio (OR) = 2.0; 95% CI: 0.7, 5.5], carbaryl (OR = 4.2; 95% CI: 0.6, 27.2), dichlorodiphenyltrichloroethane (DDT) (OR = 2.6; 95% CI: 0.5, 14.3), and propoxur (OR = 12.4; 95% CI: 1.05, 100.3); no associations were observed for malathion, chlorpyrifos, parathion, or chlordane. We previously reported positive associations between select pesticides and the occurrence of lung cancer in the Agricultural Health Study (AHS) (Alavanja et al. 2004). Of the 50 pesticides evaluated, 7 (dicamba, metolachlor, pendimethalin, carbofuran, chlorpyrifos, diazinon, and dieldrin) showed some evidence of positive associations with lung cancer incidence. Pesticide-specific analyses of diazinon (Jones et al. 2015) and metolachlor (Silver et al. 2015) that evaluated lung cancer risk, among other cancer sites, have recently been published from the AHS. Jones et al. (2015) reported increased lung cancer incidence among male pesticide applicators with the highest exposure category of lifetime

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Supplemental Material is available online (http://dx.doi.org/10.1289/EHP456).

We thank the cohort members for their willingness and commitment to participate in the Agricultural Health Study. We also thank M. Dunn, K. Torres, and A. Miller from Westat (Rockville, MD) for their outstanding research support in helping refine questionnaires and in data collection and J. Barker, from Information Management Services, Inc. (Calverton, MD) for his quality control work with the final data set.

This work was supported in part by the intramural research program of the National Institutes of Health, the National Institute of Environmental Health Sciences (Z01-ES049030), and the National Cancer Institute (Z01-CP010119).

The U.S. Environmental Protection Agency through its Office of Research and Development collaborated in the research described here. It has been subjected to agency review and approved for publication. The findings and conclusions in this report are those of the author(s) and do not necessarily represent the views of the National Institute for Occupational Safety and Health.

The authors declare they have no actual or potential competing financial interests.

Received: 7 December 2015; Revised: 3 May 2016; Accepted: 14 June 2016; Published: 6 July 2016.

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days of diazinon use [rate ratio (RR) = 1.60; 95% CI: 1.11, 2.31; p_{trend} = 0.02] as well as with intensity-weighted lifetime days of diazinon use (RR=1.41; 95% CI: 0.98, 2.04; p_{trend} = 0.08). Silver et al. (2015) found no association with either lifetime days or intensity-weighted lifetime days of metolachlor use.

Herein, we have used the AHS to investigate associations between lifetime use of 43 pesticides and the incidence of lung cancer with an additional 414 lung cancer cases and 10 years of follow-up beyond an earlier evaluation (Alavanja et al. 2004) and with updated information regarding more recent pesticide use and cigarette smoking status.

Methods and Materials

The AHS has been described previously (Alavanja et al. 1996). Briefly, we enrolled 57,310 restricted-use pesticide applicators residing in Iowa [commercial and private (farmer) = 36,792] and North Carolina (private applicators = 20,518) between 1993 and 1997 (AHS data release: P1REL201209.00, P2201209.00, and AHSREL201304.01). Vital status through 31 December 2011 was ascertained via linkage with state mortality files and the National Death Index. First primary, incident lung cancer cases that occurred between enrollment and 31 December 2010 in North Carolina and 31 December 2011 in Iowa were identified via linkage with the Iowa and North Carolina state cancer registries. Prevalent cancers (n = 1,094) and individuals who sought to obtain pesticide registration in Iowa or North Carolina but did not reside in these states (n = 341) were excluded from the analysis. Participants (n = 1,113) who moved out of Iowa or North Carolina were censored at the year they departed. All applicable Institutional Review Boards approved the protocol, and all participants provided informed consent.

Exposure Assessment

At enrollment, participants completed a self-administered questionnaire (http://aghealth.nih.gov/collaboration/questionnaires.html) indicating whether they had ever mixed or applied 50 specific pesticides. The number of years and the number of days per year the applicator personally mixed or applied a particular pesticide was also queried on the enrollment questionnaire for 22 pesticides. This detailed information about days and years of use for the remaining 28 pesticides was obtained in a supplementary take-home questionnaire completed by ~44% of the cohort. In addition, the enrollment questionnaire gathered information on pesticide application methods, mixing, repair of pesticide application equipment, and the use of personal protective equipment (PPE). Smoking history, alcohol consumption in the past 12 months, fruit and vegetable consumption, other agricultural activities, non-farm occupational exposures, family history of cancer, medical conditions, and medicines were also ascertained at enrollment. Blair and colleagues have previously shown that the reliability of reporting of pesticide use in the AHS questionnaire is similar to that for other factors routinely obtained by questionnaire for epidemiologic studies (Blair et al. 2002).

Lifetime exposure-days of use for each of the 50 pesticides was calculated from the questionnaire data as the product of the number of years a participant personally mixed or applied each specific pesticide times the number of days in an average year that pesticide was used. In addition, we used an estimate of exposure intensity based on an algorithm generated by Dosemeci et al. (2002) that was developed from a comprehensive review of the literature and was updated and supplemented by Coble et al. (2011). This algorithm also used pesticide monitoring conducted in the AHS (Hines et al. 2008; Thomas et al. 2010) to calculate an intensity-weighted lifetime exposure-days score for each pesticide [exposure intensity × lifetime exposure-days]. The exposure intensity score weights aspects of pesticide use that may modify the intensity of exposure, such as whether an applicator personally mixed pesticides for application, application methods used, repair of pesticide application equipment, and the use of PPE. Dermal absorption is generally considered the major route of exposure for many pesticides (Maroni et al. 2000). Pesticide monitoring in the AHS found that chemical-resistant glove use was a more important determinant of urinary, airborne and dermal levels of pesticides than was initially assumed (Hines et al. 2008; Thomas et al. 2010). Consequently, the updated exposure intensity score weighted the use of protective gloves more heavily (Coble et al. 2011).

Information on pesticide use was updated between 1999 and 2005 with the use of a computer-assisted telephone interview (CATI). Participants were asked to report all pesticides used in the year prior to the interview as well as the frequency of use. Because only 36,342 applicators (63%) completed both the baseline and follow-up questionnaires, we used multiple imputation with logistic regression and stratified sampling to impute missing pesticide exposure information for 20,968 applicators who did not complete the follow-up interview (Heltshe et al. 2012).

In addition to updating pesticide use information between 1999 and 2005 with the CATI, smoking status (current, past, never), but not pack-years, was also updated. To update pack-years of cigarette smoking among current smokers (n = 7,637), we multiplied the number of cigarettes smoked that was reported in the enrollment questionnaire by the number of intervening years between the enrollment and the follow-up interview. These additional pack-years of cigarette smoking were then added to the total pack-years calculated from the enrollment questionnaire to update total pack-years of cigarette smoking. For participants who reported being current smokers on the enrollment questionnaire but reported being former smokers in the follow-up interview (n = 1,712), the number of cigarettes smoked per day reported at enrollment was used in the aforementioned calculation, and the number of years of smoking during the intervening time period was estimated to comprise half the time period. This same algorithm was used for participants who reported being former smokers at enrollment, but reported smoking currently in the follow-up interview (n = 573). For participants missing information on smoking on the enrollment and the follow-up interview (n = 1,051), pack-years of smoking was not imputed. Similarly, because a small proportion of participants (n = 1,033) was missing information on the number of cigarettes smoked per day (enrollment questionnaire), pack-years of smoking was not imputed. In addition, participants missing information on other potential confounders (e.g., age, sex, total lifetime pesticide days) (n = 4,338) were also excluded. In total, 7,498 participants were excluded, leaving 49,812 (89%) participants for the statistical analysis of pesticide exposure.

Statistical Analyses

We used Cox proportional hazards to estimate hazard ratios and 95% confidence intervals, using age at risk as the time scale, to assess potential associations between pesticide use and the incidence of lung cancer. We evaluated 43 specific pesticides here. Diazinon (Jones et al. 2015) and metolachlor (Silver et al. 2015) were not evaluated because results from the evaluation of these pesticides have recently been published. In addition, 5 other pesticides (trichlorfon, carbon tetrachloride/carbon disulfide, aluminum phosphide, ziram, and 2-(2,4,5-trichlorophenoxy)propionic acid (2,4,5-TP; fenoprop) were not evaluated because there were fewer than 15 exposed lung cancer cases, which is too few for meaningful analyses. Lifetime days of exposure and intensity-weighted lifetime exposure-days were both categorized into quartiles based on the distribution among the lung cancer cases to assess exposure–response gradients where possible. For 7 pesticides (aldrin, captan, carbofuran, coumaphos, dieldrin, heptachlor, and toxaphene) tertiles were used because of

the relatively small number of exposed lung cancer cases. In addition to assessing cumulative lifetime exposure-days, we also conducted analyses in which lifetime exposure-days were lagged 5 and 15 years.

A priori covariates used in our previous report (Alavanja et al. 2004) included age, sex, pack-years of smoking separately for current and former smokers, and total lifetime days of pesticide use. We further evaluated potential confounding from cigarette smoking by including pack-years of cigarette smoking as a continuous variable; these two approaches yielded comparable risk estimates. We also assessed the potential for confounding by other covariates [education, body mass index, family history of lung cancer, race, state of residence, fruit and vegetable intake, alcohol consumption, and raising poultry and livestock, which is associated with reduced lung cancer incidence among farmers in the AHS (Beane Freeman et al. 2012)]; none of these variables meaningfully influenced the estimates of relative risk. In addition to adjusting for total lifetime days of pesticide application, we also conducted additional analyses adjusting for lifetime days of diazinon, dieldrin, and pendimethalin use because these pesticides were previously associated with lung cancer incidence in the AHS. Our final models included the *a priori* covariates only.

We used PROC MIANALYZE (SAS 9.3; SAS Institute Inc.) to account for our multiple imputation approach. For the pesticides dieldrin, 2,4,5-TP, parathion, chlordane, DDT, heptachlor, and toxaphene, there was no variability between the five imputed sets because their registrations had been canceled before the Phase 2 interviews were conducted. Therefore, standard proportional hazards models were used. *p*-Values for trend were calculated using natural log–transformed versions of the continuous exposure variables while adjusting for the covariates. We performed analyses stratified by smoking status to assess potential effect measure modification. In addition, we conducted analyses by lung cancer histologic type (adenocarcinoma vs. non-adenocarcinoma). These analyses are presented in Tables S1 and S2 only because the small number of lung cancer cases among strata limited precision and interpretation.

Results

Since our previous report (Alavanja et al. 2004), 414 additional first primary, histologically confirmed incident lung cancer cases have occurred. In total, 654 first primary incident lung cancer cases were included in the present report, with an average follow-up of 14.8 years since AHS enrollment. Selected characteristics are presented in Table 1. As expected, a higher proportion of lung cancer cases than of noncases was observed with older age and greater pack-years of cigarette smoking. The proportion of lung cancer cases was slightly higher among non-whites, among those residing in North Carolina, and among those having a history of chronic lung disease. We did not find differences with sex or family history of lung cancer. Lung cancer cases were less likely to regularly consume fruits, vegetables, and alcohol than were noncases.

Table 2 presents the HRs for lifetime days of use and intensity-weighted lifetime days for 13 pesticides and lung cancer. Results were included if they had been previously associated with lung cancer in the AHS [dicamba, pendimethalin, carbofuran, chlorpyrifos, and dieldrin (Alavanja et al. 2004)], in other epidemiologic studies [malathion (Pesatori et al. 1994), parathion (Pesatori et al. 1994), carbaryl (Pesatori et al. 1994), chlordane (MacMahon et al. 1988), DDT (Austin et al. 1989), and heptachlor (MacMahon et al. 1988)], or otherwise showed an association with lung cancer in this evaluation (chlorimuron ethyl). Table S3 depicts the hazard ratios for the remaining 30 pesticides, none of which was positively associated with lung cancer. Lifetime days of chlorimuron ethyl were associated with statistically significant increased risk in the highest exposure category only (HR = 1.74; 95% CI: 1.02, 2.96) but did not show an exposure–response trend (p_{trend} = 0.180). The highest

Table 1. Selected baseline characteristics of lung cancer cases and noncases, Agricultural Health Study (1993–1997).

Characteristic[a]	Lung cancer cases *n* = 546 (%)	Cohort members (noncases) *n* = 49,266 (%)
Age		
< 55	170 (31.1)	36,434 (74.0)
55–59	114 (20.9)	4,693 (9.5)
60–64	108 (19.8)	3,754 (7.6)
65–69	78 (14.3)	2,465 (5.0)
70–74	57 (10.4)	1,307 (2.7)
≥ 75	19 (3.5)	613 (1.2)
Smoking status (pack-years)[b]		
Never smoker	57 (10.4)	26,803 (54.4)
Former < 3.75	15 (2.8)	4,552 (9.2)
Former 3.75–15	27 (5.0)	4,128 (8.4)
Former > 15	176 (32.2)	5,405 (11.0)
Current < 11.25	26 (4.8)	1,522 (3.1)
Current 11.25–28.5	49 (9.0)	2,623 (5.3)
Current > 28.5	196 (35.9)	4,233 (8.6)
Sex		
Male	535 (98.0)	48,005 (97.4)
Female	11 (2.0)	1,261 (2.6)
Race[b]		
White	519 (95.1)	48,060 (97.8)
Black/other	27 (4.9)	1,103 (2.2)
State of residence		
Iowa	231 (42.3)	32,895 (66.8)
North Carolina	315 (57.7)	16,371 (33.2)
Education (years)[b]		
< 12	128 (24.1)	4,124 (8.5)
12	268 (50.5)	22,797 (47.2)
> 12	135 (25.4)	21,363 (44.2)
Other chronic lung disease (bronchitis and emphysema)[b]		
No	455 (89.7)	45,165 (96.4)
Yes	52 (10.3)	1,683 (3.6)
Family history of lung cancer[b]		
No	442 (90.4)	43,549 (93.7)
Yes	47 (9.6)	2,927 (6.3)
Vegetable intake (servings/week)[b]		
≤ 4	173 (35.2)	15,228 (32.8)
5–7	188 (38.3)	16,913 (36.5)
> 7	130 (26.5)	14,223 (30.7)
Fruit intake (servings/week)[b]		
≤ 2	204 (40.0)	15,313 (32.5)
3–6	189 (37.1)	18,627 (39.6)
≥ 7	117 (22.9)	13,128 (27.9)
Alcohol intake (servings/time period)[b]		
Never	227 (44.3)	14,843 (31.4)
≤ 3/month	121 (23.6)	12,928 (27.4)
≥ 4/week	165 (32.2)	19,439 (41.2)

[a]Using response categories from the Agricultural Health Study enrollment questionnaire.
[b]Numbers do not sum to total because of missing data.

quartile of lifetime days of pendimethalin use also showed a positive association with lung cancer (HR = 1.50; 95% CI: 0.98, 2.31). We further divided the 4th quartile at its median of lifetime days of pendimethalin. The HR for the lower 50% of the 4th quartile was 1.26 (95% CI: 0.65, 2.46), and the HR for those in the upper 50% of the 4th quartile was 2.52 (95% CI: 1.31, 4.83), although the p for trend was not significant (p_{trend} = 0.283). Lifetime days of dieldrin use also showed a positive association in the highest exposure tertile (HR = 1.93; 95% CI: 0.70, 5.30), as did the HR for the intensity-weighted lifetime exposure-days metric (HR = 2.06; 95% CI: 0.95, 4.43). The lowest and highest quartiles of lifetime days of DDT use showed a slight excess in risk, although a monotonic exposure–response gradient was not evident (p_{trend} = 0.695). Similarly, the highest quartile of lifetime days of malathion use showed a slight excess risk (HR = 1.35; 95% CI: 0.93, 1.97). Although parathion was only slightly associated with the risk of lung cancer in the highest quartile [(HR = 1.17; 95% CI: 0.51, 2.68) for lifetime days and (HR = 1.20; 95% CI: 0.58, 2.47) for intensity-weighted lifetime days], the test for trend was statistically significant for intensity-weighted lifetime days (p = 0.049) and borderline for lifetime days (p = 0.073). The lowest exposure category of lifetime days use for maneb/mancozeb had a statistically significant increased risk of lung cancer (HR = 3.27; 95% CI: 1.54, 2.20), but the highest exposure category was not elevated, and there was no evidence of an exposure–response gradient (p_{trend} = 0.939), nor were any of the other exposure categories significantly increased. Carbaryl, carbofuran, chlordane, chlorpyrifos, and heptachlor were not associated with the incidence of lung cancer. Dicamba showed a statistically significant inverse exposure–response trend, although the lowest risks were seen in the lower quartiles of exposure. Generally, the HRs for the intensity-weighed lifetime days for these pesticides were similar to the lifetime-days metric (Table 2).

Table 3 shows the results of lagging lifetime days of exposure 5 and 15 years. The HRs from lagging lifetime exposure-days by 5 and 15 years were somewhat lower than those from unlagged analyses for pendimethalin

Table 2. Hazard ratios and 95% confidence intervals for lung cancer by lifetime days pesticide exposure and intensity-weighted lifetime days, Agricultural Health Study.

	Lifetime days			Intensity-weighted lifetime days		
Pesticide	Cases (n)	Hazard ratio[a] (95% CI)	p for trend	Cases (n)	Hazard ratio[a] (95% CI)	p for trend
Chlorimuron ethyl (herbicide; pyrimidinylsulfonylurea)[b]						
Nonexposed	180	1.0 (Reference)		180	1.0 (Reference)	
Q1	14	1.10 (0.64, 1.90)		21	1.09 (0.69, 1.72)	
Q2	37	0.96 (0.67, 1.38)		21	0.97 (0.62, 1.51)	
Q3	11	1.17 (0.64, 2.16)		20	1.04 (0.65, 1.68)	
Q4	16	1.74 (1.02, 2.96)	0.180	16	1.69 (1.00, 2.83)	0.294
Dicamba (herbicide; benzoic acid)						
Nonexposed	293	1.0 (Reference)		293	1.0 (Reference)	
Q1	38	0.64 (0.44, 0.92)		39	0.57 (0.40, 0.82)	
Q2	45	0.57 (0.40, 0.83)		44	0.66 (0.47, 0.95)	
Q3	45	0.75 (0.55, 1.04)		36	0.73 (0.48, 1.10)	
Q4	36	0.86 (0.60, 1.24)	0.007	44	0.81 (0.59, 1.13)	0.001
Pendimethalin (herbicide; dinitroaniline)[b]						
Nonexposed	160	1.0 (Reference)		160	1.0 (Reference)	
Q1	21	1.00 (0.61, 1.62)		25	0.81 (0.52, 1.26)	
Q2	33	0.85 (0.58, 1.24)		32	0.81 (0.50, 1.31)	
Q3	29	0.91 (0.58, 1.42)		27	1.26 (0.82, 1.92)	
Q4	28	1.50 (0.98, 2.31)	0.283	26	1.47 (0.93, 2.31)	0.551
Carbaryl (insecticide; carbamate)[b]						
Nonexposed	112	1.0 (Reference)		112	1.0 (Reference)	
Q1	58	0.93 (0.66, 1.30)		47	0.94 (0.65, 1.36)	
Q2	38	0.99 (0.66, 1.49)		35	0.99 (0.67, 1.46)	
Q3	33	1.15 (0.76, 1.74)		41	1.16 (0.79, 1.40)	
Q4	28	1.17 (0.76, 1.79)	0.436	34	1.04 (0.70, 1.54)	0.757
Carbofuran (insecticide; chlorinated organic)						
Nonexposed	336	1.0 (Reference)		336	1.0 (Reference)	
Q1	40	0.76 (0.55, 1.05)		32	0.81 (0.56, 1.16)	
Q2	29	0.80 (0.54, 1.19)		31	0.80 (0.55, 1.16)	
Q3	23	1.08 (0.62, 1.89)		29	0.87 (0.59, 1.29)	
Q4	28	0.99 (0.67, 1.47)	0.299	28	0.88 (0.59, 1.30)	0.133
Chlordane (insecticide; chlorinated organic)[b]						
Nonexposed	169	1.0 (Reference)		169	1.0 (Reference)	
Q1	22	1.57 (1.01, 2.46)		17	1.64 (0.99, 2.70)	
Q2	26	1.13 (0.75, 1.71)		17	1.34 (0.81, 2.21)	
Q3	12	0.95 (0.53, 1.70)		21	0.88 (0.56, 1.39)	
Q4	13	1.13 (0.64, 2.01)	0.426	18	1.27 (0.78, 2.08)	0.403
Chlorpyrifos (insecticide; phosphorothioate)						
Nonexposed	339	1.0 (Reference)		339	1.0 (Reference)	
Q1	54	0.84 (0.63, 1.13)		44	0.91 (0.66, 1.25)	
Q2	52	1.08 (0.79, 1.48)		41	0.74 (0.53, 1.03)	
Q3	41	0.86 (0.61, 1.21)		40	1.03 (0.74, 1.44)	
Q4	46	0.98 (0.71, 1.35)	0.497	38	0.88 (0.62, 1.25)	0.210
DDT (insecticide; chlorinated organic)[b]						
Nonexposed	140	1.0 (Reference)		140	1.0 (Reference)	
Q1	20	1.45 (0.92, 2.38)		29	1.01 (0.68, 1.52)	
Q2	42	0.86 (0.61, 1.22)		27	0.96 (0.63, 1.45)	
Q3	22	1.09 (0.69, 1.72)		25	0.99 (0.64, 1.53)	
Q4	23	1.33 (0.84, 2.10)	0.695	26	1.46 (0.95, 2.25)	0.506
Dieldrin (insecticide; chlorinated organic)[b]						
Nonexposed	230	1.0 (Reference)		230	1.0 (Reference)	
T1	6	0.58 (0.26, 1.31)		5	1.01 (0.42, 2.47)	
T2	6	1.49 (0.66, 3.37)		4	0.50 (0.18, 1.34)	
T3	4	1.93 (0.70, 5.30)	0.472	7	2.06 (0.95, 4.43)	0.880
Heptachlor (insecticide; chlorinated organic)[b]						
Nonexposed	216	1.0 (Reference)		216	1.0 (Reference)	
Q1	6	1.19 (0.53, 2.68)		7	1.13 (0.53, 2.39)	
Q2	11	0.65 (0.35, 1.19)		6	0.56 (0.25, 1.26)	
Q3	10	0.89 (0.47, 1.68)		10	0.77 (0.41, 1.46)	
Q4	5	0.66 (0.27, 1.62)	0.228	9	0.82 (0.42, 1.60)	0.193
Parathion (insecticide; phosphorothioate)[b]						
Nonexposed	211	1.0 (Reference)		211	1.0 (Reference)	
Q1	5	1.60 (0.66, 3.89)		11	1.58 (0.86, 2.91)	
Q2	17	1.48 (0.90, 2.43)		9	1.37 (0.70, 2.69)	
Q3	7	1.65 (0.78, 3.52)		7	1.82 (0.86, 3.89)	
Q4	6	1.17 (0.51, 2.68)	0.073	8	1.20 (0.58, 2.47)	0.049
Malathion (insecticide; phosphorothioate)[b]						
Nonexposed	78	1.0 (Reference)		78	1.0 (Reference)	
Q1	28	0.98 (0.54, 1.78)		40	0.99 (0.61, 1.61)	
Q2	76	1.11 (0.80, 1.52)		57	1.02 (0.72, 1.43)	
Q3	35	1.00 (0.67, 1.50)		44	1.18 (0.81, 1.72)	
Q4	45	1.35 (0.93, 1.97)	0.168	43	1.37 (0.94, 2.00)	0.197
Maneb/mancozeb (fungicide; dithiocarbamate)[b]						
Nonexposed	214	1.0 (Reference)		214	1.0 (Reference)	
Q1	7	3.27 (1.54, 6.97)		11	3.21 (1.74, 5.91)	
Q2	11	1.39 (0.76, 2.57)		6	0.91 (0.40, 2.06)	
Q3	10	1.34 (0.71, 2.53)		9	1.44 (0.74, 2.81)	
Q4	5	0.72 (0.30, 1.76)	0.939	7	0.86 (0.40, 1.83)	0.436

Notes: CI, confidence interval; DDT, dichlorodiphenyltrichloroethane; Q, quartile; T, Tertile.
[a]Adjusted for age, smoking status and pack-years, sex, and total lifetime pesticide use.
[b]Lifetime-days of use were obtained from the take-home questionnaire.

and chlorimuron ethyl. The association between dieldrin and lung cancer incidence was not influenced because dieldrin use had ceased before either of these lag periods. No obvious pattern emerged from the lagged analysis of parathion.

Discussion

With an additional 10 years of follow-up and 414 additional first primary, histologically confirmed incident lung cancer cases, we reevaluated the associations between lifetime days and intensity-weighted lifetime days for 43 pesticides and relative risk for lung cancer. Independent AHS pesticide-specific analyses for diazinon (Jones et al. 2015) and metolachlor (Silver et al. 2015) were not included here because these results have been published elsewhere. We found evidence of positive, albeit imprecise, associations with lung cancer for pendimethalin and dieldrin. These two pesticides had elevated HRs in the highest exposure category, but the exposure–response gradients were neither monotonic nor statistically significant. Parathion showed some evidence of increased risk for lung cancer, but the trends were not monotonic, nor were the excesses the largest in the highest quartile of exposure. We observed an increased hazard ratio with the use of chlorimuron ethyl in the highest exposure category. Chlorimuron ethyl use was not associated with lung cancer in a previous AHS analysis (Alavanja et al. 2004). None of the other pesticides (chlorpyrifos, carbofuran, or dicamba) was associated with lung cancer risk in this reevaluation.

Pendimethalin has been shown to induce thyroid follicular cell adenomas in rats and is classified as a possible human carcinogen (Group C) by the U.S. Environmental Protection Agency (EPA) (1997). Previous analyses of pendimethalin in the AHS (Alavanja et al. 2004; Hou et al. 2006), however, have been inconsistent. There is limited experimental evidence linking pendimethalin to genotoxicity (Dimitrov et al. 2006) or carcinogenicity in rodents (Weed Society of America 2002). To our knowledge, no epidemiologic studies other than the AHS have investigated pendimethalin use and lung cancer risk. We see only weak evidence for an association from a borderline statistically significant association with lifetime days of use and intensity-weighted lifetime days. The

Table 3. Hazard ratios and 95% confidence intervals for lung cancer by 5- and 15-year lagged lifetime-days pesticide exposure, Agricultural Health Study.

	5-year lag			15-year lag		
Pesticide	Cases (*n*)	Hazard ratio[a] (95% CI)	*p* for trend	Cases (*n*)	Hazard ratio[a] (95% CI)	*p* for trend
Chlorimuron ethyl (herbicide; pyrimidinylsulfonylurea)[b]						
Nonexposed	181	1.0 (Reference)		206	1.0 (Reference)	
Q1	16	1.24 (0.75, 2.06)		16	0.87 (0.52, 1.44)	
Q2	35	0.90 (0.62, 1.31)		15	0.46 (0.27, 0.78)	
Q3	10	1.15 (0.60, 2.20)		5	0.65 (0.27, 1.59)	
Q4	16	1.61 (0.96, 2.71)	0.295	13	1.36 (0.77, 2.40)	0.222
Dicamba (herbicide; benzoic acid)						
Nonexposed	299	1.0 (Reference)		329	1.0 (Reference)	
Q1	38	0.62 (0.44, 0.88)		35	0.52 (0.37, 0.74)	
Q2	43	0.54 (0.38, 0.77)		34	0.47 (0.33, 0.67)	
Q3	45	0.73 (0.53, 1.00)		37	0.61 (0.43, 0.86)	
Q4	33	0.79 (0.55, 1.14)	0.001	21	0.59 (0.38, 0.93)	< 0.001
Pendimethalin (herbicide; dinitroaniline)[b]						
Nonexposed	161	1.0 (Reference)		201	1.0 (Reference)	
Q1	24	1.18 (0.76, 1.85)		12	0.49 (0.26, 0.90)	
Q2	30	0.78 (0.52, 1.18)		13	0.39 (0.22, 0.69)	
Q3	26	0.88 (0.55, 1.41)		8	0.33 (0.16, 0.68)	
Q4	25	1.31 (0.84, 2.05)	0.602	19	1.11 (0.68, 1.82)	0.003
Carbaryl (insecticide; carbamate)[b]						
Nonexposed	112	1.0 (Reference)		131	1.0 (Reference)	
Q1	55	0.87 (0.51, 1.22)		36	0.66 (0.46, 0.95)	
Q2	35	0.92 (0.61, 1.34)		31	1.00 (0.67, 1.48)	
Q3	29	1.20 (0.79, 1.82)		32	1.29 (0.87, 1.90)	
Q4	30	1.05 (0.70, 1.59)	0.787	16	0.61 (0.36, 1.04)	0.399
Carbofuran (insecticide; carbamate)						
Nonexposed	336	1.0 (Reference)		354	1.0 (Reference)	
Q1	40	0.76 (0.54, 1.05)		36	0.67 (0.47, 0.94)	
Q2	29	0.81 (0.55, 1.20)		23	0.67 (0.44, 1.02)	
Q3	24	1.11 (0.64, 1.91)		25	1.38 (0.78, 2.43)	
Q4	27	0.95 (0.64, 1.43)	0.261	18	0.63 (0.39, 1.02)	0.006
Chlordane (insecticide; chlorinated organic)[b]						
Nonexposed	169	1.0 (Reference)		172	1.0 (Reference)	
Q1	0	— (—)		1	8.72 (1.19, 64.22)	
Q2	48	1.30 (0.94, 1.79)		45	1.21 (0.87, 1.69)	
Q3	12	0.95 (0.53, 1.70)		11	0.89 (0.48, 1.63)	
Q4	13	1.13 (0.64, 2.01)	0.424	13	1.13 (0.64, 2.00)	0.605
Chlorpyrifos (insecticide; phosphorothioate)						
Nonexposed	344	1.0 (Reference)		401	1.0 (Reference)	
Q1	55	0.85 (0.63, 1.13)		44	0.63 (0.46, 0.86)	
Q2	49	0.98 (0.71, 1.35)		39	0.77 (0.56, 1.07)	
Q3	43	0.91 (0.66, 1.26)		20	0.43 (0.28, 0.68)	
Q4	41	0.86 (0.61, 1.20)	0.188	27	0.57 (0.38, 0.85)	< 0.001
DDT (insecticide; chlorinated organic)[b]						
Nonexposed	140	1.0 (Reference)		140	1.0 (Reference)	
Q1	20	1.48 (0.92, 2.38)		20	1.44 (0.90, 2.31)	
Q2	42	0.86 (0.61, 1.22)		42	0.87 (0.61, 1.23)	
Q3	22	1.09 (0.69, 1.72)		22	1.09 (0.69, 1.71)	
Q4	23	1.33 (0.84, 2.10)	0.695	23	1.35 (0.85, 2.13)	0.709
Dieldrin (insecticide; chlorinated organic)[b]						
Nonexposed	230	1.0 (Reference)		230	1.0 (Reference)	
T1	6	0.58 (0.26, 1.31)		6	0.59 (0.26, 1.32)	
T2	6	1.49 (0.66, 3.37)		6	1.44 (0.64, 3.26)	
T3	4	1.93 (0.70, 5.30)	0.471	4	2.09 (0.76, 5.75)	0.468
Heptachlor (insecticide; chlorinated organic)[b]						
Nonexposed	216	1.0 (Reference)		216	1.0 (Reference)	
Q1	6	1.19 (0.53, 2.68)		6	1.16 (0.51, 2.61)	
Q2	11	0.65 (0.35, 1.19)		11	0.65 (0.36, 1.20)	
Q3	10	0.89 (0.47, 1.68)		10	0.90 (0.47, 1.69)	
Q4	5	0.66 (0.27, 1.62)	0.228	5	0.67 (0.28, 1.64)	0.239
Parathion (insecticide; phosphorothioate)[b]						
Nonexposed	212	1.0 (Reference)		214	1.0 (Reference)	
Q1	4	1.22 (0.54, 3.28)		4	1.09 (0.40, 2.94)	
Q2	17	1.49 (0.90, 2.44)		15	1.44 (0.85, 2.43)	
Q3	7	1.63 (0.76, 3.47)		9	1.96 (1.00, 3.82)	
Q4	6	1.17 (0.51, 2.69)	0.083	4	0.81 (0.30, 2.24)	0.168
Malathion (insecticide; phosphorothioate)[b]						
Nonexposed	82	1.0 (Reference)		110	1.0 (Reference)	
Q1	26	0.93 (0.53, 1.62)		15	0.59 (0.34, 1.02)	
Q2	71	1.01 (0.73, 1.39)		53	0.69 (0.50, 0.96)	
Q3	35	0.67 (0.65, 1.45)		35	0.87 (0.59, 1.28)	
Q4	44	1.29 (0.89, 1.88)	0.303	32	0.85 (0.57, 1.28)	0.378
Maneb/mancozeb (fungicide; dithiocarbamate)[b]						
Nonexposed	216	1.0 (Reference)		221	1.0 (Reference)	
Q1	6	2.88 (1.27, 6.54)		6	3.20 (1.41, 7.20)	
Q2	9	1.14 (0.58, 2.23)		6	0.81 (0.36, 1.83)	
Q3	11	1.45 (0.79, 2.66)		10	1.40 (0.74, 2.64)	
Q4	5	0.71 (0.29, 1.74)	0.993	4	0.58 (0.22, 1.58)	0.566

Notes: CI, confidence interval; DDT, dichlorodiphenyltrichloroethane; Q, quartile; T, tertile.
[a]Adjusted for age, smoking status and pack-years, sex, and total lifetime pesticide use.
[b]Lifetime-days of use were obtained from the take-home questionnaire.

lung cancer excess with pendimethalin use was largely limited to the upper half of the upper quartile, but the exposure–response trends were not statistically significant.

Dieldrin is an organochlorine insecticide that was banned from agricultural use in 1970 by the U.S. EPA, although its use as a termiticide was permitted by the U.S. EPA between 1972 and 1987 (Stern 2014). There are concerns about ongoing low-level exposure because dieldrin is commonly found in hazardous waste sites and is relatively resistant to environmental degradation (Stern 2014). As with the previous analyses of the AHS cohort (Alavanja et al. 2004; Purdue et al. 2007), dieldrin was positively associated with lung cancer, but mainly in the highest tertile of use. Dieldrin has been shown to induce liver tumors in mice, but not in other rodents [International Association for Research on Cancer (IARC) 1987]. The small number of dieldrin-exposed lung cancer cases complicates interpretation here.

Parathion was recently designated by IARC as possibly carcinogenic to humans (group 2B), largely on the basis of experimental evidence (Guyton et al. 2015). To our knowledge, no previous epidemiologic studies (Pesatori et al. 1994), including our previous report (Alavanja et al. 2004), have found associations for parathion use with lung cancer specifically, although melanoma was associated with parathion use in the AHS (Dennis et al. 2010). In chronic feeding studies, parathion has been shown to be carcinogenic to Osborne-Mendel rats and to increase the incidence of alveolar/bronchiolar adenomas in B6C3F1 mice (Gulf South Research Institute et al. 1979). Furthermore, parathion has been demonstrated to damage DNA in human peripheral lymphocytes (Undeğer and Başaran 2005). In our study, the small number of exposed cases and the lack of a monotonic exposure–response gradient complicated interpretation. Although these data do not provide strong evidence to support an association, nearly all the exposure categories had excess risk and are deserving of continued investigation for a potential association between parathion and lung cancer. Malathion (Guyton et al. 2015) and DDT (Loomis et al. 2015) were also evaluated and were classified as probably carcinogenic to humans (group 2A), largely based on sufficient evidence in animals. The evidence in humans, however, was deemed limited, and the lung was not a site observed to be associated with either malathion or DDT use in the epidemiologic studies assessed. Further epidemiologic investigation of both malathion and DDT are warranted.

This is the first report from the AHS in which chlorimuron ethyl and maneb/mancozeb have been associated with lung cancer incidence. However, these new findings may be chance occurrences because they are based on relatively small numbers of exposed cases. Chlorimuron ethyl is a herbicide that was introduced in 1986 for use on soybeans. It was previously associated with wheeze among commercial applicators in the AHS (Hoppin et al. 2006). The U.S. EPA classifies chlorimuron ethyl as "not likely to be carcinogenic to humans" (U.S. EPA 2016). To our knowledge, there are no published epidemiologic reports on the relationship between chlorimuron-ethyl exposure and cancer. Maneb/mancozeb has been observed to potentiate cancer in rodents (Belpoggi et al. 2002) and to be genotoxic in cultured human lymphocytes (Srivastava et al. 2012). The U.S. EPA classifies these fungicides as probable human carcinogens (group B2) (U.S. EPA 2016). However, in the present analysis, maneb/mancozeb use was associated with lung cancer only in the lowest exposure category and did not display an exposure–response gradient.

To our knowledge, no epidemiologic studies outside of the AHS have investigated dicamba and lung cancer risk. In contrast to previous AHS evaluations, we saw no evidence of an association between dicamba and lung cancer in the present analysis with larger numbers, although *in vitro* evidence suggests that dicamba may be genotoxic (González et al. 2006, 2007). Contrary to earlier AHS evaluations, we also saw no evidence of an association between lung cancer and chlorpyrifos (Alavanja et al. 2004; Lee et al. 2004) or carbofuran (Alavanja et al. 2004; Bonner et al. 2005) use. There is experimental mechanistic evidence that chlorpyrifos can induce oxidative stress and oxidative DNA damage (Ojha and Srivastava 2014; Zafiropoulos et al. 2014) and that carbofuran may be genotoxic (Mladinic et al. 2012). The proportion of AHS cohort members using either chlorpyrifos or carbofuran has declined since enrollment (Hoppin et al. 2012). Our analysis focused on the active ingredients of formulated mixtures of commercial products. These formulations contain both active ingredients and so-called "inert ingredients," and we cannot rule out the possibility that changes in the formulated mixtures associated with dicamba, chlorpyrifos, and carbofuran products are associated with changes in observed associations. Conversely, previous associations observed between these chemicals and lung cancer with fewer cases may have been due to chance.

We observed a number of inverse associations with lagged exposures, particularly for the 15-year exposure lag. We cannot explain these inverse associations in our data; none of these inverse associations is supported by biologic evidence, however. Rather, the limited evidence that does exist suggests carcinogenic potential as previously noted for, for example, dicamba, chlorpyrifos, and maneb/mancozeb.

Several limitations are evident in the present analysis. Despite an additional 10 years of follow-up and a substantial accrual of lung cancer cases, the number of lung cancer cases exposed to some pesticides remains small and continues to hamper study precision as well as our ability to evaluate risk by histologic type of lung cancer and to explore effect modification by smoking, particularly for chemicals for which patterns of use information were collected only with the take-home questionnaire. In addition, the analysis relies on imputed pesticide use data for a substantial fraction of the cohort.

We cannot rule out the possibility for chance or multiple comparisons to explain some of our results. Although approaches to adjust for multiple comparisons exist, a number of authors have warned against using such measures in epidemiological studies (Rothman 1990; Savitz and Olshan 1995; Goldberg and Silbergeld 2011). Our goal was to describe the magnitude of associations between specific pesticides and lung cancer risk. As such, we prefer to let other epidemiological studies and other relevant evidence (e.g., toxicological data) help sort out the likely reality of the findings.

Although the reliability of information on pesticide use obtained from farmers is quite good and is comparable to that from other factors commonly obtained by questionnaire in epidemiologic studies such as smoking and alcohol consumption (Blair et al. 2002), some exposure assessment error undoubtedly occurs. In this prospective cohort study, exposure misclassification is likely to diminish estimates of relative risk and to mute any real exposure–response relationships (Blair et al. 2011).

Although information on smoking was included in the statistical models, the possibility of residual confounding by active smoking and secondhand smoke exposure should be considered. This possibility seems unlikely, however, because there was no evidence of a link between smoking and pesticide use. Links were certainly not evident with many pesticides because the use of most pesticides did not result in an increase in the relative risk of lung cancer. Thus, any residual confounding would have to be chemical-specific. We evaluated a number of factors, including use of other pesticides (diazinon, pendimethalin, dieldrin, and chlorimuron ethyl) that might potentially confound associations between specific pesticides and lung cancer, none of which meaningfully influenced the risk estimates in our analyses. Exposure to secondhand smoke was not ascertained in the AHS; however,

any confounding resulting from secondhand smoke is likely to be small in comparison to direct smoking.

There is the possibility that a healthy worker survivor effect (HWSE) may have attenuated or reversed the reported associations. Unfortunately, we cannot carefully evaluate for an HWSE because time-dependent exposure information before enrollment was not collected. Nonetheless, the likelihood of an HWSE is low in the AHS cohort because the participants are predominately farm owners/operators who have a sizable economic investment in their operation, providing an incentive to continue farming.

This study has a number of strengths. The study population comprises a large population of farmers and commercial pesticide applicators who can provide detailed and reliable information regarding their pesticide use history (Blair et al. 2002). Information on pesticide use, application practices, and other information was obtained before the onset of cancer, diminishing the chances of case–response bias. Loss to follow-up is minimal owing to the use of high-quality state cancer registries and vital records and to the low residential mobility of this cohort. An algorithm that incorporated several exposure determinants that predicted urinary pesticide levels was used to develop an intensity-weighted exposure metric in our study (Coble et al. 2011). Information on potential confounders, such as smoking and the use of other pesticides, was available and could be evaluated and controlled in the analysis.

Conclusion

Several epidemiologic studies have found associations between pesticides and lung cancer (Alavanja and Bonner 2012). In our continuing survey within the AHS, we have found that no specific class of pesticide is associated with lung cancer. Although the results were not entirely consistent, we did observe some evidence of associations with pendimethalin and dieldrin. In addition, we found possible new associations for chlorimuron ethyl and parathion with lung cancer that have not been previously observed in the AHS and deserve further evaluation.

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Environ Health Perspect
DOI: 10.1289/EHP456

Supplemental Material

Occupational Exposure to Pesticides and the Incidence of Lung Cancer in the Agricultural Health Study

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Table S1. Lung Cancer risk among applicators by lifetime exposure-days of indicated pesticide, stratified by smoking status, Agricultural Health Study

Pesticide by lifetime exposure-days	Never Smokers			Former Smokers			Current Smokers			
	Cases (*n*)	Hazard Ratio	95% confidence interval	Cases (*n*)	Hazard Ratio	95% confidence interval	Cases (*n*)	Hazard Ratio	95% confidence interval	P for Interaction
Chlorimuron-ethyl (herbicide; pyrimidinylsulfonylurea) [$]										
Non-exposed	16	1.0	Referent	84	1.0	Referent	80	1.0	Referent	
≤ median	8	2.17	0.92-5.11	10	0.58	0.30-1.12	24	1.09	0.69-1.73	
> median	7	3.80	1.53-9.48	12	1.22	0.66-2.26	17	1.10	0.59-1.73	
P_{trend}		0.001			0.987			0.872		0.107
Dicamba (herbicide; benzoic acid)										
No exposure	27	1.0	Referent	118	1.0	Referent	148	1.0	Referent	
≤ median	13	0.68	0.35-1.32	32	0.59	0.40-0.87	38	0.63	0.44-0.90	
> median	7	0.59	0.25-1.38	32	0.74	0.50-1.10	42	0.89	0.63-1.25	
P_{trend}		0.464			0.024			0.182		0.311
Pendimethalin (herbicide; dinitroaniline) [$]										
No exposure	20	1.0	Referent	70	1.0	Referent	70	1.0	Referent	
≤ median	8	1.11	0.49-2.54	18	0.74	0.44-1.25	28	0.95	0.61-1.48	
> median	2	0.41	0.10-1.80	20	1.19	0.72-1.99	35	1.27	0.82-1.96	
P_{trend}		0.585			0.678			0.206		0.141
Carbaryl (insecticide; carbamate) [$]										
No exposure	15	1.0	Referent	42	1.0	Referent	55	1.0	Referent	
≤ median	9	0.80	0.35-1.83	47	1.08	0.71-1.63	40	0.79	0.53-1.19	
> median	8	1.73	0.72-4.17	25	1.06	0.64-1.75	28	1.11	0.69-1.76	
P_{trend}		0.538			0.509			0.856		0.612
Carbofuran (insecticide; carbamate)										
No exposure	40	1.0	Referent	127	1.0	Referent	169	1.0	Referent	
≤ median	6	0.48	0.20-1.14	27	0.80	0.53-1.21	36	0.87	0.60-1.24	
> median	2	0.30	0.07-1.23	23	1.21	0.77-1.90	26	0.89	0.58-1.36	
P_{trend}		0.049			0.927			0.586		0.213

Table S1 (continued). Lung Cancer risk among applicators by lifetime exposure-days of indicated pesticide, stratified by smoking status.

Pesticide by lifetime exposure-days	Never Smokers			Former Smokers			Current Smokers			
	Cases (*n*)	Hazard Ratio	95% confidence interval	Cases (*n*)	Hazard Ratio	95% confidence interval	Cases (*n*)	Hazard Ratio	95% confidence interval	P for Interaction
Chlordane (insecticide; chlorinated organic)[$]										
No exposure	20	1.0	Referent	69	1.0	Referent	80	1.0	Referent	
≤ median	9	1.98	0.90-4.36	18	0.99	0.59-1.67	21	1.47	0.91-2.38	
> median	1	0.36	0.05-2.70	12	1.01	0.55-1.87	12	1.27	0.69-2.34	
P_{trend}		0.889			0.750			0.348		0.343
Chlorpyrifos (insecticide; phosphorothioate)										
No exposure	41	1.0	Referent	138	1.0	Referent	160	1.0	Referent	
≤ median	8	0.57	0.27-1.22	47	1.03	0.74-1.43	51	0.92	0.67-1.26	
> median	5	0.43	0.17-1.09	28	0.81	0.53-1.22	54	1.14	0.83-1.57	
P_{trend}		0.068			0.670			0.864		0.017
DDT (insecticide; chlorinated organic)[$]										
No exposure	14	1.0	Referent	55	1.0	Referent	71	1.0	Referent	
≤ median	10	1.56	0.67-3.61	28	0.87	0.55-1.37	24	1.01	0.63-1.61	
> median	6	1.85	0.68-5.02	20	1.22	0.72-2.06	19	1.06	0.62-1.80	
P_{trend}		0.336			0.881			0.965		0.453
Malathion (insecticide; phosphorothioate)[$]										
No exposure	10	1.0	Referent	27	1.0	Referent	41	1.0	Referent	
≤ median	13	0.97	0.43-2.23	51	1.42	0.89-2.27	40	0.91	0.59-1.41	
> median	11	1.24	0.52-2.94	34	1.18	0.71-1.97	35	1.17	0.74-1.86	
P_{trend}		0.551			0.596			0.299		0.940

*Hazard ratios adjusted for smoking (pack-years among current and pack-years among former smokers), age, gender, and total days of any pesticide application
[$]Lifetime-days of use were obtained from the take home questionnaire

Table S2. Lung cancer risk among applicators by lifetime exposure-days of indicated pesticide, stratified by histology, Agricultural Health Study

Pesticide by lifetime exposure-days	Adenocarcinoma			Non-Adenocarcinoma		
	Cases (*n*)	Hazard Ratio	95% confidence interval	Cases (*n*)	Hazard Ratio	95% confidence interval
Chlorimuron-ethyl (herbicide; pyrimidinylsulfonylurea) [$]						
No exposure	52	1.0	Referent	128	1.0	Referent
≤median	13	1.03	0.55-1.95	29	0.98	0.65-1.48
>median	6	0.77	0.33-1.82	30	1.49	0.99-2.25
P_{trend}		0.415			0.047	
Dicamba (herbicide; benzoic acid)						
No exposure	74	1.0	Referent	219	1.0	Referent
≤median	22	0.60	0.35-1.01	61	0.61	0.45-0.83
>median	24	0.96	0.63-1.48	57	0.81	0.62-1.05
P_{trend}		0.331			0.010	
Pendimethalin (herbicide; dinitroaniline) [$]						
No exposure	47	1.0	Referent	113	1.0	Referent
≤median	12	0.92	0.62-1.36	42	1.15	0.58-2.27
>median	12	1.45	0.92-2.30	45	0.71	0.34-1.48
P_{trend}		0.677			0.138	
Carbaryl (insecticide; carbamate) [$]						
No exposure	28	1.0	Referent	84	1.0	Referent
≤median	28	1.09	0.64-1.86	68	0.91	0.65-1.26
>median	14	1.05	0.55-2.04	47	1.20	0.84-1.75
P_{trend}		0.712			0.459	
Carbofuran (insecticide; carbamate)						
No exposure	91	1.0	Referent	245	1.0	Referent
≤median	15	0.62	0.36-1.08	54	0.83	0.62-1.12
>median	10	0.71	0.37-1.38	41	1.00	0.72-1.41
P_{trend}		0.168			0.680	
Chlordane (insecticide; chlorinated organic) [$]						
No exposure	46	1.0	Referent	123	1.0	Referent
≤median	17	1.60	0.92-2.80	31	1.19	0.80-1.76
>median	5	0.73	0.29-1.85	20	1.16	0.72-1.87
P_{trend}		0.977			0.343	
Chlorpyrifos (insecticide; phosphorothioate)						
No exposure	86	1.0	Referent	253	1.0	Referent
≤median	25	0.92	0.57-1.47	81	0.96	0.74-1.23
>median	20	0.85	0.51-1.41	67	0.94	0.71-1.24
P_{trend}		0.530			0.666	
DDT (insecticide; chlorinated organic) [$]						
No exposure	36	1.0	Referent	104	1.0	Referent
≤median	17	1.00	0.56-1.80	45	0.99	0.79-1.42
>median	15	1.52	0.82-2.84	30	1.11	0.73-1.69
P_{trend}		0.424			0.961	
Malathion (insecticide; phosphorothioate) [$]						
No exposure	22	1.0	Referent	56	1.0	Referent
≤median	32	1.07	0.60-1.90	72	1.08	0.74-1.57
>median	18	0.86	0.46-1.62	62	1.31	0.91-1.90
P_{trend}		0.528			0.041	

*Hazard ratios adjusted for smoking (pack-years among current and pack-years among former smokers), age, gender, and total days of any pesticide application.

[$]Lifetime-days of use were obtained from the take home questionnaire

Table S3. Hazard Ratios and 95% Confidence Limits for Lung Cancer by Lifetime Days of Pesticide Exposure and Intensity-Weighted Lifetime Exposure to 30 Pesticides, Agricultural Health Study

Lifetime Days Pesticide Exposure					Intensity-Weighted Lifetime Exposure Days				
Pesticide	**Cases (n)**	**Hazard* Ratio**	**95% Confidence Interval**	**P for Trend**		**Cases (n)**	**Hazard* Ratio**	**95% Confidence Interval**	**P for trend**
Atrazine									
Non-exposed	169	1.00	Ref.		Non-exposed	169	1.00	Ref.	
Q1	76	1.10	0.84-1.44		Q1	93	1.25	0.93-1.66	
Q2	119	0.95	0.73-1.24		Q2	93	0.98	0.76-1.26	
Q3	85	1.04	0.80-1.36		Q3	87	0.89	0.68-1.17	
Q4	78	0.84	0.64-1.10	0.177	Q4	85	0.86	0.65-1.15	0.187
Cyanazine									
Non-exposed	303	1.00	Ref.		Non-exposed	303	1.00	Ref.	
Q1	11	0.95	0.67-1.35		Q1	35	1.01	0.54-1.89	
Q2	81	0.89	0.64-1.22		Q2	42	0.87	0.68-1.11	
Q3	28	0.75	0.53-1.05		Q3	40	0.71	0.48-1.05	
Q4	35	0.87	0.61-1.22	0.144	Q4	38	0.91	0.63-1.30	0.250
EPTC									
Non-exposed	382	1.00	Ref.		Non-exposed	382	1.00	Ref.	
Q1	11	1.06	0.58-1.94		Q1	13	0.66	0.38-1.15	
Q2	26	0.74	0.50-1.11		Q2	17	1.00	0.61-1.62	
Q3	15	1.01	0.61-1.70		Q3	23	1.26	0.83-1.92	
Q4	13	1.08	0.62-1.90	0.602	Q4	12	0.70	0.39-1.25	0.300
Alachlor									
Non-exposed	223	1.00	Ref.		Non-exposed	223	1.00	Ref.	
Q1	58	1.00	0.74-1.33		Q1	66	0.99	0.75-1.31	
Q2	84	0.85	0.66-1.10		Q2	70	0.84	0.64-1.10	
Q3	59	1.11	0.83-1.48		Q3	60	1.08	0.81-1.43	
Q4	42	0.78	0.55-1.10	0.285	Q4	47	0.81	0.59-1.13	0.200
Metribuzin[$]									
Non-exposed	175	1.00	Ref.		Non-exposed	175	1.00	Ref.	
Q1	10	1.05	0.55-2.00		Q1	15	0.93	0.54-1.59	
Q2	39	0.80	0.57-1.14		Q2	27	0.74	0.49-1.12	
Q3	14	1.29	0.74-2.25		Q3	19	1.45	0.90-2.34	
Q4	15	1.17	0.68-2.00	0.611	Q4	17	1.07	0.64-1.78	0.373
Paraquat[$]									
Non-exposed	198	1.00	Ref.		Non-exposed	198	1.00	Ref.	
Q1	10	1.02	0.50-2.08		Q1	13	1.03	0.58-1.80	
Q2	22	1.23	0.78-1.91		Q2	15	1.33	0.75-2.35	
Q3	7	0.98	0.45-2.12		Q3	13	1.00	0.57-1.75	
Q4	16	1.24	0.73-2.10	0.678	Q4	14	1.35	0.77-2.36	0.537

Lifetime Days Pesticide Exposure					Intensity-Weighted Lifetime Exposure Days				
Pesticide	**Cases (n)**	**Hazard* Ratio**	**95% Confidence Interval**	**P for Trend**		**Cases (n)**	**Hazard* Ratio**	**95% Confidence Interval**	**P for trend**
Petroleum Oil[$]									
Non-exposed	197	1.00	Ref.		Non-exposed	197	1.00	Ref.	
Q1	4	0.79	0.29-2.14		Q1	11	0.93	0.51-1.71	
Q2	24	1.05	0.68-1.60		Q2	17	1.11	0.67-1.82	
Q3	11	1.32	0.72-2.42		Q3	12	1.20	0.67-2.15	
Q4	15	1.33	0.78-2.27	0.505	Q4	14	1.35	0.78-2.34	0.740
Imazethapyr									
Non-exposed	318	1.00	Ref.		Non-exposed	318	1.00	Ref.	
Q1	22	1.28	0.82-2.01		Q1	25	0.82	0.54-1.26	
Q2	37	0.73	0.52-1.03		Q2	35	0.83	0.58-1.19	
Q3	38	0.81	0.57-1.15		Q3	35	0.90	0.63-1.28	
Q4	38	0.88	0.62-1.24	0.616	Q4	39	0.85	0.60-1.19	0.268
Glyphosate									
Non-exposed	92	1.00	Ref.		Non-exposed	92	1.00	Ref.	
Q1	26	1.09	0.67-1.76		Q1	104	1.02	0.75-1.38	
Q2	159	1.12	0.85-1.45		Q2	120	1.21	0.91-1.61	
Q3	153	1.18	0.89-1.55		Q3	111	1.26	0.95-1.68	
Q4	101	1.15	0.85-1.55	0.693	Q4	104	1.10	0.82-1.48	0.686
Butylate[$]									
Non-exposed	192	1.00	Ref.		Non-exposed	192	1.00	Ref.	
Q1	13	0.59	0.33-1.03		Q1	17	0.77	0.47-1.27	
Q2	22	1.41	0.90-2.20		Q2	13	1.16	0.66-2.05	
Q3	11	1.72	0.94-3.17		Q3	12	1.52	0.84-2.72	
Q4	12	0.98	0.54-1.78	0.842	Q4	16	1.05	0.62-1.78	0.551
Trifluralin									
Non-exposed	247	1.00	Ref.		Non-exposed	247	1.00	Ref.	
Q1	45	0.98	0.71-1.35		Q1	46	0.86	0.62-1.19	
Q2	78	0.84	0.65-1.09		Q2	64	0.82	0.62-1.10	
Q3	40	0.80	0.56-1.15		Q3	50	0.93	0.68-1.26	
Q4	49	0.72	0.53-1.00	0.136	Q4	52	0.73	0.54-1.00	0.113
2,4-D									
Non-exposed	153	1.00	Ref.		Non-exposed	153	1.00	Ref.	
Q1	112	0.76	0.58-0.98		Q1	97	0.77	0.59-1.02	
Q2	97	0.75	0.58-0.97		Q2	99	0.70	0.54-0.91	
Q3	86	0.79	0.60-1.03		Q3	87	0.84	0.64-1.09	
Q4	84	0.74	0.56-0.97	0.41	Q4	95	0.75	0.57-0.98	0.195

Lifetime Days Pesticide Exposure					Intensity-Weighted Lifetime Exposure Days				
Pesticide	**Cases (n)**	**Hazard* Ratio**	**95% Confidence Interval**	**P for Trend**		**Cases (n)**	**Hazard* Ratio**	**95% Confidence Interval**	**P for trend**
2,4,5-T[$]									
Non-exposed	187	1.00	Ref.		Non-exposed	187	1.00	Ref.	
Q1	10	0.84	0.43-1.64		Q1	14	0.75	0.43-1.32	
Q2	21	0.71	0.45-1.12		Q2	17	0.89	0.54-1.47	
Q3	12	1.02	0.57-1.83		Q3	13	0.68	0.39-1.20	
Q4	16	1.17	0.70-1.96	0.773	Q4	15	1.47	0.86-2.51	0.939
Permethrin (crops)									
Non-exposed	394	1.00	Ref.		Non-exposed	394	1.00	Ref.	
Q1	11	1.18	0.64-2.16		Q1	13	1.18	0.68-2.05	
Q2	14	0.87	0.50-1.51		Q2	16	0.82	0.48-1.40	
Q3	18	0.90	0.54-1.50		Q3	14	1.03	0.60-1.78	
Q4	13	1.27	0.72-2.25	0.749	Q4	13	1.10	0.62-1.96	0.377
Permethrin (animals)									
Non-exposed	428	1.00	Ref.		Non-exposed	428	1.00	Ref.	
Q1	3	1.37	0.43-4.27		Q1	7	0.62	0.25-1.51	
Q2	6	0.59	0.23-1.50		Q2	9	1.02	0.48-2.14	
Q3	15	0.98	0.57-1.70		Q3	9	0.92	0.45-1.86	
Q4	6	0.65	0.27-1.59	0.718	Q4	5	0.72	0.29-1.73	0.492
Terbufos									
Non-exposed	316	1.00	Ref.		Non-exposed	316	1.00	Ref.	
Q1	64	0.93	0.71-1.21		Q1	40	0.89	0.64-1.24	
Q2	30	0.86	0.59-1.26		Q2	34	1.02	0.72-1.45	
Q3	23	0.88	0.57-1.36		Q3	34	0.88	0.62-1.26	
Q4	19	0.84	0.53-1.36	0.34	Q4	28	0.79	0.53-1.17	0.701
Fonofos									
Non-exposed	373	1.00	Ref.		Non-exposed	373	1.00	Ref.	
Q1	29	1.24	0.85-1.81		Q1	19	1.22	0.77-1.96	
Q2	17	0.83	0.51-1.36		Q2	23	0.88	0.58-1.34	
Q3	21	0.88	0.57-1.36		Q3	24	0.93	0.62-1.41	
Q4	21	1.20	0.76-1.90	0.858	Q4	22	1.23	0.80-1.90	0.669
Lindane[$]									
Non-exposed	213	1.00	Ref.		Non-exposed	213	1.00	Ref.	
Q1	11	0.69	0.37-1.31		Q1	8	0.60	0.29-1.28	
Q2	11	1.19	0.63-2.24		Q2	12	1.09	0.59-2.00	
Q3	10	1.20	0.61-2.35		Q3	7	0.94	0.44-2.01	
Q4	5	1.05	0.43-2.58	0.676	Q4	10	1.60	0.82-3.14	0.754

Lifetime Days Pesticide Exposure					Intensity-Weighted Lifetime Exposure Days				
Pesticide	**Cases (n)**	**Hazard* Ratio**	**95% Confidence Interval**	**P for Trend**		**Cases (n)**	**Hazard* Ratio**	**95% Confidence Interval**	**P for trend**
Aldicarb[$]									
Non-exposed	223	1.00	Ref.		Non-exposed	223	1.00	Ref.	
Q1	5	2.13	0.85-5.31		Q1	8	1.36	0.64-2.88	
Q2	14	0.99	0.51-1.91		Q2	5	1.18	0.48-2.92	
Q3	4	0.80	0.24-2.62		Q3	5	0.91	0.36-2.34	
Q4	5	0.81	0.26-2.51	0.379	Q4	3	0.72	0.21-2.50	0.504
Phorate[$]									
Non-exposed	182	1.00	Ref.		Non-exposed	182	1.00	Ref.	
Q1	7	0.74	0.35-1.57		Q1	13	0.69	0.39-1.21	
Q2	26	0.70	0.47-1.06		Q2	20	0.71	0.44-1.12	
Q3	16	1.05	0.63-1.75		Q3	15	1.14	0.67-1.93	
Q4	14	1.43	0.82-2.47	0.837	Q4	15	1.35	0.79-2.29	0.758
Aldrin[$]									
Non-exposed	192	1.00	Ref.		Non-exposed	192	1.00		
Tertile 1	7	1.02	0.48-2.18		Tertile 1	17	0.79	0.48-1.30	
Tertile 2	30	0.68	0.50-1.00		Tertile 2	17	0.66	0.40-1.08	
Tertile 3	15	0.81	0.47-1.40	0.535	Tertile 3	18	0.81	0.50-1.31	0.173
Toxaphene[$]									
Non-exposed	196	1.00	Ref.		Non-exposed	196	1.00	Ref.	
Tertile 1	10	2.15	1.14-4.08		Tertile 1	16	1.04	0.62-1.73	
Tertile 2	29	1.11	0.75-1.64		Tertile 2	17	1.56	0.94-2.57	
Tertile 3	12	1.19	0.65-2.17	0.712	Tertile 3	18	1.26	0.77-2.06	0.734
Coumaphos									
Non-exposed	405	1.00	Ref.		Non-exposed	405	1.00	Ref.	
Tertile 1	8	2.71	1.34-5.46		Tertile 1	12	1.05	0.59-1.87	
Tertile 2	22	1.05	0.69-1.62		Tertile 2	15	1.35	0.80-2.25	
Tertile 3	11	1.00	0.54-1.82	0.587	Tertile 3	14	1.14	0.67-1.95	0.552
DDVP									
Non-exposed	423	1.00	Ref.		Non-exposed	423	1.00	Ref.	
Tertile 1	7	2.18	1.03-4.59		Tertile 1	10	1.39	0.72-2.66	
Tertile 2	15	0.63	0.37-1.05		Tertile 2	11	0.56	0.31-1.03	
Tertile 3	10	0.89	0.47-1.67	0.855	Tertile 3	11	0.99	0.54-1.80	0.5987
Captan									
Non-exposed	412	1.00	Ref.		Non-exposed	412	1.00	Ref.	
Tertile 1	13	0.70	0.41-1.21		Tertile 1	12	0.72	0.41-1.27	
Tertile 2	15	1.07	0.58-1.96		Tertile 2	154	1.03	0.59-1.81	
Tertile 3	12	0.76	0.41-1.38	0.395	Tertile 3	154	0.79	0.44-1.42	0.301

Lifetime Days Pesticide Exposure					Intensity-Weighted Lifetime Exposure Days				
Pesticide	**Cases (n)**	**Hazard* Ratio**	**95% Confidence Interval**	**P for Trend**		**Cases (n)**	**Hazard* Ratio**	**95% Confidence Interval**	**P for trend**
Benomyl[$]									
Non-exposed	221	1.00	Ref.		Non-exposed	221	1.00	Ref.	
Q1	4	1.10	0.41-2.95		Q1	9	1.16	0.71-1.90	
Q2	10	1.00	0.53-1.90		Q2	6	1.37	0.77-2.44	
Q3	7	0.86	0.40-1.82		Q3	5	1.43	0.87-2.34	
Q4	4	1.25	0.46-3.41	0.003	Q4	5	0.76	0.41-1.39	0.659
Chlorthalonil									
Non-exposed	465	1.00	Ref.		Non-exposed	465	1.00	Ref.	
Q1	9	0.81	0.36-1.78		Q1	17	0.73	0.36-1.46	
Q2	24	1.53	1.00-2.32		Q2	15	0.80	0.45-1.44	
Q3	17	1.13	0.69-1.86		Q3	16	1.08	0.50-2.35	
Q4	10	0.97	0.49-1.92	0.873	Q4	12	0.83	0.42-1.62	0.932
Metalaxyl[$]									
Non-exposed	184	1.00	Ref.		Non-exposed	184	1.00	Ref.	
Q1	19	1.34	0.81-2.20		Q1	14	1.13	0.64-1.97	
Q2	16	1.02	0.61-1.71		Q2	16	1.20	0.70-2.05	
Q3	12	1.08	0.60-1.95		Q3	17	1.13	0.68-1.88	0.758
Q4	18	1.33	0.82-2.19	0.729	Q4	18	1.30	0.79-2.12	
Methyl Bromide									
Non-exposed	403	1.00	Ref.		Non-exposed	403	1.00	Ref.	
Q1	20	1.14	0.73-1.79		Q1	31	1.04	0.72-1.50	
Q2	44	1.01	0.74-1.37		Q2	27	0.89	0.60-1.31	
Q3	30	1.04	0.71-1.50		Q3	37	1.23	0.88-1.73	
Q4	28	1.01	0.68-1.49	0.432	Q4	26	1.01	0.67-1.52	0.706
Ethylene Dibromide[$]									
Non-exposed	228	1.00	Ref.		Non-exposed	228	1.00	Ref.	
Q1	3	0.67	0.21-2.07		Q1	5	0.95	0.39-2.30	
Q2	5	1.80	0.74-4.37		Q2	5	2.72	1.12-6.64	
Q3	4	18.8	6.85-51-58		Q3	6	2.03	0.90-4.58	
Q4	7	1.14	0.54-2.42	0.607	Q4	3	0.84	0.27-2.62	0.277

*Hazard ratios adjusted for age, smoking (pack-years among current and pack-years among former smokers) and gender, total lifetime pesticide use.

[$]Lifetime-days and Intensity-weighted lifetime Days of use were obtained from the take home questionnaire

AMERICAN JOURNAL OF INDUSTRIAL MEDICINE 39:477–486 (2001)

An Application of Hierarchical Regression in the Investigation of Multiple Paternal Occupational Exposures and Neuroblastoma in Offspring

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Background *We used hierarchical regression to study the effects of 46 paternal occupational exposures on the incidence of neuroblastoma in offspring.*
Methods *The study population included 405 cases and 302 controls. The effect of each exposure was estimated using both conventional maximum likelihood and hierarchical regression.*
Results *Using hierarchical regression, overall precision was greatly enhanced compared to the conventional analysis. In addition, adjustment of effect estimates based on prespecified prior distributions of the true effect parameters allowed a more consistent interpretation across the entire panel of exposures. Estimates for several metals and solvents were shrunk close to the null value, whereas estimates for several thinner solvents, diesel fuel, solders, wood dust, and grain dust remained moderately elevated.*
Conclusions *Hierarchical regression may mitigate some of the problems of the conventional approach by controlling for correlated exposures, enhancing the precision of estimates, and providing some adjustment of estimates based on prior knowledge.* Am. J. Ind. Med. 39:477–486, 2001.

KEY WORDS: hierarchical regression; occupation; neuroblastoma; childhood cancer; multiple comparisons

INTRODUCTION

When studying associations between rare diseases and occupational exposures, the population-based case-control study is the most practical design. In conducting these studies, information is usually collected by questionnaire on numerous occupational exposures, in order to most efficiently 'screen' a long list of chemical and physical agents. Effect estimates are traditionally calculated by including each exposure in a separate logistic regression model, along with any potential confounders. There are several problems with this approach. First, people often experience combined exposures to different agents in the

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Contract grant sponsor: National Institutes of Health; Contract grant number: CA57004 and T32-ES-07018.
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Accepted 21 January 2001

workplace, and the conventional analysis in which each exposure is included in a separate model does not account for probable correlation between occupational exposures. Second, the precision of estimates tends to vary considerably among the different exposures, depending on the number of persons exposed to each agent. Third, the occurrence of false associations by chance is of concern as in any epidemiologic study. These problems are particularly troubling in the situation in which multiple exposures are evaluated based on little prior knowledge. Presumably, results from such a preliminary study would be followed by research focusing on a few suspect agents, with epidemiologic studies using more sophisticated exposure assessment methods, or laboratory studies to determine effects of suspect agents on in vitro or animal models for the disease in question. However, when multiple effect estimates are elevated, the imprecision of some estimates in addition to potential confounding or generation of false positive associations, makes interpretation of the entire panel of results difficult. It would be helpful to minimize the total error in such case-control analyses to clarify a focus for further research.

Hierarchical regression, also known as multilevel or random-coefficient modeling, is a statistical method that can greatly improve the accuracy of unstable estimates, especially when studying effects of multiple exposures with limited data [Greenland, 1994, 2000, 2001]. In this type of analysis, disease outcomes are regressed on multiple exposures in a first-stage model. The beta coefficients from the first stage are then modeled as values of the outcome variable in a second-stage linear regression model, as a function of second-stage or 'prior' covariates that are thought to determine the magnitude of the true effects, or target parameters [Greenland, 1994, 2001]. Effect estimates and confidence limits are adjusted by an empirical-Bayes (EB) or semi-Bayes procedure that 'shrinks' unstable estimates toward estimated prior means of the target parameters. The shrinkage adjustments are made using the variance of an assumed prior distribution of the target parameter for each exposure. This variance is estimated in empirical-Bayes methods using an iterative procedure, or the variance can be prespecified in semi-Bayes methods by specifying a particular range in which a given proportion of the true parameter values are expected to lie.

Software for hierarchical regression modeling has not been widely available. However, a procedure written in SAS/IML for conducting multi-stage modeling of multiple exposures has recently been posted on the worldwide web (http://darwin.cwru.edu/~witte/episoft.html) [Witte et al., 1998], and SAS Proc GLIMMIX can also be adapted to this purpose [Witte et al., 2001]. We used these methods in our recent study of the effects of paternal occupational exposures on the incidence of neuroblastoma, a childhood cancer, in offspring. In this study, exposures to 46 specific chemical and physical agents were examined. We used the SAS/IML procedure to conduct hierarchical regression using semi-Bayes and empirical-Bayes methods to generate adjusted odds ratios and confidence limits for the effects of paternal occupational exposures on the incidence of neuroblastoma. Results from hierarchical regression models generated by specifying different prior distributions were compared to each other, and to results from a conventional analysis.

MATERIALS AND METHODS

Study population. The study population for this case-control study of neuroblastoma is described in detail elsewhere [Olshan et al., 1999]. In brief, cases were patients under the age of 19 years with a confirmed new diagnosis of neuroblastoma between 1 May 1992 and 30 April 1994, registered at any of 139 participating hospitals in the United States and English-speaking Canada. The hospitals were members of either of two pediatric collaborative clinical trials groups, the Children's Cancer Group or Pediatric Oncology Group. Of the families contacted, we enrolled 538 cases (73% of those eligible). One matched control for each of 504 cases was selected by random-digit telephone dialing (RDD). The response proportion for the RDD screening was 74%. Controls were individually caliper-matched to cases on date of birth (± 6 months for cases ≤ 3 years of age, ± 1 year for cases > 3 years of age).

Data collection. A telephone interview was conducted with each mother and with the father when available. The interview included questions on demographic characteristics such as parental age, race, and education. Occupational history was obtained, including information on dates of employment, names of employers, occupations, industries, job titles, specific duties, and hours per week. For each job held during the 2-year period prior to the child's date of birth, fathers were asked if they had been exposed to electrical equipment or radiation sources, chemicals, dusts, fumes, gases, vapors, or oils. Occupational exposure information was available for a total of 707 fathers (405 case fathers, 302 control fathers).

First-stage exposures. In this study, specific chemical and physical agents were the first-stage exposures of primary interest. A review of self-reported occupational exposures was conducted by an industrial hygienist (IH) (K. Teschke), to increase the specificity of exposure variables by reducing the number of false positives in the group classified as exposed to each agent. The review was blinded to case or control parent status. The IH review covered all reported information for each exposure including occupation, industry, hours of exposure per week, form of the substance, route of exposure, use of protective equipment or clothing, work activities, and average distance from electrical equipment. A father was coded as exposed to a chemical

substance or compound if the IH review determined 'probable' exposure to that agent in any job. A father was coded as exposed to electromagnetic fields or radiation (ionizing radiation, radiofrequency fields, or extremely low frequency fields) if the IH review determined 'probable' exposure, in any job, to equipment that produces high levels of one of these frequencies. Each occupational exposure was coded as an indicator variable (1 = exposed, 0 = unexposed). In total, 46 paternal occupational exposures were coded and analyzed.

First-stage covariates. Demographic characteristics thought to be potential confounders of associations between occupational exposures and neuroblastoma were maternal education, maternal race, and maternal age at birth of the index child. The variables were coded using indicator variables for the following categories: maternal education (less than high school graduate, high school graduate and/or some college, college degree or more as the referent), maternal race (white as the referent, black, Hispanic, other), and maternal age at birth of the index child (<18, 18–39 years as the referent, ≥ 40 years).

In order to retain information on all 405 case fathers and 302 control fathers in the analysis of occupational exposures, we decided to conduct unmatched analyses by unconditional logistic regression with adjustment for the matching factor using covariates. The matching factor, child's age, was coded as a set of indicator variables with strata as fine as numbers would allow (6-month intervals for ages ≤ 3 years; 2-year intervals for ages 3–11 years, one variable for ages >11 years).

Second-stage covariates. The second-stage matrix contained variables thought to determine the magnitude of, or explain some of the variability between, the individual target parameters. These second-stage, or prior, covariates were indicator variables representing subsets of target parameters within which the parameters were regarded as 'exchangeable', or as draws from a common prior distribution [Greenland, 1994, 2000]. We defined exchangeable categories by grouping the first-stage occupational exposures according to similarities in physicochemical properties, as halogenated hydrocarbons (HCs), nonvolatile HCs, volatile HCs, metals, paints, thinner solvents, wood-derived substances, grain-derived dusts, and non-ionizing electromagnetic field exposures (see Table I). Some exposures were included in more than one category (e.g., oil-based paints, turpentine). Other individual exposures did not have physicochemical properties that we would expect to translate into exchangeable biologic effects for neuroblastoma; these exposures were therefore not grouped together (e.g., as in groups of pesticides or dusts). There is little prior information on the potential for any of the agents to act as transgenerational carcinogens through a paternally mediated mechanism; thus, no second-stage covariates indicating toxicity were used.

TABLE I. Coding of Second-Stage Covariates: Categories of Exchangeability Between True Effect Parameters for Occupational Exposures[a,b]

Halogenated hydrocarbons	Metals
Carbon tetrachloride	Brass
Chloroform	Bronze
Freon	Galvanized steel
Methylene chloride	High-speed steel
Perchloroethylene	Mild steel
Trichloroethylene	Stainless steel
	Alloys (NOS)
Non-volatile hydrocarbons	Metals (NOS)
Cutting oil	Solders (NOS)
Diesel fuel	
Kerosene	Thinner solvents
Lubricating oil	Lacquer thinner
	Mineral spirits
Volatile hydrocarbons	Oil-based paints
Acetone	Paint thinner
Alcohols	Turpentine
Benzene	
Gasoline	Wood-derived substances
Glycols or glycol ethers	Turpentine
Lacquer thinner	Wood dust
Methyl ethyl ketone	
Mineral spirits	Grain-derived dusts
Naphtha	Flour dust
Paint thinner	Grain dust
Toluene	
Turpentine	Non-ionizing EMF
White gas	Extremely low frequency fields
Xylene	Radiofrequency fields
Paints	
Oil-based paints	
Water-based paints	

[a]Other chemicals analyzed but not included in any grouping: plastics, synthetics, or resins (NOS); cardboard dust; rubber dust; herbicides (NOS), insecticides (NOS), ionizing radiation.
[b]NOS, not otherwise specified; EMF, electric and magnetic fields.

The second-stage, or Z-matrix, was structured with one row for each of the occupational exposures, j. Each row was composed of 10 elements; column one contained a value of '1' denoting the presence of an intercept, and the following nine columns contained values for the each of the prior covariates, z_{ij}; a '1' if the exposure was present in that category and a '0' if not. An exposure that appeared in more than one category thus had multiple '1's in its row of the Z-matrix.

Statistical analyses. In our conventional analysis, each occupational exposure was evaluated in a separate unconditional logistic regression model, along with indicator variables representing child's age (the matching factor),

and the demographic covariates. Exposure odds ratios estimating incidence rate ratios were estimated using maximum likelihood.

In the first-stage model of the hierarchical regression analyses, neuroblastoma disease status was regressed simultaneously on the 46 paternal occupational exposures, child's age, and the demographic covariates. This model took the form: $\Pr(y = 1 | x, w) = \text{expit}(\alpha + X\beta + W_\gamma)$, where X represents an n-row matrix of occupational exposures and W represents an n-row matrix of potential confounders, where n represents the number of subjects in the study, and expit (●) is the logistic function exp (●)/(1 + exp (●)) [Greenland, 1998].

The estimated beta coefficients for the 46 occupational exposures in the first-stage model were then regressed in a second-stage linear regression model as a function of the prior covariates. The second-stage model should incorporate what is known about each target parameter, β_j, prior to seeing the study data [Greenland, 1994, 2001]. Therefore, a prior 'distribution' was defined for the true effect parameter for each occupational exposure, with a prior mean dependent on the joint distribution of second-stage covariates, and a prespecified prior variance for each parameter. For each occupational exposure, j, the second-stage model took the form: $\beta_j = \pi_1 z_{1j} + \pi_2 z_{2j} + \pi_3 z_{3j} + \pi_4 z_{4j} + \pi_5 z_{5j} + \pi_6 z_{6j} + \pi_7 z_{7j} + \pi_8 z_{8j} + \pi_9 z_{9j} + \delta_j = z_j\pi + \delta_j$, where π is the column vector containing the second-stage parameters, π_1 through π_9, and δ_j the deviation of the effect of occupational exposure, j, from the sum $z_j\pi$. Based on the prior distributions, the second-stage model assumed that each target parameter deviates randomly around the linear term on the right hand side of the equation [Greenland, 1992, 1993, 1994, 1998]. In other words, target parameters for occupational exposures with the same values for the second-stage covariates were assumed to have been randomly sampled from a common underlying distribution, with an unknown mean. In addition to hierarchical models using the prior covariates to determine prior means of the target parameters, we ran one hierarchical model with an intercept-only second-stage matrix, containing no prior covariates. Because our prior covariates were crudely specified categories of exchangeability, we wanted to compare a hierarchical model using no prior covariates to one using our crudely specified prior covariates, to assess the benefit of such a procedure in the face of little or no prior information. In this intercept-only model, all the target parameters were assumed to have been sampled from a common distribution with an unknown mean.

The deviation δ_j is called the residual effect of occupational exposure j; it represents effects of exposure j that are not captured by the sum $z_j\pi$ [Greenland, 1994, 1998, 2001; Witte et al., 1994], or effects above and beyond those accounted for by the 'group' effects of the second-stage covariates. Residual effects can arise from information not included in the Z-matrix, such as other information that could potentially explain variability between the target parameters; for example, detailed measures of genotoxicity and carcinogenicity. These residual effects δ_j are usually assumed to be independent random quantities having means of zero and variance τ^2, where τ^2 may be fixed in advance using background information, as in semi-Bayes analyses, or may be estimated from the data, as in empirical-Bayes analyses [Greenland, 1992, 1993, 1994, 1998, 2000, 2001; Greenland and Poole, 1994; Witte et al., 1994, 1998, 2001]. A large value for τ^2 would imply that there are likely to be substantial effects of an exposure beyond those explained by the second-stage covariates, whereas a small value for τ^2 would translate into a relatively tight range for the residual effects, and would imply a greater confidence that the effects of the exposure act through mechanisms that are almost completely mediated through the second-stage covariates. In our semi-Bayes analyses, we used different prespecified values for τ^2 in different hierarchical models to observe the sensitivity of our results to the choice. Although the hierarchical model can be generalized by allowing τ^2 to vary for different first-stage exposures [Greenland, 1994], we did not feel that we had better prior information for any specific exposure compared to the others; therefore, in each analysis, the same value for τ^2 was assigned to all exposures. Because our Z-matrix was rather crudely defined, we started with a liberal prespecified range for the residual effects of the occupational exposures. We assumed, with 95% certainty, that the rate ratio for each occupational exposure, after adjusting for the second-stage covariates, would fall within a 10-fold range (e.g., between 0.5 and 5.0), or that the δ_j is, with 95% certainty, an interval 2.3 units wide on the log rate ratio scale (e.g., ln (0.5) = −0.7 and ln (5.0) = 1.6). Thus, assuming that the δ_j are normally distributed with standard deviation of τ, $(1.96)(2)\tau = 2.3$, or $\tau = 0.59$, and the prior residual variance, τ^2, is 0.35. We also conducted two other hierarchical regression analyses using prespecified five-fold and 2.5-fold 95% ranges to estimate the prior variance, to see if the results were sensitive to the choice [Greenland, 1992, 1993, 1994, 2001; Greenland and Poole, 1994; Witte et al., 1994]. In addition, we performed a hierarchical regression analysis using empirical-Bayes estimation of the residual variance.

The first- and second-stage models together constitute a two-stage hierarchical regression model [Greenland, 1994, 1998, 2000, 2001]. The prior mean for each occupational exposure was estimated by substituting the estimated π_1 through π_9 into the equation, $\hat{\beta} = Z_j\hat{\pi}$. An average of the estimated prior mean vector and the vector of maximum likelihood estimates from the first-stage model, weighted by the covariance matrix of the first-stage estimate and τ^2, respectively, gives the estimated posterior coefficient for each exposure [Greenland, 1992, 1993, 1994; Witte and Greenland, 1996].

RESULTS

The effects of paternal occupational exposures on the incidence of neuroblastoma in offspring, estimated from the various analyses, are shown in Table II. The results from our conventional analysis using maximum likelihood estimation for each agent separately are presented as Analysis 1. Effect estimates for some exposures are relatively precise (e.g., wood dust, 95% CL ratio = 3.5 [CL ratio = upper confidence limit divided by lower confidence limit]), while others are very imprecise (e.g., turpentine, CL ratio = 18.7). The precision of these estimates is primarily dependent on the number and distribution of exposed cases and controls. While some imprecise estimates are sufficiently elevated to pique interest for further study (e.g., turpentine, OR = 10.4; CL ratio = 18.7), other imprecise estimates are only moderately elevated (e.g., high-speed steel, OR = 2.0; CL ratio = 15.4), making prioritization of future research difficult. Although each estimate can be interpreted in a straightforward manner as the observed association between the exposure and the disease outcome, some of the associations may result from confounding by combined exposures to different agents in the workplace. As in any statistical analysis, false positive associations may also have occurred by chance. Analysis 1 can be conceived of as a special case of hierarchical modeling, in which the prior residual variance is set at infinity (i.e., odds ratios of any magnitude are a priori equally likely to occur), and the true effect parameter for each occupational exposure has its own independent prior mean [Greenland, 1992]. Therefore, a second-stage model defined in this way would do nothing to adjust the estimates or confidence limits from the first stage.

The results from the first-stage model of the hierarchical regression, estimated by simultaneously modeling all occupational exposures, child's age, and the demographic covariates using maximum likelihood estimation, are presented as Analysis 2. Many of these effect estimates are wildly imprecise (e.g., turpentine, CL ratio = 54). Although the advantage of such an analysis is to control for the correlation between exposures that occur together in the workplace, the imprecision resulting from the inclusion of so many variables in one model makes the estimates virtually meaningless. This type of model is really only useful as a first-stage model for the hierarchical regression.

Analysis 3 shows the results from hierarchical regression modeling using an intercept-only second-stage matrix, with a prespecified 10-fold range for the true residual effect parameters given 95% certainty. There are no prior covariates in the second-stage equation for this analysis. The results from Analysis 3 show greatly increased precision of estimates that were previously unstable (e.g., turpentine, CL ratio = 6.5), and the effect estimates for the previously unstable estimates have been shrunk closer toward the mean of all the estimates. Analysis 3 provides somewhat of a remedy for each of the three problems of the conventional analysis (Analysis 1). Inclusion of all occupational exposures in a single model controls for potential confounding by combined occupational exposures. The enhanced precision of posterior estimates in spite of sparse data gives us greater confidence in interpreting the observed results. In addition, the shrinkage of estimates toward the mean of all estimates, within an a priori probable range for the parameters, theoretically provides some adjustment for results that may occur by chance. Any outlying association will be penalized in such a way that its posterior estimate will be closer to those of the other occupational exposures. Therefore, if the average effect of all occupational exposures is the null value, any non-null association will be somewhat attenuated. The intercept-only second-stage model implies dependency among the entire group of occupational exposures; undoubtedly, the adjustment procedure could be much improved if it was based on the distribution of prior covariates that are thought to explain some of the variability between the effects of different occupational exposures.

By adding a Z-matrix containing prior covariates to the second-stage model of the hierarchical regression analysis, we force the estimates for first-stage exposures with the same values of second-stage covariates to be more similar to each other than to those with other values. However, the results for Analysis 4 show that in this situation, with little prior information, the grouping of exposures into categories of exchangeability based on physicochemical properties adds little information to the overall analysis. With a few exceptions, the magnitude and precision of estimates from Analysis 4 are quite similar to those from Analysis 3, in which no prior covariates were added to the second stage. Exceptions generally occur where an exposure was included in more than one category of exchangeability. For example, turpentine was included in the categories of volatile hydrocarbons, thinner solvents, and wood-derived substances. The effect estimate is more elevated and more imprecise than that in Analysis 3 (OR = 4.8; CL ratio = 13.7), with the increased imprecision resulting from its complex joint distribution among second-stage covariates. Presumably, however, this estimate with the incorporated information from the second-stage matrix may be more accurate than the estimate from Analysis 3, albeit more imprecise. Effect estimates were not always elevated when included in a category of exchangeability with others that had elevated odds ratios; for example, the estimate for oil paint is near the null value, despite its inclusion in the thinner solvent category along with turpentine, lacquer thinner, mineral spirits, all of which had elevated odds ratios. The use of prior covariates in the Z-matrix theoretically improves the adjustment for associations distributed by random error, because the effect of each exposure is expected to be more similar to other exposures with the same values for the prior

TABLE II. Estimated Effects of Paternal Occupational Exposures (Odds Ratios and 95% Confidence Intervals) on the Incidence of Neuroblastoma in Offspring[a,b] Listed by Categories of Exchangeability[c]

Exposure	Number exposed		Analysis 1—ML[d], one stage, one model for each exposure	Analysis 2—ML, one stage, one model including all exposures	Analysis 3—HM[d], semi-Bayes, 10-fold range to estimate prior residual variance[e], no prior covariates	Analysis 4—HM, semi-Bayes, 10-fold range to estimate prior residual variance, with prior covariates	Analysis 5—HM, semi-Bayes, 5-fold range to estimate prior residual variance, with prior covariates	Analysis 6—HM, semi-Bayes, 2.5-fold range to estimate prior residual variance, with prior covariates
	case	control						
Halogenated hydrocarbons								
Carbon tetrachloride	4	4	0.6 (0.2, 2.6)	1.1 (0.1, 10.2)	0.9 (0.3, 2.5)	0.7 (0.3, 2.1)	0.7 (0.3, 1.7)	0.7 (0.4, 1.3)
Chloroform	3	2	1.2 (0.2, 7.5)	1.3 (0.1, 20.4)	1.0 (0.4, 2.9)	0.8 (0.2, 2.4)	0.7 (0.3, 1.8)	0.7 (0.4, 1.4)
Freon	9	13	0.5 (0.2, 1.1)	0.5 (0.1, 1.7)	0.6 (0.3, 1.4)	0.6 (0.2, 1.4)	0.6 (0.3, 1.3)	0.7 (0.4, 1.2)
Methylene chloride	4	4	0.7 (0.2, 2.8)	1.0 (0.2, 7.2)	0.9 (0.4, 2.4)	0.7 (0.3, 2.1)	0.7 (0.3, 1.7)	0.7 (0.4, 1.4)
Perchloroethylene	4	6	0.5 (0.1, 1.7)	0.3 (0.1, 2.7)	0.8 (0.3, 2.1)	0.6 (0.2, 1.7)	0.7 (0.3, 1.5)	0.7 (0.4, 1.3)
Trichloroethylene	9	7	0.9 (0.3, 2.5)	0.7 (0.1, 3.0)	0.9 (0.4, 2.1)	0.8 (0.3, 1.9)	0.8 (0.4, 1.6)	0.7 (0.4, 1.3)
Non-volatile hydrocarbons								
Cutting oil	16	7	1.7 (0.7, 4.2)	2.0 (0.5, 7.5)	1.2 (0.5, 2.8)	1.2 (0.5, 3.0)	1.1 (0.5, 2.4)	1.0 (0.6, 1.8)
Diesel fuel	42	21	1.5 (0.8, 2.6)	2.2 (0.9, 5.0)	1.5 (0.8, 2.9)	1.5 (0.8, 3.0)	1.3 (0.7, 2.4)	1.1 (0.7, 1.8)
Kerosene	16	11	1.0 (0.5, 2.2)	0.4 (0.1, 1.4)	0.8 (0.4, 1.7)	0.7 (0.3, 1.7)	0.8 (0.4, 1.7)	0.9 (0.5, 1.6)
Lubricating oil or grease	56	36	1.1 (0.7, 1.8)	0.9 (0.5, 1.9)	1.0 (0.5, 1.7)	1.0 (0.5, 1.7)	1.0 (0.6, 1.7)	1.0 (0.6, 1.5)
Volatile hydrocarbons								
Acetone	23	19	0.9 (0.5, 1.7)	0.4 (0.1, 0.9)	0.6 (0.3, 1.3)	0.6 (0.3, 1.1)	0.6 (0.4, 1.2)	0.8 (0.5, 1.2)
Alcohols	35	16	1.8 (0.9, 3.3)	1.4 (0.6, 3.4)	1.3 (0.7, 2.6)	1.3 (0.7, 2.6)	1.2 (0.7, 2.1)	1.0 (0.7, 1.6)
Benzene	5	2	2.0 (0.4, 10.3)	1.4 (0.1, 15.4)	1.1 (0.4, 3.1)	1.0 (0.4, 3.0)	1.0 (0.4, 2.2)	0.9 (0.5, 1.5)
Gasoline	45	38	0.8 (0.5, 1.3)	0.4 (0.2, 0.9)	0.6 (0.3, 1.0)	0.6 (0.3, 1.0)	0.6 (0.4, 1.1)	0.7 (0.5, 1.1)
Glycols or glycol ethers	7	4	1.3 (0.4, 4.6)	1.4 (0.2, 8.3)	1.1 (0.4, 2.9)	1.1 (0.4, 2.9)	1.0 (0.4, 2.1)	0.9 (0.5, 1.5)
Lacquer thinner	36	8	3.5 (1.6, 7.8)	3.5 (1.1, 11.6)	1.9 (0.9, 4.0)	2.1 (0.9, 4.7)	1.9 (1.0, 3.7)	1.7 (1.0, 2.8)
Methyl ethyl ketone	12	6	1.4 (0.5, 3.8)	0.8 (0.2, 3.6)	1.0 (0.4, 2.3)	0.9 (0.4, 2.1)	0.9 (0.4, 1.8)	0.9 (0.5, 1.4)
Mineral spirits	26	9	2.2 (1.0, 4.9)	2.2 (0.7, 7.2)	1.4 (0.6, 3.1)	1.8 (0.8, 4.2)	1.7 (0.8, 3.5)	1.6 (0.9, 2.8)
Naphtha	6	3	1.4 (0.4, 5.9)	1.1 (0.1, 11.4)	1.1 (0.4, 2.9)	1.0 (0.4, 2.8)	0.9 (0.4, 2.1)	0.9 (0.5, 1.5)
Paint thinner	43	17	1.9 (1.0, 3.4)	0.8 (0.3, 2.1)	1.1 (0.6, 2.2)	1.2 (0.6, 2.5)	1.3 (0.7, 2.5)	1.5 (0.9, 2.4)
Toluene	10	7	1.0 (0.4, 2.7)	0.7 (0.2, 3.2)	0.9 (0.4, 2.1)	0.8 (0.3, 2.0)	0.9 (0.4, 1.7)	0.9 (0.5, 1.4)
Turpentine	25	2	10.4 (2.4, 44.8)	17.8 (2.4, 130)	2.0 (0.8, 5.2)	4.8 (1.3, 17.9)	3.8 (1.2, 12.0)	3.1 (1.2, 7.9)
White gas	5	3	1.2 (0.3, 5.3)	1.1 (0.1, 11.0)	1.1 (0.4, 3.0)	1.0 (0.3, 2.8)	0.9 (0.4, 2.1)	0.9 (0.5, 1.5)
Xylene	10	5	1.4 (0.5, 4.3)	2.3 (0.4, 14.4)	1.1 (0.4, 2.8)	1.2 (0.5, 3.0)	1.0 (0.5, 2.1)	0.9 (0.6, 1.5)

Paints								
Oil-based paints	27	14	0.9 (0.3, 3.0)	0.9 (0.3, 3.0)	1.0 (0.5, 2.0)	0.9 (0.4, 2.2)	1.0 (0.5, 2.0)	1.0 (0.5, 1.8)
Water-based paints	24	16	1.1 (0.6, 2.2)	0.6 (0.2, 1.8)	0.8 (0.4, 1.6)	0.6 (0.3, 1.3)	0.6 (0.3, 1.2)	0.6 (0.3, 1.1)
Thinner solvents								
Lacquer thinner	36	8	3.5 (1.6, 7.8)	3.5 (1.1, 11.6)	1.9 (0.9, 4.0)	2.1 (0.9, 4.7)	1.9 (1.0, 3.7)	1.7 (1.0, 2.8)
Mineral spirits	26	9	2.2 (1.0, 4.9)	2.2 (0.7, 7.2)	1.4 (0.6, 3.1)	1.8 (0.8, 4.2)	1.7 (0.8, 3.5)	1.6 (0.9, 2.8)
Oil-based paints	27	14	0.9 (0.3, 3.0)	0.9 (0.3, 3.0)	1.0 (0.5, 2.0)	0.9 (0.4, 2.2)	1.0 (0.5, 2.0)	1.0 (0.5, 1.8)
Paint thinner	43	17	1.9 (1.0, 3.4)	0.8 (0.3, 2.1)	1.1 (0.6, 2.2)	1.2 (0.6, 2.5)	1.3 (0.7, 2.5)	1.5 (0.9, 2.4)
Turpentine	25	2	10.4 (2.4, 44.8)	17.8 (2.4, 130)	2.0 (0.8, 5.2)	4.8 (1.3, 17.9)	3.8 (1.2, 12.0)	3.1 (1.2, 7.9)
Metals								
Brass	8	4	1.5 (0.4, 5.2)	0.4 (0.1, 3.9)	1.0 (0.4, 2.5)	1.0 (0.4, 2.5)	1.0 (0.5, 2.2)	1.1 (0.7, 1.8)
Bronze	4	2	1.4 (0.3, 7.9)	1.7 (0.1, 50.9)	1.0 (0.3, 2.8)	1.1 (0.4, 3.2)	1.1 (0.5, 2.4)	1.1 (0.7, 1.8)
Galvanized iron or steel	18	8	1.6 (0.7, 3.9)	1.3 (0.3, 5.0)	1.2 (0.5, 2.7)	1.1 (0.5, 2.6)	1.1 (0.6, 2.2)	1.1 (0.7, 1.8)
High-speed steel	8	3	2.0 (0.5, 7.7)	2.8 (0.3, 27.2)	1.1 (0.4, 3.1)	1.2 (0.4, 3.2)	1.1 (0.5, 2.4)	1.1 (0.7, 1.8)
Mild steel	15	7	1.6 (0.6, 4.0)	0.4 (0.1, 2.0)	1.0 (0.4, 2.3)	0.9 (0.4, 2.1)	1.0 (0.5, 1.9)	1.1 (0.7, 1.7)
Stainless steel	13	5	1.9 (0.6, 5.4)	2.8 (0.5, 16.2)	1.2 (0.5, 3.1)	1.3 (0.5, 3.3)	1.2 (0.6, 2.5)	1.1 (0.7, 1.8)
Alloys (NOS[a])	3	5	0.4 (0.9, 1.7)	0.2 (0.1, 1.5)	0.7 (0.3, 1.9)	0.7 (0.3, 2.0)	0.9 (0.4, 1.9)	1.0 (0.6, 1.7)
Metals (NOS)	10	3	2.6 (0.7, 9.5)	2.0 (0.3, 15.5)	1.2 (0.5, 3.2)	1.3 (0.5, 3.4)	1.2 (0.6, 2.5)	1.1 (0.7, 1.9)
Solders (NOS)	17	5	2.6 (0.9, 7.1)	3.4 (0.8, 14.1)	1.4 (0.6, 3.4)	1.5 (0.6, 3.6)	1.3 (0.6, 2.7)	1.2 (0.7, 1.9)
Wood-derived substances								
Turpentine	25	2	10.4 (2.4, 44.8)	17.8 (2.4, 130)	2.0 (0.8, 5.2)	4.8 (1.3, 17.9)	3.8 (1.2, 12.0)	3.1 (1.2, 7.9)
Wood dust	47	26	1.5 (0.8, 2.8)	1.8 (0.9, 3.7)	1.4 (0.8, 2.6)	1.8 (0.9, 3.5)	1.8 (0.9, 3.5)	1.8 (0.9, 3.4)
Grain-derived dusts								
Flour dust	8	6	1.3 (0.3, 5.5)	1.4 (0.3, 6.8)	1.2 (0.5, 2.9)	1.6 (0.5, 5.3)	1.6 (0.5, 5.0)	1.6 (0.6, 4.7)
Grain dust	9	6	3.2 (0.7, 15.2)	4.4 (0.7, 29.7)	1.4 (0.5, 3.7)	2.0 (0.5, 7.3)	1.8 (0.5, 6.0)	1.7 (0.6, 5.1)
Non-ionizing EMF[4]								
Extremely low frequency fields	57	34	1.2 (0.8, 1.9)	0.8 (0.5, 1.5)	1.0 (0.6, 1.6)	1.0 (0.6, 1.7)	1.0 (0.6, 1.7)	1.1 (0.7, 1.7)
Radiofrequency fields	45	27	1.3 (0.8, 2.2)	1.3 (0.7, 2.3)	1.2 (0.7, 2.0)	1.3 (0.7, 2.1)	1.2 (0.7, 2.0)	1.2 (0.8, 1.9)
Cardboard dust	14	15	1.1 (0.4, 3.0)	0.9 (0.3, 2.8)	1.0 (0.5, 2.2)	1.0 (0.4, 2.2)	1.0 (0.5, 2.0)	1.0 (0.6, 1.8)
Herbicides (NOS)	13	8	1.3 (0.5, 3.2)	1.7 (0.4, 6.4)	1.2 (0.5, 2.7)	1.2 (0.5, 2.8)	1.1 (0.6, 2.3)	1.1 (0.6, 1.8)
Insecticides (NOS)	10	7	1.1 (0.4, 3.0)	0.8 (0.2, 3.3)	1.1 (0.5, 2.4)	1.0 (0.4, 2.4)	1.0 (0.5, 2.1)	1.0 (0.6, 1.8)
Ionizing radiation	5	4	1.2 (0.3, 4.5)	1.8 (0.4, 7.4)	1.2 (0.5, 3.0)	1.2 (0.4, 3.1)	1.1 (0.5, 2.5)	1.0 (0.6, 1.9)
Plastics, synthetics, resins (NOS)	4	5	0.6 (0.2, 2.3)	0.1 (0.0, 0.9)	0.7 (0.3, 1.9)	0.7 (0.2, 2.0)	0.8 (0.3, 2.0)	0.9 (0.5, 1.8)
Rubber dust	6	5	2.8 (0.3, 25.5)	5.6 (0.4, 88.1)	1.2 (0.4, 3.5)	1.2 (0.4, 3.9)	1.1 (0.4, 2.8)	1.1 (0.6, 2.0)

[a] n, 405 Case fathers; 302 control fathers.

[b] All estimates are adjusted for child's age, maternal race, maternal age, and maternal education.

[c] Subheadings indicate second-stage 'prior' covariates or categories of exchangeability; exposures may occur in more than one category.

[d] ML, maximum likelihood estimation; HM, hierarchical model; NOS, not otherwise specified; and EMF, electric and magnetic fields.

[e] Range in which we are 95% certain that true effect parameters will lie, after adjusting for second-stage covariates.

covariates. Therefore, if one exposure in the entire category of exchangeability had a falsely elevated or attenuated association due to chance, its estimate would be forced to be more similar to the others in the group. Obviously, more sophisticated specification of the Z-matrix would provide much better information for adjustment of estimates.

Analyses 5 and 6 are similar to the hierarchical regression specified in Analysis 4, except for the smaller values of the prespecified 95% ranges to estimate the prior variances. These models demonstrate the power the investigator has in controlling how conservative the estimates should be. The extreme situation in which the prior 95% range for the true parameters is set at a 2.5-fold range, illustrates that with this tight range, the chances of observing any but the strongest associations are greatly decreased.

In the hierarchical regression using empirical-Bayes estimation of the residual variance, τ^2 was set to zero (corresponding to a 95% certainty there are no residual effects of the first-stage exposures after adjusting for the second-stage covariates), and the SAS/IML procedure generated an automatic message indicating that there was possible underdispersion and semi-Bayes methods should be considered. Because of the potential problems in the estimation of τ^2, the results from this model are not presented.

DISCUSSION

In our conventional analysis of paternal occupational exposures and the incidence of neuroblastoma in offspring, there was some indication that exposures to hydrocarbons such as diesel fuel, lacquer thinner, mineral spirits, and turpentine, metals such as stainless and high-speed steel, and dusts such as wood dust were positively associated with neuroblastoma in offspring (Analysis 1). By adjusting the estimates based on prespecified prior distributions of the true effect parameters, a more consistent interpretation of the effects across the entire panel of exposures was possible. For example, although effect estimates were originally elevated for individual metal exposures (Analysis 1), these estimates were imprecise due to small numbers of persons exposed to each agent. After adjustment in the hierarchical regression analyses, most of these effect estimates were shrunk closer to the null value (Analyses 4). In comparison to the results for volatile hydrocarbons, of which several remain moderately elevated following the shrinkage procedure, the results for metals are less convincing. Conversely, we did not originally place much emphasis on the positive association observed for grain dust, because of its extreme imprecision and modestly elevated odds ratio (Analysis 1; OR = 3.2; CL ratio = 21.7). However, the hierarchical regression results show that the effect estimate for grain dust is somewhat elevated even after the shrinkage of the imprecise effect estimate toward a prior mean (Analysis 4; OR = 2.0; CL ratio = 14.6). These findings shed some light on our original interpretation of the data. Based on the results of the hierarchical regression analyses, further research into the effects of thinner solvents (lacquer thinner, mineral spirits, and turpentine), diesel fuel, and grain-, flour and wood dusts appear warranted, although a focus on metals seems less justifiable.

Although an appropriate value for the variance of true effect parameters was uncertain, we assumed that the true rate ratio for the effect of any occupational exposure, after adjusting for the second-stage covariates, is not likely to fall outside of a 10-fold range (e.g., between 0.5 and 5.0). Our results from the semi-Bayes analyses were sensitive to this choice; the odds ratios are shrunk closer to the null value in the models with prespecified five-fold and 2.5-fold ranges, which in some cases would affect our interpretation of results. With the measurement error and misclassification inherent in occupational exposure assessment that may bias odds ratios toward the null value when errors are independent and non-differential, we felt that interpretation of results from the more liberal analysis with 10-fold range was appropriate in this type of preliminary analysis of associations. In addition, simulation studies have observed that when conducting hierarchical regression with many first-stage exposures and few second-stage covariates, overspecifying τ^2 did not harm the confidence interval coverage or the mean squared error of estimates, whereas underspecifying τ^2 did [Greenland, 1993; Witte and Greenland, 1996]. Nonetheless, the entire array of information is useful in examining the sensitivity of results to the choice of the 95% prior range for the true residual effect parameters, and in providing further confidence in some results that remain elevated when the prior residual variance is extremely small.

In our example, use of empirical-Bayes estimation of the prior residual variance resulted in an estimate of zero for the variance of target parameters after adjusting for the second-stage covariates. Given the crude nature of our second-stage matrix, a value of zero for the prior residual variance is definitely unrealistic, and indicates a failure of the estimation procedure. Semi-Bayes methods appear to outperform empirical-Bayes methods when the ratio of subjects to parameters is not large [Greenland, 1993], as in our study (707 subjects/46 exposures $\approx$7). Thus, semi-Bayes methods seem preferable in this type of situation in which multiple effects are being estimated in a study of limited sample size, as long as the target parameters for a set of exposures are reasonably expected to fall within a definable range.

Use of a second-stage prior model can greatly improve the accuracy of effect estimates by modeling similarities among parameters of interest [Greenland, 1992, 1993, 1994, 1997, 2000; Greenland, 2001; Greenland and Poole, 1994; Witte and Greenland, 1996; Witte et al., 1994]. However, in

our study in which there was little prior information, a crudely specified second-stage covariate matrix did little to affect most of the estimates beyond the impact of an intercept-only second-stage model. Where no reliable prior information exists, a hierarchical regression analysis with an intercept-only model and prespecified residual variance at the second stage can be useful in enhancing the precision of estimates, as in our Analysis 3. Although confidence interval coverage rates tend to decrease with decreasing numbers of second-stage covariates, one simulation study found that having an intercept-only model did not harm confidence interval coverage rates as long as the prior residual variance was not underspecified [Witte and Greenland, 1996].

Even where only crude prior information exists (as in the form of categories of exchangeability), a hierarchical model with a simplified second-stage can outperform maximum likelihood estimation in enhancing the accuracy and precision of estimates [Witte and Greenland, 1996]. Some of the estimates in our analysis were changed by the inclusion of second-stage prior covariates in the model. However, where the use of prior covariates made a difference in the magnitude of effect estimates, there was also a loss of precision. These estimates may be more accurate as a result of added information provided in the second stage; however, the loss of precision may affect interpretation. Our prior covariates were all indicator variables grouping the first-stage exposures into exchangeable categories, and stratification of some of the exposures across several categories of the prior covariates was responsible for harming the precision of their posterior estimates. In general, lower precision occurs with the inclusion of a greater number of second-stage covariates, especially when the number of first-stage exposures is large [Greenland, 1993; Witte and Greenland, 1996]. Presumably, however, with the use of a carefully specified Z-matrix, the loss of precision is offset by reduction in bias. An improvement on the categories of exchangeability in our Z-matrix would be prior covariates that describe the toxicity of occupational exposures; for example, continuous measures of genotoxic potency that would determine the effects as linear functions of the continuous prior covariates. The Z-matrix could alternatively contain components of the first-stage exposures, as in studies of the effects of individual dietary items at the first-stage, mediated through the nutrient levels contained within each dietary item at the second-stage [Witte et al., 1994, 1998, 2001].

Simulation studies of hierarchical regression [Greenland, 1993, 1997; Witte and Greenland, 1996] have not addressed validity issues such as selection bias, reporting bias, and measurement error. In a population-based case-control study such as ours, these problems may greatly affect the accuracy of effect estimates. Although we have taken extensive measures to attain complete case ascertainment, collect a representative sample of population-based controls, and reduce reporting bias by carefully reviewing each reported occupational exposure to improve the specificity of our exposure measures, biases may still exist. Some of these biases could pose particular problems unique to the results of hierarchical regression analyses. For example, in our review of self-reported occupational exposures, we noticed that the frequency of reporting was higher for identifiable substances (e.g., turpentine) and chemical products with well-known common names (e.g., gasoline), compared to less frequent reporting of individual chemicals (e.g., benzene). Because these substances occur in the same category of exchangeability in the hierarchical regression models (i.e., volatile hydrocarbons), measurement error resulting from underreporting of individual chemicals may bias results in an unknown direction, not only for the substance with poor reporting, but for all other exposures in the category, since posterior estimates have been shrunk toward the common prior mean. Nothing conclusive is known about the accuracy of these self-reported occupational exposures; therefore, it is impossible to know to what extent results may have been influenced by differential reporting of the individual exposures. As in every statistical analysis, the quality of data will in part determine the quality of results.

With the development and increasing availability of software to perform hierarchical regression analyses, these procedures are becoming more accessible to application in epidemiology. The SAS/IML procedure we used is simple, and solely requires structuring as matrices in the SAS/IML language the results from the first-stage model, the prior residual variance, and second-stage covariates. However, the printed version [Witte et al., 1998] has errors which are corrected in the downloadable version posted on the Web. Also the procedure uses a weighted least-squares algorithm that tends to produce excessively wide intervals in small samples [Greenland, 1993]. The more sophisticated penalized-likelihood algorithm used by SAS GLIMMIX does not suffer from this problem [Greenland, 1997] and is also simple to use for epidemiologic analysis [Witte et al., 2001].

With relative ease of use, such analyses should become more commonplace in occupational epidemiology studies in which investigators perform preliminary screening of multiple occupational exposures without a priori hypotheses. Interpretation of results from hierarchical regression analyses may mitigate some of the problems inherent in conventional analyses, by controlling for correlated exposures, enhancing the precision of estimates, and providing some adjustment for associations occurring by chance by incorporating prior knowledge into the analysis. As in any occupational study, collection of accurate exposure information is crucial, and exposure assessment for use in hierarchical modeling requires additional thought as to the comparability of information quality across multiple

exposures, and the potential impact of data quality for individual exposures on the panel of results.

ACKNOWLEDGMENTS

The authors thank Drs. Sander Greenland and John Witte for their helpful suggestions concerning the analysis and careful review of the manuscript.

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Published in final edited form as:
Cancer Causes Control. 2012 May ; 23(5): 663–670. doi:10.1007/s10552-012-9921-1.

Poultry and Livestock Exposure and Cancer Risk among Farmers in the Agricultural Health Study

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Abstract

Purpose—The purpose of this study is to evaluate cancer risk associated with raising animals as commodities, which is associated with a variety of exposures, such as infectious agents and endotoxins.

Methods—Information was available for 49,884 male farmers in the Agricultural Health Study, who reported livestock and poultry production at enrollment (1993–1997). Cancer incidence data were obtained through annual linkage to state registries. Using Poisson regression analyses, we evaluated whether the number and type of animals raised on the farm impacted cancer risk.

Results—Overall, 31,848 (63.8%) male farmers reported raising any animals. Lung cancer risk decreased with increasing number of livestock on the farm (p-trend=0.04) and with raising poultry (Relative Risk (RR)= 0.6; 95% Confidence Interval (CI): 0.4–0.97). Raising poultry was associated with an increased risk of colon cancer (RR=1.4; 95% CI: 0.99–2.0) with further increased with larger flocks (p-trend=0.02). Risk of non-Hodgkin lymphoma was also elevated in those who raised poultry (RR=1.6; 95% CI: 1.0–2.4), but there was no evidence of increased risk with larger flocks (p-trend=0.5). Raising sheep was associated with a significantly increased risk of multiple myeloma (RR=4.9; 95% CI: 2.4–12.0). Performing veterinary services increased the risk of Hodgkin lymphoma (RR=12.2; 95% CI: 1.6–96.3).

Conclusions—We observed an inverse association between raising poultry and livestock and lung cancer risk and some evidence of increased risk of specific lymphohematopoietic malignancies with specific types of animals and performing veterinary services. Further research into associations between raising animals and cancer risk should focus on identification of etiologic agents.

Keywords

livestock; poultry; cancer; cohort study; agriculture

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Introduction

Animals raised on farms typically fall into two broad categories: poultry, which includes chickens and other birds raised for consumption and for egg production, and livestock, which includes large mammals, such as cattle (beef and dairy), hogs and sheep. Farmers who raise animals have exposures such as endotoxins, viruses, and pesticides that may influence their risk of cancer. Previous studies of individuals occupationally exposed to animals have suggested an increased risk of certain lymphohematopoietic malignancies, which may be related to their exposure to a variety of infectious agents [1]. Farmers, particularly those who raise animals, have been observed to have lower risk of lung cancer [2–3]. Although farmers smoke less than the general population, this does not appear to fully explain the lung cancer deficit. Exposure to endotoxins, which are lipopolysaccharides found in Gram-negative bacterial cell walls, is common in agricultural settings, and may be higher among those who have direct contact with animals or who handle hays and grains, which can be used as feed for animals. [4–6] Endotoxin exposure has been shown to be associated with a decreased risk of lung cancer [7], with inconsistent reports on cancer at other sites. However, cancer risks associated with specific types of farm animals or specific etiologic agents have not been clearly described. The purpose of this study is to evaluate cancer risk at a variety of sites in relation to raising livestock and poultry in a large cohort of occupationally exposed farmers.

Methods

Cohort Description

The Agricultural Health Study (AHS) is a prospective cohort that includes 57,310 licensed pesticide applicators in Iowa and North Carolina. Applicators were recruited and enrolled into the study during 1993 to 1997 when they obtained or renewed their licenses to apply restricted use pesticides. In North Carolina, only private applicators, who are primarily farmers, were recruited, while in Iowa, both private and commercial applicators (n=4,916) were included. Because the focus of this investigation was related to raising animals on the farm, we restricted these analyses to private applicators (farmers). Incident cancers were ascertained through annual linkage to state cancer registries in Iowa and North Carolina and first primary cancers diagnosed from enrollment through December 31, 2007 were included in this analysis. Annually, cohort members were matched to the National Death Index to identify vital status and to current address records of the Internal Revenue Service, motor vehicle registration offices, and pesticide license registries of state agricultural departments to determine whether they continued to reside in Iowa or North Carolina. Person-time was censored at the time of cancer incidence, death, movement out of the state or December 31, 2007, whichever was earlier. The study protocol was approved by the institutional review boards of the National Institutes of Health, the University of Iowa and other contractors.

Exposure assessment

Information about farming activities, including current raising of poultry and livestock, was collected through the completion of a self-administered questionnaire, available at http://aghealth.nci.nih.gov/questionnaires.html. Participants were asked about what major income-producing animals were currently raised on their farms (beef and dairy cattle, hogs, sheep, poultry or eggs) in the last year. They were also asked separately about the number of animals on their farm in the last year. Specifically, farmers were asked about the number of poultry and the number of all non-poultry livestock on their farms. For the purposes of this analysis, we considered anyone who reported raising poultry or eggs for income as having poultry. They were also queried about activities related to raising animals, such as performing veterinary services, butchering, grinding feed and milking cows, as well as

whether or not they worked in swine or poultry confinement areas. Participants also reported on other potential risk factors such as smoking, alcohol consumption, cancer history of first-degree relatives, diet, selected medical conditions, and demographic information.

There are relatively few females with a pesticide license in the AHS cohort (n=1,340) and only 522 who reported raising livestock or poultry on the farm. Of these, a much smaller percentage reported performing tasks related to raising animals than did their male counterparts. For these reasons, we restricted our analysis to male private applicators (n=51,036). Of these, we excluded 1,026 with a cancer diagnosis prior to enrollment and 126 who had missing or zero person-years leaving 49,884 applicators available for analysis. Relative risks (RR) and 95% confidence intervals (CI) were calculated by Poisson regression using SAS v9.1 (Cary, NC). Models were adjusted for age at enrollment (<50, 50–59, 60–69, 70+), smoking history (never, ≤12 pack-years, >12 pack-years), and state of residence (Iowa or North Carolina). We also controlled for pesticides associated with specific cancer types within the AHS. All tests were two-sided and conducted at the 0.05 alpha level. Tests for trend used the midpoint value of each exposure category treated as a continuous variable in Poisson regression models. In primary analyses, we used those who did not report exposure to each animal being studied, including those who reported raising no livestock, as the referent group. However, we also conducted analyses using farmers who did not report raising any animals in the past year as the referent group. We report results for exposures and cancer sites for which there were at least 5 exposed cases.

Results

Overall, 31,848 (63.8%) male farmers reported raising animals of any kind (Table 1). Most farmers raised more than one kind of animal. A total of 3,130 (10.8%) raised poultry, and of these only 783 (25.0%) raised poultry exclusively. Among livestock farmers who were currently raising animals, the most frequent type was beef cattle (n=18,663, 64.3%), followed by hogs (n=16,597, 57.2%). Of the 6,175 farmers who raised more than 1,000 livestock in the last year, 5,730 of them raised hogs (92.7%) and 2,890 raised cattle (46.8%). Farmers who raised animals were less likely to have ever smoked (57% never smokers) than their counterparts who did not raise animals (40% never smokers); thus all risk estimates are adjusted for smoking. Raising poultry was associated with a decreased risk of lung cancer (RR=0.6, 95% CI: 0.4–0.97) (Table 2). There was no evidence of further decreased risk with increasing numbers of poultry, although due to a small number of farmers with very large flocks, (e.g., ≥1,000 birds), we were only able to categorize flock size as <100 (RR=0.6, 95%CI 0.3–1.1) or ≥100 (RR=0.6, 95% CI: 0.3–1.2) birds (Table 3). Conversely, there was an increased risk of colon cancer among farmers who raised poultry (RR=1.4; 95% CI: 0.99–2.0), and an exposure-response association for having more poultry, with risks higher in those with more than 100 birds (RR=1.7, 95% CI 1.0–2.8; p-trend=0.02). There was some evidence of increased risk of Non-Hodgkin lymphoma (NHL) among those who raised poultry (RR=1.6; 95% CI: 1.0–2.4), but no evidence of increasing risk with more poultry (p-trend=0.48). The small number of cases of NHL (n=23) who raised poultry precluded detailed evaluation of NHL sub-types (data not shown). Working in a poultry confinement area was associated with an increased risk of NHL overall (RR=2.1; 95% CI: 1.2–3.7). There was no apparent association between raising poultry or working in a poultry confinement area and cancer at other sites.

Beef cattle were the most common type of livestock raised, and many farmers who raised dairy cattle also raised beef (Table 1). We examined these two types of cattle separately, but also considered them together since many of the exposures may be similar. There was a decreased risk of pancreatic cancer (RR=0.6; 95% CI: 03–0.9) among farmers who raised beef cattle (Table 2). Raising cattle did not appear to be associated with cancer at other sites.

Although there were no statistically significant associations between raising hogs in the last year and cancer at any site, there were suggestive positive associations with prostate cancer (RR=1.1; 95% CI: 0.99–1.2), leukemia (RR=1.3; 95%CI: 0.9–1.9) and multiple myeloma (RR=1.7; 95% CI: 0.96–3.0) (Table 2).

Although the RR for Hodgkin lymphoma for raising hogs was 1.6 (95% CI: 0.6–4.3), there was a significant increased risk among those who worked in hog confinement areas (RR=3.6; 95% CI: 1.2–10.3).

Raising sheep was also associated with a non-significantly decreased risk of lung cancer (RR=0.7; 95% CI: 0.3–1.7) based on 5 cases (Table 2). Based on 7 farmers who raised sheep, there was an increased risk of multiple myeloma (RR=4.9, 95% CI: 2.2–11.1). Permethrin, an insecticide widely used as a dip for sheep, has also been shown in a previous AHS analysis to be associated with increased risk of multiple myeloma. Further control for use of permethrin, however, did not substantively change risk estimates (RR=4.8, 95% CI: 2.1–10.7). Raising sheep was also associated with increased risk of cancers of the pancreas (RR=2.8 95% CI: 2.2–11.1) and brain (RR=2.7; 95% CI: 0.95–7.6).

There were no differences observed in risk estimates for any cancer site or animal type when we considered the referent group to be those with no livestock or poultry exposure; therefore results are reported using those who did not report exposure to each animal being evaluated as the referent group.

There was a significantly decreased risk of lung cancer among those who raised more than 1,000 head of livestock (RR=0.5, 95% CI: 0.3–0.97, p-trend=0.04) (Table 4). We were not able to link this to a specific type of animal as farmers were asked about the number of livestock on their farms and not about the number of specific types and many farmers raised more than one type. There was minimal evidence for an association between cancer and increasing number of livestock. We evaluated other factors associated with raising livestock and poultry, such as frequency of milking cows, grinding feed, butchering and performing veterinary services. The performance of veterinary services was associated with an increased risk of Hodgkin lymphoma based on 17 of the 18 cases who performed these tasks (RR=12.2; 95% CI: 1.6–96.3). Grinding feed on a monthly basis was also associated with increased risk of Hodgkin lymphoma with RR=1.7 (95% CI: 0.97–3.0). There was no evidence of an association with performing these tasks and cancer at any other site.

Discussion

The AHS provided the opportunity to evaluate cancer risks in relation to exposures associated with the rearing of poultry and livestock. In our analyses, we saw a decreased risk of lung cancer among poultry farmers. There was no additional reduction of lung cancer among those with larger numbers of birds. There was a decreasing risk, however, among those who had larger herds of livestock. Other published reports have observed deficits of lung cancer associated with dairy farming [8–9]; however, we did not observe a decrease associated with dairy cattle specifically. Of the types of livestock that we evaluated, only raising sheep was specifically associated with a decreased risk of lung cancer, although the association was not statistically significant. Unfortunately, we could not evaluate the impact of herd size and specific types of livestock since the participants were not asked to provide this information. Decreased risk of lung cancer has been consistently reported among farmers and has often been attributed to lower rates of tobacco use [2]. The decreased risks for lung cancer in our analyses persisted even after control for smoking. Exposure to endotoxins, which are associated with working with animals and handling hays and grains as feed for animals, is a primary hypothesis for the decreased risk of lung cancer among

farmers [7]. There is wide variability in endotoxin exposure levels depending on a number of factors, including the number of animals and the specific conditions in which the animals are kept [10] and amount of time spent with the animals. We were unable to account for these variations in our current analyses. Although early case-control studies conducted in Europe indicated an increased risk of lung cancer among people who had birds as pets [11–13], more recent studies, including those in the United States, have reported either no association or a decreased risk with associated with living with birds [14–15].

We observed a greater risk of NHL among those who raised poultry than among those who did not raise poultry, but no further increase among those who had more poultry on their farm. We also observed an increased risk among those who worked in hog confinements, but not among those who raised hogs. This discrepancy may indicate a spurious association, or it may indicate that there is an exposure specific to working in hog confinements that increases the risk of NHL. We saw no association with any other type of livestock. In a death certificate study, there was an increased risk of NHL mortality among those occupationally exposed to animals [1] although there was no information on specific types of animals. Results from that study also found statistically significant excesses for those with farming-related occupations with animal exposure and not statistically significant associations for non-farming occupations with possible animal exposures. In a death certificate study in Sweden, there was a non-statistically significant increased risk NHL among men who worked as livestock breeders, and among those who worked in dairy production [16]. There is some supporting evidence for our positive association with working in a hog confinement, as three case-control studies that showed increased risk of NHL with raising swine[17] or occupational exposure to pigs [18–19]. Tranah and colleagues also reported an increased risk of NHL among those who had worked with cattle more than five years [19].

In our analysis, we observed a statistically significant increased risk of multiple myeloma associated with raising sheep and a non-significant excess among those who raised hogs, but no evidence of an association with other types of animals. The association with raising sheep did not change with adjustment for use of permethrin, an insecticide used as a sheep dip that has been associated with multiple myeloma in the AHS [20].Multiple myeloma has been associated with farming in numerous studies [21], and there is increased incidence within the AHS [3]. Other studies have reported increased risk of multiple myeloma among those exposed to cattle [18, 22–23] and a meta-analysis of four case-control studies of multiple myeloma [23–26] showed statistically significant increased risk for multiple myeloma associated with raising sheep, horses and dairy cattle [21]. Svec and colleagues also reported increased risk of mortality from multiple myeloma among those who were occupationally exposed to animals, but did not specify the type of animals [1].

We found a non-statistically significant risk of leukemia among those farmers who raised hogs, but no other types of livestock or poultry. A mortality study among workers in a poultry processing plant found an excess risk of death from leukemia [27] and a case-control study showed increased risk of leukemia in those with occupational contact with beef cattle, including higher risks among those with longer duration of exposure [18]. Higher risks of lymphohematopoietic malignancies have been reported among butchers and those working in abattoirs (McLean and Pearce, 2005). However, the types of exposures experienced by workers in a processing plant may be very different than those who are engaged in the raising of chickens or livestock. Controlling for other factors such as performing veterinary services, butchering or coming in contact with animal blood did not change the association with leukemia. Additionally, we only had nine cases of leukemia among those who raised poultry, which may have limited our ability to detect an association.

With only eighteen cases of Hodgkin lymphoma overall, we had limited power to evaluate potential associations between this disease and livestock and poultry. We observed a RR of 1.6 (95% CI: 0.6–4.3) for farmers who reported raising hogs, but a statistically significant increase among those who reported working in a hog confinement. This is consistent with a case-control study that showed increased risk of Hodgkin Lymphoma among those occupationally exposed to pigs [28]. Additionally, we observed an increased risk among those who performed veterinary services. A proportionate mortality study of veterinarians in the United States observed significant excesses of Hodgkin lymphoma [29], and a case-control study based on death certificates indicated an increased risk of Hodgkin lymphoma among veterinarians [1]. The strongest etiologic hypotheses for Hodgkin lymphoma include immune response to infections [30] and individuals performing veterinary activities may have greater exposure to infectious agents than other farmers.

We observed an increased risk of colon cancer for farmers who raised poultry and higher risks among those with more birds. We are unaware of any other literature that has suggested such an association. Colon cancer incidence is generally lower in studies of farmers (Blair and Beane Freeman, 2009). An earlier study evaluating colon cancer and pesticide use in the AHS showed an association between the use of specific pesticides and colon cancer risk (Lee 2007). Controlling for use of these pesticides did not impact the observed risk estimates.

Strengths of this study include its prospective design, and more detailed information on the type and number of animals raised than is available in many other studies. We were able to assess risk for incident cancer cases diagnosed and reported to the state cancer registries. We were able to control for possible confounders for the various cancers, including use of various pesticides and lifestyle factors. Limitations include our inability to fully link the number of livestock to the types of livestock raised. For example, we saw some evidence of an inverse exposure-response with increasing numbers of livestock and lung cancer, but we were unable to attribute this to a specific type of animal because the question on the number of animals was not tied to the specific type of livestock. We do note, however, that most of the farmers with larger number of livestock raised hogs. We were also limited by small numbers to fully evaluate risks among farmers with only one type of livestock and by the fact that farmers were asked about the livestock and poultry that they were currently raising, which may not be indicative of previous exposures. Finally, while raising certain types of livestock may influence cancer risk, the identification of etiologic agents requires more detailed exposure assessment than was possible in this analysis.

Conclusions

We observed a decreased risk of lung cancer among farmers who raised poultry compared to those who did not. We also observed that lung cancer risk decreased with increasing numbers of livestock. These observations are consistent with increased exposure to endotoxins, which have been shown to decrease lung cancer risk and are elevated in agricultural settings, but other factors could be involved. We also observed increased risk of NHL among poultry farmers and increased risk of multiple myeloma among sheep farmers. We also observed an increased risk of Hodgkin lymphoma among those who performed veterinary services and worked in hog confinements. Colon cancer was also increased among poultry farmers, with evidence of an exposure-response trend. Possible exposures from rearing and tending of poultry and livestock have received less attention than chemical exposures in agricultural settings. Our findings indicate that further research into associations between livestock, poultry, and cancer risk are warranted and that they should focus on identification of possible etiologic agents.

Acknowledgments

This work was supported by funds from the Intramural Research Program of the National Institutes of Health (National Cancer Institute Z01 CP010119 and the National Institute of Environmental Health Sciences Z01 ES049030). All data used in these analyses were based on AHS data release P1REL201005.00.

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Table 1

Frequency of selected demographic and farm activities among AHS farmers who do and do not raise livestock

	Farmers with no livestock or poultry		Farmers with livestock or poultry		χ^2 p-value
	n	%	n	%	
	18,036	**36.2**	**31,848**	**63.8**	
Age at enrollment					
<35	2690	14.9	6492	20.4	
40–49	6562	36.4	13967	43.9	
50–59	4103	22.7	6511	20.4	
60–69	3332	18.5	3832	12.0	
70+	1349	7.5	1046	3.3	
					p<0.001
State					
Iowa	7085	39.3	23568	74.0	
North Carolina	10951	60.7	8280	26.0	
					p<0.001
Education					
< High school	2418	13.4	2486	7.8	
High school	7459	41.4	15602	49.0	
> High school	6570	36.4	13208	41.5	
					p<0.001
Smoking					
Never	7269	40.3	18262	57.3	
Former	5774	32.0	9024	28.3	
Current	3413	18.9	4362	13.7	
					p<0.001
Type of Livestock					
Poultry			3,130	10.8	
Beef			18,663	64.3	
Dairy			2,784	9.6	
Either Beef or Dairy			20,441	70.4	

Cancer Causes Control. Author manuscript; available in PMC 2013 May 01.

	Farmers with no livestock or poultry		Farmers with livestock or poultry		χ^2 p-value
	n	%	n	%	
	18,036	36.2	31,848	63.8	
Hogs			16,597	57.2	
Sheep			1,593	5.5	

Table 2

Major income producing animals on the farm and risk of cancer among male farmers in the AHS

	Poultry		Beef		Dairy	
Cancer Site	Number exposed (%)	RR 95 %CI	Number exposed (%)	RR 95 %CI	Number exposed (%)	RR 95 %CI
Lung	22 (4.9)	0.63 0.4–0.97	130 (28.7)	1.0 0.8–1.3	17 (4.0)	1.3 0.8–2.0
Pancreas	7 (8.3)	1.1 0.5–2.5	18 (21.4)	0.6 0.3–0.9	6 (3.0)	0.3 0.1–2.2
Colon	36 (10.4)	1.4 0.99–2.0	108 (31.1)	0.9 0.7–1.1	14 (4.1)	1.0 0.6–1.6
Rectum	10 (6.2)	0.7 0.4–1.4	58 (35.8)	1.0 0.7–1.4	4 (2.5)	NA
Renal	9 (6.0)	0.8 0.4–1.5	55 (36.4)	1.1 0.8–1.5	3 (2.0)	NA
Bladder	12 (6.0)	0.8 0.5–1.5	67 (33.5)	1.0 0.7–1.3	6 (3.0)	0.8 0.3–1.7
Prostate	130 (6.9)	1.0 0.8–1.1	705 (37.4)	1.1 0.98–1.2	77 (4.1)	0.9 0.7–1.2
Brain	1	NA	17 (30.4)	0.8 0.4–1.4	2 (3.6)	NA
Melanoma	14 (7.8)	0.9 0.5–1.6	62 (34.4)	0.9 0.7–1.3	7 (3.9)	0.8 0.4–1.6
Oral Cavity	1 (0.09)	NA	35 (32.4)	0.8 0.5–1.2	3 (2.8)	NA
Leukemia	9 (6.4)	0.9 0.5–1.7	47 (33.6)	0.9 0.6–1.2	3 (2.1)	NA
Hodgkin lymphoma	0	NA	9 (50.0)	1.3 0.5–3.4	2 (11.1)	NA
Non-Hodgkin lymphoma	23 (11.4)	1.6 1.0–2.4	75 (37.3)	1.1 0.8–1.4	10 (5.0)	1.1 0.6–2.0
Multiple myeloma	5 (7.2)	1.0 0.4–2.4	19 (27.5)	0.7 0.4–1.2	0	NA
Soft tissue sarcoma	0	NA	6 (37.5)	1.1 0.4–3.0	4 (25.0)	NA

Adjusted for age, state, smoking and education

Table 3

Number of poultry and risk of cancer among male farmers in the AHS

Cancer site	Number of Poultry	n	RR	95% CI	
Lung	None	324	1.0	REF	
	<100	11	0.6	0.3	1.1
	≥100	8	0.6	0.3	1.2
				p-trend=0.10	
Colon	None	252	1.0	REF	
	<100	18	1.3	0.8	2.0
	≥100	16	1.7	1.0	2.8
				p-trend=0.02	
Bladder	None	157	1.0	REF	
	<100	6	0.7	0.3	1.6
	≥100	6	1.2	0.5	2.6
				p-trend= 0.79	
Prostate	None	1427	1.0	REF	
	<100	83	1.1	0.9	1.3
	100–999	14	0.9	0.5	1.4
	≥1,000	27	0.8	0.6	1.2
				p-trend=0.31	
Non-Hodgkin lymphoma		153	1.0	REF	
	<100	13	1.4	0.8	2.5
	≥100	7	1.3	0.6	2.8
				p-trend=0.48	

Adjusted for age, state, smoking and education

Table 4

Number of livestock and risk of cancer among male farmers in the AHS

Cancer site	Number of livestock	n	RR	95% CI	
Lung	None	171	1.0	REF	
	<100	115	1.1	0.9	1.4
	100–999	60	0.9	0.7	1.3
	≥1,000	12	0.5	0.3	1.0
				p-trend=0.04	
Pancreas	None	32	1.0	REF	
	<100	17	0.9	0.5	1.6
	≥100	19	0.7	0.4	0.6
				p-trend=0.32	
Colon	None	128	1.0	REF	
	<100	73	0.9	0.7	1.2
	100–999	61	0.7	0.5	1.0
	≥1,000	26	0.8	0.5	1.3
				p-trend=0.15	
Rectum	None	55	1.0	REF	
	<100	34	1.0	0.6	1.5
	100–999	34	0.9	0.5	1.4
	≥1,000	15	1.0	0.5	1.8
				p-trend=0.72	
Renal	None	50	1.0	REF	
	<100	39	1.2	0.8	1.9
	100–999	31	0.8	0.5	1.3
	≥1,000	10	0.7	0.3	1.4
				p-trend=0.10	
Bladder	None	77			
	<100	41	0.9	0.6	1.3
	100–999	38	0.9	0.6	1.3
	≥1,000	15	0.9	0.5	1.7
				p-trend=0.66	

Cancer site	Number of livestock	n	RR	95% CI
Brain	None	22	1.0	REF
	<100	9	0.6	0.3 1.4
	100–999	13	0.7	0.3 1.4
	≥1,000	5	0.6	0.2 1.8
				p-trend=0.43
Cutaneous melanoma		54	1.0	REF
	<100	29	0.8	0.5 1.3
	100–999	45	1.2	0.7 1.8
	≥1,000	23	1.4	0.9 2.4
				p-trend=0.09

Int. J. Environ. Res. Public Health **2011**, *8*, 2320-2330; doi:10.3390/ijerph8062320

OPEN ACCESS

International Journal of
Environmental Research and Public Health
ISSN 1660-4601
www.mdpi.com/journal/ijerph

Article

Exposure to Multiple Pesticides and Risk of Non-Hodgkin Lymphoma in Men from Six Canadian Provinces

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Received: 20 May 2011; in revised form: 9 June 2011 / Accepted: 9 June 2011 / Published: 21 June 2011

Abstract: Non-Hodgkin lymphoma (NHL) has been linked to several agricultural exposures, including some commonly used pesticides. Although there is a significant body of literature examining the effects of exposure to individual pesticides on NHL, the impact of exposure to multiple pesticides or specific pesticide combinations has not been explored

in depth. Data from a six-province Canadian case-control study conducted between 1991 and 1994 were analyzed to investigate the relationship between NHL, the total number of pesticides used and some common pesticide combinations. Cases (n = 513) were identified through hospital records and provincial cancer registries and controls (n = 1,506), frequency matched to cases by age and province of residence, were obtained through provincial health records, telephone listings, or voter lists. In multiple logistic regression analyses, risk of NHL increased with the number of pesticides used. Similar results were obtained in analyses restricted to herbicides, insecticides and several pesticide classes. Odds ratios increased further when only 'potentially carcinogenic' pesticides were considered (OR[one pesticide] = 1.30, 95% CI = 0.90–1.88; OR[two to four] = 1.54, CI = 1.11–2.12; OR[five or more] = 1.94, CI = 1.17–3.23). Elevated risks were also found among those reporting use of malathion in combination with several other pesticides. These analyses support and extend previous findings that the risk of NHL increases with the number of pesticides used and some pesticide combinations.

Keywords: occupational cancer; non-Hodgkin lymphoma; pesticides; case-control study

1. Introduction

Non-Hodgkin lymphoma (NHL) has been associated with several agricultural and farm-specific exposures, including some phenoxy herbicide, organochlorine, organophosphate and carbamate pesticides [1-3]. Although a number of studies have examined the relationship between individual pesticides and NHL, few studies investigate the impact of exposure to multiple pesticides or specific pesticide combinations. This is necessary because most pesticide applicators use multiple chemicals throughout the year or in combination for individual applications.

DeRoos and colleagues pooled data from three NHL case-control studies conducted in the 1980s in four American mid-western states in one of the first attempts to examine the impact of exposure to multiple pesticides [4]. They found that, although the risk of NHL increased marginally with the number of pesticides used, it increased substantially when analyses were restricted to 'potentially carcinogenic' pesticides. Further, they found a super-additive effect whereby use of atrazine amplified risk of NHL when used in combination with several other pesticides including alachlor, diazinon and carbofuran [4].

In order to further evaluate the findings reported by DeRoos [4] we used data from a multi-provincial Canadian study to examine the impact of exposure to multiple pesticides, and common use combinations of pesticides, on the risk of NHL [5].

2. Methods

2.1. Data Source

The data used in these analyses were part of the Cross-Canada Study of Pesticides and Health, a case-control study of Canadian men 19 years of age or older, conducted between 1991 and 1994 in six Canadian provinces (Alberta, British Columbia, Manitoba, Ontario, Quebec, and Saskatchewan) [5].

Cases of NHL, Hodgkin lymphoma, soft tissue sarcoma, and multiple myeloma were identified through hospital records in Quebec and from cancer registries in all other provinces. A common control group for all cancer sites was assembled using provincial health insurance records (Alberta, Saskatchewan, Manitoba, and Quebec), computerized telephone listings (Ontario) and voter lists (British Columbia). Controls were frequency matched to cases by age (±2 years) and province of residence [5].

Information on demographic characteristics, medical and occupational history, exposure to selected substances, and other potentially confounding variables was obtained from all participants via a postal questionnaire. Detailed information on pesticide use was collected by telephone interview from all participants indicating they had ten or more hours of pesticide use during their lifetime and a 15% random sample of those with less than 10 hours. Specific pesticides were included in the questionnaire if the compound was ever registered for use in Canada and reviewed by the International Agency for Research on Cancer (IARC); if it was recently restricted or banned in Canada; or, if it was commonly used in Canada. Included pesticides were listed in table format, along with variables for number of days used and number of hours per day at home or work. This method of collecting pesticide use data was validated in a pilot study whereby twenty-seven volunteer farmers completed the questionnaire and subsequently provided purchase records. Investigators found excellent concordance between the two sources [5].

Questionnaires used in both portions of the study were modified versions of the questionnaire developed for a study of pesticide exposure, NHL and other tumors in Kansas and Nebraska, which were included in the analyses presented by DeRoos [4]. A detailed description of the data collection procedures for the Cross-Canada Study of Pesticides and Health has been published elsewhere [5,6]. The data used here are slightly different from previous publications because a pathology review resulted in the exclusion of four cases of NHL.

2.2. Statistical Analyses

2.2.1. Exposure to Multiple Pesticides

A brief examination of the impact of exposure to multiple pesticides on NHL has been reported previously in this population [5]. To expand upon these analyses, the total number of pesticides individuals reported using was categorized into four groups: no pesticide use, and use of one, two to four, or five or more pesticides. Additional analyses were conducted looking at number of insecticides, herbicides and fungicides used; the number of phenoxy herbicides, organochlorines, and organophosphates used; and the number of 'potentially carcinogenic' pesticides used. A pesticide was considered 'potentially carcinogenic' if it was classified as possibly carcinogenic to humans (group 2B) or higher by IARC [7], or suggestive evidence of carcinogenic potential or more severe by the United States Environmental Protection Agency (US EPA) Integrated Risk Assessment System or Office of Pesticides Program [8,9] (for a complete list of pesticides determined to be 'potentially carcinogenic' see Appendix A). All analyses were conducted using the statistical package SAS, version 9.2. Trends were examined using the Cochrane-Armitage test. Dose and duration information were not utilized in this analysis due to sample size limitations, which restricted further stratification.

2.2.2. Combinations of Pesticides

For the purpose of this analysis, a pesticide combination was defined as any two pesticides used by the same person. Commonly used pesticide combinations were determined by generating a correlation matrix of all pesticides used by twenty or more participants. All combinations yielding a correlation coefficient of 0.4 or greater were examined. In addition, combinations containing either malathion or mecoprop with a correlation coefficient of 0.3 or greater were examined based on hypotheses generated from associations found in preliminary analyses conducted using this dataset.

Unconditional logistic regression models were generated with variables for use of either individual pesticide in the combination, use of both pesticides, and use of neither pesticide. Where the odds ratio for joint exposure was higher than the odds ratio for exposure to either pesticide in the combination alone, interaction on the additive scale was evaluated using an interaction contrast ratio ($ICR = OR_{\text{both pesticides}} - OR_{\text{pesticide 1 only}} - OR_{\text{pesticide 2 only}} + 1$). ICR values above 0.5 were interpreted as indicating super-additivity. Models were developed which include a variety of potentially confounding factors suggested by the literature, including exposure to diesel exhaust, ultra-violet rays, and chemicals such as benzene; and family history of cancer in a first-degree relative.

The University of Toronto Health Sciences Research Ethics Board reviewed and approved the protocol for these secondary analyses. Ethics approval for data collection in the original study was obtained from research ethics boards in each province.

3. Results

The dataset used in this analysis contains information on 513 NHL cases and 1,506 controls. This represents 66.6% of contacted cases and 48.0% of contacted controls. As reported by McDuffie *et al*., potential subjects from urban and rural areas were equally likely to respond, and a greater proportion of responders were in the middle-age group than at either extreme among both cases and controls [5].

Cases were slightly older than controls and, proportional to their population size, the greatest number of cases and controls were obtained from Ontario and Quebec (Table 1). Proxy respondents were required for 21% of the cases and 15% of the controls. Nearly half of the participants had lived or worked on a farm in their lifetime. Additional demographic information on the participants has been published previously [5].

Table 1. Comparison of non-Hodgkin lymphoma cases and controls in the Cross-Canada Study of Pesticides and Health.

	Cases (n = 513)		**Controls (n = 1,506)**	
	Mean	**SD**	**Mean**	**SD**
Age	57.71	14.26	54.08	16.35
	N	**%**	**N**	**%**
Province				
Alberta	65	12.67	196	13.01
British Columbia	126	24.56	230	15.27
Manitoba	34	6.63	113	7.50
Ontario	142	27.68	585	38.84
Quebec	117	22.81	291	19.32
Saskatchewan	29	5.65	91	6.04

Table 1. *Cont.*

	Cases (n = 513)		Controls (n = 1,506)	
	Mean	SD	Mean	SD
	N	%	N	%
Ever lived or worked on a farm				
Yes	235	45.81	673	44.69
No	278	54.19	833	55.31
Respondent				
Self-respondent	403	78.56	1286	85.39
Proxy respondent	110	21.44	220	14.61

3.1. Multiple Pesticides

Risk of NHL tended to be greater among individuals who reported use of an increasing number of any type of pesticide (Table 2). This pattern was also evident for subgroups of herbicides, insecticides and fungicides. Odds ratios in the highest pesticide use category were 1.63 (95% CI: 1.20–2.21, p[trend] = 0.01) for any pesticide, 1.57 (95% CI: 0.96–2.57, p[trend] = 0.02) for herbicides, 1.70 (95% CI: 0.95–3.05, p[trend] < 0.01) for insecticides and 1.72 (95% CI: 1.07–2.77, p[trend] = 0.04) for fungicides. Odds ratios were also typically elevated for the use category of two to four pesticides, but less so than in the upper category. NHL risk also increased with number of pesticides used by chemical class (Table 3). Odds ratios tended to be the largest among participants using two or more pesticides in these categories with 1.78 (95% CI: 1.27–2.50, p[trend] = 0.01) for phenoxy herbicides, 1.36 (95% CI: 0.92–2.02, p[trend] = 0.15) for organochlorines, and 1.69 (95% CI: 1.04–2.74, p[trend] < 0.01) for organophosphates.

Table 2. Effect of exposure to multiple pesticides by pesticide type and carcinogenicity on NHL.

	Cases N (%)	Controls N (%)	OR *	95% CI
All pesticides			p(trend) = **0.01**	
0	352 (68.62)	1,095 (72.71)	1.00	–
1	14 (2.73)	56 (3.72)	0.80	0.44–1.47
2–4	67 (13.06)	176 (11.69)	**1.39**	**1.02–1.91**
5+	80 (15.59)	179 (11.89)	**1.63**	**1.20–2.21**
Herbicides			p(trend) = **0.02**	
0	369 (71.93)	1,147 (76.16)	1.00	–
1	45 (8.77)	127 (8.43)	1.24	0.86–1.80
2–4	73 (14.23)	167 (11.09)	**1.62**	**1.18–2.22**
5+	26 (5.07)	65 (4.32)	1.57	0.96–2.57
Insecticides			p(trend) < **0.01**	
0	367 (71.54)	1,153 (76.56)	1.00	–
1	43 (8.38)	126 (8.37)	1.22	0.84–1.77
2–4	85 (16.57)	189 (12.55)	**1.67**	**1.25–2.24**
5+	18 (3.51)	38 (2.52)	1.70	0.95–3.05

Table 2. *Cont.*

Fungicides			p(trend) = **0.04**	
0	453 (88.30)	1,361 (90.37)	1.00	–
1	30 (5.85)	90 (5.98)	1.03	0.67–1.60
2+	30 (5.85)	55 (3.65)	**1.72**	**1.07–2.77**
	Cases N (%)	**Controls N (%)**	**OR ***	**95% CI**
'Potentially carcinogenic' pesticides			p(trend) = **0.01**	
0	374 (72.90)	1,164 (77.29)	1.00	–
1	46 (8.97)	132 (8.76)	1.30	0.90–1.88
2–4	67 (13.06)	160 (10.62)	**1.54**	**1.11–2.12**
5+	26 (5.07)	50 (3.32)	**1.94**	**1.17–3.23**

* Adjusted for age, province and use of a proxy respondent.

Table 3. Effect of exposure to multiple pesticides by selected classes on NHL.

	Cases N (%)	**Controls N (%)**	**OR ***	**95% CI**
Phenoxy herbicides			p(trend) = **0.01**	
0	384 (74.85)	1,188 (78.88)	1.00	–
1	66 (12.87)	185 (12.28)	1.33	0.97–1.82
2+	63 (12.28)	133 (8.83)	**1.78**	**1.27–2.50**
Organochlorines			p(trend) = 0.15	
0	407 (79.34)	1,230 (81.67)	1.00	–
1	66 (12.87)	169 (11.22)	1.33	0.97–1.81
2+	40 (7.80)	107 (7.10)	1.36	0.92–2.02
Organophosphates			p(trend) < **0.01**	
0	421 (82.07)	1,337 (88.78)	1.00	–
1	65 (12.67)	115 (7.64)	**2.10**	**1.50–2.94**
2+	27 (5.26)	54 (3.59)	**1.69**	**1.04–2.74**

* Adjusted for age, province and use of a proxy respondent.

When analyses were restricted to those pesticides determined to be 'potentially carcinogenic', odds ratios increased further to 1.30 (95% CI: 0.90–1.88) in those reporting use of one pesticide, 1.54 (95% CI: 1.11–2.12) in those using two to four pesticides and 1.94 (95% CI: 1.17–3.23) in those using five or more pesticides (p[trend] = 0.01) (Table 2). This odds ratio is greater than any produced when examining use of any single pesticide [5]. Odds ratios were not significantly impacted by adjusting for potentially confounding factors such as exposure to ultra-violet rays, farm animals, or diesel exhaust (not presented).

3.2. Combinations of Pesticides

The correlation matrix yielded thirty-six pesticide combinations for analysis (for complete list of combinations examined see Appendix B). Several pesticide combinations produced higher odds ratios among participants using both pesticides than those reporting use of either one (Tables 4). These combinations always included malathion: malathion and 2,4-D, malathion and mecoprop, malathion

and glyphosate, malathion and DDT, and malathion and carbaryl. None of the interaction terms in these models were statistically significant, and only malathion and carbaryl had a super-additive joint effect (ICR > 0.5). Similar to analyses on multiple pesticides, these findings were not impacted by adjusting for potentially confounding factors.

Table 4. Individual and joint effects of commonly used pesticide combinations on NHL.

	Cases N (%)	Controls N (%)	OR *	95% CI
Malathion and 2,4-D			p = 0.59, ICR = 0.39	
Malathion only	11 (2.14)	21 (1.39)	1.73	0.81–3.66
2,4-D only	49 (9.55)	187 (12.42)	0.94	0.67–1.33
Malathion and 2,4-D	61 (11.89)	106 (7.04)	**2.06**	**1.45–2.93**
Malathion and carbaryl			p = 0.45, ICR = **1.42**	
Malathion only	52 (10.14)	106 (7.04)	**1.75**	**1.22–2.52**
Carbaryl only	5 (0.97)	13 (0.86)	1.17	0.41–3.36
Malathion and carbaryl	20 (3.90)	21 (1.39)	**3.34**	**1.77–6.31**
Malathion and DDT			p = 0.30, ICR = −0.64	
Malathion only	52 (10.14)	95 (6.31)	**2.03**	**1.41–2.94**
DDT only	13 (2.53)	27 (1.79)	1.72	0.86–3.42
Malathion and DDT	20 (3.90)	32 (2.12)	**2.11**	**1.17–3.80**
Malathion and glyphosate			p = 0.69, ICR = 0.23	
Malathion only	41 (7.99)	72 (4.78)	**1.95**	**1.29–2.93**
Glyphosate only	19 (3.70)	78 (5.18)	0.92	0.54–1.55
Malathion and glyphosate	31 (6.04)	55 (3.65)	**2.10**	**1.31–3.37**
Malathion and mecoprop			p = 0.64, ICR = 0.19	
Malathion only	44 (8.58)	92 (6.11)	**1.76**	**1.20–2.60**
Mecoprop only	23 (4.48)	46 (3.05)	**2.09**	**1.23–3.54**
Malathion and mecoprop	28 (5.46)	35 (2.32)	**3.04**	**1.80–5.15**

* Adjusted for age, province and use of a proxy respondent.

4. Discussion

Investigations of pesticides and cancer have, quite appropriately, focused on potential effects of individuals chemicals whenever possible for ease of analysis and policy and regulation purposes. Multiple exposures, however, complicate assessment of relationships between pesticides and cancer and more accurately reflect how pesticides are used in practice. McDuffie [5] previously reported that the risk of NHL in the Cross-Canada Study of Pesticides and Health tended to increase with the number of pesticides used. In a study from the United States, DeRoos [4] reported similar results in some cases, noting that risk increases when only pesticides with some evidence of carcinogenicity were included in the analysis and that risk were also increased for several specific combinations. Our results extend these findings.

The risk of NHL rose with increasing numbers of pesticides used and tests for trend were almost always statistically significant. Two additional findings stand out. First, the rising trend did not appear to be associated with any particular pesticide class and was observed for herbicides, insecticides, and fungicides. These analyses, however, are not on mutually exclusive exposure groups because many

individuals used pesticides from all three classes. Second, odds ratios increased further when only pesticides with some evidence of carcinogenicity were considered in the summation. Risk rose to nearly two-fold among those reporting use of five or more potentially carcinogenic pesticides.

Our findings and those from earlier studies [4,5] might be explained in a several ways. It could be that several pesticides each contribute a small risk that sums to a larger relative risk when they are considered in combination. Another explanation might be that as the number of pesticides used increases, the chances of including one or more that has considerable carcinogenic properties may also increase. Finally, use of multiple pesticides may be acting as a proxy measure for a more complex farming operation that may present some unique exposures that could be related to NHL.

DeRoos [4] had found that specific combinations of pesticides led to higher risks than would have been predicted from additive models, particularly those combinations that included atrazine. We were unable to evaluate findings for atrazine because its use was only reported by five individuals in the Cross-Canada Study of Pesticides and Health. Our analyses of specific combinations of pesticides did find some evidence of increased risk related to use of malathion in combination with 2,4-D, mecoprop, carbaryl, glyphosate, and DDT, where odds ratios increased beyond that from use of either pesticide alone. Interaction odds ratios should be interpreted cautiously because odds ratios for most combinations are not much larger than for malathion alone and were not statistically significant, and only the combination of malathion and carbaryl appeared to have a super-additive effect.

Findings indicating increased risk with reported use of pesticide combinations including malathion, a common organophosphate insecticide used on a wide range of crops and gardens and for public health-related mosquito control, are somewhat unexpected given that there is limited evidence of its carcinogenicity in human and animal studies. IARC categorized malathion as a group 3 substance (not classifiable as to its carcinogenicity to humans), and the US EPA classified it as having "suggestive evidence of carcinogenicity" [10,11]. There are several hypothesized mechanisms of carcinogenicity for malathion but they are not well-established, particularly for NHL [12].

A major limitation of our analysis is that our proxy measures for pesticide exposure were based on self-reported lifetime use. It is not clear whether use of combinations of pesticides were from actual tank mixtures, combinations used during the same growing season, or use in different years over a lifetime. These are quite different exposure scenarios and, even if the pesticides were carcinogenic, we might expect quite different biologic effects from these different exposure patterns. Moreover, we have no direct information on pesticide exposure or absorbed dose because analyses were based on self-reported pesticide use, which was measured in a binary fashion. This may result in exposure measurement error and depending on the underlying distribution of true exposure, and the presence of confounding and other factors, risk estimates can be biased in unpredictable ways.

Furthermore, recall bias for exposures is a concern in case-control studies because cases may have spent more time thinking about past exposures than controls. This could lead to differential misclassification and bias relative risks away from null. We lack direct information to address this issue, however, results from a methodological analysis of this issue in a similar case-control study in the United States did not uncover any evidence of case-response bias [13].

This study has several strengths. Information was obtained on pesticide use for a relatively large number of cases and controls. About 45% of cases and controls had lived or worked on a farm and occupational pesticide use was largely confined to this group. Accuracy of past events from

questionnaires is always a concern, but farmer's recall of pesticide has been found to be as good as for many other factors traditionally obtained by interview for epidemiologic studies [14]. Finally, information on many potential confounders for NHL was obtained and used in the models where appropriate but did not have a significant impact on risk.

5. Conclusions

These analyses confirm and extend previously reported results suggesting that the risk of NHL increases with the number of pesticides used, particularly when pesticides with some evidence of carcinogenicity are considered. Risk with reported use of combinations of pesticides showed few situations where risks were increased with pair wise use, although joint use of malathion and carbaryl appeared to have a super-additive effect. Additional work is needed to determine the role of exposure and dose, duration of exposure and factors modifying exposures such as protective clothing, respirators and glove use on these multiple-use situations.

Conflict of Interest

The authors declare no conflict of interest.

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Appendices

Appendix A. List of 'potentially carcinogenic' pesticides reportedly used by participants of the Cross-Canada Study of Pesticides and Health.

1. 2,4,5-T
2. 2,4-D
3. 2,4-DB
4. Arsenic
5. Asulam
6. Benomyl
7. Bromoxynil
8. Carbaryl
9. Cypermethrin
10. DDT
11. Dicamba
12. Diclofop-methyl
13. Dieldrin
14. Dimethoate
15. Dinoseb
16. Formaldehyde
17. Heptachlor
18. Lindane
19. Linuron
20. Mancozeb
21. MCPA
22. Mecoprop
23. Methidathion
24. Paraquat
25. Propoxur
26. Toxaphene
27. Triallate
28. Trichloroacetic acid
29. Trifluralin

Appendix B. Complete list of pesticide combinations evaluated.

1. Bromoxynil and diallate
2. Bromoxynil and glyphosate
3. Carbathin and bromoxynil
4. Carbathin and glyphosate
5. Carbofuran and diallate
6. Diallate and bromoxynil
7. Diallate and carbathin
8. Diclofop methyl and bromoxynil
9. Diclofop methyl and carbathin
10. Diclofop methyl and diallate
11. Difenzoquat and bromoxynil
12. Difenzoquat and carbathin
13. Difenzoquat and diclofop methyl
14. Difenzoquat and sethoxydim
15. Difenzoquat Trifluralin
16. Glyphosate and 2,4-D
17. Malathion and 2,4-D
18. Malathion and carbaryl
19. Malathion and DDT
20. Malathion and dimethoate
21. Malathion and glyphosate
22. Malathion and mecoprop
23. Malathion and methoxychlor
24. Mecoprop glyphosate
25. Mecoprop and methoxychlor
26. Mecoprop and 2,4-D
27. Methoxychlor and 2,4-D
28. Sethoxydim and bromoxynil
29. Sethoxydim and carbathin
30. Sethoxydim and carbofuran
31. Sethoxydim and diclofop-methyl
32. Triallate and diclofop-methyl
33. Triallate and trifluralin
34. Trifluralin and bromoxynil
35. Trifluralin and carbathin
36. Trifluralin and difenzoquat

American Journal of Epidemiology
Published by Oxford University Press on behalf of the Johns Hopkins Bloomberg School of Public Health 2012.

Vol. 177, No. 1
DOI: 10.1093/aje/kws225
Advance Access publication:
November 21, 2012

Original Contribution

Risk of Total and Aggressive Prostate Cancer and Pesticide Use in the Agricultural Health Study

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Initially submitted November 17, 2011; accepted for publication April 19, 2012.

Because pesticides may operate through different mechanisms, the authors studied the risk of prostate cancer associated with specific pesticides in the Agricultural Health Study (1993–2007). With 1,962 incident cases, including 919 aggressive prostate cancers among 54,412 applicators, this is the largest study to date. Rate ratios and 95% confidence intervals were calculated by using Poisson regression to evaluate lifetime use of 48 pesticides and prostate cancer incidence. Three organophosphate insecticides were significantly associated with aggressive prostate cancer: fonofos (rate ratio (RR) for the highest quartile of exposure (Q4) vs. nonexposed = 1.63, 95% confidence interval (CI): 1.22, 2.17; $P_{trend} < 0.001$); malathion (RR for Q4 vs. nonexposed = 1.43, 95% CI: 1.08, 1.88; $P_{trend} = 0.04$); and terbufos (RR for Q4 vs. nonexposed = 1.29, 95% CI: 1.02, 1.64; $P_{trend} = 0.03$). The organochlorine insecticide aldrin was also associated with increased risk of aggressive prostate cancer (RR for Q4 vs. nonexposed = 1.49, 95% CI: 1.03, 2.18; $P_{trend} = 0.02$). This analysis has overcome several limitations of previous studies with the inclusion of a large number of cases with relevant exposure and detailed information on use of specific pesticides at 2 points in time. Furthermore, this is the first time specific pesticides are implicated as risk factors for aggressive prostate cancer.

aggressive prostate cancer; cohort study; farming; organophosphate insecticides; pesticide exposure; prostate cancer

Abbreviations: CI, 95% confidence interval; Q4, highest quartile of exposure; RR, rate ratio.

Occupational exposure to pesticides has been associated with increased prostate cancer risk in many epidemiologic studies (1–6). In the Agricultural Health Study, the largest prospective cohort study to examine this association, a significant excess of prostate cancer has been observed for both private (farmer) and commercial applicators, with standardized incidence ratios = 1.19 (95% confidence Interval (CI): 1.14, 1.25) and 1.28 (95% CI: 1.00, 1.61), respectively, compared with rates expected in the 2 study states (7). Although several groups or chemical classes have been linked to prostate cancer, including triazine herbicides (1, 8, 9), organochlorine insecticides (9–12), and organophosphate insecticides (9, 13, 14), none of the associations is conclusive, and it is unclear which specific pesticides might be driving the group findings. Alteration of hormonal signaling pathways or induction of DNA damage is each postulated as a mechanism (15–19).

Investigation of the role of pesticides in prostate cancer development is complicated because of the need to obtain information on exposure to specific individual pesticides, to track changes in pesticide use patterns over time, and, because prostate cancer is so common in older men, to consider whether pesticides are associated with clinically significant or aggressive disease. We are aware of only 2 reports that considered tumor characteristics, one that reported no association between any pesticide exposure and

Table 1. Characteristics of Incident Prostate Cancer Cases in the Agricultural Health Study, 1993–2007

Characteristic	Cohort Person-Years (Total = 638,628.4)	Total Prostate Cancer (*n* = 1,962)		Aggressive Prostate Cancer[a] (*n* = 919)		Family History of Prostate Cancer (*n* = 305)	
		No.	%	No.	%	No.	%
Age at diagnosis, years[b]							
<60	614,045.6	406	20.7	179	19.5	78	25.6
60–64	5,043.1	360	18.4	159	17.3	60	19.7
65–69	6,573.0	489	24.9	227	24.7	77	25.3
70–74	5,885.6	382	19.5	181	19.7	52	17.1
≥75	7,081.1	325	16.6	173	18.8	38	12.5
State							
Iowa	415,184.0	1,153	58.8	588	64.0	212	69.5
North Carolina	638,628.4	809	41.2	331	36.0	93	30.5
Race							
White	602,100.5	1,797	91.6	852	92.7	296	97.1
Black	21,923.0	74	3.8	42	4.6	8	2.6
Other/missing	14,604.9	91	4.6	25	2.7	1	0.3
Family history of prostate cancer							
No	532,438.5	1,399	71.3	661	72.0	N/A	
Yes	48,709.6	305	15.6	139	15.1	305	100
Missing	57,480.3	258	13.2	118	12.9	N/A	
Smoking status							
Never	331,056.9	922	47.0	442	48.1	153	50.1
Former	170,340.9	709	36.1	328	35.7	115	37.7
Current	104,753.8	198	10.1	90	9.8	28	9.2
Missing	32,476.8	133	6.8	59	6.4	9	3.0
Fruit servings							
<1/day	437,300.6	1,229	62.6	580	63.1	203	66.5
≥1/day	157,242.9	533	27.2	243	26.4	93	30.5
Missing	44,084.9	200	10.2	96	10.5	9	3.0
Leisure-time physical activity in the winter							
None	72,048.4	359	18.3	157	17.1	62	20.3
>0–2 hours/week	119,336.9	418	21.3	209	22.7	72	23.6
≥3 hours/week	79,519.3	234	11.9	107	11.6	32	10.5
Missing	367,723.9	951	48.5	446	48.5	139	45.6

Table continues

risk of localized or advanced prostate cancer (20) and another that reported a larger proportion of later stage tumors among men with "significant" exposure to pesticides compared with men with no exposure (21).

We used data from the Agricultural Health Study, a large cohort study of pesticide applicators with pesticide use data at 2 points in time, to evaluate the association between specific pesticide exposure and prostate cancer. We previously reported on pesticide use and prostate cancer risk among 566 incident cancer cases that occurred through 1999 (13). In the current study, we extend follow-up through 2007 and update analyses to include 1,962 incident cases of prostate cancer (including 919 cases of aggressive prostate cancer).

MATERIALS AND METHODS

Study population

The Agricultural Health Study is a prospective cohort study of 52,394 licensed private pesticide applicators in Iowa and North Carolina and 4,916 licensed commercial

Table 1. Continued

Characteristic	Cohort Person-Years (Total = 638,628.4)	Total Prostate Cancer (n = 1,962)		Aggressive Prostate Cancer[a] (n = 919)		Family History of Prostate Cancer (n = 305)	
		No.	%	No.	%	No.	%
Stage							
Localized	10,502.8	1,499	76.4	596	64.9	238	78.0
Regional	2,044.5	324	16.5	230	25.0	51	16.7
Distant	447.6	59	3.0	59	6.4	6	2.0
Unknown	517.5	80	4.1	34	3.7	10	3.3
Grade							
Well differentiated, Gleason score 2–4	381.1	88	4.5	2	0.2	14	4.6
Moderately differentiated, Gleason score 5–6	6,220.6	935	47.7	22	2.4	147	48.2
Poorly differentiated, Gleason score 7–10	6,465.5	875	44.6	875	95.2	138	45.2
Not graded	445.2	64	3.3	20	2.2	6	2.0
Gleason score							
2–6	5,813.7	840	42.8	17	1.8	141	46.2
7	4,381.1	583	29.7	583	63.4	96	31.5
8–10	1,787.7	232	11.8	232	25.2	37	12.1
Missing	1,530.0	307	15.7	87	9.5	31	10.2
Fatal prostate cancer, yes	556.8	106	5.4	106	11.5	10	3.3
Age at diagnosis, years							
<60	614,045.6	406	20.7	179	19.5	78	25.6
60–64	5,043.1	360	18.4	159	17.3	60	19.7
65–69	6,573.0	489	24.9	227	24.7	77	25.3

Abbreviations: N/A, not available; SD, standard deviation.

[a] Distant stage or poorly differentiated (after January 1, 2003, Gleason score 7–10) or Gleason score ≥7 or fatal (underlying cause: prostate cancer).

[b] Mean age at diagnosis: total prostate cancer, 66.5 (SD, 8.3) years; aggressive prostate cancer, 67.1 (SD, 8.5) years; family history of prostate cancer, 65.2 (SD, 7.9) years.

applicators from Iowa. The cohort has been described in detail by Alavanja et al. (22). Briefly, the cohort included individuals seeking licenses for restricted use pesticides from December 1993 through December 1997 (82% of the target population enrolled). All participants provided informed consent, and the protocol was approved by relevant institutional review boards. We obtained cancer incidence information by annual linkage to cancer registry files in Iowa (Surveillance, Epidemiology, and End Results Program) and North Carolina (National Program of Cancer Registries). In addition, we annually matched cohort members to state mortality registries and the National Death Index to identify vital status and to address records of the Internal Revenue Service, motor vehicle registration files, and pesticide license registries of state agricultural departments to determine residence in Iowa or North Carolina. The current analysis included all incident prostate cancers (n = 1,962) diagnosed from enrollment (1993–1997) through December 31, 2007. We censored follow-up at the time of death, movement out of state, or December 31, 2007. Among the 57,310 applicators, we excluded 2,898 participants (1,563 females, 1,071 prevalent cancers of all types, 264 with no follow-up information), leaving 54,412 individuals.

Tumor characteristics

Information on tumor characteristics was obtained from state cancer registries. Cases were characterized by stage (localized, regional, distant, or unknown extension or metastasis), histologic grade (well differentiated, moderately differentiated, and poorly differentiated), and Gleason score. Tumors that were not classified by pathologists were listed as having unknown grade. Gleason scores are currently equated with the 3 grade categories as follows: tumors with Gleason scores of 2–4 are classified as well differentiated, scores of 5–6 as moderately differentiated, and scores of 7–10 as poorly differentiated (23). For cases diagnosed prior to January 1, 2003, when the grading procedure was modified (23), we reabstracted Gleason scores

Table 2. Phase I and Phase II Data for Cumulative Lifetime Pesticide Exposure to Organophosphate Insecticides and Risk of Total and Aggressive Prostate Cancer in the Agricultural Health Study, 1993–2007

	Total Prostate Cancer			Aggressive Prostate Cancer[a]		
	No. of Cases[b]	RR[c]	95% CI	No. of Cases[b]	RR[c]	95% CI
Chlorpyrifos						
Nonexposed	1,129	1.00	Referent	511	1.00	Referent
Q1	167	1.08	0.92, 1.28	83	1.02	0.81, 1.29
Q2	168	1.03	0.87, 1.21	83	1.10	0.87, 1.39
Q3	166	0.94	0.80, 1.11	82	1.15	0.90, 1.46
Q4	167	0.89	0.75, 1.05	82	1.01	0.80, 1.28
P_{trend}			0.11			0.84
Coumaphos						
Nonexposed	1,506	1.00	Referent	710	1.00	Referent
Q1	35	1.18	0.84, 1.65	14	0.85	0.49, 1.46
Q2	35	0.81	0.58, 1.13	14	0.64	0.38, 1.08
Q3	35	0.93	0.66, 1.30	14	0.89	0.52, 1.54
Q4	34	1.02	0.72, 1.43	14	0.90	0.53, 1.53
P_{trend}			0.97			0.59
Dichlorvos						
Nonexposed	1,515	1.00	Referent	705	1.00	Referent
Q1	43	1.07	0.79, 1.45	22	0.92	0.59, 1.44
Q2	43	1.01	0.74, 1.36	22	1.15	0.76, 1.75
Q3	43	0.85	0.63, 1.15	22	0.90	0.58, 1.39
Q4	43	0.91	0.67, 1.24	21	0.95	0.62, 1.48
P_{trend}			0.50			0.80
Diazinon[d]						
Nonexposed	727	1.00	Referent	343	1.00	Referent
Q1	66	1.30	1.01, 1.68	31	1.24	0.84, 1.85
Q2	63	1.15	0.88, 1.49	29	1.00	0.67, 1.48
Q3	66	1.04	0.81, 1.35	30	0.89	0.59, 1.34
Q4	63	0.94	0.72, 1.24	30	1.31	0.87, 1.96
P_{trend}			0.59			0.27
Fonofos						
Nonexposed	1,305	1.00	Referent	581	1.00	Referent
Q1	97	0.89	0.74, 1.17	55	0.96	0.72, 1.28
Q2	95	1.38	1.11, 1.70	50	1.20	0.89, 1.61
Q3	96	1.13	0.91, 1.39	52	1.16	0.86, 1.55
Q4	96	1.21	0.98, 1.49	52	1.63	1.22, 2.17
P_{trend}			0.03			<0.001

Table continues

and harmonized the classification scheme with current practice. For 35 cases from Iowa and 24 cases from North Carolina, Gleason score information conflicted with the reported grade category; in these instances, we used the abstracted Gleason score to assign an appropriate grade code. Gleason score was missing for 62 of 1,153 (5.4%) incident cases from Iowa and 245 of 809 (30.3%) incident cases from North Carolina. If the Gleason score was missing, the original histologic grade variable delivered from the yearly cancer registry link was used (22 well differentiated, 161 moderately differentiated, 60 poorly differentiated, and 64 not graded). For the current analysis, aggressive prostate cancer was defined as having 1 or more of the following tumor characteristics: distant stage, poorly differentiated grade, Gleason score of ≥7, or fatal prostate cancer (underlying cause, prostate cancer). Two alternative definitions of aggressive prostate cancer were also considered in analysis (using a Gleason score cutoff of ≥4 + 3 or a Gleason score

Table 2. Continued

	Total Prostate Cancer			Aggressive Prostate Cancer[a]		
	No. of Cases[b]	RR[c]	95% CI	No. of Cases[b]	RR[c]	95% CI
Malathion[d]						
Nonexposed	328	1.00	Referent	140	1.00	Referent
Q1	189	1.03	0.84, 1.26	95	1.19	0.89, 1.59
Q2	187	1.13	0.94, 1.36	93	1.27	0.97, 1.67
Q3	184	1.11	0.93, 1.34	93	1.28	0.98, 1.68
Q4	186	1.08	0.90, 1.29	93	1.43	1.08, 1.88
P_{trend}			0.62			0.04
Parathion[d]						
Nonexposed	878	1.00	Referent	413	1.00	Referent
Q1	25	1.21	0.81, 1.81	12	1.96	1.10, 3.50
Q2	25	1.37	0.92, 2.05	12	1.04	0.58, 1.86
Q3	25	1.21	0.81, 1.81	12	1.51	0.82, 2.77
Q4	24	0.85	0.56, 1.28	11	0.98	0.53, 1.79
P_{trend}			0.51			0.97
Phorate[d]						
Nonexposed	675	1.00	Referent	314	1.00	Referent
Q1	76	0.96	0.76, 1.23	37	0.78	0.55, 1.12
Q2	76	1.11	0.87, 1.41	36	1.26	0.89, 1.79
Q3	77	0.88	0.69, 1.13	37	0.80	0.56, 1.14
Q4	75	1.12	0.88, 1.42	36	1.36	0.96, 1.93
P_{trend}			0.46			0.10
Terbufos						
Nonexposed	1,042	1.00	Referent	466	1.00	Referent
Q1	162	1.05	0.88, 1.24	81	1.06	0.83, 1.36
Q2	158	1.08	0.91, 1.28	80	1.06	0.83, 1.35
Q3	161	1.06	0.89, 1.25	80	1.15	0.90, 1.47
Q4	158	1.04	0.88, 1.23	80	1.29	1.02, 1.64
P_{trend}			0.63			0.03

Abbreviations: CI, confidence interval; Q1, quartile 1; Q2, quartile 2; Q3, quartile 3; Q4, quartile 4; RR, rate ratio.

[a] Distant stage or poorly differentiated (after January 1, 2003, Gleason score 7–10) or Gleason score ≥7 or fatal (underlying cause: prostate cancer).

[b] Numbers do not sum to total because of missing data.

[c] Adjusted for age, state, race, family history of prostate cancer, smoking, fruit servings, and leisure-time physical activity in the winter.

[d] Detailed information for these chemicals was collected on the take-home questionnaire at enrollment.

of ≥8) in combination with the other factors listed above (stage, fatal disease).

Exposure assessment

Information on lifetime use of 50 pesticides was captured in 2 self-administered questionnaires (http://aghealth.org/questionnaires.html) completed during cohort enrollment (phase 1). All 57,310 applicators completed the first enrollment questionnaire, which inquired about ever/never use of the 50 pesticides, as well as duration (years) and frequency (average days/year) of use for a subset of 22 pesticides. In addition, 25,291 of 57,310 (44.1%) of the applicators returned the second (take-home) enrollment questionnaire, which inquired about duration and frequency of use for the remaining 28 pesticides. We used 2 exposure metrics to assess cumulative exposure to each pesticide: 1) lifetime days of pesticide use, that is, the product of years of use of a specific pesticide and the number of days used per year; and 2) intensity-weighted lifetime days of use, that is, the product of lifetime days of use and a measure of exposure intensity. Intensity was derived from an algorithm using questionnaire data on mixing status, application method,

Table 3. Phase I and Phase II Data for Cumulative Lifetime Pesticide Exposure to Organochlorine Insecticides and Risk of Total and Aggressive Prostate Cancer in the Agricultural Health Study, 1993–2007

	Total Prostate Cancer			Aggressive Prostate Cancer[a]		
	No. of Cases[b]	RR[c]	95% CI	No. of Cases[b]	RR[c]	95% CI
Aldrin						
Nonexposed	715	1.00	Referent	328	1.00	Referent
Q1	65	1.04	0.80, 1.35	33	0.97	0.67, 1.41
Q2	64	0.94	0.72, 1.22	33	1.09	0.75, 1.57
Q3	64	1.14	0.88, 1.48	34	1.21	0.84, 1.74
Q4	64	1.25	0.97, 1.63	31	1.49	1.03, 2.18
P_{trend}			0.07			0.02
Chlordane						
Nonexposed	740	1.00	Referent	356	1.00	Referent
Q1	59	0.79	0.61, 1.04	26	0.73	0.48, 1.10
Q2	58	1.29	0.99, 1.69	26	1.07	0.72, 1.60
Q3	58	0.96	0.73, 1.25	26	0.91	0.61, 1.37
Q4	58	1.02	0.78, 1.34	25	1.17	0.77, 1.77
P_{trend}			0.80			0.49
DDT						
Nonexposed	578	1.00	Referent	267	1.00	Referent
Q1	96	0.98	0.78, 1.22	47	1.06	0.76, 1.48
Q2	97	1.27	1.02, 1.58	46	1.17	0.85, 1.61
Q3	96	1.27	1.02, 1.58	46	1.56	1.13, 2.15
Q4	95	1.18	0.95, 1.48	46	1.30	0.94, 1.80
P_{trend}			0.14			0.10
Dieldrin						
Nonexposed	918	1.00	Referent	429	1.00	Referent
Q1	19	0.94	0.60, 1.49	8	0.83	0.41, 1.68
Q2	19	0.86	0.54, 1.36	7	2.00	0.94, 4.23
Q3	18	0.93	0.58, 1.49	8	0.68	0.33, 1.37
Q4				7	1.39	0.65, 2.94
P_{trend}			0.68			0.54

Table continues

equipment repair, and use of personal protective equipment (24). A follow-up questionnaire, which ascertained pesticide use since enrollment, was administered 5 years after enrollment (phase 2) and completed by 36,342 (63%) of the original participants. For participants who did not complete a phase 2 questionnaire (20,968 applicators, 37%), a data-driven multiple imputation procedure was used to impute use of specific pesticides in phase 2. A detailed description of the imputation process and validation is described by Heltshe et al. (25). Briefly, logistic regression and stratified sampling were used to impute use of specific pesticides in phase 2. All variables from phase 1 that had the potential to be associated with either missingness or pesticide use were considered. The variables most strongly predictive of use of any pesticide on the phase 2 questionnaire were gender, marital status, farm ownership, farm size, days/year mixing pesticides, percent time personally mixing pesticides, percent time personally applying pesticides, and application of any pesticide in the prior year. Covariates associated with nonresponse to phase 2 were age, education, state, applicator type, and years mixing chemicals. Covariates from participants with complete data from both phases were modeled and then applied to the model for participants missing phase 2 data to obtain estimates of the missing data. To assess the imputation procedure, a 20% random sample of participants was withheld for comparison. The observed and imputed prevalences of any pesticide use in the holdout data set were 85.7% and 85.3%, respectively, indicating that the logistic regression model for the multiple imputation performed well.

We combined phase 1 and phase 2 information to generate cumulative intensity-weighted and unweighted days of use. Web Table 1 (available at http://aje.oxfordjournals.org/)

Table 3. Continued

	Total Prostate Cancer			Aggressive Prostate Cancer[a]		
	No. of Cases[b]	RR[c]	95% CI	No. of Cases[b]	RR[c]	95% CI
Heptachlor						
Nonexposed	809	1.00	Referent	369	1.00	Referent
Q1	45	1.08	0.80, 1.47	24	1.29	0.83, 2.00
Q2	44	1.05	0.77, 1.44	24	1.65	1.08, 2.52
Q3	45	1.03	0.76, 1.40	24	1.17	0.77, 1.76
Q4	44	1.05	0.78, 1.44	23	0.88	0.57, 1.35
P_{trend}			0.73			0.62
Lindane						
Nonexposed	840	1.00	Referent	395	1.00	Referent
Q1	43	0.88	0.63, 1.23	19	0.81	0.50, 1.32
Q2	36	1.06	0.76, 1.49	19	0.91	0.56, 1.49
Q3	39	1.06	0.76, 1.48	19	1.45	0.91, 2.30
Q4	39	1.16	0.84, 1.60	19	1.24	0.77, 2.00
P_{trend}			0.33			0.23
Toxaphene						
Nonexposed	831	1.00	Referent	386	1.00	Referent
Q1	39	0.91	0.66, 1.26	19	1.02	0.64, 1.65
Q2	38	1.06	0.77, 1.46	19	1.32	0.83, 2.09
Q3	38	1.28	0.92, 1.78	19	1.30	0.82, 2.07
Q4	38	0.97	0.70, 1.35	19	1.14	0.71, 1.83
P_{trend}			0.95			0.48

Abbreviations: CI, confidence interval; DDT, dichlorodiphenyltrichloroethane; Q1, quartile 1; Q2, quartile 2; Q3, quartile 3; Q4, quartile 4; RR, rate ratio.

[a] Distant stage or poorly differentiated (after January 1, 2003, Gleason score 7–10) or Gleason score ≥7 or fatal (underlying cause: prostate cancer).

[b] Numbers do not sum to total because of missing data.

[c] Adjusted for age, state, race, family history of prostate cancer, smoking, fruit servings, and leisure-time physical activity in the winter.

provides the complete list of pesticides and their prevalence of use. Data were obtained from Agricultural Health Study data release versions P1REL201005.00 (for phase 1) and P2REL201007.00 (for phase 2).

Statistical analyses

We conducted analyses using unlagged exposure and 15-year lagged exposure, which excluded the most recent 15 years of exposure for both lifetime and intensity-weighted days. For each chemical, we categorized exposure into nonexposed and quartiles or tertiles of exposure on the basis of the distribution of exposed cases. This was done separately for total and aggressive prostate cancer. We used Poisson regression to calculate rate ratios and 95% confidence intervals and used the MIANALYZE procedure in SAS, version 9.2, software (SAS Institute, Inc., Cary, North Carolina) to obtain the appropriate variance when using phase 2 imputed data in the 95% confidence interval calculation. We evaluated only pesticides with 15 or more exposed cases of prostate cancer, thereby excluding trichlorfon and ziram. Rate ratios were adjusted for statistically significant ($\alpha = 0.05$) predictors of prostate cancer in the Agricultural Health Study. We evaluated several lifestyle and demographic measures and identified the following as potential confounding variables: age at enrollment (<40, 40–49, 50–59, 60–69, ≥70); race (white, black, other, missing); state (Iowa, North Carolina); family history of prostate cancer in first-degree relatives (yes, no, missing); cigarette smoking history (never, former, current, missing); fruit servings (<1/day, ≥1/day); and leisure-time physical activity in the winter (none, >0–2 hours/week, ≥3 hours/week). We further adjusted models for other pesticides shown to be associated with prostate cancer in the current analysis. Separate analyses were conducted by disease aggressiveness, family history of prostate cancer (yes, no), state, applicator type (private, commercial), age at enrollment (<65, ≥65), and for analyses of organochlorines with additional adjustment for body mass index. Likelihood ratio tests were

Table 4. Phase I and Phase II Data for Cumulative Lifetime Pesticide Exposure to Triazine Herbicides and Risk of Total and Aggressive Prostate Cancer in the Agricultural Health Study, 1993–2007

	Total Prostate Cancer			Aggressive Prostate Cancer[a]		
	No. of Cases[b]	RR[c]	95% CI	No. of Cases[b]	RR[c]	95% CI
Atrazine						
Nonexposed	507	1.00	Referent	228	1.00	Referent
Q1	336	0.97	0.84, 1.12	163	0.93	0.75, 1.16
Q2	335	1.05	0.91, 1.21	162	1.00	0.81, 1.24
Q3	336	0.97	0.84, 1.12	163	1.12	0.90, 1.39
Q4	335	0.98	0.85, 1.12	162	1.05	0.85, 1.30
P_{trend}			0.68			0.39
Cyanazine						
Nonexposed	1,015	1.00	Referent	462	1.00	Referent
Q1	169	0.90	0.76, 1.06	85	0.91	0.71, 1.16
Q2	169	0.99	0.83, 1.17	84	0.92	0.72, 1.17
Q3	169	0.87	0.73, 1.03	84	0.93	0.73, 1.18
Q4	168	0.94	0.79, 1.11	84	0.98	0.76, 1.25
P_{trend}			0.51			0.97

Abbreviations: CI, confidence interval; Q1, quartile 1; Q2, quartile 2; Q3, quartile 3; Q4, quartile 4; RR, rate ratio.

[a] Distant stage or poorly differentiated (after January 1, 2003, Gleason score 7–10) or Gleason score ≥7 or fatal (underlying cause: prostate cancer).

[b] Numbers do not sum to total because of missing data.

[c] Adjusted for age, state, race, family history of prostate cancer, smoking, fruit servings, and leisure-time physical activity in the winter.

used to assess differences between strata ($P_{interaction}$). We also analyzed phase 1 data only to assess the impact of the additional information collected or imputed from phase 2. All tests were 2 sided and conducted at the $\alpha = 0.05$ level. Tests for trend used the midpoint value of each exposure category treated as grouped linear in regression models.

RESULTS

The mean age at prostate cancer diagnosis for applicators with a family history of prostate cancer was younger (65.2 years) compared with aggressive cases (67.1 years) or overall prostate cancer (66.5 years) (Table 1).

Results were comparable (not shown) for both metrics (lifetime and intensity-weighted lifetime days) for both lagged and unlagged exposures. Therefore, we present rate ratios for unlagged intensity-weighted lifetime days only. The association between cumulative exposure to selected pesticides and risk of total and aggressive prostate cancer is presented in Tables 2–4. There was no significant association between any specific pesticide and risk of total prostate cancer. Four insecticides were, however, associated with aggressive prostate cancer: fonofos (rate ratio (RR) for the highest quartile of fonofos exposure (Q4) vs. nonexposed = 1.63, 95% CI: 1.22, 2.17; $P_{trend} < 0.0001$); aldrin (RR for aldrin Q4 vs. nonexposed = 1.49, 95% CI: 1.03, 2.18; $P_{trend} = 0.02$); malathion (RR for Q4 vs. nonexposed = 1.43, 95% CI: 1.08, 1.88; $P_{trend} = 0.04$); and terbufos (RR for Q4 vs. nonexposed = 1.29, 95% CI: 1.02, 1.64; $P_{trend} = 0.03$). The observed risk for each chemical persisted when they were analyzed together (simultaneous adjustment for fonofos, malathion, terbufos, and aldrin and aggressive prostate cancer). There was no association between the use of other organochlorine insecticides, triazine herbicides, or any other pesticides not presented and prostate cancer risk. Web Table 2 provides a list of rate ratios and 95% confidence intervals for the remainder of the 48 pesticides examined that are not presented in Tables 2–4. Results from analyses of phase 1 data only yielded similar results (data not shown).

Tables 5–7 show the association between pesticide exposure and total prostate cancer stratified by family history of prostate cancer. In the Agricultural Health Study, previous analyses suggested an increased risk of prostate cancer associated with selected pesticides for those with a family history of prostate cancer (13). Here, we observed significant interactions between family history of prostate cancer and the use of fonofos ($P_{interaction} = 0.04$) and aldrin ($P_{interaction} = 0.04$). A significantly increased risk of prostate cancer was also observed for men with exposure to lindane who had a family history of cancer, while there was no increased risk among men without a family history, although this interaction was not statistically significant ($P = 0.26$). We observed no other significant interactions between pesticide exposure and family history of prostate cancer. Web Table 3 provides a list of rate ratios and 95% confidence

intervals for the remainder of the 48 pesticides examined that are not presented in Tables 5–7.

Separate analyses by state, applicator type (private, commercial), age (<65, ≥65), and organochlorine models with additional adjustment for body mass index were not statistically significant and are therefore not shown. Results for alternative definitions of aggressive prostate cancer were similar to those presented and are therefore not shown. Limited statistical power precluded detailed analysis of family history of prostate cancer with exposures to fonofos, malathion, terbufos, or aldrin among those with aggressive prostate cancer.

DISCUSSION

In this analysis, we observed significant increases in the risk of aggressive prostate cancer associated with 4 insecticides: fonofos (organophosphate), malathion (organophosphate), terbufos (organophosphate), and aldrin (organochlorine). Further, we observed significant increases in risk of total prostate cancer with increasing use of fonofos and aldrin among those with a family history of prostate cancer but no increased risk among those without a family history. These findings are consistent with some findings from an earlier follow-up of these data from the Agricultural Health Study and offer new insights about risk of aggressive prostate cancer.

An earlier report from the Agricultural Health Study that included 566 prostate cancer cases occurring from enrollment until 1999 identified only the use of the fumigant methyl bromide to be significantly associated with prostate cancer risk (aggressive prostate cancer was not evaluated). This risk does not persist with additional follow-up (26), although methyl bromide use has declined from 1993–2005 because of a US Environmental Protection Agency phase-out. Here, we found the strongest associations for aggressive prostate cancer and use of fonofos, terbufos, malathion, and aldrin. Fonofos and terbufos have previously been associated with prostate cancer in earlier follow-up analyses in the Agricultural Health Study, although these associations were observed only among men with a family history of prostate cancer (14, 27). A recent Canadian prostate cancer case-control study reported no association with fonofos (5 exposed cases) but a significant increased risk with malathion (82 exposed cases) (1). Another study from California reported no risk associated with malathion (222 exposed cases) (9). We are not aware of other epidemiologic studies that have reported on the use of terbufos and prostate cancer risk. An association between aldrin and prostate cancer was observed previously in the Agricultural Health Study (13) but not after subsequent follow-up (28). Several occupational studies have implicated organochlorine insecticide use and prostate cancer risk (11, 13, 29–31); however, risk associated with specific organochlorine insecticides was less clear. None of these studies focused specifically on aggressive prostate cancer.

Fonofos (*O*-ethyl *S*-phenyl ethylphosphonodithioate), which as of 1998 is no longer registered for use in the United States (32), and terbufos (*S*-*tert*-butylthiomethyl *O*, *O*-diethyl phosphorodithioate) are classified by the US Environmental Protection Agency as group E for carcinogenicity (evidence of noncarcinogenicity for humans) (33). Organophosphate insecticides such as fonofos and terbufos are metabolized to their highly toxic oxon intermediate. The oxon form of the compound is more toxic than the parent compound and has been associated with a number of biologic endpoints including the generation of reactive oxygen species and DNA damage (34–36). Alternatively, these pesticides might impact other important cellular functions. In the Agricultural Health Study, we observed a significant interaction between terbufos and fonofos exposure and genetic variants on chromosome 8q24 and risk of prostate cancer (37). Recent studies have suggested that 8q24 variants might be related to the nearest coding region, the *MYC* gene, and its expression (38), suggesting that these pesticides might influence prostate cancer risk by altering important cancer signaling pathways involved in cellular adhesion, proliferation, and differentiation. The US Environmental Protection Agency concluded in 2000 that there was "suggestive" evidence of carcinogenicity for malathion, while the International Agency for Research on Cancer lists malathion in group 3, or not classifiable as to its carcinogenicity to humans. Like the other organophosphate insecticides, purported mechanisms of action include direct genotoxicity (of either malathion or malaoxon) (39, 40) and potential endocrine disruption (41, 42). The International Agency for Research on Cancer lists aldrin as not classifiable as to its carcinogenicity to humans (group 3). Organochlorine insecticides are putative endocrine disruptors that accumulate and persist in adipose tissue, providing a background of continuous endocrine perturbation that may increase prostate cancer risk (43, 44). Because these compounds are stored in fat, we additionally considered body mass index as an adjustment factor in these models (not shown). Body mass index was not a confounder or effect modifier of the relation between organochlorine insecticide use and prostate cancer in our study.

Of the 9 organophosphate insecticides evaluated for risk, 4 are dithioates: fonofos, malathion, phorate, and terbufos (http://www.alanwood.net/pesticides/class_insecticides.html), and we observed significant increased risks with 3 of the 4. Interestingly, a recent study reported another dithioate insecticide, azinphos-methyl, with an increased risk of prostate cancer (1). Although these pesticides might be similar with respect to their structure, there is still little information overall about their role in the carcinogenic process. Our observation for associations between these pesticides and aggressive prostate cancer suggests they may play a role in prostate cancer progression rather than at the earlier initiation stage of transformation. Future work on the mechanisms by which dithioate insecticides might impact prostate carcinogenesis would be valuable.

An alternative explanation for the lack of association between total prostate cancer and a positive association for aggressive cancer may be screening bias. It has been suggested that pesticide applicators would have lower prostate-specific antigen screening rates than the general population on account of greater variability in the availability of health insurance or access to care in rural areas (45, 46). This would result in a bias of risk estimates toward the null for

Table 5. Phase I and Phase II Data for Cumulative Lifetime Pesticide Exposure to Organophosphate Insecticides and Risk of Total Prostate Cancer by Family History of Prostate Cancer in the Agricultural Health Study, 1993–2007

	No Family History			Yes Family History		
	No. of Cases[a]	RR[b]	95% CI	No. of Cases[a]	RR[b]	95% CI
Chlorpyrifos						
Nonexposed	823	1.00	Referent	170	1.00	Referent
Q1	118	1.04	0.86, 1.27	32	1.20	0.81, 1.76
Q2	123	1.00	0.82, 1.21	30	1.08	0.73, 1.60
Q3	131	0.98	0.82, 1.18	24	0.77	0.50, 1.18
Q4	125	0.90	0.74, 1.09	30	0.86	0.58, 1.29
P_{trend}			0.24			0.32
$P_{interaction}$				0.81		
Coumaphos						
Nonexposed	1,187	1.00	Referent	235	1.00	Referent
Q1	26	1.09	0.73, 1.62	8	1.64	0.81, 3.33
Q2	19	0.60	0.39, 0.93	14	1.59	0.90, 2.82
Q3	25	0.84	0.57, 1.25	8	1.35	0.67, 2.75
Q4	24	0.92	0.61, 1.38	8	1.41	0.70, 2.87
P_{trend}			0.51			0.26
$P_{interaction}$				0.07		
Dichlorvos						
Nonexposed	1,185	1.00	Referent	240	1.00	Referent
Q1	31	1.02	0.71, 1.46	10	1.29	0.68, 2.44
Q2	31	1.00	0.70, 1.44	12	1.21	0.67, 2.18
Q3	36	0.93	0.67, 1.29	6	0.61	0.27, 1.37
Q4	29	0.77	0.53, 1.12	13	1.76	1.00, 3.09
P_{trend}			0.16			0.07
$P_{interaction}$				0.15		
Diazinon[c]						
Nonexposed	531	1.00	Referent	121	1.00	Referent
Q1	51	1.34	1.00, 1.79	11	1.15	0.62, 2.14
Q2	49	1.20	0.89, 1.61	9	0.93	0.46, 1.86
Q3	45	0.96	0.71, 1.31	15	1.26	0.72, 2.20
Q4	48	1.08	0.79, 1.47	8	0.88	0.42, 1.83
P_{trend}			0.78			0.82
$P_{interaction}$				0.84		
Fonofos						
Nonexposed	1,022	1.00	Referent	197	1.00	Referent
Q1	75	0.89	0.70, 1.12	18	0.91	0.55, 1.49
Q2	72	1.30	1.02, 1.65	20	1.70	1.07, 2.72
Q3	71	1.06	0.83, 1.36	18	1.22	0.74, 1.99
Q4	61	1.02	0.78, 1.32	30	2.01	1.36, 2.99
P_{trend}			0.70			0.0004
$P_{interaction}$				0.04		

Table continues

total prostate cancer and may explain the lack of association and/or smaller effect sizes observed for total prostate cancer in our study. Conversely, we would also have to consider whether the observed pesticide associations for

Table 5. Continued

	No Family History			Yes Family History		
	No. of Cases[a]	RR[b]	95% CI	No. of Cases[a]	RR[b]	95% CI
Malathion[c]						
Nonexposed	242	1.00	Referent	45	1.00	Referent
Q1	138	0.99	0.78, 1.25	44	1.37	0.87, 2.15
Q2	137	1.11	0.89, 1.37	34	1.12	0.72, 1.76
Q3	126	1.01	0.81, 1.26	35	1.23	0.79, 1.92
Q4	144	1.17	0.95, 1.44	21	0.70	0.42, 1.18
P_{trend}			0.15			0.15
$P_{interaction}$				0.15		
Parathion[c]						
Nonexposed	647	1.00	Referent	143	1.00	Referent
Q1	16	1.14	0.69, 1.87	5	1.32	0.54, 3.23
Q2	18	1.36	0.85, 2.19	5	1.54	0.63, 3.80
Q3	16	1.08	0.66, 1.79	6	1.58	0.65, 3.84
Q4	20	0.99	0.63, 1.55	3		
P_{trend}			0.98			0.88
$P_{interaction}$				0.51		
Phorate[c]						
Nonexposed	497	1.00	Referent	94	1.00	Referent
Q1	53	0.88	0.66, 1.18	21	1.39	0.85, 2.28
Q2	63	1.17	0.89, 1.54	9	0.71	0.35, 1.42
Q3	55	0.85	0.64, 1.13	18	0.99	0.59, 1.66
Q4	52	1.07	0.80, 1.43	21	1.53	0.94, 2.49
P_{trend}			0.73			0.12
$P_{interaction}$				0.15		
Terbufos						
Nonexposed	802	1.00	Referent	153	1.00	Referent
Q1	123	1.04	0.85, 1.26	34	1.34	0.90, 2.00
Q2	122	1.09	0.90, 1.32	29	1.12	0.74, 1.70
Q3	126	1.10	0.91, 1.33	29	1.09	0.73, 1.63
Q4	117	1.05	0.86, 1.27	36	1.27	0.88, 1.85
P_{trend}			0.57			0.30
$P_{interaction}$				0.72		

Abbreviations: CI, confidence interval; Q1, quartile 1; Q2, quartile 2; Q3, quartile 3; Q4, quartile 4; RR, rate ratio.
[a] Numbers do not sum to total because of missing data.
[b] Adjusted for age, state, race, smoking, fruit servings, and leisure-time physical activity in the winter.
[c] Detailed information for these chemicals was collected on the take-home questionnaire at enrollment.

aggressive prostate cancer reflect a true underlying risk factor that has increased the occurrence of more aggressive disease or whether this increase might be a result of decreased prostate-specific antigen screening. To explore this possibility, we calculated the prevalence of prostate-specific antigen screening in a subgroup of Agricultural Health Study men ($n = 23,265$) who provided this information from a follow-up questionnaire completed between 2005 and 2010. A large proportion of Agricultural Health Study men from Iowa (73.9%) and North Carolina (76.0%) reported having a prostate-specific antigen test within the past 5 years. This is similar to the proportion reported by the Behavioral Risk Factor Surveillance System data from Iowa (69.0%) and North Carolina (72.7%) (47). We additionally explored whether prostate-specific antigen screening might act as a confounder of the observed significant association and found no change in risk estimate with this additional adjustment. Taken together, this suggests that screening

Table 6. Phase I and Phase II Data for Cumulative Lifetime Pesticide Exposure to Organochlorine Insecticides and Risk of Total Prostate Cancer by Family History of Prostate Cancer in the Agricultural Health Study, 1993–2007

	No Family History			Yes Family History		
	No. of Cases[a]	RR[b]	95% CI	No. of Cases[a]	RR[b]	95% CI
Aldrin[c]						
Nonexposed	538	1.00	Referent	95	1.00	Referent
Q1	50	0.99	0.74, 1.33	12	1.29	0.70, 2.40
Q2	38	0.72	0.51, 1.00	20	1.95	1.17, 3.25
Q3	45	1.06	0.78, 1.45	17	1.83	1.08, 3.09
Q4	45	1.13	0.83, 1.54	16	2.13	1.22, 3.72
P_{trend}			0.42			0.005
$P_{interaction}$				0.04		
Chlordane[c]						
Nonexposed	544	1.00	Referent	118	1.00	Referent
Q1	39	0.70	0.50, 0.96	15	1.15	0.67, 1.98
Q2	45	1.35	0.99, 1.83	11	1.33	0.71, 2.47
Q3	45	1.01	0.75, 1.38	8	0.72	0.35, 1.48
Q4	43	0.99	0.72, 1.36	9	1.12	0.57, 2.23
P_{trend}			0.88			0.91
$P_{interaction}$				0.52		
DDT[c]						
Nonexposed	421	1.00	Referent	93	1.00	Referent
Q1	73	0.96	0.74, 1.24	17	1.08	0.63, 1.85
Q2	76	1.37	1.06, 1.76	17	1.43	0.83, 2.44
Q3	70	1.25	0.97, 1.62	15	1.43	0.81, 2.51
Q4	67	1.22	0.93, 1.59	15	1.04	0.58, 1.83
P_{trend}			0.15			0.98
$P_{interaction}$				0.76		
Dieldrin[c]						
Nonexposed	675	1.00	Referent	148	1.00	Referent
T1	15	0.90	0.54, 1.51	4		
T2	13	0.73	0.42, 1.26	5	1.55	0.63, 3.82
T3	13	0.90	0.52, 1.56	5	1.54	0.62, 3.83
P_{trend}			0.56			0.29
$P_{interaction}$				0.69		
Heptachlor[c]						
Nonexposed	592	1.00	Referent	132	1.00	Referent
Q1	37	1.20	0.86, 1.69	7	0.81	0.37, 1.75
Q2	35	1.11	0.78, 1.57	7	0.83	0.39, 1.80
Q3	31	0.94	0.65, 1.36	11	1.17	0.63, 2.21
Q4	32	1.01	0.70, 1.44	8	0.91	0.44, 1.88
P_{trend}			0.93			0.91
$P_{interaction}$				0.73		

Table continues

bias is not likely an issue in the Agricultural Health Study and that pesticide exposure may truly increase aggressive prostate cancer risk.

We also observed an association between fonofos and aldrin use and risk of total prostate cancer that was modified by family history of prostate cancer. This is consistent

Table 6. Continued

	No Family History			Yes Family History		
	No. of Cases[a]	RR[b]	95% CI	No. of Cases[a]	RR[b]	95% CI
Lindane[c]						
Nonexposed	622	1.00	Referent	127	1.00	Referent
Q1	30	0.87	0.59, 1.27	11	1.02	0.52, 2.01
Q2	30	1.20	0.82, 1.75	6	0.97	0.45, 2.08
Q3	27	0.98	0.66, 1.45	10	1.48	0.77, 2.84
Q4	25	0.95	0.64, 1.42	10	2.17	1.13, 4.17
P_{trend}			0.84			0.01
$P_{interaction}$				0.26		
Toxaphene[c]						
Nonexposed	617	1.00	Referent	137	1.00	Referent
Q1	23	0.71	0.47, 1.09	10	1.18	0.62, 2.24
Q2	30	1.14	0.80, 1.64	7	1.17	0.54, 2.51
Q3	25	1.20	0.80, 1.80	7	1.36	0.63, 2.94
Q4	27	0.92	0.62, 1.36	6	1.22	0.52, 2.84
P_{trend}			0.82			0.57
$P_{interaction}$				0.96		

Abbreviations: CI, confidence interval; DDT, dichlorodiphenyltrichloroethane; Q1, quartile 1; Q2, quartile 2; Q3, quartile 3; Q4, quartile 4; RR, rate ratio; T1, tertile 1; T2, tertile 2; T3, tertile 3.

[a] Numbers do not sum to total because of missing data.

[b] Adjusted for age, state, race, smoking, fruit servings, and leisure-time physical activity in the winter.

[c] Detailed information for these chemicals was collected on the take-home questionnaire at enrollment.

Table 7. Phase I and Phase II Data for Cumulative Lifetime Pesticide Exposure to Triazine Herbicides and Risk of Total Prostate Cancer by Family History of Prostate Cancer in the Agricultural Health Study, 1993–2007

	No Family History			Yes Family History		
	No. of Cases[a]	RR[b]	95% CI	No. of Cases[a]	RR[b]	95% CI
Atrazine						
Nonexposed	375	1.00	Referent	54	1.00	Referent
Q1	242	0.94	0.80, 1.12	53	1.07	0.72, 1.58
Q2	244	0.98	0.83, 1.16	57	1.25	0.85, 1.84
Q3	236	0.90	0.76, 1.07	67	1.26	0.87, 1.83
Q4	250	0.96	0.81, 1.13	65	1.27	0.88, 1.83
P_{trend}			0.73			0.29
$P_{interaction}$				0.64		
Cyanazine						
Nonexposed	788	1.00	Referent	150	1.00	Referent
Q1	128	0.87	0.71, 1.06	30	0.89	0.59, 1.34
Q2	129	0.98	0.81, 1.19	30	0.96	0.64, 1.46
Q3	132	0.91	0.75, 1.10	31	0.90	0.60, 1.35
Q4	125	0.90	0.74, 1.10	40	1.23	0.85, 1.77
P_{trend}			0.37			0.21
$P_{interaction}$				0.67		

Abbreviations: CI, confidence interval; Q1, quartile 1; Q2, quartile 2; Q3, quartile 3; Q4, quartile 4; RR, rate ratio.

[a] Numbers do not sum to total because of missing data.

[b] Adjusted for age, state, race, smoking, fruit servings, and leisure-time physical activity in the winter.

with the observed effect modification by family history for fonofos within the Agricultural Health Study (13, 14) and provides new information about potential effect modification from a family history of prostate cancer among individuals with exposure to aldrin. These observations suggest that selected insecticides may interact with genetic determinants or that nongenetic factors that track in families might account for the observed association.

Our study is able to address several limitations common in other studies of pesticide use and prostate cancer. It included a large number of prostate cancer cases with exposure to pesticides and detailed information on use of specific pesticides that was available at 2 points in time. We also provided risk estimates, for the first time, for specific pesticides and clinically significant prostate cancer. Some limitations of our study should also be acknowledged. For example, information on the Gleason score was missing for 30% of the cases in North Carolina, which most likely led to an underestimation of advanced cases from this state. If these underestimated cases were more likely to have high exposure to the observed chemicals with an association for prostate cancer, the true risk may be higher than we observed here. Furthermore, Gleason scores were not standardized by centralized pathologic review. Moreover, because detailed information on some pesticides was collected only from the take-home questionnaire, missing data on these chemicals could introduce selection bias. We believe this is unlikely however, since individuals completing the take-home questionnaire were comparable to nonrespondents (48). In addition, although information on pesticide use provided by farmers in the Agricultural Health Study is quite reliable (49, 50), exposure misclassification undoubtedly occurred. In a prospective study such as the Agricultural Health Study, such misclassification is likely to be nondifferential and would tend to bias relative risks toward the null and diminish any "real" exposure-response gradients (51). Finally, given the large number of pesticides examined, we cannot rule out the possibility that some of our findings might be due to chance.

In conclusion, we observed significant increases in the risk of aggressive prostate cancer associated with 4 insecticides: fonofos (organophosphate), malathion (organophosphate), terbufos (organophosphate), and aldrin (organochlorine). This is the first time specific pesticides have been studied as risk factors for aggressive prostate cancer. These pesticide-specific findings need to be supported by mechanistic studies where there is still limited information about how pesticides impact carcinogenesis. Future follow-up in the Agricultural Health Study to further evaluate the relation between pesticides and aggressive prostate cancer is anticipated.

ACKNOWLEDGMENTS

Author affiliations: Division of Cancer Epidemiology and Genetics, National Cancer Institute, National Institutes of Health, Department of Health and Human Services, Bethesda, Maryland (Stella Koutros, Laura E. Beane Freeman, Jay H. Lubin, Gabriella Andreotti, Kathryn Hughes Barry, Curt T. DellaValle, Aaron Blair, Michael C. R. Alavanja,); Epidemiology Branch, National Institute of Environmental Health Sciences, National Institutes of Health, Department of Health and Human Services, Research Triangle Park, North Carolina (Jane A. Hoppin, Dale P. Sandler); Department of Pediatrics, School of Medicine, University of Washington, Seattle, Washington (Sonya L. Heltshe); and College of Public Health, University of Iowa, Iowa City, Iowa (Charles F. Lynch).

This work was supported by the Intramural Research Program of the National Institutes of Health; the Division of Cancer Epidemiology and Genetics, National Cancer Institute (Z01CP010119); and the National Institute of Environmental Health Sciences (Z01ES0490300).

Conflict of interest: none declared.

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Web Table 1. List and Prevalence of 48 Chemicals Examined for Risk of Total Prostate Cancer

Pesticide	Type	Prevalence of use (% Ever)		Quartile Medians-Total Prostate Cancer[b]			
		Prostate Cases n (%)	Rest of Cohort[a] n (%)	Q1	Q2	Q3	Q4
Atrazine	Herbicide (TR)	1342 (68.4)	36,752 (70.1)	12.0	48.8	108.5	336.0
Dicamba	Herbicide	838 (42.7)	25,516 (48.7)	8.8	24.5	58.8	224.8
Cyanazine	Herbicide (TR)	675 (34.4)	19,542 (37.3)	8.8	24.5	56.0	224.8
Chlorimuron-ethyl	Herbicide	303 (15.4)	8,238 (15.7)	8.8	30.0		
Metolachlor	Herbicide	768 (39.1)	23,757 (45.3)	8.8	24.5	56.0	224.8
EPTC	Herbicide	300 (15.3)	9,552 (18.2)	8.8	56.0		
Alachlor	Herbicide	917 (46.7)	24,548 (46.8)	8.8	24.5	86.0	236.0
Metribuzin	Herbicide	372 (19.0)	8,750 (16.7)	8.8	20.0	24.5	103.3
Paraquat	Herbicide	178 (9.1)	4,453 (8.5)	2.5	8.8	24.5	108.5
Petroleum Oil	Herbicide	206 (10.5)	4,838 (9.2)	8.8	24.5	56.0	236.0
Pendimethalin	Herbicide	408 (20.1)	12,323 (23.5)	7.0	15.0	24.5	105.0
Imazethapyr	Herbicide	642 (32.7)	20,810 (39.7)	8.8	24.5	56.0	
Glyphosate	Herbicide	1464 (74.6)	42,420 (80.9)	8.8	24.0	56.0	224.8
2,4,5 T P	Herbicide	51 (2.6)	1,061 (2.0)	8.8	24.5	108.5	
Butylate	Herbicide	241 (12.3)	5,764 (11.0)	8.8	24.5	56.0	116.0
Trifluralin	Herbicide	893 (45.5)	25,245 (48.1)	14.5	50.8	108.5	236.0
2,4-D	Herbicide	1469 (74.9)	39,677 (75.7)	10.0	50.8	118.5	396.0
2,4,5 T	Herbicide	245 (12.5)	3,860 (7.4)	8.8	50.8		
Permethrin (crop)	Insecticide	196 (10.0)	7,587 (14.5)	8.8	55.7		
Permethrin (animal)	Insecticide	177 (9.0)	6,540 (12.5)	8.8	56.0		
Terbufos	Insecticide (OP)	639 (32.6)	17,838 (34.0)	12.0	48.8	108.5	336.0
Fonofos	Insecticide (OP)	384 (19.6)	9,681 (18.5)	8.8	24.5	50.8	116.0
Lindane	Insecticide (OC)	157 (8.0)	3,215 (6.1)	8.8	20.0	24.5	108.0
Carbofuran	Insecticide	534 (27.2)	12,292 (23.4)	8.8	24.5	108.5	
Chlorpyrifos	Insecticide (OP)	668 (34.1)	20,233 (38.6)	8.8	24.0	50.8	116.0
Malathion	Insecticide (OP)	746 (38.1)	17,212 (32.8)	8.8	20.0	38.8	116.5
Parathion	Insecticide (OP)	99 (5.1)	1,592 (3.0)	8.8	24.5	116.0	
Carbaryl	Insecticide	558 (28.4)	11,601 (22.1)	8.8	20.0	45.0	175.0
Diazinon	Insecticide (OP)	258 (13.2)	5,626 (10.7)	8.8	38.8		
Aldicarb	Insecticide	92 (4.7)	2,315 (4.4)	8.0	24.5	103.3	
Phorate	Insecticide (OP)	304 (15.5)	6,418 (12.2)	8.8	24.5	56.0	116.0
Aldrin	Insecticide (OC)	257 (13.1)	3,315 (6.3)	8.8	24.5	50.8	103.3
Chlordane	Insecticide (OC)	233 (11.9)	3,917 (7.5)	8.8	24.5		
Dieldrin	Insecticide (OC)	56 (2.9)	725 (1.4)	8.8	24.5		
DDT	Insecticide (OC)	384 (19.6)	4,332 (8.3)	8.8	24.5	116.0	
Heptachlor	Insecticide (OC)	178 (9.1)	2,402 (4.6)	8.8	24.5	56.0	
Toxaphene	Insecticide (OC)	153 (7.8)	2,319 (4.4)	8.8	24.5	108.5	
Coumaphos	Insecticide (OP)	139 (7.1)	3,614 (6.9)	8.8	20.0	38.8	176.5
DDVP	Insecticide (OP)	172 (8.8)	4,563 (8.7)	8.8	24.5	103.3	752.3
Methyl Bromide	Fumigant	281 (14.3)	7,374 (14.1)	3.5	15.5	35.0	122.5
Aluminum Phosphide	Fumigant	30 (1.5)	1,271 (2.4)	3.5	24.5		
Mix 80/20	Fumigant	72 (3.4)	877 (1.7)	3.5	12.3	54.3	
Ethylene Dibromide	Fumigant	37 (1.9)	929 (1.8)	3.5	15.5	87.5	
Benomyl	Fungicide	82 (4.2)	1,790 (3.4)	3.5	24.5	108.5	
Chlorothalonil	Fungicide	142 (7.2)	4,395 (8.4)	7.9	28.0	64.0	200.0
Captan	Fungicide	170 (8.7)	4,879 (9.3)	0.3	7.8	64.0	
Maneb/Mancozeb	Fungicide	79 (4.0)	1,720 (3.3)	7.0	30.0	224.8	
Metalaxyl	Fungicide	197 (10.0)	4,884 (9.3)	1.0	12.3	25.0	59.3

Abbreviations: Triazine (TR); Organophosphate (OP); Organochlorine (OC); quartile 1 (Q1); quartile 2 (Q2); quartile 3 (Q3); quartile 4 (Q4).

[a] Male applicators with no previous history of cancer and complete follow-up.

[b] Tertile cutpoints or median cutpoints provided for some chemicals.

Web Table 2. Cumulative Lifetime Pesticide Exposure and Risk of Total and Aggressive Prostate Cancer in the AHS, 2007

	Intensity Weighted Days		Intensity Weighted Days	
	Total PCA		Aggressive PCA[a]	
	Cases[b]	RR[c] (95% CI)	Cases[b]	RR[c] (95% CI)
Dicamba				
Nonexposed	837	Ref	380	Ref
Q1	212	0.97 (0.82, 1.14)	102	0.85 (0.67, 1.07)
Q2	208	0.89 (0.75, 1.05)	99	0.92 (0.72, 1.16)
Q3	209	0.90 (0.76, 1.06)	100	0.82 (0.64, 1.04)
Q4	209	1.04 (0.88, 1.22)	100	0.96 (0.75, 1.22)
p-trend		0.50		0.98
Chlorimuron				
Nonexposed	718	Ref	348	Ref
Q1	76	1.01 (0.79, 1.29)	32	0.90 (0.62, 1.31)
Q2	76	1.09 (0.86, 1.39)	31	1.20 (0.83, 1.74)
Q3	76	1.02 (0.80, 1.32)	31	0.80 (0.53, 1.21)
Q4	75	0.89 (0.70, 1.13)	31	0.74 (0.51, 1.08)
p-trend		0.36		0.10
Metolachlor				
Nonexposed	910	Ref	427	Ref
Q1	192	1.04 (0.88, 1.23)	93	1.03 (0.81, 1.31)
Q2	192	1.05 (0.89, 1.23)	89	1.00 (0.79, 1.26)
Q3	192	0.98 (0.83, 1.16)	91	0.95 (0.74, 1.20)
Q4	192	0.91 (0.78, 1.07)	91	0.98 (0.78, 1.24)
p-trend		0.21		0.81
EPTC				
Nonexposed	1352	Ref	624	Ref
Q1	75	1.00 (0.79, 1.26)	39	1.01 (0.72, 1.41)
Q2	76	1.25 (0.99, 1.58)	37	1.29 (0.93, 1.81)
Q3	74	0.93 (0.73, 1.17)	38	0.97 (0.70, 1.35)
Q4	75	0.93 (0.73, 1.17)	38	1.01 (0.73, 1.41)
p-trend		0.48		0.98
Alachlor				
Nonexposed	745	Ref	362	Ref
Q1	230	0.96 (0.83, 1.12)	104	0.89 (0.71, 1.12)
Q2	231	1.03 (0.89, 1.20)	104	0.95 (0.76, 1.18)
Q3	227	0.99 (0.85, 1.15)	104	0.96 (0.77, 1.20)
Q4	229	0.99 (0.86, 1.15)	103	0.90 (0.73, 1.13)
p-trend		0.96		0.49
Metribuzin				
Nonexposed	633	Ref	295	Ref
Q1	93	0.97 (0.77, 1.21)	45	0.86 (0.62, 1.19)
Q2	93	1.17 (0.93, 1.47)	42	1.03 (0.73, 1.45)
Q3	93	0.94 (0.75, 1.17)	44	0.99 (0.71, 1.38)
Q4	93	1.08 (0.87, 1.35)	43	1.11 (0.80, 1.54)
p-trend		0.57		0.46
Paraquat				
Nonexposed	844	Ref	391	Ref
Q1	45	0.96 (0.71, 1.31)	21	0.92 (0.57, 1.49)
Q2	44	0.97 (0.71, 1.33)	20	0.93 (0.58, 1.49)
Q3	45	0.93 (0.68, 1.27)	21	1.32 (0.83, 2.08)
Q4	44	1.03 (0.75, 1.42)	20	1.30 (0.80, 2.10)
p-trend		0.88		0.22
Petroleum Oil				

Nonexposed	796	Ref	364	Ref
Q1	52	0.96 (0.71, 1.28)	27	0.87 (0.57, 1.31)
Q2	52	1.17 (0.88, 1.55)	26	1.34 (0.89, 2.03)
Q3	51	1.05 (0.79, 1.39)	26	1.30 (0.87, 1.96)
Q4	51	1.14 (0.86, 1.52)	26	1.30 (0.87, 1.93)
p-trend		0.34		0.14
Pendimethalin				
Nonexposed	649	Ref	305	Ref
Q1	102	0.84 (0.68, 1.05)	50	0.80 (0.59, 1.10)
Q2	102	0.92 (0.74, 1.15)	46	1.02 (0.74, 1.42)
Q3	102	0.90 (0.72, 1.13)	48	0.97 (0.70, 1.34)
Q4	102	1.16 (0.93, 1.43)	48	1.32 (0.97, 1.80)
p-trend		0.15		0.06
Imazethapyr				
Nonexposed	1019	Ref	470	Ref
Q1	161	1.00 (0.84, 1.20)	78	0.95 (0.73, 1.23)
Q2	160	1.06 (0.88, 1.26)	77	0.89 (0.69, 1.16)
Q3	161	1.11 (0.92, 1.32)	77	0.97 (0.75, 1.25)
Q4	160	1.00 (0.84, 1.20)	77	1.08 (0.84, 1.38)
p-trend		0.86		0.51
Glyphosate				
Nonexposed	385	Ref	188	Ref
Q1	366	0.91 (0.79, 1.06)	170	0.93 (0.74, 1.16)
Q2	366	0.96 (0.83, 1.12)	169	0.91 (0.73, 1.13)
Q3	366	1.01 (0.87, 1.17)	170	1.01 (0.82, 1.25)
Q4	366	0.99 (0.86, 1.15)	169	0.94 (0.75, 1.18)
2,4,5-TP				
Nonexposed	939	Ref	434	Ref
Q1	17	0.73 (0.44, 1.21)	8	0.93 (0.44, 1.95)
Q2	18	1.14 (0.72, 1.79)	8	1.49 (0.74, 3.01)
Q3	16	0.83 (0.52, 1.35)	8	1.04 (0.51, 2.09)
Q4			7	1.31 (0.62, 2.77)
p-trend		0.50		0.46
Butylate				
Nonexposed	756	Ref	348	Ref
Q1	62	0.74 (0.57, 0.96)	30	0.71 (0.48, 1.05)
Q2	61	0.90 (0.69, 1.17)	30	1.03 (0.71, 1.51)
Q3	58	1.24 (0.95, 1.63)	30	1.38 (0.94, 2.02)
Q4	60	1.20 (0.92, 1.56)	29	1.28 (0.87, 1.87)
p-trend		0.08		0.08
Trifluralin				
Nonexposed	784	Ref	357	Ref
Q1	224	0.94 (0.81, 1.09)	109	0.95 (0.76, 1.19)
Q2	223	1.05 (0.89, 1.22)	107	1.04 (0.83, 1.30)
Q3	223	0.98 (0.84, 1.14)	108	1.05 (0.84, 1.31)
Q4	223	0.97 (0.83, 1.13)	108	0.99 (0.79, 1.24)
p-trend		0.78		0.96
2,4-D				
Nonexposed	392	Ref	186	Ref
Q1	369	0.99 (0.85, 1.15)	173	0.95 (0.76, 1.18)
Q2	366	0.97 (0.83, 1.14)	173	0.85 (0.67, 1.07)
Q3	367	1.01 (0.87, 1.18)	173	0.88 (0.71, 1.10)
Q4	367	0.95 (0.82, 1.11)	172	0.96 (0.76, 1.20)
p-trend		0.52		0.79

2,4,5-T				
Nonexposed	744	Ref	343	Ref
Q1	62	1.32 (1.02, 1.71)	31	1.15 (0.78, 1.70)
Q2	62	1.09 (0.84, 1.42)	31	1.20 (0.81, 1.77)
Q3	60	0.96 (0.73, 1.25)	29	1.00 (0.68, 1.46)
Q4	61	0.82 (0.63, 1.06)	30	0.87 (0.59, 1.26)
p-trend		0.10		0.42
Permethrin crop				
Nonexposed	1468	Ref	693	Ref
Q1	49	0.93 (0.69, 1.23)	22	0.95 (0.60, 1.49)
Q2	49	1.07 (0.80, 1.43)	22	1.07 (0.69, 1.66)
Q3	49	0.86 (0.64, 1.16)	22	0.65 (0.41, 1.03)
Q4	49	1.05 (0.79, 1.40)	21	1.33 (0.86, 2.05)
p-trend		0.85		0.38
Permethrin animal				
Nonexposed	1529	Ref	709	Ref
Q1	45	0.98 (0.69, 1.37)	24	1.07 (0.67, 1.70)
Q2	44	1.16 (0.83, 1.63)	23	1.11 (0.71, 1.74)
Q3	44	1.11 (0.81, 1.51)	24	1.09 (0.72, 1.64)
Q4	44	0.84 (0.62, 1.13)	23	1.11 (0.73, 1.68)
p-trend		0.32		0.58
Carbofuran				
Nonexposed	1128	Ref	508	Ref
Q1	140	0.97 (0.81, 1.16)	69	1.11 (0.85, 1.44)
Q2	127	1.23 (1.02, 1.47)	66	1.11 (0.87, 1.43)
Q3	134	1.13 (0.94, 1.36)	64	1.13 (0.86, 1.47)
Q4	133	0.88 (0.74, 1.05)	66	1.22 (0.95, 1.58)
p-trend		0.24		0.12
Carbaryl				
Nonexposed	483	Ref	237	Ref
Q1	140	1.04 (0.86, 1.26)	67	1.05 (0.78, 1.42)
Q2	139	1.13 (0.92, 1.38)	60	1.03 (0.76, 1.40)
Q3	140	1.25 (1.02, 1.53)	64	1.19 (0.88, 1.60)
Q4	139	0.91 (0.73, 1.13)	63	0.99 (0.71, 1.39)
p-trend		0.18		0.85
Aldicarb				
Nonexposed	913	Ref	436	Ref
Q1	23	0.95 (0.62, 1.44)	10	0.87 (0.39, 1.93)
Q2	23	1.73 (1.13, 2.65)	10	2.99 (1.52, 5.87)
Q3	23	0.98 (0.63, 1.53)	10	1.06 (0.50, 2.27)
Q4	23	1.00 (0.64, 1.56)	9	0.72 (0.33, 1.57)
p-trend		0.97		0.47
Methyl bromide				
Nonexposed	1570	Ref	750	Ref
Q1	72	0.94 (0.73, 1.20)	30	1.17 (0.80, 1.71)
Q2	69	0.90 (0.70, 1.16)	30	0.79 (0.53, 1.18)
Q3	70	0.94 (0.73, 1.21)	30	1.10 (0.74, 1.63)
Q4	70	0.94 (0.73, 1.21)	29	0.93 (0.63, 1.38)
p-trend		0.66		0.78
Aluminum Phosphide				
Nonexposed	959	Ref	445	Ref
T1	10	1.07 (0.57, 1.99)	10	1.58 (0.84, 2.96)
T2	11	0.64 (0.35, 1.17)	8	0.78 (0.39, 1.57)
T3	9	0.85 (0.44, 1.65)		

p-trend		0.49		0.53
Mix 80/20				
Nonexposed	912	Ref	425	Ref
Q1	25	1.73 (1.16, 2.58)	10	1.71 (0.88, 3.32)
Q2	24	1.15 (0.76, 1.73)	9	1.54 (0.79, 2.99)
Q3	23	1.05 (0.69, 1.59)	9	0.84 (0.42, 1.69)
Q4			9	1.44 (0.74, 2.79)
p-trend		0.79		0.33
Ethylene Dibromide				
Nonexposed	953	Ref	447	Ref
T1	13	1.16 (0.67, 2.01)	7	0.89 (0.39, 2.00)
T2	13	1.18 (0.68, 2.05)	7	0.56 (0.26, 1.20)
T3	11	0.44 (0.24, 0.81)		
p-trend		0.009		0.13
Benomyl				
Nonexposed	904	Ref	424	Ref
Q1	21	1.06 (0.69, 1.64)	10	1.33 (0.71, 2.50)
Q2	20	1.00 (0.64, 1.57)	9	0.93 (0.47, 1.81)
Q3	21	1.36 (0.88, 2.12)	10	1.20 (0.61, 2.35)
Q4	20	0.71 (0.45, 1.11)	9	1.18 (0.62, 2.24)
p-trend		0.19		0.59
Chlorothalonil				
Nonexposed	1720	Ref	797	Ref
Q1	37	0.87 (0.61, 1.24)	18	1.09 (0.63, 1.89)
Q2	34	1.25 (0.88, 1.76)	17	1.82 (1.09, 3.03)
Q3	36	0.86 (0.62, 1.21)	17	1.00 (0.61, 1.65)
Q4	35	0.88 (0.62, 1.23)	17	1.09 (0.67, 1.79)
p-trend		0.39		0.72
Captan				
Nonexposed	1508	Ref	692	Ref
Q1	44	1.07 (0.78, 1.45)	23	0.86 (0.57, 1.32)
Q2	41	1.05 (0.77, 1.44)	23	1.90 (1.21, 2.98)
Q3	43	1.10 (0.81, 1.49)	23	1.39 (0.91, 2.12)
Q4	42	0.96 (0.70, 1.33)	23	1.35 (0.87, 2.08)
p-trend		0.86		0.14
Maneb/Mancozeb				
Nonexposed	907	Ref	425	Ref
Q1	20	0.86 (0.54, 1.36)	11	1.14 (0.62, 2.10)
Q2	20	0.89 (0.55, 1.42)	8	1.10 (0.54, 2.23)
Q3	20	0.84 (0.54, 1.31)	9	1.59 (0.81, 3.14)
Q4	19	0.83 (0.52, 1.32)	9	0.67 (0.34, 1.31)
p-trend		0.41		0.29
Metalaxyl				
Nonexposed	791	Ref	379	Ref
Q1	50	0.96 (0.72, 1.28)	23	1.04 (0.68, 1.58)
Q2	49	1.01 (0.74, 1.37)	22	1.04 (0.65, 1.66)
Q3	49	1.11 (0.82, 1.51)	22	1.19 (0.75, 1.88)
Q4	49	1.03 (0.76, 1.40)	22	1.13 (0.70, 1.82)
p-trend		0.78		0.59

Abbreviations: Agricultural Health Study (AHS); Prostate Cancer (PCA); quartile 1 (Q1); quartile 2 (Q2); quartile 3 (Q3); quartile 4 (Q4).

[a]Distant Stage OR Poorly differentiated (after 1/1/2003 Gleason 7-10) OR Gleason ≥7 OR Fatal (underlying cause-prostate cancer)

[b]Numbers do not sum to total due to missing data.

[c]Adjusted for age, state, race, family history of prostate cancer, smoking, fruit servings, and leisure time physical activity in the winter

Web Table 3. Cumulative Lifetime Pesticide Exposure and Risk of Total Prostate Cancer by Family History of Prostate Cancer in the Agricultural Health Study

	Intensity Weighted Days		Intensity Weighted Days		
FAMILY HISTORY	NO		YES		
	Cases[a]	RR[b] (95% CI)	Cases[a]	RR[b] (95% CI)	p-interaction
Dicamba					
Nonexposed	638	Ref	122	Ref	
Q1	163	0.97 (0.81, 1.17)	42	1.17 (0.80, 1.71)	
Q2	163	0.93 (0.76, 1.13)	37	0.90 (0.60, 1.33)	
Q3	154	0.88 (0.73, 1.07)	44	1.02 (0.70, 1.49)	
Q4	160	1.05 (0.87, 1.27)	42	1.20 (0.82, 1.75)	
p-trend		0.55		0.37	0.22
Chlorimuron					
Nonexposed	530	Ref	114	Ref	
Q1	58	0.99 (0.75, 1.32)	17	1.29 (0.77, 2.15)	
Q2	53	0.98 (0.74, 1.30)	18	1.56 (0.92, 2.62)	
Q3	56	1.02 (0.76, 1.37)	9	0.65 (0.32, 1.32)	
Q4	54	0.90 (0.68, 1.20)	11	0.79 (0.43, 1.48)	
p-trend		0.51		0.32	0.11
Metolachlor					
Nonexposed	716	Ref	134	Ref	
Q1	137	0.94 (0.78, 1.13)	40	1.43 (0.99, 2.08)	
Q2	151	1.05 (0.87, 1.25)	30	0.99 (0.65, 1.51)	
Q3	140	0.92 (0.76, 1.10)	41	1.15 (0.78, 1.69)	
Q4	144	0.86 (0.72, 1.03)	37	1.04 (0.72, 1.52)	
p-trend		0.10		0.96	0.53
EPTC					
Nonexposed	1064	Ref	207	Ref	
Q1	60	1.03 (0.79, 1.34)	14	0.95 (0.55, 1.64)	
Q2	54	1.16 (0.88, 1.53)	21	1.70 (1.08, 2.68)	
Q3	50	0.86 (0.65, 1.15)	20	1.12 (0.70, 1.79)	
Q4	50	0.81 (0.61, 1.08)	20	1.28 (0.80, 2.03)	
p-trend		0.12		0.29	0.22
Alachlor					
Nonexposed	587	Ref	105	Ref	
Q1	172	0.92 (0.77, 1.09)	45	1.21 (0.85, 1.73)	
Q2	175	1.01 (0.85, 1.20)	47	1.26 (0.89, 1.79)	
Q3	186	1.04 (0.88, 1.22)	30	0.82 (0.54, 1.24)	
Q4	165	0.92 (0.77, 1.09)	49	1.38 (0.98, 1.94)	
p-trend		0.46		0.16	0.11
Metribuzin					
Nonexposed	469	Ref	87	Ref	
Q1	67	0.90 (0.69, 1.18)	18	1.05 (0.62, 1.78)	
Q2	65	1.07 (0.82, 1.40)	24	1.61 (1.00, 2.58)	
Q3	70	0.94 (0.73, 1.22)	18	0.88 (0.51, 1.49)	
Q4	69	1.03 (0.79, 1.33)	20	1.43 (0.87, 2.37)	
p-trend		0.85		0.27	0.33
Paraquat					
Nonexposed	618	Ref	146	Ref	
Q1	37	1.08 (0.77, 1.52)	5	0.62 (0.25, 1.52)	
Q2	31	0.93 (0.64, 1.35)	4	**	
Q3	28	0.83 (0.56, 1.24)	9	1.13 (0.56, 2.28)	
Q4	32	0.96 (0.65, 1.40)	6	1.13 (0.48, 2.66)	
p-trend		0.74		0.67	0.59

Petroleum Oil					
Nonexposed	587	Ref	124	Ref	
Q1	34	0.81 (0.56, 1.16)	13	1.39 (0.78, 2.48)	
Q2	40	1.15 (0.83, 1.59)	10	1.31 (0.68, 2.51)	
Q3	38	1.02 (0.73, 1.41)	10	1.14 (0.60, 2.18)	
Q4	36	1.11 (0.80, 1.56)	9	0.97 (0.49, 1.91)	
p-trend		0.49		0.89	0.66
Pendimethalin					
Nonexposed	483	Ref	101	Ref	
Q1	76	0.86 (0.67, 1.10)	22	0.92 (0.54, 1.55)	
Q2	73	0.87 (0.67, 1.13)	21	1.19 (0.73, 1.94)	
Q3	76	0.87 (0.66, 1.14)	13	0.70 (0.38, 1.28)	
Q4	69	1.03 (0.80, 1.33)	21	1.61 (1.00, 2.60)	
p-trend		0.78		0.07	0.62
Imazethapyr					
Nonexposed	796	Ref	153	Ref	
Q1	122	0.97 (0.79, 1.19)	32	1.09 (0.72, 1.65)	
Q2	127	1.07 (0.87, 1.31)	26	0.92 (0.59, 1.43)	
Q3	112	1.04 (0.84, 1.28)	41	1.26 (0.87, 1.84)	
Q4	127	1.01 (0.82, 1.23)	27	0.91 (0.58, 1.43)	
p-trend		0.90		0.81	0.99
Glyphosate					
Nonexposed	280	Ref	48	Ref	
Q1	255	0.89 (0.75, 1.06)	61	1.00 (0.65, 1.53)	
Q2	251	0.93 (0.77, 1.12)	65	1.01 (0.68, 1.50)	
Q3	270	1.01 (0.85, 1.20)	61	1.00 (0.68, 1.48)	
Q4	280	1.02 (0.86, 1.21)	60	0.95 (0.64, 1.40)	
p-trend		0.27		0.71	0.86
2,4,5-TP					
Nonexposed	687	Ref	157	Ref	
T1	11	0.64 (0.34, 1.20)	6	1.10 (0.45, 2.68)	
T2	15	1.20 (0.72, 2.01)	3	**	
T3	13	0.92 (0.54, 1.57)	1	**	
p-trend		0.82		0.19	0.33
Butylate					
Nonexposed	561	Ref	110	Ref	
Q1	42	0.66 (0.48, 0.91)	16	1.02 (0.60, 1.74)	
Q2	51	1.05 (0.78, 1.40)	7	0.48 (0.22, 1.02)	
Q3	35	1.01 (0.72, 1.43)	21	2.20 (1.35, 3.56)	
Q4	46	1.23 (0.91, 1.66)	9	1.03 (0.52, 2.04)	
p-trend		0.14		0.40	0.01
Trifluralin					
Nonexposed	611	Ref	112	Ref	
Q1	172	0.95 (0.80, 1.13)	41	1.01 (0.69, 1.47)	
Q2	170	1.06 (0.88, 1.26)	41	1.00 (0.69, 1.45)	
Q3	170	1.04 (0.87, 1.24)	41	0.85 (0.58, 1.23)	
Q4	161	0.93 (0.78, 1.12)	43	1.04 (0.72, 1.49)	
p-trend		0.52		0.92	0.69
2,4-D					
Nonexposed	290	Ref	43	Ref	
Q1	262	0.93 (0.78, 1.11)	60	1.21 (0.80, 1.82)	
Q2	256	0.88 (0.74, 1.05)	68	1.29 (0.85, 1.95)	
Q3	287	1.03 (0.86, 1.22)	51	0.86 (0.56, 1.31)	
Q4	260	0.87 (0.73, 1.03)	73	1.17 (0.78, 1.75)	

p-trend		0.25		0.90	0.42
2,4,5-T					
Nonexposed	544	Ref	121	Ref	
Q1	43	1.23 (0.90, 1.68)	18	2.03 (1.22, 3.37)	
Q2	45	1.05 (0.77, 1.43)	11	1.13 (0.61, 2.10)	
Q3	45	0.96 (0.71, 1.30)	10	0.93 (0.48, 1.78)	
Q4	50	0.90 (0.68, 1.21)	6	0.49 (0.21, 1.11)	
p-trend		0.43		0.06	0.22
Permethrin crop					
Nonexposed	1141	Ref	239	Ref	
Q1	37	0.93 (0.66, 1.29)	11	0.92 (0.47, 1.81)	
Q2	40	1.12 (0.82, 1.54)	7	0.77 (0.32, 1.87)	
Q3	36	0.79 (0.56, 1.11)	11	1.30 (0.69, 2.44)	
Q4	33	0.91 (0.64, 1.29)	11	1.52 (0.83, 2.78)	
p-trend		0.46		0.15	0.16
Permethrin animal					
Nonexposed	1183	Ref	246	Ref	
Q1	37	1.10 (0.76, 1.58)	6	0.52 (0.19, 1.41)	
Q2	32	1.16 (0.80, 1.67)	10	1.27 (0.61, 2.64)	
Q3	33	1.14 (0.80, 1.62)	9	1.05 (0.52, 2.14)	
Q4	35	0.86 (0.61, 1.21)	7	0.69 (0.34, 1.40)	0.78
p-trend		0.48		0.35	
Carbofuran					
Nonexposed	891	Ref	169	Ref	
Q1	112	1.00 (0.82, 1.22)	20	0.75 (0.46, 1.22)	
Q2	91	1.10 (0.89, 1.36)	28	1.79 (1.21, 2.66)	
Q3	102	1.13 (0.92, 1.39)	29	1.33 (0.89, 1.99)	
Q4	98	0.88 (0.71, 1.08)	32	1.11 (0.76, 1.61)	
p-trend		0.30		0.49	0.28
Carbaryl					
Nonexposed	357	Ref	86	Ref	
Q1	100	1.04 (0.83, 1.31)	30	1.08 (0.70, 1.67)	
Q2	107	1.15 (0.91, 1.47)	23	1.09 (0.67, 1.76)	
Q3	100	1.23 (0.96, 1.56)	22	1.32 (0.79, 2.18)	
Q4	101	0.98 (0.75, 1.27)	12	0.49 (0.25, 0.97)	
p-trend		0.59		0.03	0.19
Aldicarb					
Nonexposed	674	Ref	154	Ref	
Q1	16	0.87 (0.53, 1.44)	3	**	
Q2	17	1.65 (1.01, 2.72)	1	**	
Q3	17	0.98 (0.59, 1.63)	3	**	
Q4	15	0.83 (0.47, 1.45)	3	**	
p-trend		0.56			
Methyl bromide					
Nonexposed	1166	Ref	245	Ref	
Q1	48	0.88 (0.65, 1.19)	16	1.58 (0.92, 2.73)	
Q2	48	0.84 (0.62, 1.14)	11	1.22 (0.64, 2.34)	
Q3	45	0.81 (0.60, 1.11)	13	2.00 (1.10, 3.67)	
Q4	55	1.00 (0.75, 1.33)	10	1.10 (0.55, 2.17)	
p-trend		0.97		0.79	0.10
Aluminum Phosphide					
Nonexposed	710	Ref	157	Ref	
T1	6	0.82 (0.37, 1.84)	4	**	
T2	8	0.60 (0.29, 1.23)	3	**	

T3	6	0.83 (0.37, 1.86)	1	**	
p-trend		0.50			
Mix 80/20					
Nonexposed	675	Ref	147	Ref	
T1	18	1.71 (1.07, 2.74)	7	2.34 (1.08, 5.06)	
T2	17	0.98 (0.61, 1.59)	6	2.07 (0.90, 4.72)	
T3	16	0.98 (0.60, 1.62)	4	0.96 (0.35, 2.67)	
p-trend		0.93		0.92	0.69
Ethylene Dibromide					
Nonexposed	699	Ref	160	Ref	
T1	10	1.22 (0.65, 2.28)	3	**	
T2	11	1.50 (0.82, 2.75)	0	**	
T3	8	0.46 (0.23, 0.93)	2	**	
p-trend		0.04			
Benomyl					
Nonexposed	665	Ref	156	Ref	
Q1	15	1.02 (0.61, 1.71)	2	**	
Q2	14	0.98 (0.57, 1.67)	1	**	
Q3	16	1.33 (0.80, 2.20)	2	**	
Q4	16	0.76 (0.46, 1.26)	3	**	
p-trend		0.37			
Chlorothalonil					
Nonexposed	1262	Ref	275	Ref	
Q1	27	0.86 (0.57, 1.30)	8	1.92 (0.91, 4.03)	
Q2	24	1.21 (0.80, 1.82)	6	1.69 (0.74, 3.87)	
Q3	23	0.75 (0.49, 1.15)	8	1.41 (0.68, 2.92)	
Q4	28	0.96 (0.65, 1.40)	3	**	
p-trend		0.65		0.29	0.16
Captan					
Nonexposed	1174	Ref	246	Ref	
Q1	37	1.11 (0.79, 1.54)	7	0.88 (0.39, 1.99)	
Q2	31	0.98 (0.68, 1.41)	7	1.25 (0.61, 2.54)	
Q3	33	1.14 (0.81, 1.62)	7	0.96 (0.44, 2.12)	
Q4	32	1.00 (0.69, 1.45)	8	0.96 (0.47, 1.96)	
p-trend		0.93		0.90	0.84
Maneb/Mancozeb					
Nonexposed	663	Ref	155	Ref	
Q1	17	1.04 (0.63, 1.73)	2	**	
Q2	15	0.92 (0.53, 1.60)	3	**	
Q3	15	0.88 (0.53, 1.47)	4	**	
Q4	14	0.93 (0.54, 1.60)	3	**	
p-trend		0.75			
Metalaxyl					
Nonexposed	588	Ref	142	Ref	
Q1	34	0.9 (0.63, 1.27)	9	1.1 (0.56, 2.18)	
Q2	37	1.12 (0.78, 1.61)	4	**	
Q3	36	1.13 (0.79, 1.63)	5	0.86 (0.34, 2.20)	
Q4	35	1.07 (0.74, 1.54)	6	0.95 (0.40, 2.25)	
p-trend		0.66		0.89	0.67

Abbreviations: quartile 1 (Q1); quartile 2 (Q2); quartile 3 (Q3); quartile 4 (Q4).

[a]Numbers do not sum to total due to missing data.

[b]Adjusted for age, state, race, smoking, fruit servings, and leisure time physical activity in the winter

International Journal of Epidemiology, 2016, 792–805
doi: 10.1093/ije/dyv195
Advance Access Publication Date: 27 September 2015
Original article

Environmental Exposures and Cancer

Occupational exposure to pesticides and bladder cancer risk

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Accepted 3 September 2015

Abstract

Background: In the developed world, occupational exposures are a leading cause of bladder cancer. A few studies have suggested a link between pesticide exposures among agricultural populations and bladder cancer.

Methods: We used data from the Agricultural Health Study, a prospective cohort study which includes 57 310 pesticide applicators with detailed information on pesticide use, to evaluate the association between pesticides and bladder cancer. We used Poisson regression to calculate rate ratios (RRs) and 95% confidence intervals (CIs) to estimate the association between each of 65 pesticides and 321 incident bladder cancer cases which accrued over the course of follow-up (1993–2011), adjusting for lifestyle and demographic and non-pesticide farm-related exposures, including those previously linked to bladder cancer. We conducted additional analyses stratified by smoking status (never, former, current).

Results: We observed associations with bladder cancer risk for two imidazolinone herbicides, imazethapyr and imazaquin, which are aromatic amines. Ever use of imazaquin (RR = 1.54, 95% CI: 1.05, 2.26) was associated with increased risk whereas the excess risk among users of imazethapyr was evident among never smokers (RR in highest quartile vs non-exposed = 3.03, 95% CI: 1.46, 6.29, *P*-interaction = 0.005). We also observed increased risks overall and among never smokers for use of several chlorinated pesticides including chlorophenoxy herbicides and organochlorine insecticides.

Conclusions: Several associations between specific pesticides and bladder cancer risk were observed, many of which were stronger among never smokers, suggesting that

International Journal of Epidemiology, 2016, Vol. 45, No. 3

possible risk factors for bladder cancer may be more readily detectable in those unexposed to potent risk factors like tobacco smoke.

Key words: Pesticides, bladder cancer, epidemiology

Key Messages

- Occupational exposures are a leading cause of bladder cancer, but occupational pesticide exposure has been little explored as a possible risk factor.
- We observed increased risks for two aromatic amine herbicides, chlorophenoxy herbicides and organochlorine insecticides.
- Several associations were more apparent among never smokers, suggesting that pesticide exposure may be an overlooked exposure in bladder carcinogenesis.
- Our results highlight the difficulty in trying to understand the impact of other exposures on smoking-related cancers.

Introduction

In the developed world, bladder cancer is the fourth and twelfth most common cancer in men and women, respectively.[1] The leading risk factors are cigarette smoking and occupational exposures.[2] Aromatic amines, including 2-naphthylamine, 4-aminobiphenyl, benzidine, ortho-toluidine and others, are established bladder carcinogens that have been described in the occupational setting.[3] Agricultural populations have a lower prevalence of smoking than the general population,[4–6] which may explain why several studies have found either no association or a decreased risk of bladder cancer in this occupational group.[7–13] On the other hand, two studies have shown a link between farming and bladder cancer among non-smokers,[14,15] which suggests a complexity in interpreting the effect of other exposures in the presence of smoking, the primary risk factor for bladder cancer. In addition, some studies have suggested a link between farming, herbicide exposure or specific agricultural settings and risk of bladder cancer.[14–22] Bladder cancer risk might be explained by the urogenous contact hypothesis which proposes that active carcinogens dissolved in urine come into contact with and transform cells of the bladder epithelium.[23] Many pesticides and their metabolites are readily excreted from the body via the urine. Thus, the potential exists for pesticides to adversely affect the bladder. We previously reported an increased risk of bladder cancer[24] in a cohort of farmers occupationally exposed to the aromatic amine herbicide, imazethapyr. Other specific pesticides, however, have been little explored as possible risk factors for bladder cancer. Thus, we used data from the Agricultural Health Study (AHS), a large prospective cohort study of pesticide applicators with detailed pesticide use data, to evaluate the association between several specific pesticides and bladder cancer risk.

Methods

Study population

The AHS is a prospective cohort study that includes 52 394 licensed private pesticide applicators in Iowa and North Carolina and 4916 licensed commercial applicators in Iowa. The cohort has been described in detail.[6,24,25] Briefly, individuals seeking licenses for restricted-use pesticides were recruited from December 1993 through December 1997 (82% of the target population enrolled). The protocol was approved by all relevant institutional review boards. We obtained cancer incidence information by regular linkage to cancer registry files in Iowa and North Carolina. In addition, the cohort is matched to state mortality registries and the National Death Index to identify vital status, and to home address records of the Internal Revenue Service, motor vehicle registration files and pesticide license registries of state agricultural departments to determine residence in Iowa or North Carolina. The current analysis included all incident bladder cancers (invasive and *in situ*) diagnosed from enrolment (1993–97) through 31 December 2010 in North Carolina and 31 December 2011 in Iowa. We censored follow-up at the date of cancer diagnosis, time of death, movement out of state or at the end of the current follow-up time. Because there was only one case of bladder cancer diagnosed among female applicators, we excluded women from the analysis ($n = 1562$), as well as 1071 individuals with prevalent cancer at enrolment and 333 with no follow-up information, leaving

54 344 men for analysis among whom a total of 321 incident bladder cancers were diagnosed.

Exposure assessment

Information on use of individual pesticides was captured in two self-administered questionnaires [http://www.aghealth.nih.gov/collaboration/questionnaires.html] completed during cohort enrolment. All applicators completed the first enrolment questionnaire, which enquired about ever/never use of 50 pesticides, as well as duration (years) and frequency (average days/year) of use for a subset of 22 pesticides. In addition, 44.1% of the applicators returned the second (take-home) enrolment questionnaire, which enquired about duration and frequency of use for the remaining 28 additional pesticides and ever/never use of additional pesticides. A follow-up questionnaire, which ascertained pesticide use since enrolment and last year applied, was administered 5 years after enrolment and completed by 36 342 (63%) of the original participants. For participants who did not complete a follow-up questionnaire (20 968 applicators, 37%), a data-driven multiple imputation procedure was used to impute use of specific pesticides at follow-up. A detailed description of the imputation process and validation is described by Heltshe *et al.*[26] Enrolment and follow-up information were combined to generate cumulative lifetime days of use and intensity-weighted lifetime days of use.

We restricted analyses to those pesticides with 10 or more exposed cases ($n = 65$). Among these, 44 had detailed data to explore associations between cumulative exposure and bladder cancer risk, using two exposure metrics: (i) lifetime days of pesticide use, that is the product of years of use of a specific pesticide and the number of days used per year; and (ii intensity-weighted lifetime days of use, which is the product of lifetime days of use and a measure of exposure intensity. Intensity was derived from an algorithm using questionnaire data on mixing status, application method, equipment repair and use of personal protective equipment.[27] We also used 15-year lagged cumulative exposure, discounting the most recent 15 years of use. Supplementary Table 1 (available as Supplementary data at *IJE* online) provides the complete list of pesticides evaluated and their prevalence of use. Data were obtained from Agricultural Health Study data release versions P1REL201209.00 and P2REL201209.00.

Statistical analyses

For each pesticide, we categorized exposure based on the distribution of use among exposed cases. Depending on the prevalence of exposure, we created categories based on the median exposure, tertiles or quartiles. We used Poisson regression to calculate rate ratios (RRs) and 95% confidence intervals (CIs) and used the MIANALYZE procedure in SAS, version 9.3 (SAS Institute, Inc., Cary, NC, USA) to obtain the appropriate variance for the imputed data. Analyses were conducted using ever/never use, the lifetime days, intensity-weighted lifetime days and the 15-year lagged metrics. We evaluated several lifestyle, demographic and non-pesticide farm-related exposures, including those previously linked to bladder cancer (diesel exhaust exposure, welding, painting, grinding metal) as possible confounders of the relationship between pesticides and bladder cancer, and ultimately included the following variables which were independently related to bladder cancer in our population for adjustment of all models: attained age (10-year intervals), race (White, other), cigarette smoking (status, pack-years among former and current smokers) and pipe smoking (ever/never). Smoking status [never, former (smoked at least 100 cigarettes in the past], current) was ascertained at enrolment and subsequently upon cohort follow-up. Duration (years) and intensity (cigarettes/day) of smoking were assessed at enrolment. To fully explore possible confounding due to smoking, we explored adjusting for smoking in two ways: (i) status (never, former, current) and pack-years smoked; and (ii) status and duration (years) of smoking. We also conducted analyses stratified by smoking status (never, former, current). We also explored adjustment for ever use of pesticides most highly associated with a given individual pesticide in multivariate models, as well as mutual adjustment for pesticides that were associated with bladder cancer risk. Likelihood ratio tests were used to assess differences between strata (P-interaction). All tests were two-sided and conducted at the $\alpha = 0.05$ level. Tests for trend used the midpoint value of each exposure category in regression models.

Results

In all, 321 cases of bladder cancer were diagnosed among male applicators through the current follow-up period. Of these, 96% ($n = 307$) were urothelial carcinomas and the majority of these were localized tumours ($n = 272$) (data not shown); 83 cancers were diagnosed among never smokers, 161 among former smokers and 69 among current smokers (Table 1); 13% of cases also reported a history of pipe use (Table 1); and all of these men were former cigarette smokers at enrolment.

Table 2 shows the rate ratios of bladder cancer associated with ever use of specific herbicides, insecticides, fumigants and fungicides. Increased risks of bladder cancer were observed among ever users of the herbicides bentazon

Table 1. Characteristics of incident bladder cancer cases among men in the Agricultural Health Study

Characteristic	Cohort Person-years (total = 802,905.7)	Total Bladder Cancer $n = 321$ n (%)[a]
Age at the end of current follow-up		
<60	402510.437 (50.1)	57 (17.8)
60–69	203258.327 (25.3)	100 (31.2)
70–79	138180.408 (17.2)	114 (35.5)
80+	58956.5777 (7.3)	50 (15.6)
Mean (SD)		69.6 (10.4)
State		
Iowa	534349.517 (66.6)	185 (57.6)
North Carolina	268556.233 (33.4)	136 (42.4)
Applicator Type		
Private/farmer	729393.3 (91.0)	300 (93.5)
Commercial	70440.4 (8.8)	21 (6.5)
Exposed to engine exhaust		
No	268975.2 (33.5)	123 (38.3)
Yes	80786.8 (10.1)	50 (15.6)
Missing	450071.6 (56.1)	148 (46.1)
Paint at least once a year		
No	257887.4 (32.2)	153 (47.7)
Yes	541946.2 (67.5)	168 (52.3)
Missing		
Grind metal in summer and/or winter		
Monthly	93414.5 (11.6)	57 (17.8)
Weekly	145398.4 (18.2)	63 (19.6)
Other	68232.9 (8.5)	36 (11.1)
Missing	490545.0 (61.1)	165 (51.4)
Race		
White	767652.107 (95.6)	317 (98.8)
Black/Other	35253.6427 (4.4)	4 (1.2)
Smoking Status[b]		
Never	416616.101 (51.9)	83 (25.9)
Former	231281.971 (28.8)	161 (50.2)
Current	130657.717 (16.3)	69 (21.5)
Missing	24349.9603 (3.0)	8 (2.5)
Pipe Smoker		
Never	764677.153 (95.2)	278 (86.6)
Ever	38228.5969 (4.8)	43 (13.4)

[a]Percents may not sum to 100 due to rounding.
[b]Assessed at enrolment and follow-up.

(RR = 1.55, 95% CI: 1.10, 2.19), bromoxynil (RR = 1.51, 95% CI: 1.04, 2.20), chloramben (RR = 1.56, 95% CI: 1.10, 2.22), diclofop-methyl (RR = 1.85, 95% CI: 1.01, 3.42) and imazaquin (RR = 1.54, 95% CI: 1.05, 2.26). Additional associations were observed between ever use of 2,4-D (RR = 1.46, 95% CI: 0.98, 2.18) and ever use of sethoxydim (RR = 0.65, 95% CI: 0.43, 1.00), with a positive and an inverse association observed, respectively. The organochlorine insecticides dichlorodiphenyltrichloroethane (DDT) and heptachlor were positively associated with bladder cancer risk (RR = 1.40, 95% CI: 1.10, 1.80 and RR = 1.30, 95% CI: 0.98, 1.74, respectively).

Table 3 shows the associations between cumulative intensity-weighted lifetime days of herbicide use and risk of bladder cancer overall and stratified by smoking status. We observed positive trends for 2,4,5-T [RR in tertile 3 (T3) vs non-exposed = 2.64, 95% CI: 1.23, 5.68, *P*-trend = 0.02], 2,4-D [RR in quartile 4 (Q4) vs non-exposed = 1.88, 95% CI: 0.94, 3.77, *P*-trend = 0.02], glyphosate (RR in Q4 vs non-exposed = 1.93, 95% CI: 0.95, 3.91, *P*-trend = 0.03), and imazethapyr (RR in Q4 vs. non-exposed = 3.03, 95% CI: 1.46, 6.29, *P*-trend = 0.004) among never smokers. There was evidence of effect modification by smoking on the relationship between cumulative intensity-weighted days of imazethapyr and bladder cancer (*P*-interaction = 0.005). An inverse trend with 2,4,5-T among former smokers, and a borderline inverse trend with dicamba among current smokers, were also observed.

Table 4 shows the associations between cumulative intensity-weighted lifetime days of insecticide use and risk of bladder cancer overall and stratified by smoking status. Overall, there were no positive trends in risk with increasing levels of insecticide use. Among never smokers, positive gradients in risk were observed with increasing use of two carbamate insecticides, aldicarb [RR high (M2) vs non-exposed = 4.04, 95% CI: 1.20, 13.57, *P*-trend = 0.03] and carbofuran (RR in T2 vs non-exposed = 1.99, 95% CI: 1.06, 3.75, *P*-trend = 0.03), two organochlorine insecticides, chlordane (RR T3 vs non-exposed = 2.83. 95% CI: 1.16, 6.90, *P*-trend = 0.02) and toxaphene (RR high vs non-exposed = 3.75, 95% CI: 1.57, 8.97, *P*-trend = 0.003), one organophosphate insecticide, fonofos (RR T3 vs non-exposed = 2.01, 95% CI: 1.01, 4.00, *P*-trend = 0.05) and one pyrethroid insecticide, permethrin use (RR high vs non-exposed = 2.28, 95% CI: 1.08, 4.82, *P*-trend = 0.04). No trends were observed between bladder cancer and pesticides among former or current smokers. The interaction between exposure and smoking was only evident for carbofuran (*P*-interaction = 0.04) and chlorpyrifos (*P*-interaction = 0.01).

There were no associations overall or among any of the smoking strata for use of any fumigants or fungicides evaluated (Supplementary Table 2, available as Supplementary data at *IJE* online) and bladder cancer, with the exception of a positive association among smokers using carbon tetrachloride/carbon disulfide, which was based on only three exposed cases. In addition, Supplementary Table 3 (available as Supplementary data at *IJE* online) provides stratified risks of bladder cancer by smoking status for those pesticides with no cumulative use information. No notable differences in observed associations emerged from analyses of lifetime days or from lagged exposures and these are, therefore, not shown.

Table 2. Ever use of pesticides and risk of bladder cancer in the Agricultural Health Study

Pesticide	Exposed Cases	RR[a] (95% CI)
Herbicides		
2,4,5-T[b]	91	1.15 (0.84, 1.59)
2,4,5-TP[b,c]	40	1.07 (0.74, 1.56)
2,4-D	245	**1.46 (0.98, 2.18)**
Acifluorfen, sodium salt[c]	28	1.21 (0.79, 1.85)
Alachlor	158	1.15 (0.86, 1.52)
Atrazine	220	1.22 (0.88, 1.69)
Bentazon[c]	67	**1.55 (1.10, 2.19)**
Bromoxynil[c]	51	**1.51 (1.04, 2.20)**
Butylate	86	0.86 (0.63, 1.19)
Chloramben[b,c]	46	**1.56 (1.10, 2.22)**
Chlorimuron-ethyl	91	0.85 (0.62, 1.17)
Clomazone[c]	24	0.99 (0.64, 1.54)
Cyanazine	101	0.90 (0.67, 1.21)
Dicamba	125	0.84 (0.62, 1.14)
Diclofop-methyl[c]	11	**1.85 (1.01, 3.42)**
EPTC	49	0.98 (0.70, 1.37)
Ethalfluralin[c]	10	0.77 (0.40, 1.45)
Fluazifop-butyl[b,c]	26	1.06 (0.68, 1.64)
Glyphosate	248	1.17 (0.78, 1.77)
Imazaquin[c]	38	**1.54 (1.05, 2.26)**
Imazethapyr	104	1.03 (0.76, 1.40)
Linuron[c]	21	0.97 (0.60, 1.55)
Metolachlor	113	0.86 (0.65, 1.13)
Metribuzin	107	0.75 (0.54, 1.04)
Propachlor[b,c]	27	1.20 (0.78, 1.83)
Paraquat	71	0.86 (0.61, 1.20)
Pendimethalin	113	0.75 (0.55, 1.02)
Petroleum Oil/Petroleum Distillates	130	0.88 (0.65, 1.21)
Sethoxydim[c]	28	**0.65 (0.43, 1.00)**
Simazine[b,c]	16	1.04 (0.61, 1.77)
Thifensulfuron-methyl[c]	14	1.04 (0.59, 1.82)
Trifluralin	139	1.08 (0.80, 1.45)
Insecticides		
Acephate[c]	21	0.91 (0.55, 1.50)
Aldicarb	35	0.88 (0.59, 1.32)
Aldrin[b]	88	1.20 (0.92, 1.57)
Carbaryl	192	1.04 (0.70, 1.54)
Carbofuran	67	0.86 (0.63, 1.16)
Chlordane[b]	97	0.95 (0.74, 1.22)
Chlorpyrifos	108	0.88 (0.67, 1.14)
Coumaphos	19	0.95 (0.59, 1.54)
DDT[b]	136	**1.40 (1.10, 1.80)**
DDVP[b]	25	1.01 (0.65, 1.55)
Diazinon	98	0.74 (0.54, 1.02)
Dieldrin[b,c]	32	1.19 (0.82, 1.72)
Disulfoton[b,c]	15	0.94 (0.54, 1.65)
Ethoprop[c]	11	0.73 (0.39, 1.37)
Fonofos[b]	53	1.09 (0.78, 1.52)
Heptachlor[b]	72	**1.30 (0.98, 1.74)**
Lindane[b]	69	1.08 (0.82, 1.42)
Malathion	223	1.01 (0.65, 1.58)
Methomyl[c]	13	1.17 (0.64, 2.12)
Parathion[b]	62	1.14 (0.81, 1.61)
Permethrin	44	0.75 (0.53, 1.07)
Phorate	96	0.99 (0.72, 1.37)
Terbufos	92	1.05 (0.79, 1.41)
Toxaphene[b]	56	0.96 (0.72, 1.30)
Fumigants		
Aluminum Phosphide	20	1.13 (0.70, 1.83)
Carbon Tetrachloride/Carbon Disulfide[b]	32	1.39 (0.93, 2.09)
Ethylene Dibromide[b,c]	17	0.86 (0.51, 1.46)
Methyl Bromide	48	0.86 (0.60, 1.23)
Fungicides		
Benomyl[b]	42	1.09 (0.74, 1.60)
Captan	32	1.19 (0.81, 1.74)
Chlorothalonil	27	1.09 (0.71, 1.66)
Maneb/Mancozeb	35	0.86 (0.57, 1.29)
Metalaxyl	65	0.66 (0.47, 0.94)

[a]Model adjusted for age, race, state, pack-years of cigarettes and pipe smoking.

[b]No longer registered for use in the USA.

[c]Results available on ever use only.

Discussion

In this analysis, we saw associations between two imidazolinone herbicides, imazethapyr and imazaquin which are aromatic amines, and bladder cancer risk. Ever use of other herbicides, including the general use pesticides bentazon and bromoxynil, the chlorophenoxy herbicide diclofop-methyl and another chlorinated herbicide chloramben, were also associated with bladder cancer. Increased risks of bladder cancer were also observed with regard to use of the chlorinated insecticide DDT; however, no consistent exposure-response relationship was observed in expanded analyses.

Imazethapyr is an imidazolinone herbicide used to control weeds in corn, soybean, dry bean, alfalfa and other crops.[28] Imazaquin is a general-use pesticide used to control grasses and broadleaf weeds.[29] In a previous analysis in the AHS focusing on risk of all cancer in a subcohort of applicators that used imazethapyr, we reported a relationship between imazethapyr and bladder cancer based on 41 exposed cases. In this analysis, which includes 6–7 years of additional follow-up and an additional 100 exposed cases, we did not observe an overall association with imazethapyr. An exposure-response relationship, however, was observed (P-trend = 0.004) among never smokers, with the highest category of exposure experiencing a 3-fold risk.

We also observed that ever use of another imidazolinone herbicide, imazaquin, was associated with bladder cancer risk. Although neither herbicide has demonstrated evidence of carcinogenicity in mice or rats, there is some plausibility for a possible link between exposure to imazethapyr and imazaquin and risk of bladder cancer because these herbicides are aromatic amine compounds, a chemical class which has been linked to bladder cancer, and animal metabolism studies show that these pesticides are readily excreted in the urine predominantly as the parent aromatic compounds.[28,29] The risk associated with imazethapyr exposure, however, was predominantly observed only among a smaller group of never smokers and it was not possible to evaluate quantitative exposure for imazaquin, and thus findings are unclear. Neither imazethapyr nor imazaquin have undergone a complete evaluation for evidence of human carcinogenic potential by the USA Environmental Protection Agency (U.S. EPA) or the International Agency for Research on Cancer (IARC). We are unaware of any other epidemiological study outside the AHS that has evaluated exposure to these pesticides as possible risk factors for cancer.

We also observed an increased risk of bladder cancer associated with ever use of the herbicides bentazon and bromoxynil. Bentazon and bromoxynil are used on a variety of food crops but are also used on lawns, turfs and golf courses. In our data, ever use of bentazon and bromoxynil were moderately correlated ($r = 0.54$). When we mutually adjusted models for these two herbicides, the results for both became non-significant. However, whereas the magnitude of the effect for bromoxynil diminished, the effect of bentazon was similar to that observed overall, and additional analyses stratified by smoking status also showed a strong association between bentazon and bladder cancer among never smokers (RR = 2.14, 95% CI: 1.09, 4.21, Supplementary Table 3, available as Supplementary data at *IJE* online), suggesting the effect is unlikely to be due to smoking and that bentazon might be more important in driving the observed bladder cancer risk than bromoxynil. There are limited experimental data on bentazon as a bladder carcinogen. In a combined chronic toxicity-carcinogenicity study in rats,[30] bentazon was found to result in increases in urine volume along with reduced urinary specific gravity, which may be related to bladder cancer risk.[31] Although there are few other data to support our findings regarding bentazon and bromoxynil, the use of these pesticides in both agricultural and general-use purposes indicates additional evaluation is warranted. Bentazon has been classified as a Group E carcinogen, evidence of non-carcinogenicity to humans, by the U.S. EPA based on animal models[30] and bromoxynil has been classified as a Group C, possible human carcinogen, based on observed liver tumours in animals;[32] neither have been evaluated by IARC.

Several chlorinated pesticides were also shown to influence bladder cancer risk in our analyses. Chloramben is an herbicide used to control weeds on soybean and other crops. No information is available on the carcinogenic effects of chloramben in humans, although a US study reported that oral exposure to chloramben caused liver tumours in mice but not in rats.[33] We also found that ever use of the organochlorine insecticide DDT increased bladder cancer risk, but no trend in risk with increasing use was observed. This may be due, in part, to the lack of detailed information from more than half of those reporting being ever exposed to DDT (only 46% reported days and years of use). Two other organochlorine insecticides, chlordane and toxaphene, showed evidence of increased bladder cancer risk but only among never smokers. Organochlorine insecticides have been linked to several cancer sites,[34] but we are unaware of any studies suggesting a link with bladder cancer.

In subgroup analyses, we also observed some interesting associations between several herbicides and insecticides and bladder cancer among never smokers. Never smoking applicators with the highest use of the chlorophenoxy herbicides 2,4,5-T and 2,4-D had higher risk of bladder cancer, and heavy users of the herbicide glyphosate had increased risk as well. Recently, a cohort of chlorophenoxy herbicide manufacturing workers in The Netherlands was observed to have excess bladder cancer mortality, in particular among workers involved in the manufacture of 2,4,5-T.[35] Because the numbers of observed bladder cancer deaths in this and other manufacturing cohorts was small,[36,37] it is difficult to draw a definitive conclusion. Observational studies in dogs showed that exposure to herbicide-treated lawns, in particular those treated with phenoxy herbicides, was associated with higher bladder cancer risk.[38,39] Interestingly we also observed a positive association between another chlorophenoxy herbicide, diclofop-methyl, and bladder cancer, albeit among few exposed cases ($n = 11$). Diclofop-methyl is classified as likely to be carcinogenic to humans by the U.S. EPA[40] and IARC ranks chlorophenoxy herbicides as possibly carcinogenic to humans (Group 2B). Taken together, these data suggest a possible link between chlorophenoxy herbicide exposure and bladder cancer. Several insecticides showed higher risk of bladder cancer among the never smokers as well, but power was limited to draw conclusions as the numbers of exposed cases were often small, given their lower prevalence of use.

An interesting element of this analysis is the observed differences in risk among never smokers for multiple chemicals. Since cigarette smoking is the major risk factor for

Table 3. Cumulative intensity-weighted days for herbicide use and risk of bladder cancer, overall and stratified by smoking status

Pesticide	OVERALL		NEVER		FORMER		CURRENT		
	n = 321 cancers		*n* = 83 cancers		*n* = 161 cancers		*n* = 69 cancers		
	Cases	RR[a] (95% CI)	Cases	RR[b] (95% CI)	Cases	RR[c] (95% CI)	Cases	RR[b] (95% CI)	p-interaction
2,4,5-T[d]									
Non-exposed	122	Ref	28	Ref	70	Ref	22	Ref	
T1	14	1.35 (0.77, 2.36)	4	1.73 (0.60, 4.99)	8	1.16 (0.56, 2.43)	1	**	
T2	14	0.99 (0.56, 1.73)	2	0.63 (0.15, 2.66)	9	1.00 (0.50, 2.02)	3	1.54 (0.46, 5.23)	
T3	15	0.83 (0.48, 1.42)	9	2.64 (1.23, 5.68)	3	0.25 (0.08, 0.81)	3	1.12 (0.33, 3.77)	
p-trend		0.45		0.02		0.02		0.82	0.02
2,4-D									
Non-exposed	61	Ref	13	Ref	31	Ref	17	Ref	
Q1	60	1.25 (0.86, 1.82)	13	0.99 (0.44, 2.25)	34	1.26 (0.74, 2.14)	13	1.41 (0.67, 2.94)	
Q2	61	1.01 (0.70, 1.47)	18	1.19 (0.58, 2.44)	30	0.87 (0.51, 1.48)	13	1.16 (0.54, 2.48)	
Q3	61	0.89 (0.61, 1.30)	16	0.90 (0.42, 1.90)	30	0.75 (0.43, 1.31)	15	1.30 (0.63, 2.69)	
Q4	62	1.25 (0.87, 1.81)	23	1.88 (0.94, 3.77)	31	1.12 (0.66, 1.91)	8	0.83 (0.33, 2.04)	
p-trend		0.31		0.02		0.69		0.45	0.65
Alachlor									
Non-exposed	126	Ref	33	Ref	61	Ref	32	Ref	
Q1	37	1.10 (0.75, 1.60)	10	1.10 (0.54, 2.25)	22	1.25 (0.76, 2.07)	5	0.71 (0.26, 1.91)	
Q2	39	0.90 (0.63, 1.30)	12	1.06 (0.54, 2.06)	18	0.83 (0.49, 1.41)	9	0.94 (0.44, 2.03)	
Q3	38	1.23 (0.85, 1.77)	11	1.33 (0.67, 2.63)	21	1.41 (0.85, 2.32)	6	0.82 (0.34, 1.97)	
Q4	39	1.00 (0.70, 1.43)	14	1.43 (0.77, 2.68)	18	0.99 (0.59, 1.68)	7	0.67 (0.29, 1.51)	
p-trend		0.94		0.25		0.99		0.37	0.84
Atrazine									
Non-exposed	89	Ref	23	Ref	52	Ref	14	Ref	
Q1	53	1.30 (0.91, 1.86)	23	1.04 (0.51, 2.11)	29	1.10 (0.68, 1.76)	11	2.39 (1.09, 5.27)	
Q2	55	0.94 (0.65, 1.36)	22	0.63 (0.29, 1.36)	23	0.67 (0.40, 1.12)	21	2.72 (1.32, 5.62)	
Q3	56	0.98 (0.69, 1.39)	26	0.95 (0.5,0 1.83)	28	0.78 (0.48, 1.27)	12	1.67 (0.77, 3.62)	
Q4	55	0.95 (0.67, 1.34)	28	1.03 (0.54, 1.96)	27	0.80 (0.50, 1.29)	10	1.28 (0.56, 2.89)	
p-trend		0.46		0.69		0.43		0.52	0.13
Butylate[d]									
Non-exposed	115	Ref	35	Ref	58	Ref	19	Ref	
Q1	16	1.29 (0.76, 2.19)	3	0.65 (0.20, 2.13)	11	1.81 (0.94, 3.49)	2	1.13 (0.26, 4.92)	
Q2	15	1.44 (0.84, 2.49)	3	0.87 (0.26, 2.84)	10	1.84 (0.93, 3.64)	2	1.39 (0.32, 6.04)	
Q3	16	0.98 (0.58, 1.66)	3	0.57 (0.18, 1.88)	10	1.38 (0.70, 2.73)	3	0.96 (0.28, 3.29)	
p-trend		0.98		0.36		0.32		0.98	0.64
Chlorimuron-ethyl[d]									
Non-exposed	121	Ref	27	Ref	71	Ref	20	Ref	
T1	15	1.07 (0.62, 1.83)	6	1.66 (0.68, 4.07)	6	0.75 (0.32, 1.73)	3	1.30 (0.38, 4.40)	
T2	15	0.88 (0.51, 1.54)	3	0.76 (0.23, 2.52)	7	0.82 (0.37, 1.79)	5	1.31 (0.44, 3.89)	
T3	17	0.79 (0.47, 1.31)	8	1.75 (0.79, 3.88)	6	0.54 (0.23, 1.24)	3	0.62 (0.18, 2.09)	
p-trend		0.33		0.21		0.15		0.43	0.34
Cyanazine									
Non-exposed	175	Ref	48	Ref	87	Ref	40	Ref	
Q1	25	0.71 (0.46, 1.10)	6	0.59 (0.24, 1.46)	17	0.88 (0.51, 1.51)	2	0.33 (0.08, 1.40)	
Q2	25	0.66 (0.42, 1.03)	9	0.90 (0.43, 1.89)	10	0.46 (0.23, 0.94)	6	0.87 (0.36, 2.09)	
Q3	24	1.25 (0.80, 1.95)	5	0.90 (0.35, 2.31)	12	1.22 (0.65, 2.30)	7	1.90 (0.82, 4.40)	
Q4	26	0.81 (0.53, 1.24)	9	1.03 (0.49, 2.15)	14	0.89 (0.49, 1.59)	3	0.42 (0.13, 1.37)	
p-trend		0.59		0.76		0.94		0.31	0.27
Dicamba									
Non-exposed	150	Ref	30	Ref	74	Ref	37	Ref	
Q1	31	0.92 (0.61, 1.38)	9	0.83 (0.38, 1.78)	15	0.85 (0.47, 1.54)	7	1.14 (0.48, 2.74)	
Q2	32	0.70 (0.45, 1.08)	7	0.56 (0.23, 1.34)	20	0.85 (0.49, 1.47)	5	0.54 (0.19, 1.58)	

(continued)

Table 3. Continued

Pesticide	OVERALL		NEVER		FORMER		CURRENT		
	n = 321 cancers		*n* = 83 cancers		*n* = 161 cancers		*n* = 69 cancers		
	Cases	RR[a] (95% CI)	Cases	RR[b] (95% CI)	Cases	RR[c] (95% CI)	Cases	RR[b] (95% CI)	p-interaction
Q3	32	0.81 (0.54, 1.22)	9	0.84 (0.39, 1.83)	15	0.70 (0.39, 1.28)	8	1.05 (0.45, 2.42)	
Q4	32	0.77 (0.51, 1.16)	13	1.12 (0.56, 2.27)	17	0.84 (0.48, 1.49)	2	0.23 (0.05, 0.98)	
p-trend		0.31		0.50		0.62		0.05	0.32
EPTC									
Non-exposed	226	Ref	66	Ref	116	Ref	44	Ref	
T1	15	0.72 (0.42, 1.23)	3	0.50 (0.15, 1.60)	8	0.68 (0.33, 1.4)	4	1.29 (0.45, 3.70)	
T2	15	1.33 (0.79, 2.27)	3	0.83 (0.26, 2.67)	5	0.86 (0.35, 2.13)	7	3.75 (1.64, 8.58)	
T3	17	0.96 (0.58, 1.58)	5	1.02 (0.41, 2.55)	11	1.23 (0.65, 2.30)	1	**	
p-trend		0.94		0.93		0.49		0.44	0.09
Glyphosate									
Non-exposed	60	Ref	14	Ref	31	Ref	15	Ref	
Q1	62	1.28 (0.86, 1.89)	19	1.64 (0.75, 3.58)	31	1.22 (0.72, 2.08)	12	1.00 (0.46, 2.13)	
Q2	62	0.96 (0.65, 1.41)	11	0.79 (0.35, 1.77)	36	1.07 (0.64, 1.78)	15	0.88 (0.41, 1.87)	
Q3	62	0.85 (0.58, 1.26)	14	0.85 (0.37, 1.95)	30	0.83 (0.49, 1.39)	16	0.86 (0.40, 1.82)	
Q4	62	1.07 (0.73, 1.56)	23	1.93 (0.95, 3.91)	29	1.00 (0.58, 1.72)	10	0.58 (0.25, 1.34)	
p-trend		0.99		0.03		0.67		0.17	0.19
Imazethapyr									
Non-exposed	167	Ref	41	Ref	87	Ref	39	Ref	
Q1	24	0.82 (0.51, 1.31)	7	1.00 (0.41, 2.27)	12	0.77 (0.40, 1.47)	5	0.79 (0.27, 2.32)	
Q2	26	0.96 (0.61, 1.49)	13	1.88 (0.96, 3.71)	10	0.71 (0.35, 1.42)	3	0.51 (0.15, 1.74)	
Q3	23	0.92 (0.58, 1.46)	3	0.46 (0.14, 1.53)	16	1.27 (0.72, 2.26)	4	0.70 (0.24, 2.05)	
Q4 bottom	14	2.08 (1.18, 3.66)	4	2.12 (0.74, 6.10)	6	1.83 (0.78, 4.28)	4	0.76 (0.26, 2.23)	
Q4 top	13	0.94 (0.52, 1.68)	10	3.03 (1.46, 6.29)	3	0.47 (0.15, 1.53)	0	**	
p-trend		0.63		0.004		0.61		0.20	0.005
Metolachlor									
Non-exposed	168	Ref	40	Ref	86	Ref	42	Ref	
Q1	27	0.88 (0.58, 1.34)	8	0.99 (0.44, 2.20)	17	1.09 (0.63, 1.86)	2	0.28 (0.07, 1.17)	
Q2	27	0.74 (0.49, 1.12)	6	0.69 (0.29, 1.64)	13	0.69 (0.38, 1.28)	8	0.92 (0.43, 1.99)	
Q3	28	0.66 (0.44, 0.99)	14	1.29 (0.69, 2.42)	14	0.65 (0.36, 1.17)	0	**	
Q4	28	0.95 (0.63, 1.44)	10	1.50 (0.74, 3.01)	14	0.97 (0.54, 1.75)	4	0.47 (0.15, 1.46)	
p-trend		0.73		0.18		0.78		0.12	0.01
Metribuzin[d]									
Non-exposed	108	Ref	29	Ref	63	Ref	15	Ref	
Q1	12	1.09 (0.59, 2.01)	3	0.88 (0.26, 2.94)	5	0.72 (0.29, 1.83)	4	3.14 (1.00, 9.86)	
Q2	15	0.85 (0.49, 1.48)	3	0.56 (0.16, 1.89)	7	0.64 (0.29, 1.43)	5	2.37 (0.82, 6.87)	
Q3	10	0.89 (0.46, 1.72)	3	0.86 (0.26, 2.88)	6	0.89 (0.38, 2.09)	1	**	
Q4	17	0.72 (0.43, 1.22)	6	0.89 (0.37, 2.19)	8	0.56 (0.27, 1.20)	2	0.73 (0.16, 3.32)	
p-trend		0.21		0.86		0.17		0.48	0.44
Paraquat[d]									
Non-exposed	130	Ref	33	Ref	70	Ref	24	Ref	
T1	10	0.96 (0.49, 1.89)	3	1.30 (0.39, 4.26)	4	0.63 (0.20, 2.03)	3	1.66 (0.49, 5.67)	
T2	13	1.64 (0.91, 2.96)	5	2.97 (1.10, 8.03)	8	1.96 (0.92, 4.19)	0	**	
T3	12	1.29 (0.69, 2.40)	3	2.20 (0.71, 6.87)	7	1.45 (0.64, 3.28)	2	0.45 (0.06, 3.48)	0.08
p-trend		0.65		0.54		0.45		0.57	
Pendimethalin[d]									
Non-exposed	106	Ref	26	Ref	61	Ref	17	Ref	
T1	19	1.00 (0.60, 1.67)	3	0.59 (0.18, 1.96)	12	1.13 (0.58, 2.20)	3	0.97 (0.28, 3.35)	
T2	22	0.62 (0.39, 0.99)	5	0.67 (0.25, 1.82)	12	0.58 (0.31, 1.09)	5	0.73 (0.25, 2.10)	
T3	23	1.11 (0.67, 1.84)	10	2.08 (0.91, 4.75)	9	0.89 (0.42, 1.86)	4	0.92 (0.30, 2.82)	
p-trend		0.67		0.11		0.80		0.93	0.49

(continued)

Table 3. Continued

Pesticide	OVERALL		NEVER		FORMER		CURRENT		
	$n = 321$ cancers		$n = 83$ cancers		$n = 161$ cancers		$n = 69$ cancers		
	Cases	RR[a] (95% CI)	Cases	RR[b] (95% CI)	Cases	RR[c] (95% CI)	Cases	RR[b] (95% CI)	p-interaction
Petroleum Oil/Petroleum Distillates[d]									
Non-exposed	132	Ref	36	Ref	73	Ref	20	Ref	
T1	10	0.90 (0.46, 1.77)	2	0.68 (0.16, 2.84)	5	0.71 (0.26, 1.95)	3	2.17 (0.64, 7.33)	
T2	10	0.70 (0.37, 1.34)	1	**	6	0.78 (0.34, 1.80)	3	1.34 (0.39, 4.58)	
T3	11	1.10 (0.59, 2.04)	3	1.17 (0.36, 3.80)	6	1.09 (0.47, 2.51)	2	1.40 (0.32, 6.03)	
p-trend		0.78		0.82		0.83		0.70	0.63
Trifluralin									
Non-exposed	133	Ref	36	Ref	71	Ref	26	Ref	
Q1	34	1.23 (0.83, 1.81)	13	1.39 (0.68, 2.82)	14	1.02 (0.57, 1.84)	7	1.48 (0.60, 3.64)	
Q2	33	0.76 (0.50, 1.17)	9	0.76 (0.34, 1.68)	16	0.64 (0.36, 1.15)	8	1.10 (0.49, 2.49)	
Q3	35	0.89 (0.61, 1.30)	7	0.63 (0.28, 1.43)	21	0.95 (0.57, 1.58)	7	1.17 (0.50, 2.76)	
Q4	34	0.86 (0.58, 1.27)	12	1.14 (0.59, 2.23)	15	0.72 (0.41, 1.29)	7	0.92 (0.37, 2.25)	
p-trend		0.39		0.86		0.35		0.75	0.80

[a]Model adjusted for age, race, state, pack-years of cigarettes and pipe smoking.
[b]Model adjusted for age, race, state.
[c]Model adjusted for age, race, state, pipe smoking.
[d]Detailed information for these chemicals was collected on the take-home questionnaire at enrolment.

bladder cancer, it is perhaps not surprising that smoking may obscure the effect of another exposure, particularly if that effect is weaker than the smoking effect. Recently, a study of agricultural workers in Egypt found that the associations between farming and bladder cancer were more evident among those who never smoked, and there are other historical examples of positive risks for bladder cancer in association with several factors among never smokers.[14,41–43] A common challenge in these studies, as in ours, is the low precision of estimated associations and lack of statistical interaction, given that the number of never smokers who develop bladder cancer is small. Thus, much larger studies will be needed to fully evaluate a relationship between pesticides, smoking and risk of bladder cancer. Along the same lines, studies have also suggested an interaction with smoking for some exposures, where risk can either be potentiated[42] or diminished[44] across smoking strata. These data and ours suggest that evaluating possible bladder cancer risk factors such as pesticides across strata of smoking may provide valuable insights into bladder cancer risk; however, large studies will be needed to be able to detect risks among specific subgroups and true interactions.

Our study had both strengths and limitations. Detailed self-reported pesticide use information, at two points in time, was used to evaluate cancer risk. Information on pesticide use provided by farmers in the AHS has been found to be accurate and reliable,[45,46] allowing for this exploration of the relationship between specific pesticide exposures and bladder cancer risk. Nonetheless, there is potential for exposure misclassification though it is probably non-differential and would bias relative risks toward the null, diminishing any real exposure-response gradients.[47] Smoking status information was collected at enrolment for use in analyses but also reconciled with data from two follow-up questionnaires that allowed us to carefully characterize this important bladder cancer risk factor. In addition, we performed several sensitivity analyses related to smoking, including exploring adjustment for status and intensity and status and duration, which provided comparable results. We also had information on the ever use of other tobacco products reported at enrolment. Using detailed questionnaire data, we were also able to control for several other suggested bladder cancer risk factors, including exposure to diesel exhaust[48] and grinding metal,[49] none of which changed the estimates between pesticide exposures and bladder cancer risk. In addition we were able to take into consideration the use of pesticides that were correlated with the pesticide of interest and, except for where stated (bentazon and bromoxynil), we found only weak correlation among pesticides, whcih did not influence the calculated risk estimates. Although we evaluated a large number of pesticides ($n = 65$), we observed more positive associations than would have been expected by chance alone (6 observed less than $P = 0.05$ and 3 additional borderline positive associations, wheras 3.25 (or 5%) would have been expected by chance, Table 2). Still, we cannot rule out the possibility that some of our findings

Table 4. Cumulative intensity-weighted days for insecticide use and risk of bladder cancer, overall and stratified by smoking status

Pesticide	OVERALL		NEVER		FORMER		CURRENT		
	n = 321 cancers		*n* = 83 cancers		*n* = 161 cancers		*n* = 69 cancers		
	Cases	RR[a] (95% CI)	Cases	RR[b] (95% CI)	Cases	RR[c] (95% CI)	Cases	RR[b] (95% CI)	p-interaction
Aldicarb[d,h]									
Non-exposed	153	Ref	39	Ref	85	Ref	26	Ref	
M1	8	1.18 (0.56, 2.48)	2	1.75 (0.39, 7.94)	3	0.73 (0.22, 2.39)	2	1.42 (0.30, 6.65)	
M2	8	1.25 (0.56, 2.79)	4	4.04 (1.20, 13.57)	2	0.71 (0.17, 2.98)	2	0.81 (0.09, 6.88)	
p-trend		0.58		0.03		0.61		0.84	0.23
Aldrin[e,h]									
Non-exposed	113	Ref	30	Ref	59	Ref	21	Ref	
T1	15	0.88 (0.50, 1.53)	6	1.38 (0.55, 3.48)	9	0.94 (0.46, 1.94)	0	**	
T2	18	1.61 (0.96, 2.68)	1	**	11	1.75 (0.90, 3.40)	6	2.98 (1.15, 7.71)	
T3	17	1.51 (0.89, 2.55)	6	2.30 (0.92, 5.75)	9	1.44 (0.71, 2.96)	2	1.01 (0.23, 4.40)	
p-trend		0.08		0.12		0.21		0.57	0.05
Carbaryl[d,h]									
Non-exposed	73	Ref	23	Ref	34	Ref	14	Ref	
Q1	25	1.10 (0.68, 1.78)	6	0.82 (0.31, 2.17)	15	1.25 (0.66, 2.38)	4	1.25 (0.41, 3.82)	
Q2	28	1.93 (1.21, 3.09)	5	1.06 (0.36, 3.12)	16	2.35 (1.25, 4.41)	7	2.77 (1.10, 7.00)	
Q3	26	1.49 (0.92, 2.41)	6	1.50 (0.57, 3.91)	13	1.38 (0.68, 2.81)	6	1.94 (0.69, 5.42)	
Q4	27	0.91 (0.55, 1.50)	6	0.90 (0.32, 2.53)	18	1.19 (0.60, 2.34)	2	0.34 (0.07, 1.61)	
p-trend		0.29		0.84		0.90		0.08	0.45
Carbofuran[d]									
Non-exposed	206	Ref	50	Ref	110	Ref	46	Ref	
T1	21	0.52 (0.33, 0.82)	4	0.39 (0.14, 1.09)	13	0.55 (0.31, 0.97)	4	0.62 (0.22, 1.73)	
T2	23	0.98 (0.64, 1.51)	12	1.99 (1.06, 3.75)	8	0.65 (0.32, 1.33)	3	0.60 (0.19, 1.92)	
T3	22	0.90 (0.58, 1.40)	11	1.81 (0.94, 3.50)	7	0.55 (0.26, 1.19)	4	0.73 (0.26, 2.05)	
p-trend		0.77		0.03		0.12		0.51	0.04
Chlordane[e,h]									
Non-exposed	120	Ref	33	Ref	60	Ref	24	Ref	
T1	14	1.21 (0.69, 2.12)	1	0.35 (0.05, 2.56)	12	1.75 (0.94, 3.26)	1	**	
T2	15	0.78 (0.45, 1.34)	3	0.62 (0.19, 2.03)	10	0.93 (0.47, 1.82)	2	0.66 (0.16, 2.83)	
T3	15	1.46 (0.85, 2.52)	6	2.83 (1.16, 6.90)	8	1.34 (0.64, 2.84)	1	**	
p-trend		0.24		0.02		0.55		**	0.27
Chlorpyrifos[f]									
Non-exposed	200	Ref	45	Ref	117	Ref	38	Ref	
Q1	22	0.67 (0.43, 1.05)	8	1.02 (0.47, 2.21)	7	0.34 (0.16, 0.73)	7	1.34 (0.60, 3.00)	
Q2	23	0.84 (0.54, 1.31)	6	0.86 (0.37, 2.01)	7	0.43 (0.18, 0.99)	10	2.08 (1.03, 4.17)	
Q3	23	0.99 (0.64, 1.54)	11	1.86 (0.96, 3.61)	10	0.74 (0.37, 1.46)	2	0.55 (0.13, 2.31)	
Q4	23	0.69 (0.45, 1.06)	10	1.23 (0.62, 2.44)	9	0.50 (0.25, 0.98)	4	0.54 (0.19, 1.53)	
p-trend		0.14		0.42		0.06		0.19	0.01
Coumaphos[f]									
Non-exposed	245	Ref	74	Ref	121	Ref	50	Ref	
M1	8	0.49 (0.24, 0.99)	2	0.36 (0.09, 1.49)	4	0.46 (0.17, 1.25)	2	0.78 (0.19, 3.20)	
M2	11	1.79 (0.98, 3.27)	0	**	9	2.91 (1.48, 5.73)	2	1.66 (0.40, 6.86)	
p-trend		0.09		**		0.003		0.50	0.07
Diazinon[f,h]									
Non-exposed	133	Ref	39	Ref	70	Ref	22	Ref	
T1	11	0.76 (0.41, 1.40)	1	**	8	0.99 (0.47, 2.06)	2	0.97 (0.23, 4.11)	
T2	10	0.52 (0.26, 1.04)	1	**	6	0.40 (0.14, 1.15)	3	1.56 (0.47, 5.21)	
T3	13	1.03 (0.56, 1.90)	2	0.78 (0.18, 3.35)	7	1.06 (0.47, 2.37)	3	1.07 (0.24, 4.66)	
p-trend		0.96		**		0.95		0.86	0.34

(continued)

Table 4. Continued

Pesticide	OVERALL		NEVER		FORMER		CURRENT		
	$n = 321$ cancers		$n = 83$ cancers		$n = 161$ cancers		$n = 69$ cancers		
	Cases	RR[a] (95% CI)	Cases	RR[b] (95% CI)	Cases	RR[c] (95% CI)	Cases	RR[b] (95% CI)	p-interaction
DDT[e,h]									
Non-exposed	102	Ref	31	Ref	48	Ref	21	Ref	
Q1	15	0.96 (0.55, 1.66)	4	0.98 (0.34, 2.86)	11	1.19 (0.61, 2.32)	0	**	
Q2	16	1.43 (0.84, 2.44)	1	**	13	1.97 (1.05, 3.67)	2	1.25 (0.29, 5.41)	
Q3	15	0.76 (0.43, 1.32)	4	0.80 (0.27, 2.34)	6	0.56 (0.24, 1.33)	4	1.24 (0.41, 3.72)	
Q4	16	1.11 (0.64, 1.90)	4	1.29 (0.44, 3.79)	11	1.40 (0.71, 2.73)	1	**	
p-trend		0.78		0.59		0.48		0.34	0.18
DDVP[f]									
Non-exposed	253	Ref	69	Ref	129	Ref	55	Ref	
M1	12	0.85 (0.47, 1.54)	3	0.65 (0.20, 2.08)	8	1.04 (0.51, 2.15)	1	**	
M2	12	0.93 (0.52, 1.67)	4	1.05 (0.38, 2.89)	7	0.97 (0.45, 2.09)	1	**	
p-trend		0.82		0.92		0.94		**	0.77
Fonofos[f]									
Non-exposed	220	Ref	57	Ref	116	Ref	47	Ref	
T1	15	0.72 (0.42, 1.22)	5	0.88 (0.35, 2.23)	7	0.57 (0.26, 1.24)	3	0.93 (0.29, 3.05)	
T2	17	0.92 (0.56, 1.53)	5	1.01 (0.40, 2.57)	9	0.86 (0.43, 1.71)	3	0.92 (0.28, 2.99)	
T3	18	0.92 (0.57, 1.50)	10	2.01 (1.01, 4.00)	7	0.64 (0.30, 1.39)	1	**	
p-trend		0.78		0.05		0.28		0.20	0.37
Heptachlor[e,h]									
Non-exposed	139	Ref	34	Ref	76	Ref	26	Ref	
M1	14	0.82 (0.46, 1.44)	4	0.91 (0.31, 2.66)	7	0.65 (0.30, 1.44)	3	1.49 (0.44, 5.11)	
M2	14	1.10 (0.63, 1.93)	6	1.91 (0.78, 4.70)	8	1.06 (0.51, 2.23)	0	**	
p-trend		0.75		0.15		0.89		**	0.21
Lindane[e]									
Non-exposed	139	Ref	36	Ref	77	Ref	23	Ref	
M1	12	0.77 (0.43, 1.37)	4	0.82 (0.29, 2.32)	5	0.56 (0.22, 1.39)	3	1.49 (0.44, 5.03)	
M2	12	1.43 (0.78, 2.62)	4	2.00 (0.71, 5.63)	6	1.21 (0.53, 2.81)	2	1.62 (0.38, 6.97)	
p-trend		0.27		0.20		0.72		0.45	0.54
Malathion[f,h]									
Non-exposed	49	Ref	17	Ref	24	Ref	7	Ref	
Q1	28	1.00 (0.62, 1.59)	4	0.35 (0.11, 1.11)	17	1.16 (0.62, 2.17)	6	1.88 (0.62, 5.67)	
Q2	27	1.15 (0.71, 1.86)	9	1.09 (0.49, 2.43)	13	1.03 (0.52, 2.04)	5	1.80 (0.57, 5.72)	
Q3	29	1.14 (0.71, 1.83)	9	1.05 (0.45, 2.44)	15	1.13 (0.59, 2.15)	4	1.26 (0.33, 4.90)	
Q4	29	0.95 (0.60, 1.52)	6	0.66 (0.26, 1.71)	19	1.11 (0.60, 2.04)	4	1.17 (0.34, 4.01)	
p-trend		0.73		0.63		0.85		0.82	0.44
Parathion[f,h]									
Non-exposed	148	Ref	41	Ref	77	Ref	27	Ref	
M1	7	1.05 (0.49, 2.26)	2	1.09 (0.26, 4.60)	5	1.28 (0.51, 3.19)	0	**	
M2	8	1.13 (0.55, 2.36)	1	**	5	1.39 (0.54, 3.54)	2	1.54 (0.35, 6.84)	
p-trend		0.74		**		0.90		**	0.62
Permethrin[g]									
Non-exposed	239	Ref	64	Ref	123	Ref	52	Ref	
T1	13	0.92 (0.52, 1.61)	4	0.96 (0.36, 2.65)	7	0.90 (0.42, 1.93)	2	0.79 (0.19, 3.26)	
T2	13	0.45 (0.25, 0.81)	4	0.46 (0.17, 1.28)	5	0.33 (0.13, 0.81)	4	0.75 (0.25, 2.25)	
T3	15	1.11 (0.65, 1.87)	8	2.28 (1.08, 4.82)	5	0.72 (0.30, 1.77)	2	0.62 (0.15, 2.58)	
p-trend		0.93		0.04		0.31		0.49	0.44
Phorate[f,h]									
Non-exposed	115	Ref	30	Ref	62	Ref	21	Ref	
T1	16	0.74 (0.43, 1.27)	4	0.61 (0.21, 1.76)	8	0.66 (0.31, 1.42)	4	1.24 (0.41, 3.73)	
T2	16	0.99 (0.58, 1.69)	3	0.64 (0.19, 2.13)	10	1.13 (0.57, 2.26)	2	0.89 (0.21, 3.87)	

(continued)

Table 4. Continued

Pesticide	OVERALL		NEVER		FORMER		CURRENT		
	$n = 321$ cancers		$n = 83$ cancers		$n = 161$ cancers		$n = 69$ cancers		
	Cases	RR[a] (95% CI)	Cases	RR[b] (95% CI)	Cases	RR[c] (95% CI)	Cases	RR[b] (95% CI)	p-interaction
T3	17	0.98 (0.58, 1.64)	7	1.42 (0.62, 3.28)	8	0.89 (0.42, 1.88)	2	0.71 (0.17, 3.07)	
p-trend		0.96		0.36		0.90		0.62	0.76
Terbufos[f]									
Non-exposed	182	Ref	47	Ref	96	Ref	39	Ref	
T1	29	0.83 (0.56, 1.24)	7	0.76 (0.34, 1.71)	14	0.68 (0.38, 1.20)	8	1.48 (0.68, 3.20)	
T2	30	0.93 (0.63, 1.38)	16	1.77 (0.99, 3.15)	10	0.59 (0.31, 1.14)	4	0.69 (0.24, 1.94)	
T3	30	0.82 (0.55, 1.21)	8	0.80 (0.38, 1.71)	18	0.92 (0.55, 1.55)	4	0.57 (0.20, 1.59)	
p-trend		0.35		0.74		0.81		0.22	0.11
Toxaphene[e,h]									
Non-exposed	135	Ref	30	Ref	77	Ref	25	Ref	
M1	13	1.13 (0.64, 2.01)	6	2.34 (0.97, 5.68)	5	0.74 (0.30, 1.84)	2	1.14 (0.27, 4.86)	
M2	16	1.40 (0.82, 2.39)	7	3.75 (1.57, 8.97)	8	1.10 (0.52, 2.33)	1	**	
p-trend		0.24		0.003		0.80		**	0.09

[a]Model adjusted for age, race, state, pack-years of cigarettes and pipe smoking.
[b]Model adjusted for age, race, state.
[c]Model adjusted for age, race, state, pipe smoking.
[d]Carbamate insecticide.
[e]Organochlorine insecticide.
[f]Organophosphate insecticide.
[g]Pyrethroid insecticide.
[h]Detailed information for these chemicals was collected on the take-home questionnaire at enrolment.

might be due to chance, in particular in some of the stratified analyses where the number of exposed cases is small. Thus, future follow-up in the AHS to further evaluate the relationship between pesticides and bladder cancer, and to evaluate whether smoking modifies this relationship, are anticipated.

In conclusion, we observed increased risk of bladder cancer with two aromatic amine herbicides, the imidazolinone herbicides imazethapyr and imazaquin. The relationship between bladder cancer and imazethapyr, as well as for several other agricultural and general use herbicides, was more apparent among never smokers and highlights the complexity of trying to understand the impact of other exposures on smoking-related cancers. Associations with bladder cancer incidence and use of several chlorinated pesticides, including chlorophenoxy herbicides and organochlorine insecticides, were observed for the first time. Because farmers generally have lower rates of bladder cancer compared with the general population, few studies have explored whether pesticides, which readily pass through the bladder, might be risk factors for bladder cancer. Collectively, our data suggest that pesticide exposure may be an overlooked exposure in bladder carcinogenesis. Future studies with detailed pesticide information on specific active ingredients and those that explore risks across smoking status are needed.

Supplementary Data

Supplementary data are available at *IJE* online.

Funding

This work was supported by the Intramural Research Program of the National Institutes of Health, NCI, Division of Cancer Epidemiology and Genetics (Z01CP010119), NIEHS (Z01ES0490300), the Iowa Cancer Registry (HHSN261201300020I) and Iowa's Holden Comprehensive Cancer Center (P30CA086862) as well as the NIEHS-funded Environmental Health Sciences Research Center at the University of Iowa (P30ES005605).

Acknowledgement

We thank the participants of the Agricultural Health Study.

Conflict of interest: None declared.

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