

**Short Communication****Work in Agriculture, Childhood Residence, Nitrate Exposure, and Testicular Cancer Risk: A Case-Control Study in Denmark<sup>1</sup>****Henrik Møller<sup>2</sup>**

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**Abstract**

A population-based case-control study in Denmark investigated the hypothesis that parental occupation in agriculture increases the risk of testicular cancer in the offspring. Other factors investigated were: childhood residence on a farm, in the country, or in an area with high nitrate concentration in ground water, and the subjects' own occupation in agriculture. The only association that emerged was with childhood residence in the high-nitrate area. The excess risk was, however, confined entirely to men who did not grow up on a farm or in the country. This makes it very unlikely that nitrate exposure *per se* should be responsible. Further analyses revealed that the excess risk was confined largely to men who grew up in nonrural areas within Århus County, where Denmark's second largest city is located, and an excess risk was also seen among men who grew up in Copenhagen Municipality, which is the center of the most urban area in Denmark. The geographic pattern of incidence is stronger for the area of childhood residence than for the area of residence at the time of diagnosis. This supports indirectly the idea that testicular cancer is caused by unidentified factors early in life.

**Introduction**

Descriptive epidemiological data show that the incidence of testicular cancer varies strongly over time (1) and between countries (2, 3). Analytical studies, however, have not been successful in identifying strong causal factors for testicular cancer, apart from the consistently reported association with cryptorchidism (4, 5). A recent article in *Cancer Epidemiology, Biomarkers and Prevention* by researchers from Norway (6) showed an unexpected association between testicular cancer risk and a parent's occupation in agriculture ( $RR^3 = 1.24$ ), particularly on a farm with a high level of use of nitrate fertilizer relative to phosphate fertilizer ( $RR = 1.99$ ). The association was stronger for nonseminoma testicular cancer ( $RR = 4.53$ ) than for seminoma ( $RR = 2.14$ ). The present

paper describes data from a Danish case-control study of testicular cancer on parental occupation in agriculture and residence in childhood and the subjects' own occupation in agriculture.

**Materials and Methods**

This population-based case-control study was conducted in Denmark. The details of the study design have been described elsewhere (4, 7). In brief, we enrolled 514 men, born between 1916 and 1970, who developed testicular cancer in the period from 1986 to 1988, and 720 controls drawn at random from the computerized register of the Danish population and frequency matched to the case group by year of birth. The men were interviewed by telephone. For the subset of the men who were born in the period 1946–1970, self-administered questionnaires were mailed to their mothers. Completed questionnaires were received from 296 case mothers and 287 control mothers. The population coverage of testicular cancer cases (*i.e.*, the proportion of all testicular cancer cases in the relevant birth cohorts who actually participated in the study) was 74% for the men and 58% for the mothers. The participation rates (*i.e.*, the proportion of all persons invited to participate who finally participated) were 88% and 69% in cases and controls, and 87% and 74% in case mothers and control mothers. All information was obtained directly from the relevant persons. Proxy interviews or proxy questionnaires were not used.

Analyses were made of parental occupation in agriculture, as stated by the mothers, and of area of residence (*e.g.*, "in the country" or "on a farm"), based on information from the men. Information on parental occupation was obtained specifically for the period of the pregnancy with the boy. Information on childhood residence was obtained by several questions such as: "Did you ever live on a farm for more than 6 months during your childhood?" and "In which county did you live for the largest part of your childhood?"

Due to susceptible soils and high nitrate output from manure and artificial fertilizers, three counties in Denmark, Århus, Viborg, and Nordjylland Counties, have particularly high average concentrations of nitrate in the ground water, which is used for drinking water in both rural and urban areas. In 1994, 22% of water works in this high-nitrate area produced water with more than 25 mg nitrate per liter, compared with 6% in the rest of the country. There is evidence that drinking water concentrations in the area have been relatively high back to the 1930s (8). The effect of birthplace and childhood residence in the high-nitrate area on testicular cancer risk were analyzed.

The subjects' own occupation in agriculture was investigated. Results are presented for subjects who were ever employed in agriculture and for those employed in agriculture in 1985, the latter corresponding to a period just prior to the time of diagnosis of the cases and a comparable period for the controls.

Supplementary analyses were carried out for rural and

Received 7/26/96; accepted 11/12/96.

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<sup>1</sup> This work was supported by the Danish Cancer Society and the Danish Medical Research Council.

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<sup>3</sup> The abbreviation used is: RR, rate ratio.

**Table 1** Case-control study in Denmark of testicular cancer risk in relation to parental occupation in agriculture, residence during childhood, and the subject's own occupation in agriculture

	Total				Nonseminoma			Seminoma		
	296 cases (%)	287 controls (%)	Odds ratio	95% confidence interval	164 cases (%)	Odds ratio	95% confidence interval	125 cases (%)	Odds ratio	95% confidence interval
<b>Parental occupations during pregnancy</b>										
Mother employed in agriculture	5	4	1.23	0.56–2.69	5	1.32	0.51–3.40	6	1.34	0.50–3.57
Father employed in agriculture with animals	15	21	0.64	0.42–0.99	15	0.68	0.40–1.16	16	0.61	0.34–1.08
Father employed in agriculture without animals	2	3	0.73	0.25–2.14	2	0.81	0.21–3.18	2	0.79	0.20–3.08
	514 cases (%)	720 controls (%)	Odds ratio	95% confidence interval	239 cases (%)	Odds ratio	95% confidence interval	262 cases (%)	Odds ratio	95% confidence interval
<b>Residence in childhood</b>										
Born in high-nitrate area <sup>a</sup>	31	27	1.27	0.99–1.64	30	1.28	0.91–1.80	32	1.27	0.93–1.74
Lived in high-nitrate area for the larger part of childhood	32	26	1.40	1.09–1.81	33	1.49	1.06–2.08	33	1.36	1.00–1.86
Lived in the country for the larger part of childhood	45	52	0.79	0.63–1.00	42	0.71	0.52–0.98	49	0.85	0.63–1.13
Lived on a farm more than 6 months	22	30	0.72	0.55–0.94	18	0.57	0.39–0.83	27	0.86	0.62–1.19
<b>Occupation</b>										
Ever employed in agriculture	18	24	0.76	0.57–1.02	16	0.73	0.48–1.11	20	0.79	0.55–1.14
Employed in agriculture in 1985	5	6	0.99	0.59–1.65	5	0.86	0.42–1.78	6	1.09	0.60–2.01

<sup>a</sup> Århus, Viborg, and Nordjylland Counties.

**Table 2** Interaction between the variable "Lived in high-nitrate area" for the larger part of childhood" and two other variables on childhood residence, "Lived on a farm more than 6 months" and "Lived in the country for the larger part of childhood"

Residence in childhood	Total				Nonseminoma			Seminoma		
	514 cases (%)	720 controls (%)	Odds ratio	95% confidence interval	239 cases (%)	Odds ratio	95% confidence interval	262 cases (%)	Odds ratio	95% confidence interval
In high-nitrate area and on a farm	8	10	0.89	0.59–1.35	7	0.71	0.39–1.30	9	1.03	0.63–1.69
In high-nitrate area, not on a farm	24	16	1.59	1.18–2.15	26	1.76	1.19–2.59	23	1.54	1.06–2.23
On a farm, not in high-nitrate area	14	19	0.77	0.55–1.07	11	0.63	0.39–1.02	17	0.93	0.62–1.38
Not in high-nitrate area, not on a farm	54	55	1.00		56	1.00		50	1.00	
In high-nitrate area and in the country	18	16	1.10	0.79–1.53	17	1.04	0.66–1.62	19	1.14	0.76–1.72
In high-nitrate area, not in the country	15	10	1.51	1.03–2.20	16	1.74	1.07–2.82	14	1.37	0.86–2.20
In the country, not in high-nitrate area	28	36	0.78	0.59–1.03	25	0.72	0.49–1.05	30	0.80	0.56–1.14
Not in high-nitrate area, not in the country	40	38	1.00		42	1.00		37	1.00	

<sup>a</sup> Århus, Viborg, and Nordjylland Counties.

nonrural areas in 13 individual counties and in the Copenhagen area.

All analyses were done using unconditional logistic regression analysis, adjusting for year of birth.

## Results

No association was seen between a mother's or father's occupation in agriculture and the risk of testicular cancer in the offspring (Table 1). For the most prevalent of these exposure indices, "father employed in agriculture with animals," the odds ratio was below the expected and was borderline statistically significant.

Regarding the men's own occupation in agriculture, no significant association was seen with testicular cancer risk (Table 1). The tendency was in the direction of a reduction in risk among those employed in agriculture.

Living in the country or on a farm in childhood was negatively associated with testicular cancer risk (Table 1). In

the separate analyses of nonseminoma, the negative associations were statistically significant. Birthplace or residence in childhood in one of the three counties with relatively high nitrate concentrations in ground water was positively associated with testicular cancer risk; residence in this area carried an estimated relative risk of 1.40 overall, and 1.49 and 1.36 for nonseminoma and seminoma, respectively.

Due to the unexpected opposite effects of childhood residence on a farm and in the high-nitrate area (a predominantly rural area), the interaction between these factors was explored. Table 2 shows that the excess risk of testicular cancer in the high-nitrate area has no association with residence on a farm or in the countryside within that area. Indeed, it seems that childhood residence in nonrural areas within these counties is the factor most strongly associated with the highest risk of testicular cancer.

Table 3 shows how the relative risk of testicular cancer varies between rural and nonrural parts of Denmark's 13 coun-

Table 3 Testicular cancer risk in relation to childhood residence: county and urbanization

Location	Urbanization					
	Total		In the country		Not in the country	
	Odds ratio	95% confidence interval	Odds ratio	95% confidence interval	Odds ratio	95% confidence interval
Copenhagen Municipality	1.60	0.82–3.14	2.14	0.30–15.08	1.48	0.64–3.42
Remaining Copenhagen area	0.87	0.47–1.63	0.91	0.33–2.56	0.80	0.35–1.81
Frederiksborg County	1.00		1.00		0.87	0.29–2.60
Roskilde County	0.54	0.20–1.46	0.43	0.11–1.67	0.63	0.15–2.65
Vestsjælland County	0.92	0.43–1.95	1.05	0.40–2.75	0.56	0.16–1.91
Storstrøm County	0.87	0.42–1.80	0.70	0.28–1.79	1.18	0.37–3.74
Bornholm County	1.12	0.31–4.07	1.08	0.20–5.90	1.03	0.14–7.34
Fyn County	0.90	0.45–1.78	0.80	0.32–1.99	0.92	0.34–2.48
Sønderjylland County	0.68	0.31–1.49	0.62	0.22–1.76	0.66	0.20–2.16
Ribe County	0.61	0.28–1.35	0.49	0.15–1.55	0.65	0.23–1.85
Vejle County	0.70	0.35–1.42	0.53	0.20–1.41	0.84	0.32–2.25
Ringkøbing County	0.92	0.45–1.88	0.67	0.26–1.76	1.23	0.44–3.44
Århus County	1.46	0.78–2.76	0.80	0.33–1.92	2.37	0.98–5.71
Viborg County	1.06	0.52–2.18	1.14	0.45–2.86	0.73	0.23–2.29
Nordjylland County	1.16	0.61–2.21	1.23	0.52–2.90	0.89	0.35–2.26

ties and the Copenhagen area. The highest risks are found among men who grew up in Copenhagen Municipality or in nonrural parts of Århus County, corresponding to the two largest cities in the country, Copenhagen and Århus.

The results were generally consistent between nonseminoma and seminoma (Tables 1 and 2) and between broad age groups of men. Statistical adjustment for cryptorchidism [which is associated with testicular cancer risk (4)] and socioeconomic status [which is only marginally associated with testicular cancer risk in Denmark (7)] had no material influence on the results.

## Discussion

The incidence of testicular cancer in Denmark is among the highest in the world. Denmark is traditionally an agricultural country characterized by intensive production with heavy use of fertilizers and pesticides. It was therefore of interest to see the recent paper from Norway linking agricultural occupations in parents and testicular cancer risk in the offspring. If such an association exists, it should be demonstrable in the Danish population as well. The results of this analysis are clear on this point: in Denmark, occupation in agriculture *per se* is not associated with an increased risk of testicular cancer either in those employed themselves or in their offspring. Most of the estimates are in fact in the opposite direction, suggesting a slightly reduced incidence of testicular cancer in men employed in agriculture and their sons. A previous investigation, based on the occupational information collected routinely at the Danish Cancer Registry, similarly showed no association between testicular cancer incidence and occupation in agriculture (9).

In the Norwegian analysis, there was a strong association between a high level of use of nitrate fertilizer, relative to the use of phosphate fertilizer, on the farm where a parent worked (6). The authors state that this pattern of fertilizer use may indicate intense production of livestock on the farm. The available Danish data are less detailed. We have no information on specific agricultural practices on the farms, only the question of whether animals were kept on the farm (which is the most common situation, and which shows the lowest association with testicular cancer risk in these data).

The use of three counties to indicate an area of relatively high nitrate exposure is a very crude measure. On one hand, this may explain the much weaker association with nitrate exposure in Denmark than in Norway, because, if an effect of nitrate exposure is real, it would be much more diluted in the Danish analysis. On the other hand, it cannot be ruled out with confidence that some aspect of life other than nitrate exposure is driving the observed association in these counties. It is of particular interest to note that, even within the high-nitrate area, residence on a farm or in the countryside does not add to the excess risk of testicular cancer associated with residence in the area *per se*. This adds weight against the hypothesis that the effect of residence in this area is due to nitrate exposure, which typically is higher on farms and in the countryside (where many households obtain water from a private well) than in more urban areas (where communal water works are more common).

The analyses on childhood residence in rural and nonrural parts of individual counties in Denmark show that the excess risk of testicular cancer in the high-nitrate area is confined largely to men who grew up in nonrural parts of Århus county, the location of Denmark's second largest city. An excess risk is seen also among men who grew up in Copenhagen Municipality, the center of the largest urban area in Denmark. For reasons we do not know, testicular cancer thus seems to occur most frequently in men who grew up in the most urban areas of the country. It is of interest to note that degree of urbanization in adulthood is only weakly associated with testicular cancer risk (10). These results therefore support indirectly the idea that testicular cancer risk is influenced by unidentified factors early in life (1).

## Acknowledgments

Lars Grønberg assisted with the data analysis.

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