Scand J Work Environ Health 2019;45(1):82-89

<https://doi.org/10.5271/sjweh.3756>

Published online: 19 Jul 2018, Issue date: 01 Jan 2019

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This study demonstrates that non-occupational asbestos exposure accounts for up to 66% of malignant mesothelioma (MM) cases among women and identifies a 10,000 meter "hotspot" around asbestos industries where the risk of MM is significantly elevated. These findings empower the notion of compensation for non-occupationally asbestos exposed women with MM.

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Refers to the following text of the Journal: [2007;33\(4\):286-292](#)

Key terms: [asbestos](#); [asbestos exposure](#); [asbestos industry](#); [Denmark](#); [exposure](#); [hotspot](#); [malignant mesothelioma](#); [non-occupational exposure](#); [North Jutland](#); [risk for malignant mesothelioma](#); [women](#)

This article in PubMed: www.ncbi.nlm.nih.gov/pubmed/30025147

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Non-occupational exposure to asbestos is the main cause of malignant mesothelioma in women in North Jutland, Denmark

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Panou V, Vyberg M, Meristoudis C, Hansen J, Bøgsted M, Omland Ø, Møller Weinreich U, Røe OD. Non-occupational exposure to asbestos is the main cause of malignant mesothelioma in women in North Jutland, Denmark. *Scand J Work Environ Health*. 2019;45(1): 82–89. doi:10.5271/sjweh.3756

Objectives Diffuse malignant mesothelioma (MM) is mainly caused by asbestos inhalation. The malignancy is rare among women and studies of the prevalence and causative role of non-occupational asbestos exposure among women with MM are scarce. This observational study aimed to elucidate the asbestos exposure patterns among women with MM.

Methods All histological and cytological specimens from women diagnosed with MM between 1974–2015 at the Institute of Pathology, Aalborg University Hospital in Denmark, were re-evaluated. Occupational and habitation information were obtained from Danish registries and medical journals based on record linkage via the unique person ID. The number of MM cases in each parish in the region of North Jutland was determined and the incidence density in parishes was used to calculate the spatial relative risk (RR) of MM among women.

Results Diagnosis of MM was confirmed in 91 women. Exposure types were classified as occupational (9%), domestic (10%), environmental (22%), combination of domestic and environmental (34%) and unknown (25%). Twenty continuous parishes formed a MM “hotspot” around the asbestos-consuming industries in the city of Aalborg. Of these, the maximum RR was found in a parish housing an asbestos factory [RR 10.5, 95% confidence interval (CI) 5.5–19.4, environmental exposure in particular RR 2.9, 95% CI 0.7–9.1].

Conclusion Non-occupational asbestos exposure is the main cause of MM and may account for up to 66% of MM cases among women in North Jutland, Denmark.

Key terms asbestos exposure; asbestos industry; hotspot; risk for malignant mesothelioma.

Diffuse malignant mesothelioma (MM) is an aggressive malignancy that derives from the mesothelial lining of the pleura (MPM), peritoneum (MPeM), pericardium and tunica vaginalis testis and is mainly caused by asbestos inhalation (1). Asbestos is classified into two major categories: the amphiboles, including crocidolite, amosite and tremolite, and the serpentines, namely

chrysotile (2). Asbestos exposure is not only occupational but can also be domestic, by sharing a residence with an asbestos worker, as well as environmental, by living in proximity to an asbestos-emitting facility. All such exposures are carcinogenic (3–6). MM incidence among men is significantly higher than among women, but it seems to be a matter of exposure pattern and

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extent, since the male:female MM ratio is close to 1:1 when equally exposed to asbestos (2).

The North Jutland region in Denmark and particularly the largest city in the region, Aalborg, has a long-term history of large-scale asbestos use from two enterprises: the major Danish asbestos cement product factory (DAF) that has operated in the city of Aalborg since 1928, and a large shipyard (AaS) that used asbestos until 1986 (7). The predominant type of asbestos used in DAF was chrysotile (89%), while smaller quantities of amosite (10%) and crocidolite (1%) were used between 1946–1968 (8, 9). The factory was located in a densely inhabited area in the city of Aalborg, even neighboring four primary schools, and employed approximately 8000 male and 590 female workers (9, 10). Therefore, not only occupational, but domestic and environmental asbestos exposure, as well as combinations thereof might be expected as a cause of MM in that area.

In general, MM is a rare disease among men and even rarer among women, and there are only few detailed epidemiological studies of women with MM (5, 6, 11). Thus, the aim of this observational study was to investigate the scale of domestic and environmental asbestos exposure for the female MM patients.

Methods

All archival histological and cytological specimens obtained from female patients diagnosed with MM during 1974–2015 in the Institute of Pathology, Aalborg University Hospital, were reviewed. Two experienced pathologists individually reclassified the diagnoses based on available slides supplemented (when relevant and possible) with additional immunostains according to the international guidelines for mesothelioma diagnosis (12). For standardized classification, a 5-tiered scheme was applied: (i) definitely, (ii) probably, (iii) likely, (iv) unlikely and (v) definitely not MM. All the biopsies classified as unlikely and definitely not MM were excluded from the study, as these patients had probably or definitely other diagnoses.

The asbestos exposure information was obtained by linking individual identification numbers applied to all residents in Denmark from the Danish Supplementary Pension Fund Register (SPFR), the Danish Civil Registration System (CRS), medical journals and the national statistical service of Denmark (Statistics Denmark). The SPFR contains information on all employments, including company name and a unique company code, start and end dates for each employment since 1964, as well as a unique personal identification number, which makes it possible to link information on employment history to information on individuals in nationwide registers (13).

The CRS was established in 1968 and covers information for all residents living in Denmark that includes among others the unique personal identification number, family relations (father, mother, siblings and children), and historical address since 1971 and parish at birth (14). Supplementary information about occupational and domestic exposure to asbestos was acquired from the medical journals, and particularly the assessments of occupational health specialist and pulmonologist were used accordingly.

The type of potential asbestos exposure was classified into four categories. *Occupational*, for the patients who worked with asbestos; *domestic*, referring to women who shared a residence with a family member who was an asbestos worker; *environmental*, defined as living or working in a radius of <10 000 meters from DAF or AaS while asbestos was being used [the distance cutoff being based on previous studies (4–6, 15)]; *unknown*, where no source of asbestos exposure prior to development of MM could be identified. All the women who were occupationally exposed to asbestos were previously evaluated by an occupational health specialist in order to verify the asbestos exposure. The domestic asbestos exposure was either self-reported (N=22, 55%) or following an assessment from an occupational health specialist (N=18, 45%). The SPFR confirmed all information about occupational and domestic exposure to asbestos. The CRS was used to determine the environmental asbestos exposure. The duration of asbestos exposure and disease latency were defined by the period from the first reported asbestos exposure until the MM diagnosis was established.

Fisher's exact and t-tests were used to test differences between groups of categorical and continuous variables, respectively. Incidence density per 100 000 person-years in each parish of the North Jutland region in the period 1974–2015 was used to calculate the spatial relative risk (RR) of MM among women. An estimate of the number of female residents at risk in each parish and Denmark in total was calculated using the median number of residents for each five years from 1980–2015. When MM patients changed address inside the 10 000 meter radius from the asbestos industries, the parish where they lived the longest was included in the analysis. Hereafter, the incidence density of MM in the period 1974–2015 for each parish and Denmark in total was calculated by the ratio of the number of cumulated cases and the estimated number of females at risk multiplied by 41 years of observation. Clopper-Pearson's method was used to calculate confidence interval (CI) for the risk. The RR over the period 1974–2015 for each parish was calculated by dividing the incidence density of the parish with the incidence density for Denmark in total, referred to 100 000. Approximate CI for the RR, was obtained by dividing the endpoint of the risk CI with

the risk of Denmark. No age adjustment was made either for the North Jutland region or Denmark due to lack of information regarding age distribution in the parishes. Results were considered statistically significant when $P < 0.05$. The statistical software “R” (www.r-project.org) was used for all the analyses. Data were kept and registered according to the regulations of the Danish Protection Authorities. Approval of the Ethical Committee of the Region of North Jutland was granted prior to the start of the study (approval number N20140032).

Results

The pathology records revealed 101 women with a MM diagnosis. After the reclassification, 91 patients had a morphologic MM diagnosis (definitely MM, $N=56$, probably MM, $N=30$, likely MM, $N=5$), all of whom also had a clinical MM diagnosis. Among the 91 cases, 75 (82%) had MPM and 16 (18%) MPeM.

For the 10 patients reclassified as unlikely or definitely not MM, the review diagnoses were metastasis from carcinoma or fibrosing pleuritis.

Mesothelioma and exposure types

Complete or partial employment history backdating to 1964 was available for 87 (96%) women, where 8 (9%) had occupational exposure to asbestos (table 1). Domestic exposure was recorded in 40 (44%) women, via their husbands ($N=26$), fathers ($N=7$), sons ($N=4$) or both husbands and sons ($N=3$) who were employed in jobs involving asbestos. Occupational records of the relatives of the MM patients were available for all the cases (table 2). Potential environmental exposure was found in 54 (59%) of the patients; in 31 (34%) there was combined domestic and environmental exposure; in 3 (3%) there was combined occupational and environmental exposure, and in 20 (22%) there was no other asbestos exposure identified, implying environmental exposure alone. For 22 (25%) women with MM, no source of asbestos exposure could be identified. One woman was identified as being environmentally exposed for 24 years through damaged building material containing asbestos at her place of work outside the city of Aalborg. The types of asbestos exposure for the MM patients are summarized in figure 1. Analysis of the type of MM, MPM or MPeM, related to type of exposure showed that the women with secondary exposure to asbestos developed pleural ($N=54$) rather than peritoneal disease ($N=7$); on the contrary, women occupationally exposed to asbestos developed MPeM ($N=3$) rather than MPM ($N=5$) ($P=0.046$).

Table 1. Employment data for the malignant mesothelioma patients with occupational asbestos exposure. The asbestos exposure information resulted from assessment by an occupational health expert. [AaS=Aalborg shipyard; DAF=Danish asbestos cement product factory]

Number of cases	Workplace	Type of work
3	DAF	Manufacture of asbestos cement products
1	AaS	Cleaning duties
2	Laboratory	Laboratory work that involved daily use of small amounts of asbestos
1	Sacks Rental Company	Handling sacks that were rented to DEF and used for asbestos transport
1	Occupational asbestos exposure not specified	Occupational asbestos exposure not specified

Table 2. Employment data for the relatives of the malignant mesothelioma patients. The asbestos exposure information were either self-reported or resulted from assessment by an occupational health specialist; all information was checked by use of the Danish Registries. [AaS= Aalborg shipyard; DAF=Danish asbestos cement product factory.]

Number of cases	Relatives workplace or employment type
11	AaS
11	DAF
1	DAF and AaS
2	Electrician
8	Construction worker
1	Car mechanic
1	Engineer
2	Insulator
2	Worker at pipe factory
1	Worker installing asbestos roof

Duration of asbestos exposure and age at diagnosis

The median duration of the potential environmental asbestos exposure was 45 (range 1–72) years and the median time since first asbestos exposure was 61.5 (range 11–95) years. The start and duration of asbestos exposure was not registered for all the patients and thus, the MM latency could not be estimated for 37 cases ($N=7$ cases with domestic, $N=2$ cases with environmental, $N=5$ cases with occupational, $N=1$ case with domestic and environmental, and $N=22$ cases with unknown asbestos exposure). The median age at diagnosis for the occupationally and non-occupationally exposed patients was 65 (range 55–80) and 69 (range 32–95) years, respectively.

Spatial risk and mesothelioma

The heavy asbestos burden in the region of North Jutland is reflected by a crude incidence for MPM of 6.2/100 000 for men and 1.6/100 000 for women during the period 2010–2015, which is particularly high and still increasing, compared to the national average of 3.6/100 000 and 0.7/100 000 respectively (figure 2) (data from the Danish

Figure 1. Types of asbestos exposure for the 91 women with malignant mesothelioma (MM), as well as malignant pleural mesothelioma (MPM) and malignant peritoneal mesothelioma (MPeM)

Figure 2. Crude incidence rate for malignant mesothelioma (MM) for the women (black color) and both men and women (grey color) of the North Jutland region, Denmark.

Table 3. Malignant mesothelioma incidence in Danish regions and relative risk ratio (RRR) as to the North Jutland region. The incidence is calculated per 100 000 inhabitants and refers to the period 2010–2014

Danish Regions	Crude rate	RRR: North Jutland/ other Danish Region
North Jutland	1.3	
Central Jutland	0.7	1.9
South Jutland	0.7	1.9
Capital	0.6	2.2
Zealand	0.5	2.6

Cancer Registry, unpublished data from the registry of the Institute of Pathology, Aalborg University Hospital). The RR for developing MPM for the women in North Jutland in comparison to the other Danish regions was 1.9–2.6 times higher (table 3) [(16), data from NORDCAN and the DCR]. A map over the North Jutland region revealed that 20 continuous parishes with shared borders in the city area of Aalborg, within a 10 000 meter radius around the asbestos emitting enterprises, constituted a "hotspot" with 59 MM cases in total (figure 3a and 3c). Of the 59 patients, 4 were exposed to asbestos occupationally, 1 domestically, 20 environmentally, 31 had combined domestic and environmental and 3 combined occupational and environmental exposure to asbestos (appendix, table S1, www.sjweh.fi/show_abstract.php?abstract_id=3756).

The MM incidence density for women in Denmark during 1974–2015 was 0.69/100 000 person-years, while the MM incidence density for these parishes in North Denmark region during the same period ranged 0.72–7.21/100 000 person-years, with the highest incidence density recorded in parish number 1 where DAF was located (data from Statistics Denmark and the Danish Cancer Registry, Figure 3a and 3c). In that particular parish, there were a total of 11 MM cases (crude incidence density 7.21 per 100 000 person-years 95% CI 3.80–13.31, RR 10.5, 95% CI 5.5–19.4) where 2 had occupational, 6 domestic exposure history, and 3 solely environmental exposure (crude incidence density 2.0 per 100 000 person-years, 95% CI 0.51–6.26, RR 2.9, 95% CI 0.7–9.1). Apart from three outliers, all 20 environmentally exposed MM cases lived in the closest parishes to DAF and the RR for MM was increased in all these parishes (figures 3b and 3d).

Discussion

Our data suggest that environmental and domestic exposure to asbestos is the main cause of MM for women in the North Jutland region.

Figure 3. Malignant mesothelioma incidence for female residents in the parishes of the North Jutland region (a, b) and the city of Aalborg (c, d). The parishes depicted in figures 3a/3c and 3b/3d correspond to the parishes that are included in tables S1 and S2 in the appendix, respectively. The Aalborg shipyard (upper) and the Danish asbestos cement factory (lower) are depicted as blue triangles. White areas on the map have zero malignant mesothelioma cases registered in women in 1974–2015. The numbers in the map areas correspond to parishes, please see also the appendix.

Environmental asbestos exposure by residential proximity has previously been documented to increase the risk of MM (3–6, 15) and pathology studies of environmentally exposed persons had a surprisingly high mean asbestos fiber burden (17). A hotspot of 20 parishes in the North Jutland region that included or were nearest neighbors to the asbestos emitting facilities, had a higher MM incidence compared to the general Danish female population (16). The results were striking, as the highest incidence, rate and RR of MM was recorded in parish number 1 where DAF was located, and out of the 59 MM cases in the 20 parishes, only four women were solely occupationally exposed to asbestos (figure 3). There were a few isolated MM cases in other parishes in the North Jutland region. However, these parishes were spread out, not forming hotspots, most of them had few residents ($N < 900$) with only 1–2 cases per parish, making statistics unreliable. Widespread use of asbestos cement roofs in houses and farms of the countryside could account for these cases. Our study indicates that environmentally induced MM may be caused not only by living in proximity to an asbestos-emitting location, but also by living in a city or geographic area with airborne asbestos contamination due to heavy asbestos industry, with the hotspot ranging up to a 10 000-meter radius from the asbestos industry, in line with other studies on environmental exposure (3–5, 18). These findings substantiate previous research supporting that environmental exposure alone is sufficient to increase the risk of

MM (4, 5, 18). A big fraction of the non-occupationally exposed women had a history of domestic exposure (44%), that is often described in the literature (5, 18). On the contrary, the high rate of combined domestic and environmental exposure (34%) has rarely been reported and it could also represent a rationale for the high incidence of MM among women in Aalborg (19). The low rate of occupational history of asbestos exposure (9%) in spite of complete occupational history is unusual (20, 21). This discrepancy may have to do with the fact that working in the asbestos industry in Aalborg was a typical male work (male: female ratio was 13:1). With 8000 male employees at the asbestos factory, naturally thousands of women would be living with an asbestos worker and domestically exposed to dust. At the same time, these workers' families lived in the neighbourhood of the factory at some time point, and therefore an additional environmental exposure could be anticipated.

Previous research has suggested that occupational asbestos exposure, inferring more intense exposure, may predispose to MPeM while supposedly lighter asbestos exposure predominantly induces MPM (18, 22, 23). Our research indicates that the type of asbestos exposure could influence the development of either MPM or MPeM and that it should be taken into consideration when evaluating risk. However, other studies have shown that the type of asbestos exposure is not decisive for MM location and no definite conclusions can be drawn on the basis of retrospective studies (24).

Moreover, in our cohort one year's environmental exposure was enough to be associated with MM, which is consistent with the literature; a dose–effect relationship between asbestos exposure and MM has been recognized but no minimum threshold of exposure to asbestos identified (25–27). The disease latency varies from one to seven decades, but the median latency is higher than observed in previous studies (28, 29). Latency depends on exposure intensity and duration as well as unknown factors that probably are genetic (2). In low exposures, the latency increases, and in a recent article in 35 persons, exposed to asbestos domestically, the mean latency for MM was 59 years, where the offspring had a latency of 73 years (30). As the patients in our cohort had mostly a low-grade exposure over time, it is expected that the latency is higher than previously observed. Moreover median latency could depend on the time when asbestos was banned and consequently, high exposure stopped in the different countries. It is interesting, though, that even though asbestos has not been used in the North Jutland region since 1986, MM cases are still increasing (figure 2). The "new" cases could be attributed to prolonged latency due to the longstanding low-grade domestic and environmental asbestos exposure (30). Our findings highlight that clinicians need to be alert when it comes to MM and thoroughly look into asbestos exposure even several decades prior to the diagnosis.

Both domestic and environmental asbestos exposure are well-known, but often neglected risk factors for MM, also reflected by the fact that compensation is granted only to occupationally exposed MM cases in most countries (5, 25, 26, 31). In Denmark, since 2016, women living with an asbestos worker can get partly compensated. Identifying a hotspot around asbestos industries where the MM risk for women is high, as well as uncovering the major role of non-occupational exposure to asbestos in MM, empowers the notion of full compensation for the affected population.

Retrospective studies are by nature associated with methodological limitations. As such, in our study, there were limited or no data concerning the occupational history of some of the women and their relatives before 1964 and no detailed residence information before 1970. Furthermore, there were incomplete data regarding the residence and work address of seven women. Therefore, some of the MM cases that were classified as environmentally induced could have a mixed domestic or primary asbestos exposure. Due to the popularity of asbestos products in Denmark until the prohibition in 1986 (data from the Danish Occupational Health Authorities) undocumented asbestos exposure could also occur from damaged asbestos-bearing constructions or recreational use of these materials (5, 26). Reliable information about such types of asbestos exposure could not be acquired for all the patients and thus, the authors

chose not to include this potential type of asbestos exposure in the study. Lastly, rare causes of MM are erionit inhalation and heredity. The first is not a mineral used in Denmark and hereditary MM has not been reported in Denmark. In total our data are reliable, as they were collected from the Danish Cancer Registry, the SPFR and CRS that are high quality, compulsory and validated registries and complete information was available for the majority of our study population (14, 32).

In conclusion, this study strongly suggests that non-occupational asbestos exposure is the main cause of MM among women in North Jutland. A high-incidence hotspot of 20 parishes within 10 000 meter of the asbestos emitting industry was detected with increased risk of more than ten times than the rest of Denmark. Combined domestic and environmental was the single largest of the exposure groups and needs further study as well. These observations indicate that asbestos industry contamination of a large area took place and highlights the need to reevaluate the rules for compensation.

The authors declare no competing interests. This research did not receive any specific grant from funding agencies in the public, commercial, or not-for-profit sectors. Dr. Røe is in part funded by the Liaison Committee between the Central Norway Regional Health Authority (RHA) and the NTNU, grant no 46060915.

Acknowledgements

The authors would like to thank researcher Evgenios Vlachos for his contribution to the statistical analyses, the secretaries of the Department of Occupational Medicine, Aalborg University Hospital, and the employers of Aalborg University Hospital's archive for assisting in acquiring medical journals.

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Received for publication: 15 March 2018