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Aberrations of chromosome 19 in asbestos-associated lung cancer and in asbestos-induced micronuclei of bronchial epithelial cells in vitro

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Introduction

Occupational exposure to asbestos is known to induce lung cancer, and our previous studies have suggested that specific chromosomal regions, such as 19p13, are preferentially aberrant in lung tumours of asbestos-exposed patients. Here, we further examined the association between the 19p region and exposure to asbestos using array comparative genomic hybridization and fluorescence in situ hybridization (FISH) in lung tumours and FISH characterization of asbestos-induced micronuclei (MN) in human bronchial epithelial BEAS 2B cells in vitro. We detected an increased number of 19p losses in the tumours of asbestos-exposed patients in comparison with tumours from non-exposed subjects with similar distribution of tumour histology in both groups (13/33; 39% versus 3/25; 12%, P = 0.04). In BEAS 2B cells, a 48 h exposure to crocidolite asbestos (2.0 µg/cm²) was found to induce centromere-negative MN-harboung chromosomal fragments. Furthermore, an increased frequency of rare MN containing a 19p fragment was observed after the crocidolite treatment in comparison with untreated controls (6/6000 versus 1/10 000, P = 0.01). The results suggest that 19p has significance in asbestos-associated carcinogenesis and that asbestos may be capable of inducing specific chromosome aberrations.

Materials and methods

Tumour specimens

Fresh-frozen tumour specimens and paraffin-embedded samples mounted on tissue microarrays were included in the study (Table I). The samples originated from Finnish asbestos-exposed and non-exposed lung tumour patients whose exposure status had been determined based on work history and measurement of pulmonary asbestos fibre concentration (17). Asbestos fibre concentrations of 2–5 million are assumed to represent roughly a 2-fold increase in risk of lung cancer (18,19). Patients included in the non-exposed group were without exposure history, and their pulmonary fibre count was <0.5 million fibres per gram of dry lung tissue. Patients with a history of occupational exposure to asbestos and pulmonary fibre concentration exceeding 1 million fibres per gram of dry lung tissue were considered to be exposed.

The study protocols were approved by the Ethical Review Board for Research in Occupational Health and Safety and the Coordinating Ethical Review Board, Helsinki and Uusimaa Hospital District (75/E2/2001). The permission to use diagnostic samples for the research purpose was given by the National Agency for Medicolegal Affairs (4476/33/300/05), and the collection of patient information for research was permitted by the Ministry for Social Affairs and Health (STM/2474/2005).

Oligonucleotide array CGH

Array CGH was performed on 10 lung tumours, 5 from exposed and 5 from non-exposed individuals (Table I). The samples were selected for the analysis on the basis of our previous results on the AI (16) and high tumour content (>70%). Cases with differing AI profiles were chosen, aiming at detecting microlevel aberrations that could be associated with genes targeted by copy number changes. DNA extraction was performed according to the standard protocols from fresh-frozen tissue samples that were cut into 10 µm sections. Array CGH was performed using Agilent 60mer oligonucleotide-based microarray (Human Genome CGH Microarray Kit 44B). Labelling, hybridization and scanning of the arrays were done according to the manufacturer’s instructions (Agilent Technologies, Version 2).

Data analysis was performed by using CGH Analytics software (Agilent Technologies) and MATLAB 6.5 (MathWorks, Natick, MA). In this study, a minimum of three consecutive clones showing a change was considered a reliable aberration. To determine whether a tumour harboured a chromosomal aberration in chromosome 19, the array data measured from 19p were compared with 19q. Two-sided t-test was used to evaluate whether the means of the copy number data distributions of the p and q arms were the same, and an aberration was called when the P-value was <0.0001.

FISH

A bacterial artificial chromosome (BAC) RP11-333F10 obtained from Dr Mariano Rocchi (University of Bari) targeting the telomeric region of chromosomal arm 19p was used in FISH analyses. The sample set included 25 cases used in the array CGH analyses of this study or our previous study (15) and 33 additional cases (Table I)

The BAC DNA was isolated using Qiagen plasmid midi kit (Qiagen, Valencia, CA) according to the manufacturer’s recommendations with the following modifications: the volumes of buffers P1, P2 and P3 were increased to 10 ml, and the DNA was eluted using five aliquots of 1 ml buffer QF. The DNA was

Abbreviations: AI, allelic imbalance; BAC, bacterial artificial chromosome; CGH, comparative genomic hybridization; FISH, fluorescence in situ hybridization; MN, micronuclei.

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Asbestos-exposed and non-exposed BEAS 2B cells were treated with different concentrations of crocidolite fibres and in control cultures. MN are formed from chromosomal abnormalities. FISH was applied to analyze the content of MN in BEAS 2B cells treated with crocidolite fibres. Metaphase spreads were prepared according to the air-drying method, and the multicolour FISH probe mixture (24XCyte-MetaSystems, France and KREATECH Biotechnology BV, Amsterdam, The Netherlands), chromosome 9 (FISH Bright, Qbiogene, Illkirch, France and KREATECH Biotechnology BV, Amsterdam, The Netherlands), chromosomes 10 and 15 (Vysis/Abbott Molecular) and chromosome 3 (21) labelled directly with SpectrumRed-dUTP using a Vysis nick translation kit (Abbott Molecular, Downers Grove, IL). Hybridization of the probe to the fresh-frozen sections was performed using standard procedures recommended by Vysis preceded by a 2 h pretreatment in 0.01 M citrate (pH 6.0) at 80°C and a 10 min digestion at 37°C using DigestAll (Zymed Laboratories/Invitrogen Inmuno detection, San Francisco, CA). Hybridization onto tissue microarrays was performed as described previously (20) except that pretreatment and digestion were done using the same procedures as for the fresh-frozen sections.

Tumour ploidy was assessed by a combination of two to five centromeric probes using the above hybridization protocols. Probes for chromosome 2, either chromosome 2 centromere-specific probe (Vysis/Abbott Molecular, Downers Grove, IL) or satellite probe (Qbiogene, KREATECH Biotechnology BV, Amsterdam, The Netherlands), chromosome 9 (FISH Bright, Qbiogene, Illkirch, France and KREATECH Biotechnology BV, Amsterdam, The Netherlands), chromosomes 10 and 15 (Vysis/Abbott Molecular) and chromosome 3 (21) were applied.

For the detection of the hybridization signals, Zeiss Axioplan fluorescence microscope (Zeiss, Jena, Germany) equipped with a SpectrumRed filter was used. At least 75 cells were scored for each tumour sample. To identify copy number aberrations, the 19p copy number counts were divided by the estimated tumour ploidy, i.e. the average of the centromeric counts. A specimen with a signal-to-ploidy ratio ≥ 1.3 or with a signal-to-ploidy difference ≥ 0.9 was considered to carry a gain. On the contrary, a specimen with signal-to-ploidy ratio of ≤ 0.75 or with a signal-to-ploidy difference ≤ −0.9 was considered to harbour a loss.

Asbestos exposure of BEAS 2B cells

Immortalized human bronchial epithelial BEAS 2B cells were exposed to crocidolite fibres in vitro, and dual FISH was performed to determine whether the fibres induce damage preferentially in 19p. The BEAS 2B cells were obtained from the National Cancer Institute Laboratory of Human Carcinogenesis, Bethesda, MD. The cells were cultured in bronchial epithelial basal medium (BEBM; Cambrex, Walkersville, MD) supplemented with BEGM SingleQuots (Cambrex) supplements. For the exposure, 20 000 cells were plated onto Lab-Tek 2-well chamber slides (Nalge Nunc International, Naperville, IL) and maintained in the growth medium for 48 h. At 48 h, one-third of the slides were exposed to 2.0 μg/cm² crocidolite fibres, one-third to 250 ng/ml mitomycin C (positive control) and the remaining third received no exposure (negative control). All the cultures were incubated for 48 h in the presence of 9 μg/ml cytochalasin-B, which blocks the cells from performing cytokinesis after nuclear division (22). After the incubation period, the slides were washed in phosphate-buffered saline, fixed in absolute methanol, air-dried and stored at −20°C.

Multicolour FISH

The non-treated BEAS 2B cells were karyotyped by multicolour FISH on metaphase spreads to confirm that significant structural and numerical chromosomal abnormalities were not present and that the cell line had two copies of chromosome 19. Metaphase spreads were prepared according to the air-drying method, and the multicolour FISH probe mixture (24XCyte-MetaSystems 24-colour kit, with B-tect kit; MetaSystems GmbH, Altlussheim, Germany) was used as recommended by the manufacturer.

Locus-specific FISH in BEAS 2B cells

FISH was applied to analyze the content of MN in BEAS 2B cells treated with crocidolite fibres and in control cultures. MN are formed from chromosomal fragments or whole chromosomes that lag behind during cell division (23). Two DNA probes were used: the BAC probe RP11-333F10 (labelled directly with SpectrumRed-dUTP using a Vysis nick translation kit) identifying 19p telomere used also in the studies of tumour specimens and a Human Chromosome Pan-Centromeric probe (Cambio, Cambridge, UK), which labels all human centromeres. The two probes were hybridized to the target simultaneously to allow separation of chromosomal fragments (no centromere signal present) from whole chromosomes (centromere signal present). FISH was performed similarly as described previously for lymphocytes (24). After overnight incubation, the slides were washed in 2× saline-sodium citrate and 50% formamide in 2× saline-sodium citrate at 37°C.

MN analyses

The experiments were repeated twice, and in each experiment the frequency of MN was scored from 1000 binucleated cells for each treatment (crocidolite, mitomycin C and control; making a total of 2000 binucleated cells per treatment) by one scorer on coded slides using Zeiss Axioplan fluorescence microscope. The frequency of MN was determined using a ×40 objective and 4’,6-diamidino-2-phenylindole filter. MN containing whole chromosomes (centromere positive, C+) and chromosomal fragments (centromere negative, C−) were separately recorded using the fluorescein isothiocyanate filter and a ×100 objective.

To evaluate the role of chromosome 19 in crocidolite-induced MN in binucleate BEAS 2B cells, 169–206 MN per treatment were classified for having 19p fragment (only 19p signal), whole chromosome 19 (both 19p and centromeric signals), another fragment (no signal) or another whole chromosome present) from whole chromosomes (centromere signal present). FISH was performed similarly as described previously for lymphocytes (24). After overnight incubation, the slides were washed in 2× saline-sodium citrate and 50% formamide in 2× saline-sodium citrate at 37°C.

Table I. Characteristics of lung cancer patients and lung tumours studied by array CGH and FISH

<table>
<thead>
<tr>
<th>Variable</th>
<th>Oligonucleotide array CGH</th>
<th>FISH</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Asbestos exposed (n = 5)</td>
<td>Non-exposed (n = 5)</td>
</tr>
<tr>
<td>Gender</td>
<td>Male/female</td>
<td>5/5</td>
</tr>
<tr>
<td>Asbestos fibre count</td>
<td>8.4 (5.9–35)</td>
<td>0.0 (0.0–0.0)</td>
</tr>
<tr>
<td>Histology</td>
<td>AC</td>
<td>2</td>
</tr>
<tr>
<td></td>
<td>SCC</td>
<td>2</td>
</tr>
<tr>
<td></td>
<td>LCLC</td>
<td>—</td>
</tr>
<tr>
<td></td>
<td>SCLC</td>
<td>1</td>
</tr>
<tr>
<td></td>
<td>Others</td>
<td>—</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Variable</th>
<th>Oligonucleotide array CGH</th>
<th>FISH</th>
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<tr>
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<td>Asbestos exposed (n = 5)</td>
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<td></td>
<td>SCC</td>
<td>2</td>
</tr>
<tr>
<td></td>
<td>LCLC</td>
<td>—</td>
</tr>
<tr>
<td></td>
<td>SCLC</td>
<td>1</td>
</tr>
<tr>
<td></td>
<td>Others</td>
<td>—</td>
</tr>
</tbody>
</table>

aPulmonary asbestos fibre count in million per gram of dried lung.

bAC, adenocarcinoma; SCC, squamous cell carcinoma; LCLC, large cell carcinoma; SCLC, small cell carcinoma.

Others include two adenosquamous carcinomas and one pleomorphic carcinoma.

Results

19p alterations in lung tumour specimens

Fine-resolution oligoarrays were performed on 10 lung tumours, 5 from exposed and 5 from non-exposed individuals, to reveal both microlevel and larger chromosomal aberrations, which occur at the 19p region. No aberrations <1 Mb were detected in 19p. Aberration breakpoints were, however, detected at the centromeric region of chromosome 19 in 80% (4/5) of the exposed and 40% (2/5) of the non-exposed cases. These aberrations commonly involved either loss or gain of the whole arm of chromosome 19. The complementary DNA microarray data of Nymark et al. (15) from additional seven exposed and nine non-exposed lung tumour patients were also analyzed for chromosome 19 aberrations. In the combined data set, a centromeric aberration was detected in 67% (8/12) of the exposed and 36% (5/14) of the non-exposed cases. The loss of 19p or chromosome 19 was more common in tumours of the asbestos-exposed patients (42%; 5/12) than in those of the non-exposed patients (14%; 2/14). The results are summarized in Table II. Although suggestive, the differences were not statistically significant (Fisher’s exact test).

The putative association between asbestos exposure and loss of 19p was further analyzed by FISH using a BAC probe targeting the...
telomeric region of 19p on 58 lung tumours. As the microarray results indicated that the whole chromosome arm was usually involved in the aberration, this analysis was probably representative also for 19p13, which was previously found to be significantly associated with asbestos exposure by us (16). We also determined the ploidy levels for the tumour specimens using two to five centromeric probes as ploidy alterations are detected in ~45% of lung adenocarcinomas and squamous cell carcinomas (25). To detect specimens with copy number losses and gains, the 19p copy numbers were scaled against the estimated tumour ploidy, i.e. the average of the centromeric counts. Losses were detected in 39% (13/33) of the exposed and in 12% (3/25) of the non-exposed cases (P = 0.04; Fisher’s exact test). Interestingly, gains were more common in the non-exposed than exposed (36%; 9/25 versus 18%; 6/33), but the difference was not statistically significant. The results are summarized in Table III. The aberration frequencies were observed to vary between different histological tumour types with losses being most common in small cell carcinomas and large cell carcinomas and gains in squamous cell carcinomas (Table III). However, thorough comparisons of the subtypes are not possible due to limited number of samples analyzed.

**MN induced by crocidolite**

To assess whether asbestos fibres are capable of inducing chromosomal damage in 19p in vitro, human bronchial epithelial BEAS 2B cells were exposed to crocidolite. Prior to the treatment, multinucleated cells were used. FISH analysis was performed to ascertain that the BEAS 2B cell line harboured two copies of chromosome 19. In the 12 analyzed metaphases, chromosome 19 was present in two copies, and no rearrangements affecting the chromosome were observed. However, recurrent translocations and copy number changes affecting autosomes 3, 9, 14, 15, 20 and 22 were observed in at least one of the studied metaphases (data not shown).

Crocidolite significantly (P = 0.008) increased the frequency of binucleate cells with C− MN, with a 1.7-fold difference to the control cultures (Figure 1). Thus, our results indicated that crocidolite induces MN-harbouiring chromosomal fragments. The positive control substance, mitomycin C, induced a 4.8-fold increase in C− MN (P < 0.001), in accordance with its well-known clastogenicity. The frequency of binucleate cells with C− MN (harbouring whole chromosomes) remained fairly similar between the different treatments.

Figure 2 shows examples of micronucleated binucleate cells stained with the dual FISH technique used to evaluate the presence of 19p signals and centromeric signals in MN. Fragments containing 19p DNA (only 19p signal present) were detected, among C− MN of binucleate cells, in 3.4% (6/176) in the crocidolite treatment, 1.4% (2/139) in the mitomycin C treatment and 0.6% (1/159) in the untreated control cultures (Table IV). Thus, the proportion of MN containing a 19p fragment was 5.4 times higher in crocidolite-treated cells than in the unexposed control, but the difference was not quite statistically significant (P = 0.079; Fisher’s exact test). When the total number of cells scored to obtain the numbers of MN shown in Table IV were considered, the frequency of cells harbouring MN with a 19p fragment was ~10 times higher in the crocidolite-treated cultures (~6000 cells scored) in comparison with the untreated controls (~10 000 cells scored; P = 0.01, Fisher’s exact test), despite the total frequency of cells with C− MN was only doubled. Assuming that chromosomal breakage occurs randomly, relative to the length of each chromosome, the expected contribution of 19p to chromosomal breakage can be calculated from the physical lengths of human chromosomes (26). The observed 3.4% prevalence of 19p fragment in C− MN in crocidolite-treated cultures was 3.6 times higher than the expected value (0.945%), but the difference was not statistically significant (Fisher’s exact test). Cells with MN harbouring a whole chromosome 19 (both 19p and C+ signals) were rare (Table IV).

**Discussion**

In this study, array CGH and FISH on lung tumour samples and MN test in BEAS 2B cells were performed to assess 19p aberrations in asbestos-related carcinogenesis. Studies of 19p were motivated by our recent findings, which implied that AI of 19p was significantly associated with asbestos exposure in lung cancer (16). However, as both allelic gains and losses may underlie AI, FISH and array CGH analyses were now carried out to reveal which type of aberation was related to asbestos exposure.

The present characterization specified that centromeric breaks in chromosome 19, and especially the loss of 19p, may be related with asbestos exposure. Here, copy number changes of 19p could also be distinguished from ploidy alterations, which is not possible in typical

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**Table II.** Array CGH results for lung tumour samples from 12 asbestos-exposed and 14 non-exposed patients regarding chromosome 19

<table>
<thead>
<tr>
<th>Aberration</th>
<th>Subjects</th>
<th>Asbestos exposed, n (%)</th>
<th>Non-exposed, n (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Centromeric aberration</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Break</td>
<td>8 (67)</td>
<td>5 (36)</td>
<td></td>
</tr>
<tr>
<td>No break</td>
<td>4 (33)</td>
<td>9 (64)</td>
<td></td>
</tr>
<tr>
<td>19p aberration</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>19p loss</td>
<td>4 (33)</td>
<td>1 (7)</td>
<td></td>
</tr>
<tr>
<td>Chromosome 19 loss</td>
<td>1 (8)</td>
<td>1 (7)</td>
<td></td>
</tr>
<tr>
<td>19p gain</td>
<td>0 (0)</td>
<td>0 (0)</td>
<td></td>
</tr>
<tr>
<td>Chromosome 19 gain</td>
<td>0 (0)</td>
<td>3 (21)</td>
<td></td>
</tr>
</tbody>
</table>

**Table III.** 19p aberrations in lung carcinomas determined by FISH

<table>
<thead>
<tr>
<th>Variable</th>
<th>Subjects with</th>
<th>19p loss, n (%)</th>
<th>19p gain, n (%)</th>
<th>No change in 19p, n (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Exposure status</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Exposed</td>
<td>13 (39)</td>
<td>6 (18)</td>
<td>14 (42)</td>
<td></td>
</tr>
<tr>
<td>Non-exposed</td>
<td>3 (12)</td>
<td>10 (40)</td>
<td>12 (48)</td>
<td></td>
</tr>
<tr>
<td>Histologyb</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>All</td>
<td>16 (28)</td>
<td>16 (28)</td>
<td>26 (45)</td>
<td></td>
</tr>
<tr>
<td>AC</td>
<td>5 (22)</td>
<td>8 (35)</td>
<td>10 (43)</td>
<td></td>
</tr>
<tr>
<td>SCC</td>
<td>3 (17)</td>
<td>6 (33)</td>
<td>9 (50)</td>
<td></td>
</tr>
<tr>
<td>LCLC</td>
<td>4 (57)</td>
<td>0 (0)</td>
<td>3 (43)</td>
<td></td>
</tr>
<tr>
<td>SCLC</td>
<td>4 (57)</td>
<td>0 (0)</td>
<td>3 (43)</td>
<td></td>
</tr>
<tr>
<td>Others</td>
<td>0 (0)</td>
<td>2 (67)</td>
<td>1 (33)</td>
<td></td>
</tr>
</tbody>
</table>

*P = 0.04, in comparison with non-exposed cases; Fisher’s exact test.

bSee Table I footnotes for definitions of the histological tumour types.
Crocidolite (2.0 μg/cm²) was found to induce a significant increase in the frequency of MN harbouring 19p fragment in comparison with the untreated control culture, further indicating that the 19p alterations may be related to asbestos exposure. The finding suggests that loss of 19p occurs early in asbestos-related carcinogenesis. Although the frequency of cells that had lost 19p to MN was rather low (1/1000 cells) after the 48 h asbestos treatment, loss of 19p is likely to increase if this alteration resulted in growth advantage.

Earlier studies support the ability of asbestos fibres to induce chromosomal aberrations (44–47). However, if damage occurred randomly, asbestos-related aberration hot spots should not be observed. Yet, previous in vitro studies have shown that asbestos fibres cause breaks in chromosomes 1 and 9 (41–43). Furthermore, lung tumours of asbestos-exposed patients have been suggested to show preferentiality for aberrations of distinct chromosomal loci (15,16,48), including regions in chromosomes 1, 9 and 19 (15). The apparent non-random targeting of distinct chromosomal loci could indicate that some of the driver genes may be different in the carcinogenic process, if influenced by asbestos. Although this study further indicated that 19p appears a likely preferential site of asbestos-induced aberrations, further studies are needed to confirm the relevance of 19p and the other putative asbestos-related aberration hot spots in lung carcinogenesis.

In summary, our studies suggest that asbestos exposure has importance in causing chromosomal aberrations in chromosome 19 and especially losses of 19p. Knowledge of specific features involved in asbestos-related lung carcinogenesis is a prerequisite for improved diagnosis as well as treatment of the disease, thus the role of 19p should be carefully examined.

Table IV. Percentage of MN in BEAS 2B cells found to harbour 19p signals by FISH

<table>
<thead>
<tr>
<th>Treatment</th>
<th>MN scored, n (%)</th>
<th>MN with 19p signals among</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>C⁺ MN</td>
</tr>
<tr>
<td>Untreated control</td>
<td>177</td>
<td>5.6% (1/18)</td>
</tr>
<tr>
<td>Crocidolite (2.0 μg/cm²)</td>
<td>206</td>
<td>3.3% (1/30)</td>
</tr>
<tr>
<td>Mitomycin C (250 ng/ml)</td>
<td>169</td>
<td>0.0% (0/30)</td>
</tr>
</tbody>
</table>

MN with (C⁺) and without (C⁻) centromere signals were analyzed separately.

AI assays, such as the one used by us (16), Indeed, ~20% of our AI cells appeared to be caused by a change in ploidy, whereas the remaining 80% were derived from both gains and losses.

Differences in aberration frequencies were observed between different histological tumour types, with losses being most common in small cell and large cell carcinomas. As regions that are preferentially aberrant in specific histological tumour types have been suggested in most chromosomes, the differential aberration rates detected here are not surprising (27–31). In accordance with our findings, these studies have also shown that both gains and losses occur in the 19p region (27–31) with differing frequencies across tumour types (27,30,31).

Interestingly, AI of 19p has been shown previously to be associated with smoking in lung adenocarcinomas (32). Furthermore, mutations of the presumed target gene of 19p, LKB1 (33), have been reported to occur preferentially in lung tumours of smoking males (34). Also other important differences, such as preferentiality for mutations in either K-RAS or EGFR, have been recognized between adenocarcinomas of smokers and non-smokers, respectively ([35–37], reviewed in ref. 38). The frequent 19p losses in tumours of exposed patients found in this study indicate that asbestos exposure, in addition to smoking, could play an important role in driving aberrations of this region. In accordance with our finding, monosomy of chromosome 19 has been suggested to have relevance in asbestos-induced tumorigenic transformation of human bronchial epithelial BEP2D cells (39).

Previously, asbestos fibres have been shown to induce MN in different types of cells, and labelling of MN with kinetochore antibodies has demonstrated that asbestos produces MN both via clastogenic and aneugenic mechanisms (40–43). In the present study, asbestos-induced MN were for the first time characterized by pan-centromeric FISH. Crocidolite was found to induce a significant increase in the frequency of MN harbouring 19p fragment in comparison with the untreated control culture, further indicating that the 19p alterations may be related to asbestos exposure. The finding suggests that loss of 19p occurs early in asbestos-related carcinogenesis. Although the frequency of cells that had lost 19p to MN was rather low (1/1000 cells) after the 48 h asbestos treatment, loss of 19p is likely to increase if this alteration resulted in growth advantage.

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References

Abberrations of chromosome 19 in asbestos-associated lung cancer


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