545 in 1994, thereby surpassing the number of mesotheliomas ($n=350$ in 1992 and $n=495$ in 1994). For 1999, some 776 cases of lung + laryngeal cancer were classified as asbestos-related in comparison to 617 mesotheliomas. This ratio (1.26:1) corresponds to the proportions of excess lung cancer cases and mesotheliomas observed in cohort studies (see Table 1).[36,238]

Further data on the German system of dose estimation have been reported[239] for 3498 male lung cancer cases in comparison to 3541 population controls, in a pooled analysis based on two sub-studies[240,241] (see also Jöckel *et al.*[242]). A detailed smoking and occupational history was obtained by a personal standardised interview where asbestos exposure was assessed on the basis of 17 job-specific supplementary questionnaires in a semi-automated fashion. Ever exposure to asbestos after adjustment for smoking was associated with an OR_{LCA} of 1.41 (95%CI = 1.24–1.60), and a clear dose-response relationship with an OR_{LCA} of 1.79 (95%CI = 1.39–2.30) was found for >2500 days of exposure. For a sub-sample of 301 cases and 313 controls, estimates of fibre-years of exposure based on the convention of the *Berufsgenossenschaften*[215] were performed by two experts. In a logistic regression model adjusted for smoking and stratified for age and origin of the patients, the OR_{LCA} was associated with log (fibre-years + 1); 25 fibre-years corresponded to an OR_{LCA} of 1.99 (95%CI = 1.20–3.30). In a two-phase case-referent study, Pohlabeln *et al.*[243] derived results 'consistent with a doubling of the lung cancer risk with 25 fibreyears asbestos exposure'.

In an analysis of two German case-referent studies, Hauptmann *et al.*[89] found that the OR_{LCA} was 1.8 (95%CI = 1.2–2.7) for subjects who had worked for 3–7 years in a job with potential exposure to asbestos, and was 2.4 (95%CI = 1.7–3.4) for those who worked in similar jobs for ≥8 years, in comparison to never-exposed subjects.

ASBESTOS FIBRE CONCENTRATIONS IN LUNG TISSUE, ESTIMATED CUMULATIVE EXPOSURE, AND THE RISK OF LUNG CANCER

In The Helsinki Criteria,[102] the following lung tissue concentrations were delineated to identify workers with a high probability of exposure to asbestos in the workplace:

(a) >1000 ABs/g dry lung (equivalent to >100 ABs/g wet lung);
(b) >100 000 amphibole fibres >5 μm in length/g dry lung;
(c) >1 000 000 amphibole fibres >1 μm in length/g dry lung;
(d) >1 AB/mL BAL fluid.

Each laboratory should establish its own reference values, and the median values of those exposed occupationally should be substantially above the reference values. Besides other criteria (discussed also in The Helsinki Criteria), a lung fibre count exceeding this background range should be sufficient for probabilistic attribution of mesothelioma to asbestos exposure.

The basis for these concentrations of ABs and asbestos and amphibole fibres is tabulated in a review by Tossavainen,[17] for lung tissue samples and BAL fluid from the general population or from patients not exposed in the workplace. Different fibre definitions, different measuring methods and different statistical parameters complicate comparison of these data. In Fig. 2A–C (data for BAL fluid not shown), the data are presented as the percentage of measurements below a certain concentration value according to the following rules:

(i) Geometric mean and median values: <50%
(ii) Arithmetic mean values: <70%
(iii) Upper limit of the range: <100%

If several of these parameters were given for a series of measurements, they are presented side by side.

With the exception of two series of mesothelioma patients, the median values of the concentrations of short and long amphibole fibres and ABs ranged below the limit values given by The Helsinki Criteria. In most of the studies, less than 20% of the measured values exceed these limits. An increased percentage of counts exceeding the limits is observed for short amphibole fibres among Australian and, probably, Japanese patients. For ABs, an increased percentage is observed for one of the French and the Belgian series, as well as for Canadian patients living near the Quebec mines.

In a German mesothelioma case-referent study, 15% of 66 hospital referents who underwent lung resections mainly for lung cancer exceeded the limit value for long amphibole fibres (length >5 μm), in comparison to about 70% of the cases.[244,245] The same percentages of measurements above the delimiting value were obtained for short fibres (length >1 μm). AB counts were also available for 147 referents and 66 cases: the limit value of 100 ABs/g wet lung (≅1000 ABs/g dry) was exceeded for 18% of the referents in comparison to 73% of the cases, and this percentage for referents diminished to 8.7% when evaluation was restricted to 69 unexposed referents.

In a mesothelioma case-referent study on patients from Yorkshire,[246] the concentration of total amphibole fibres longer than 0.5 μm was measured. Twenty-two per cent of 122 referents exceeded the limit value in comparison to 80% of 147 cases; when evaluation is restricted to referents not exposed occupationally to asbestos according to the judgement of surviving relatives ($n=61$; Table 4 in Howel *et al.*[246]), the percentage is slightly less than 20% (Fig. 1 in Howel *et al.*[246]). For controls and workers from the textile factory in South Carolina, fibres were counted at a magnification of ×20 000 without specification of a minimum fibre length.[86] Among 31 controls, the delimiting value for amphibole fibres >1 μm in length was exceeded for 9.7% of the tremolite counts, 6.4% of the anthophyllite counts and 12.9% of the amosite and crocidolite counts. It may be assumed that some of these counts were obtained from the same patients.

In a study of 33 patients from Texas with no history of occupational exposure to asbestos, Dodson *et al.*[247,248] found that all had no more than 20 ABs/g wet lung and 26 had no detectable ABs; chrysotile was undetectable in 19 cases, and 10 of the 33 had no asbestos fibres within the detection limits of the study (the total uncoated asbestos fibre burden was in the range of 0–290 000 fibres/g dry, for fibres >0.5 μm with an aspect ratio of ≥3:1). Although amosite and crocidolite fibres were found occasionally,

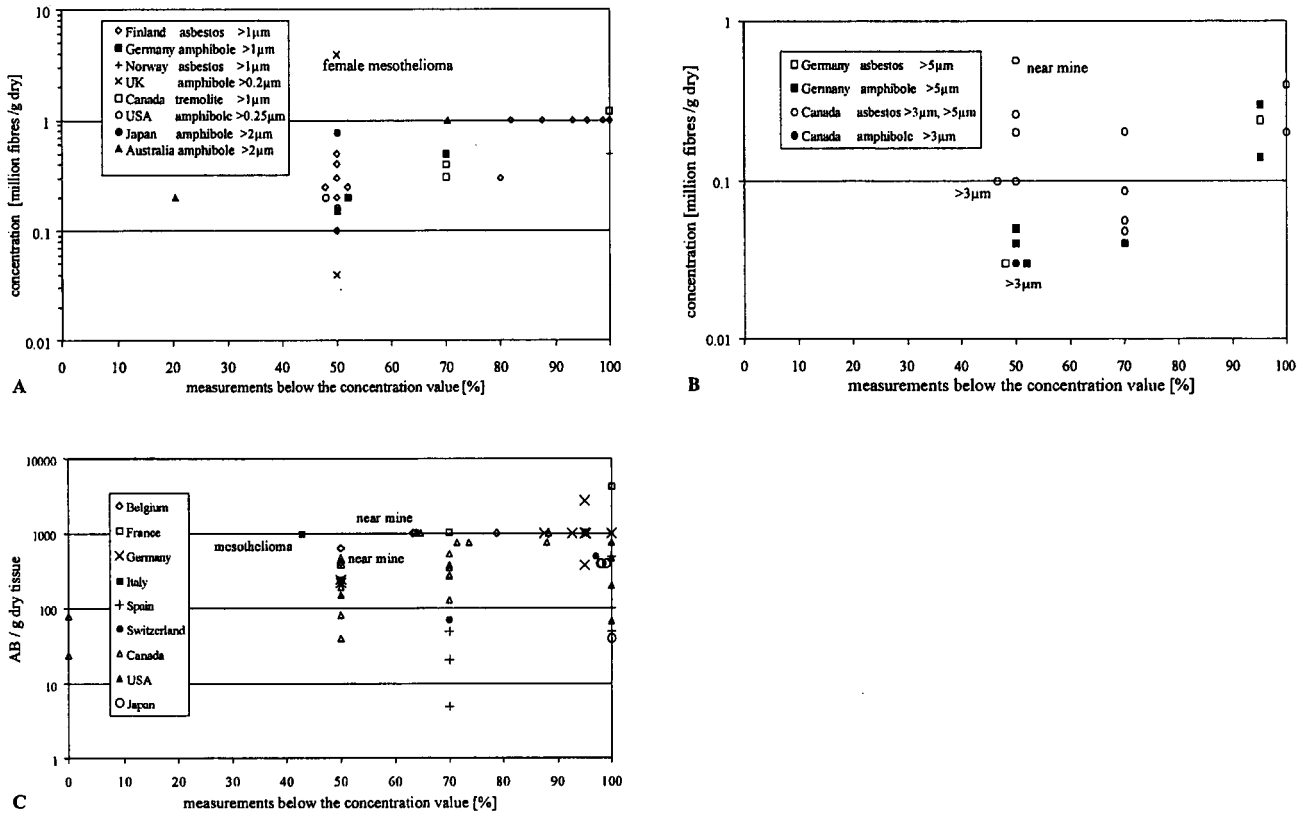

Fig. 2 (A) Amphibole fibres longer than a minimum value of 0.2–2 µm in lung tissue samples from the general population or from patients not exposed at the workplace.[17] Measurements from Finland and Norway represent asbestos fibres but because of scanning electron microscopy (SEM) was used at a magnification of × 5000, predominantly amphibole fibres were registered. In the German measurements, fibres originally were counted if they were longer than 0.3 µm but with the magnification (× 10 000) of this study, only very few fibres shorter than 1 µm were recorded. (B) Asbestos and amphibole fibres longer than a minimum value of 3–5 µm in lung tissue samples from the general population or from patients not exposed in the workplace.[17] (C) Asbestos bodies in lung tissue samples from the general population or from patients not exposed in the workplace.[17].

they were few in number: anthophyllite (12 of 33 cases) was almost as likely.

It is also notable that in mesothelioma case-referent studies,[86,245,249–251] increased ORs are found at fibre concentrations immediately above the delimiting values for occupational exposure given in The Helsinki Criteria. In comparison to a reference group for whom the tissue concentration was less than 50 000 fibres/g dry lung, Rödelsperger *et al.*[245] found that the OR for mesothelioma (OR_{MESO}) increased in an almost linear fashion according to the relationship:

$$OR_{MESO} = \frac{\text{Concentration of amphibole fibres longer than 5 µm/g dry lung}}{\text{25 000 fibres/g dry lung}}$$

In this study, a significantly increased OR_{MESO} of 4.5 (95%CI = 1.1 – 17.9) was observed, even at the low fibre concentration range between 100 000 and 200 000 fibres longer than 5 µm/g dry lung.

Roggli and Sanders[111] studied 234 cases of lung cancer with some history of asbestos exposure, but with no quantitation of exposure as fibre-years. For 70 patients with asbestosis they recorded a median total asbestos fibre concentration of 2.53 million fibres/g dry for fibres 5 µm in length or more (converted from wet weight figures), which included a median count of 2.53 million commercial amphiboles (crocidolite/amosite) and 220 000 non-commercial amphiboles, and a median count of 270 000 ABs/g dry; although this AB count is well above (18 times) the upper limit of 5000–15 000 ABs specified in The Helsinki Criteria, the uncoated fibre count is roughly comparable to the figure of 2 million in The Criteria. The Helsinki figure of 5 million fibres/g dry (for fibres > 1 µm in length) also bears comparison to the geometric mean fibre concentration of 2.5 million fibres/g dry for Western Australian asbestosis cases whose exposure occurred other than at Wittenoom.[158,159] The number of ABs in The Helsinki Criteria is about 23 times above the upper limit of the range of AB concentrations, and the uncoated fibre count is almost 79 times above the upper limit for the range of uncoated total fibres and crocidolite/amosite fibres, reported for the control group in Roggli and Sanders[111] (220 ABs/g dry and 25 400 fibres/g dry, respectively).

In 1994, Karjalainen *et al.*[109] reported a case-referent study that examined the relationship between lung fibre burden and the risk of lung cancer based on 113 surgically treated lung cancer patients in comparison to 297 autopsy referents from the Finnish population. Lung tissue fibre analysis was carried out for fibres longer than 1 µm by scanning electron microscopy (SEM) at a magnification of × 5000 and included mainly amphibole fibres. In comparison to a reference group with a tissue concentration of

less than 1 million fibres/g dry, the OR_{LCA} increased to 1.7 for concentrations in the range 1.0–4.99 million fibres/g dry and to 5.3 for concentrations of 5.0 million or more fibres/g dry. Karjalainen *et al.*[109] stated that when two cases of asbestosis and seven cases of minor 'histological fibrosis compatible with asbestosis' were excluded, an elevated OR_{LCA} was still associated with asbestos fibre concentrations of 5.0 million or more fibres/g dry lung (age-adjusted $OR_{LCA}=2.8$; 95%CI = 0.9–8.7; $P=0.07$) and for asbestos fibre counts in the range 1.0–4.99 million fibres/g dry ($OR_{LCA}=1.5$; 95%CI = 0.8–2.9; $P=0.19$). One criticism directed at this study is that the results fail to achieve significance in terms of P values, thereby proving that 'significance' lies only with the cases of fibrosis.[115] This objection overlooks the fact that the limit $P \leq 0.05$ is an arbitrary statistical convention and that reality lacks sharp boundaries of this type: what is important in this study is the trend from a low to a higher OR_{LCA} with transition from an intermediate fibre count (1.0–4.99 million) to the higher value (≥ 5.0 million). If one excludes the nine cases of fibrosis and assumes that seven were in the high fibre group (≥ 5.0 million fibres/g dry) and two were in the intermediate fibre group (1.0–4.99 million fibres/g dry),** one can calculate the crude lung cancer ORs to be 2.85 and 1.8, respectively, as consistent as possible with the age-adjusted ORs of 2.8 and 1.5 in the original paper; trend testing then yields χ^2_1 (trend) = 7.2 ($P<0.01$). In addition, it is possible from the published data to recalculate the OR for adenocarcinoma only, after exclusion of all cases with any fibrosis: assuming that all were in the high fibre group, the OR is still significantly elevated for a count >1.0 million compared with <1.0 million ($OR_{LCA}=2.65$; 95%CI = 1.11–6.26; $P<0.001$).[1]

Much steeper dose–response relationships were obtained from mesothelioma case-referent studies;[86,245,249–251] e.g., Rödelsperger *et al.*[245] calculate the mesothelioma OR to be about 100 when patients with a burden of 2.5 million amphibole fibres/g dry (for fibres longer than 5 μm) are compared with the reference group.

In assessing the significance of asbestos lung fibre burdens for attribution of lung cancer, it should be emphasised that the 'controls' for case-referent studies represent individuals without the disease in question, sampled randomly and independently of exposure.[29,31] This is a critical necessity for the validity of a case-referent study. Thus, the 'control' group will generally comprise both exposed and unexposed individuals. In using data from 'control' groups in case-referent studies for assessing likely lung fibre levels in the unexposed in comparison to those exposed, only data from the unexposed fraction of the 'controls' should be used.

Estimates of cumulative exposure as fibre-years apply equally to all types and mixtures of asbestos. In contrast, fibre analysis of lung tissue applies mainly to amphiboles because of the lower biopersistence of chrysotile in lung tissue.[54,252,253] Therefore, the concentrations of asbestos and amphibole fibres that correspond to 25 fibre-years of exposure are largely dependent on the proportion of amphiboles in the relevant asbestos-containing materials.

**Based upon an assumption that the clinical asbestosis cases were in the heaviest exposure group and that the mild histological fibrosis cases were in the intermediate exposure group.

From historical national data on the consumption of the different types of asbestos and the known composition of various products (e.g., asbestos-cement products), there is abundant evidence that chrysotile comprised about 94–95% or more of asbestos consumption, and amphiboles about 5% or less.[54,254,255] However, in some industries—e.g., workers at the Nottingham gas mask factory[256] and the Wittenoom crocidolite miners/millers in Western Australia[257]—the exposures involved a far higher proportion of amphiboles (notably crocidolite for both of these industries, so that exposure at Wittenoom unaffected by other exposures was to virtually 100% crocidolite). It follows that for these workers, much higher tissue concentrations of amphibole fibres are equivalent to an exposure of 25 fibre-years than for those exposed to a small percentage of amphibole fibres during their lives.

Table 6 gives summary estimates of lung tissue concentrations of amphibole fibres and ABs that may be related to a cumulative exposure of 25 fibre-years. As expected, the concentrations increase according to the percentage of the amphibole used, so that the smallest amount is encountered among 38 workers from the South Carolina textile plant.[86]

In the South Carolina textile industry, chrysotile contaminated with less than 1% tremolite was the only type of asbestos processed as raw material, besides a small amount of crocidolite yarn. The concentrations of asbestos fibres of all lengths (without a specified minimum length) per gram dry lung were compared with individual fibre-years, which were available for the same patients from an extensive industrial hygiene survey.[260] Roughly 40 million asbestos fibres/g dry lung correspond to an exposure of 25 fibre-years, but this result is influenced by a high number of small chrysotile fibres; nevertheless, the quantity of amphibole fibres may be estimated to be 4.5 million fibres/g dry lung using geometrical mean values given for the single types of asbestos (Table 3 in Green *et al.*[86]). Figure 3 in this paper represents the relationship between tremolite as the main type of amphibole fibre and estimated fibre-years of exposure, and shows concordance with The Helsinki Criteria.

Somewhat greater amounts of amphiboles may be expected for the cases and controls in Rödelsperger *et al.*[194,244] and for the cohort reported by Albin *et al.*[258,259] However, Rödelsperger[244] reported that: 'A relationship is demonstrated between asbestos fibre dose estimated from the interview and concentration of amphibole fibres from lung tissue analysis. From this a dose of 25 fibre-years corresponds to an amphibole fibre concentration of 2 fibres/μg' (in other words, 2 million amphibole fibres/g dry lung for fibres longer than 5 μm; abstract and p. 307).

In Rödelsperger's study on mesothelioma patients,[244] 25 fibre-years and the count of 2 million uncoated fibres/g dry lung corresponded roughly to an AB count of 15 000/g dry lung given in The Helsinki Criteria (see also Thimpont and De Vuyst[233]); for obvious reasons, these values could not be derived for the control patients.

By far the largest amount of amphibole is expected for 90 crocidolite miners/millers from Wittenoom. A strong correlation between analysis of the lung burden and the estimate of fibre-years was observed.[257,261] For these workers, concentrations of 21 000 ABs/g wet lung and

TABLE 6 Concentrations of amphibole fibres and ABs from fibre analysis of lung tissue, relative to an estimated exposure of 25 fibre-years from occupational histories

Study	Patients	Exposure		Lung tissue fibre analysis	Concentration related to 25 fibre-years		Remarks
		Type of asbestos	Fibre-years estimate by (ref)		Million f/g dry	ABs/g wet	
258	Swedish asbestos cement factory: 76 workers	More than 85% chrysotile; up to 4% crocidolite until 1966; up to 17% amosite before 1956	259	TEM; fibres of all lengths	Asbestos: 189 Amphibole: 55 Asbestos: 96 Amphibole: 9		Seven mesothelioma cases, from median values Sixty-nine other workers, from median values
86	South Carolina asbestos textile factory: 54 workers	Chrysotile with <1% tremolite; very little crocidolite (difference in consumption >4000:1)	260	TEM; fibres of all lengths	Asbestos: 40 Amphibole: 4.5		See Fig. 1; from geometrical mean values of Table 3 in original, the ratio of amphibole to all asbestos fibres is ~9:1
244 194	Germany: 66 mesothelioma cases; 66 and 147* controls respectively with lung resection	Mixed, according to consumption of ~94% chrysotile in Germany	194	TEM; fibres >5 μm in length	Amphibole: 2	1500	Sixty-six cases and 66 (147*) controls: comparison of different types by regression analysis
261	Wittenoom: 32 miners/millers	Almost 100% crocidolite	262	LM		4400	
257	Wittenoom: 90 miners/millers	Almost 100% crocidolite	262	TEM; length >0.4 μm	Crocidolite: 220	21	From geometrical mean values, Fig. 1 in original. The AWARD Criteria specify a count of 100 million crocidolite fibres longer than 1 μm to correspond to 25 fibres/mL-yr.[225]

ABs, asbestos bodies; Ref, reference; TEM, transmission electron microscopy.
*ABs only counted by light microscopy, per gram wet lung.

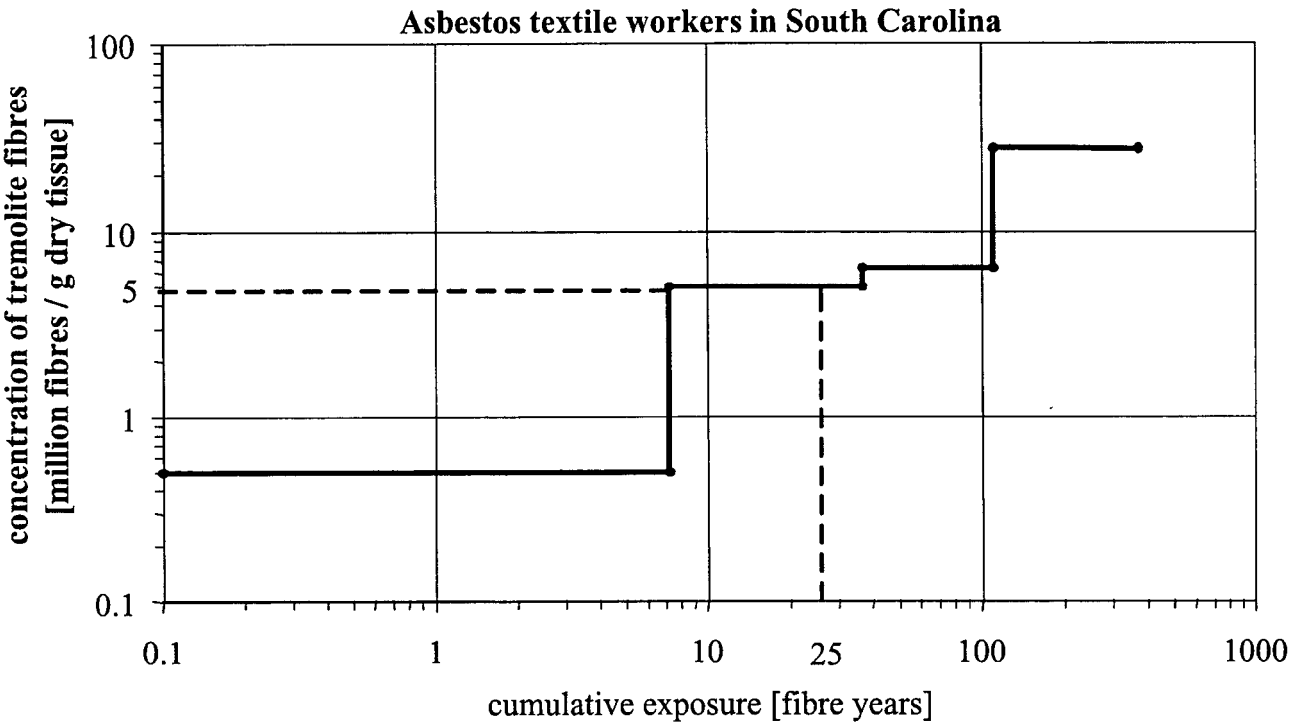

Fig. 3 Relationship between the concentration of tremolite fibres in the lung tissue and the estimate of the fibre-years for 39 textile workers from the cohort from South Carolina, after Table 5 in Green *et al.*[86].

220 million crocidolite fibres longer than 0.4 μm/g dry lung (~100 million fibres longer than 1.0 μm)[225] correspond to an exposure of 25 fibre-years. These concentrations are respectively 20- and 45-fold greater than the AB and fibre concentrations specified by The Helsinki Criteria. They support the proposition that the percentage of amphiboles used in the workplace is crucial if the concentration of asbestos fibres in the lung tissue forms the basis for estimation of fibre-years of cumulative exposure.

LUNG CANCER AND THE CLASTOGENICITY AND MUTAGENICITY OF ASBESTOS

Detailed discussion of the molecular and genetic aberrations inducible by asbestos in experimental animals and cultured cell lines lies outside the scope of this review (see references 1, 96, 167, 263–266). However, asbestos is known to be genotoxic and clastogenic, with the capacity to induce DNA strand breaks, anaphase-telophase abnormalities and sister chromatid exchanges in cell lines *in vitro*—where fibrosis cannot be implicated—and free radicals generated from the surface of asbestos fibres or macrophages are implicated in these aberrations. Both crocidolite and chrysotile have been shown to disturb cell division, producing binucleated cells, which may lead to aneuploidy or polyploidy.[267] Asbestos fibres can also induce oncogene expression—such as *c-fos* and *c-jun* proto-oncogenes—in cultured rodent mesothelial cells.[268] Asbestos-related adenocarcinoma of lung is also associated with *p53* and *k-ras* mutations.[96,265,269–272]

In a study of 84 male patients with a histological diagnosis of adenocarcinoma of lung, Nelson *et al.*[272] found a higher prevalence of *k-ras* mutations in those with a history of asbestos exposure than in those without, after adjustment for age and pack-years smoked, and that the estimated intensity of exposure was greater for the patients with *k-ras* mutations than those without. There was no detectable association with the duration of exposure, but the time since first exposure was associated with mutation status; in addition, the association was not dependent on radiographic evidence of asbestos-related disease. Nelson *et al.*[272] concluded that their data were suggestive of an increased likelihood of *k-ras* codon 12 mutations as a consequence of asbestos exposure and that 'this process occurs independently of the induction of interstitial fibrosis'.

Wang *et al.*[273] have also reported that chrysotile and cigarette smoke in solution act synergistically to produce DNA damage in a dose-dependent fashion and to activate *c-ras* in human embryo lung cells as assessed by *p21* expression. Jung *et al.*[274] found that amosite and cigarette smoke each produced an increase in DNA strand breaks and necrosis in rat bronchiolar epithelial cells *in vivo*, both alone and in additive fashion when in combination.

Using a papillomavirus-immortalised human bronchial epithelial cell line, Hei *et al.*[275] found that a single 7-day treatment of the cells with chrysotile induced stepwise transformation, with altered growth kinetics, resistance to terminal differentiation and anchorage-independent growth, to produce progressive tumorigenic growth in nude mice.[276] Hei *et al.*[277] also found that treatment of the same cell line with α-particles to simulate the effects of radon, induced a similar pattern of apparent neoplastic transformation in the same cell line. The same group of researchers[278] had shown earlier that chrysotile is

mutagenic for cultured mammalian cells—with the production of large deletions—and comparable with the mutagenicity of γ-rays.

The fragile histidine triad (FHIT) tumour suppressor gene located at 3p14.2[279–283] appears to represent a site of genomic fragility relevant to carcinogenesis: FHIT protein is expressed in most non-neoplastic tissues, and the highest levels of expression occur in epithelial cells. FHIT appears to be subject to deletion or loss of heterozygosity (LOH) by cigarette smoke and asbestos.[279,280,282,283] Diminished expression of FHIT has been recorded in up to 80% of cigarette smoke-associated lung cancers,[279] and in both asbestos-associated lung cancers (~69%) and non-exposed cases (~59%) in one study,[282] and in ~54% of mesotheliomas[283] (Pylkkänen *et al.*[283] suggest that LOH affecting FHIT can be concealed by normal cells present in mesotheliomas). Genomic instability affecting FHIT has also been identified in cases of idiopathic pulmonary fibrosis.[284]

GENETIC SUSCEPTIBILITY TO LUNG CANCER

It is well known that genetic factors play a major causal role in the genesis of some cancers, notably those related to mutations in tumour suppressor genes or DNA repair genes, with high penetrance of the mutated gene(s):[285,286] such cancers include gastrointestinal cancers among families with familial adenomatous polyposis (APC gene), and cancers related to mutations affecting DNA repair genes, such as hereditary non-polyposis colon cancer (HNPCC) and xeroderma pigmentosum (XP[A-D] genes),[286] and it has been estimated that genetic abnormalities of this type may account for about 1–4% of all cancers.[286,287]

It is also known that in some circumstances there is an interplay between genetic predisposition to cancer and environmental factors.[286,288] One classical example is xeroderma pigmentosum (XP), where the mutated DNA repair genes XP(A-D) produce extreme susceptibility (>1000-fold above 'normal'[289]) to skin cancers (basal and squamous cell carcinomas and melanoma),[286] because of an impaired capacity to repair DNA damage induced in the skin by ultraviolet radiation in sunlight; management of patients with XP includes isolating them from sunlight to minimise the DNA damage and hence to reduce the otherwise virtually certain risk of skin cancer.

Delineation of the genetic component for cancers related to multiple gene variants of low penetrance poses far greater difficulties than for high-penetrance single-gene disorders, and familial aggregation of some cancers is complicated by the fact, that apart from some shared genes, family members frequently share environmental factors, including diet, lifestyle, recreations and occupations.

Although lung cancer risk is highly dependent on environmental factors such as cigarette smoke (and less commonly asbestos and other occupational/environmental factors), it is a truism that that only a minority of tobacco smokers ever develop lung cancer during their lifetimes (about one in $10^{287,290}$), and only a minority of those exposed to asbestos ever develops lung cancer. Chance alone might be invoked as the explanation for cancer/not-cancer—for example the 'correct' combination of mutational events may not occur at all or in the 'correct' order, or a mutational event may be lethal to the cell—however, there is evolving evidence for modulation of cancer risk by genetic susceptibility/resistance (G_S and G_R) factors.[287,290–295]

In studies based on the Swedish Family-Cancer Database,[296–298]†† the 'proportion of cancer susceptibility, accounted for by genetic effects' was estimated at 14%[285] and later at 8%[299] for lung cancer, with shared and childhood environmental components of 9 and 4%, respectively, and 79% for non-shared environmental factors.[299] A further study on the same database gave an estimated familial population attributable fraction (PAF) of ~3% for lung cancer, with a familial percentage proportion of ~6% (defined as the percentage of affected offspring with affected parents).[300] A further study on the Swedish Database also yielded a higher familial risk for large cell carcinoma and adenocarcinoma of lung (SIRs = 2.29 and 2.18, respectively) than for other histological types (small cell carcinoma = 1.74 and squamous cell carcinoma = 1.78).[296]

Apart from gatekeeper genes such as *p53* and *k-ras*, a number of studies have focused on polymorphisms for caretaker genes[301]—for example, those encoding the cytochrome p450 superfamily,[288,302,303] such as CYP1A1,[302,303] as well as *N*-acetyltransferase, glutathione *S*-transferase M1 (GSTM1), microsomal epoxide hydrolase (*m*EH)[290,304] NAD(P)H:quinone oxidoreductase ($^{609}C \rightarrow T$ polymorphism)[290,305] and myeloperoxidase (MPO)[306]—which are involved in the activation or detoxification of carcinogens,[290,307] and on DNA repair genes[290,308] (about 130 DNA repair genes have been recorded, divisible into base excision repair, nucleotide excision repair and mismatch repair genes).[309] For example, in relation to DNA repair genes it has also been reported that polymorphisms affecting exons 10 and 23 of XPD modulate risks for lung cancer among never-smokers, so that the presence of one or two variant alleles was associated with an OR_{LCA} of 2.6 for exon 10 (95%CI = 1.1–6.5) and 3.2 for exon 23 (95%CI = 1.3–8.0);[289] in addition, current or recent smokers had higher aromatic DNA adduct levels than former/never smokers, and the same study[289] found that subjects with exon 10 AA and exon 23 CC had significantly higher aromatic DNA adduct levels than subjects with any other genotype, with an increased risk of lung cancer.

In all probability, many potential G_S/G_R genes have yet to be identified,[290] and analysis of the interplay between multiple G_S and G_R genes and environmental carcinogens constitutes a problem of great complexity; nonetheless, it seems likely that 'everyone may have a unique combination of polymorphic traits that modify genetic susceptibility and response to ... carcinogens',[290] especially for multifactorial diseases such as lung cancer.[290] To simplify matters, the following discussion concentrates mainly on the MPO gene.

MPO is a lysosomal enzyme found in both neutrophils

††The largest database of its type in the World, the Swedish Family-Cancer Database contains data on people born in Sweden after 1931, including their parents; by 2002, the Database comprised information on 10.2 million individuals across 3.2 million families, with data on more than 1 million tumours.[296–298]

and macrophages, and it catalyses the reaction between H_2O_2 and Cl^-, generating hypochlorous acid (HOCl)[310] and other reactive oxygen species (ROS); MPO is involved in the metabolism of several DNA-damaging intermediary factors that include tobacco smoke mutagens, and MPO appears to contribute to lung carcinogenesis by activation of procarcinogens such as benzo[*a*]pyrene intermediates, 4-aminobiphenyl and arylamines.[311] The MPO gene is localised to the long arm of chromosome 17 and comprises 11 introns and 12 exons.

Multiple investigations have evaluated the potential protective effect of the variant A allele for MPO in comparison to the wild-type genotype G/G ($^{-463}$MPO G→A) on the risk of lung cancer.[311–321] Although two studies[316,319] did not detect any significant association between the A allele in comparison to G/G, most found that the A allele was associated with up to a 70% reduced RR_{LCA}/OR_{LCA} at equivalent levels of smoking; in one study[314] the reduced risk was confined to the homozygous AA polymorphism and not to the heterozygous G/A form, but others detected a reduced risk for G/A,[311,313,317,318,320] and one[315] reported the findings as the risk for G/A+A/A only. Most studies reported the protective effect of the A allele in terms of RR_{LCA}/OR_{LCA} relative to G/G, but Lu *et al.*[321] and Schabath *et al.*[322] reported their results as an increased OR_{LCA} for G/G relative to G/A+A/A. The proportions of G/G versus G/A and A/A appear not to differ greatly from lung cancer cases in comparison to controls: across all studies cited above,[311–321] G/G was found in 62% of controls versus 65% of cases; for G/A and A/A for controls versus cases, the percentage proportions were 33 vs 31% and 5 vs 4%; when the two studies that found no effect of MPO polymorphisms on lung cancer risk[316,319] are removed, the proportions for controls versus cases become 61 vs 68% for G/G, 29 vs 33% for G/A and 3 vs 6% for A/A.

Evidence for a component of genetic susceptibility for asbestos-related mesothelioma[323–325] and for lung cancer is much less extensive than the evidence for cigarette smoke-related lung cancer. Nonetheless, this notion has biological plausibility,[326] and is supported by the following observations: (i) only a minority of asbestos-exposed individuals, even those exposed heavily to crocidolite, develop mesothelioma during their lifetimes[327,328] (see preceding discussion); (ii) familial clusters of asbestos-associated mesothelioma are well documented;[329–341] (iii) one study[323] found that patients with mesothelioma have a greater frequency of non-mesothelioma cancers among their parents than non-mesothelioma cases; and (iv) genomic variants have been described in mesothelioma, such as inactivating mutations of the neurofibromatosis type 2 (NF2) gene[342] and simian virus 40 (SV40) transcripts incorporated into the genome (although the evidence for a contributory causal role of SV40 in the development of asbestos-related mesothelioma remains unproven[343,344]).

So far as we are aware, there are only two reports on G_S/G_R for asbestos-associated lung cancer, relative to polymorphisms for the GSTM1[345] and MPO genes.[322] Stucker *et al.*[345] found no evidence that the risk of lung cancer after asbestos exposure differed according to the GSTM1 genotype, although this study had 'low statistical power'.[345] Conversely, in a molecular case-referent study, Schabath *et al.*[322] found that subjects with self-reported asbestos exposure and with the MPO genotype G/G had an OR_{LCA} of 1.72 for asbestos exposure compared with no exposure after controlling for age, gender and smoking, whereas subjects with a G/A+A/A genotype had a lower OR_{LCA} of 0.89. Subjects with G/G had an OR_{LCA} of 1.69 for ≥45 pack-years of smoking (heavy) compared with <45 (light), whereas the OR_{LCA} for those with G/A+A/A was <1.0. For GG, the joint effect of asbestos and heavy smoking in comparison to no asbestos and light smoking was 2.19, and the analogous OR_{LCA} for G/A+A/A was 1.18.

Given the emerging evidence on G_S/G_R for lung cancer, for both cigarette smoke and (to a far lesser extent) asbestos, and taking into account the complexity of the multiple genes and polymorphisms implicated so far, it seems that individuals comprising any population will vary in their susceptibility to (and risk from) these carcinogens. Therefore, one can deduce that the risk derived as an average or mean across entire cohorts/populations will tend to underestimate the risk for those with a G_S profile (RR_{GS}) and to overestimate risk for those with G_R (RR_{GR}). It also follows that those with the disease in question are more likely to have G_S for that disease and therefore to be at greater risk than either: (i) those who are resistant (G_R); or (ii) the average/mean risk (i.e., $RR_{GS} > [RR_{GS}+RR_{GR}]/2$), even if the variation in risk from the mean is only very small.

Assessing the significance of interaction between genetic and environmental factors in disease causation involves a new type of epidemiological study, the case-only study,[345,346] in which departure from a purely multiplicative model of joint effect can be assessed by computing the case-only OR (OR_{C-O}), derived for cases with and without the susceptibility gene and with and without exposure from a 2×2 table; if OR_{CS} represents the OR among control subjects related to exposure and susceptibility genotype, then:

$$OR_{C-O} = [OR_{GE}/(OR_E \cdot OR_G)] \cdot OR_{CS}$$

where OR_{GE}, OR_G and OR_E are conventional case–control ORs for combined genetic susceptibility plus exposure, genetic susceptibility, and exposure separately.[346] Because the genotype and the exposure are generally independent variables in the source population from which the cases arise, the expected value of OR_{CS} is unity; if the joint effect is more than multiplicative, OR_{C-O} is greater than 1.0, and it is less than 1.0 if the joint effect is less than multiplicative.[346] Applied to the data in Table III of Schabath *et al.*[322] (asbestos and genotype), the above analysis gives an OR_{C-O} of 0.96, indicating near-multiplicativity.

If such findings[322] are validated in other analogous investigations, they would suggest that the asbestos-related lung cancer risk derived as an average across groups might be revised upwards for those with a susceptibility genotype, so that cumulative exposures lower than the average (e.g., <25 fibres/mL-years) could be accepted as imposing an OR ≥2.0, and the risk would be correspondingly revised downward for those with a genetic resistance profile, with the requirement for a greater cumulative exposure to impose the same risk. We consider that this approach to carcinogenesis by environmental factors in general has a sound theoretical and, to a lesser extent,

empirical basis, and we expect that molecular epidemiological studies that address these issues will lead to further refinement of approaches to causation by cigarette smoke, asbestos, and other environmental carcinogens. Nonetheless, we consider that at present it is not possible to apply existing data on G_S/G_R for the attribution of lung cancer to asbestos in the individual patient, or to modify existing cumulative exposure approaches to causation, because of: (i) contradictory and inadequate G_S/G_R data, even for single gene polymorphisms; (ii) uncertainties surrounding G_S/G_R profile effects overall; (iii) inadequate data on net G_S/G_R interactivity with asbestos; and, as a consequence, (iv) unquantifiability of any such effects. We also emphasise that these theorisings do not detract from the critical role of the exogenous carcinogens in causation of the disease:[287] in the absence of the carcinogen, it would be less likely that genetic susceptibility (G_S/no-exposure) would be expressed as a particular cancer at the time of occurrence of the cancer, than for a G_S/exposure situation (in other words, the carcinogens produce an increment in risk above 'background' G_S).

We emphasise that although 'traditional' epidemiology has been highly effective for the detection and quantitation of the net or average causal effects of various carcinogens across populations or groups as reflected in cohort or case-referent studies, it becomes less precise for the quantitation of causal effects when applied to assessment of causation in an individual, because of the following factors among many others:

1. Differential exposures to the carcinogen within the cohort or within the cases group for case-referent studies (unless the exposure estimates are individualised or stratified for different patterns of work and exposure). (See discussion of the study by Carel *et al.*,[165] p. 529.)

2. Changes over time in exposures and smoking habits across the cohort/group unless the parameters of exposure/smoking are evaluated longitudinally over time.

3. Differential clearance of asbestos fibres from bronchopulmonary tissues, related to differences in the proportions of asbestos fibre types for mixed asbestos exposures and fibre dimensions, and the efficacy of host clearance mechanisms as influenced by a variety of factors that include innate and acquired differences in the capacity for fibre clearance.

4. Differential genetic susceptibility to the carcinogen(s).

In general, these factors will tend to depress unquantifiably the slope of the dose–response line in comparison to the real effects for those who have asbestos-associated lung cancer, and thereby underestimate probability of causation.

EXPOSURE ASSESSMENT: NATIONAL APPROACHES AND FUTURE DIRECTIONS

The cumulative exposure standard of 25 fibre-years or more for lung cancer attribution is also applied in Denmark, and equivalent job histories elsewhere in Scandinavia, with no requirement for asbestosis.[1] Occupational histories similar to those delineated by The Helsinki Criteria[102] also form the basis for attribution in France and Belgium.[49,233] In Australia, the courts have ruled in favour of the cumulative exposure model as a basis for attribution, and similar criteria were also endorsed by the AWARD Workshop.[225,235]

Because decision-making on compensation now appears to favour The Helsinki Criteria approach, construction of databases such as those described by Burdorf and Swuste[228] or *Faserjahre*[64] will be essential for equitable compensation of lung cancer due to asbestos, when evidence of quantified exposure must be based on history.[2] The approach in The Netherlands is more qualitative than the German system, with probabilistic assessments of the likelihood of different exposure levels. Without such systems, boards and tribunals will continue to spend inordinate time evaluating uncertainties over past exposures and conflicting opinions from expert witnesses. The aim of databased systems of these types is to create a matrix that defines asbestos exposure by industry, occupation and time. In association with each value, one can then assign a level of confidence ranging from:

1. Direct measurement.
2. Interpolated measurement.
3. Measurement in a similar facility.
4. Interpolation from a similar facility.
5. Consensus estimate.
6. Estimate for which no consensus can be reached.

In practice, when there are no direct measurements of airborne fibre levels in a particular workplace, as is often the case in nations such as Australia, experts often express estimated cumulative exposure as a low/high range in fibre-years, based on: (i) the number and duration of work shifts which together comprise about 20% of calendar time; and (ii) published low and high values for airborne fibre concentrations generated by the same or similar types of work in other workplaces, and with derivation of a likely mean estimate.

On the basis of prevailing evidence, the cumulative exposure model for lung cancer induction by asbestos appears to conform to modern approaches to assessment of causality,[29,59,221,326,347,348] with coherence of data across multiple different types of investigation that include dose-response data from epidemiological studies and case-referent studies based on lung tissue fibre measurements; the evidence also encompasses a variety of pathological observations that include the separate and combined clastogenic and mutagenic effects of asbestos and tobacco smoke on cell lines *in vitro* and on bronchiolar epithelium *in vivo*. In terms of generalisability,[29] the cumulative exposure model appears to have explanatory-predictive value: after the 25 fibres/mL-year standard was introduced in Germany—where attribution is primarily an administrative exercise, so that decision-making is less likely to be skewed than by adversarial court-based systems of compensation—the excess lung cancer to mesothelioma ratio has shown close agreement with the same ratio obtained from multiple epidemiological investigations.

Finally, we emphasise that estimates of cumulative exposure (25 fibre-years or an equivalent job history) set forth in The Helsinki Criteria are applicable to amphibole and asbestos textile exposures and, we believe, mixed exposures (notably exposures to asbestos-cement and insulation materials that contained chrysotile and amphiboles);