Experimental and epidemiological studies

Although epidemiological studies have attached special significance to crocidolite in the induction of mesotheliomal tumours, other forms (amosite and chrysotile) have produced tumours in experimental animals, and chrysotile has been claimed to produce tumours in man (*ibid* 1968, 6, 657; *ibid* 1970, 8, 208). All three forms produced mesotheliomas when administered intrapleurally to SPF or Standard rats in the form of a suspension containing 20 mg dust (Wagner & Berry, *Br. J. Cancer* 1969, 23, 567). The percentage incidence of mesotheliomas was 64% for chrysotile, 59% for crocidolite, 59% for crocidolite from which oils had been removed by extraction and 40% from amosite in SPF rats and 69, 68, 64 and 31% in the corresponding groups of Standard rats. In general tumour incidence was inversely related to survival times, but the group differences were more marked in SPF than in Standard rats. Deaths from mesothelioma occurred earliest in the chrysotile group (353 days in Standard rats), followed by the groups treated with extracted crocidolite (376 days), crocidolite (417 days) and amosite (557 days) but after this time lag there was a rapid onset of mesotheliomas. It was necessary to distinguish between mortality from mesotheliomas and from natural causes and this was achieved using life table methods, but a mathematical model has been tested and found suitable for analysis of experiments involving smaller groups of animals (Berry & Wagner, *ibid* 1969, 23, 582). The study described above was initiated in 1962 and work is now in progress on several additional projects, including an attempt to establish a dose-response relationship in rats by injecting crocidolite and chrysotile in a range of doses and a study using the inhalation route, which is more relevant to human experience.

Epidemiological studies are gradually helping to ascertain which forms of asbestos are carcinogenic. In Australia, 15 cases of pleural mesothelioma have been reported in workers exposed to asbestos for 6 months–30 yr (Milne, *Med. J. Aust.* 1969, 2, 669). Some workers had a history of exposure to crocidolite. In an informative review, Wright (*Am. Rev. resp. Dis.* 1969, **100**, 467) cites unpublished data presented at a symposium on pneumoconiosis held in South Africa in 1969. Reference is made to the inability of anthophyllite and chrysotile to produce mesotheliomas in Finnish and Canadian workers respectively. The Finnish workers exhibited asbestosis and a three- to fivefold increase in the incidence of bronchial cancer, but Canadian workers showed no evidence of an increase in bronchial cancer. Mesotheliomas have been found in workers in some but not all of the crocidolite mines in South Africa so far examined, the striking exceptions being the crocidolite mines in the Transvaal. Less surprisingly, no cases were associated with the chrysotile mines in this area, but a few mesothelioma cases from the Transvaal amosite mines have been recorded. Vigliani *et al.* (*Medna Lav.* 1968, 59, 481) could find no evidence of increased cancer of the lungs or pleura in Italian workers exposed to chrysotile. Wright (*loc. cit.*) also refers to a report indicating that chrysotile penetrates to the deep lung structures less readily and is cleared from the lung faster than either amosite or crocidolite. Summing up these epidemiological findings, it would appear that crocidolite and, to a much lesser degree, amosite produce mesotheliomas, while anthophyllite and chrysotile do not. In considering some of the apparently anomalous findings, Wright (*loc. cit.*) suggests that asbestos acts as a cocarcinogen rather than a true carcinogen, but the justification for reaching this conclusion on the supporting evidence must be questioned. The possible existence of a dose–response relationship between severity of exposure and bronchial cancer is also discussed and seems a more tenable proposition.

In a survey of the causes of death among 4500 workers in an asbestos factory during 1933–1964, Newhouse (*Br. J. ind. Med.* 1969, 26, 294) found that the mortality rate started to exceed the norm after 16 yr from the first exposure to mixtures of amosite, crocidolite and chrysotile. A significant excess of deaths from cancer of the lung and pleura even among workers with less than a 2-yr period of service and from respiratory disease in workers with more prolonged service was confined to the heavily-exposed groups. A retrospective examination of death certificates, autopsy reports and histological specimens pertaining to 301 workers from an asbestos factory revealed an underestimate of mesotheliomas in the certified causes of death, 15 additional cases being discovered in 84 histological re-examinations, but the incidence of bronchial cancer had not been greatly underestimated (Newhouse & Wagner, *ibid* 1969, 26, 302). Moderate to severe asbestosis was found in all confirmed cases of lung carcinoma.

Asbestos-body formation

The mechanism of formation and the aetiological significance of asbestos bodies have been discussed earlier (*Cited in F.C.T.* 1970, 8, 209). More light has been thrown on the development of an asbestos body from an asbestos fibre by electronmicroscopic studies involving either intratracheal instillation of soft or harsh chrysotile or amosite into hamsters (Suzuki & Churg, *Envir. Res.* 1969. 3, 107) or intrapleural injection of chrysotile into guinea-pigs (Davis, *Expl. mol. Path.* 1970, 12, 133). These and earlier studies (*Cited in F.C.T.* 1968, 6, 659) suggest that the process entails the coating of the asbestos fibre with acid mucopolysaccharide before its phagocytosis by the alveolar macrophages, and the subsequent transfer into the phagosome of haemosiderin granules which have accumulated in the surrounding cytoplasm. The fibre thus becomes coated with a layer, or sometimes several layers, of dense granules to form an immature and subsequently a mature asbestos body. The bodies, which are produced largely in giant cells, are first seen about 2 wk after asbestos administration and their numbers increase to a maximum at about 6 wk. Asbestos bodies are formed from fibres longer than 5 μ and the process may involve no more than 0·1% of all the fibres present. Davis (*loc. cit.*) also provides evidence suggesting that in guinea-pigs the surface of the acid mucopolysaccharide coat serves as a depository for the laying down of calcium (in the form of apatite crystals) in the development of the fibrous lesions, which eventually calcify to produce plaques similar to those seen in human asbestosis.

The significance of asbestos bodies in the development of asbestosis and malignancy remains to be defined. Dicke & Naylor (*Dis. Chest* 1969, 56, 122) identified asbestos bodies in scrapings of the lungs and lymph nodes from 19 out of 100 autopsies of members of the general population in Michigan, although the presence of asbestos fibres in the bodies was not confirmed. In only four of these cases were asbestos bodies found in the sections of lung tissue. Morphological evidence of asbestos bodies in the lungs of asbestos workers with mesothelioma was obtained at each of nine autopsies but asbestos bodies were not found in lung tissue removed from similar workers at thoracotomy (Milne, *loc. cit.*). In seven of the nine autopsy cases, the asbestos bodies were found in routinely stained sections of lung tissue. Otto & von Fragstein (*Arch. Gewerbepath. Gewerbehyg.* 1969, 25, 193) observed sporadic needle-shaped particles in normal and silicotic lungs and an abundance of these particles in the lungs of men with a history of asbestos exposure. Histological similarities between asbestos bodies and these needles became evident when the needles exceeded 20 μ in length. Differences in the prevalence of these needle-shaped particles in the lungs are claimed to be of diagnostic significance.