AN EXPERIMENTAL MODEL FOR RISK ASSESSMENT OF COMBINED EXPOSURE TO RADON AND OTHER AIRBORNE POLLUTANTS

G. Monchaux, J.P. Morlier, M. Morin and R. Maximilien
Laboratoire de Cancérologie Expérimentale
CEA-DSV-DRR, BP 6, 92265 Fontenay aux Roses Cedex

INTRODUCTION

Combined exposure to radon and its progeny and various occupational or environmental airborne pollutants may lead to synergistic effects for lung cancer induction. In humans, an increased incidence of pulmonary neoplasia has been observed in different groups exposed to radon and its daughters, including uranium miners (1), iron miners (2) and other miners (3) especially cigarette smokers (4). These findings suggest that co-carcinogenic mechanisms may be involved in the pathogenesis of lung cancer. In laboratory animals, a co-carcinogenic effect results in increased tumour rates after combined administration of the potential carcinogens (5). A standardized protocol has been developed in Sprague-Dawley rats to identify potential co-carcinogenic agents. In this, rats are exposed to 1000 WLM of radon followed by exposure to the agent to be studied (6). Exposure to 1000 WLM of radon alone results in a lung cancer incidence of 20%. About 30% of the tumours are squamous cell carcinoma, 50% adenocarcinoma and 20% bronchioloalveolar carcinoma. In these lifespan studies, after exposure, rats were allowed to live until they died or were moribund and then killed and the latency period for tumour induction was about 700 days. The effects in rats of combined exposure to radon and various environmental or industrial airborne pollutants such as cigarette smoke, mineral fibres, diesel exhausts, minerals associated with metallic mine ores and chlorinated compounds are reviewed in relation with the possibility of combined exposure for workers in different industries.

RESULTS

Combined exposure to radon and tobacco smoke

The first experiments were carried out to investigate the effects of inhalation of radon and its daughters at various cumulative doses, before or after various passive exposures to tobacco smoke (7), using cigarettes with and without filters. For a 1000 WLM radon exposure, the incidence of lung carcinomas was slightly lower in rats exposed to tobacco smoke before radon exposure than in rats exposed to radon alone, but the distribution of the different histological types of tumours were similar in the two groups. In contrast, a highly significant excess of lung carcinomas, mainly of the squamous cell type, was observed in the group exposed to tobacco smoke after radon exposure. In this group, the incidence of lung carcinomas was almost four times greater than in the group exposed to radon alone.

The results of further studies in which rats were exposed to cigarette smoke following exposure to radon showed that for the same tobacco smoke exposure, the incidence of lung carcinomas increased with the cumulative dose of radon. In the same way, for an identical cumulative dose of radon and its daughters, the incidence of lung carcinomas increased with the cumulative exposure to tobacco smoke.

The induction of lung carcinomas was less efficient in rats exposed to tobacco smoke produced by filter cigarettes than in those exposed to cigarettes without filters. The incidence of lung carcinomas were higher, but not statistically significant in the groups exposed to radon and tobacco smoke combined than in the group exposed to radon alone and the proportion of lung carcinomas was lower in the group exposed to filter cigarettes than in the group exposed to unfiltered cigarettes. The increased incidence of lung carcinomas in the group exposed to radon and non filter cigarettes was mainly related to an increased incidence of squamous cell carcinomas. These findings suggested a stronger synergistic effect of radon and non filter cigarettes compared to that of radon and filter cigarettes.

In rats exposed to radon and tobacco smoke combined, for the same radon exposure, the incidence of lung carcinomas was greatly increased in the group exposed to radon and tobacco smoke compared with the group exposed to radon only. Tumours observed in the

groups exposed to radon and tobacco smoke were larger and more invasive than in the groups exposed to radon alone. These tumours also spread more to the pleura and the presence of intrapulmonary metastases or of multiple tumours in the lung was observed. For the same radon exposure, the mean latent period for lung carcinomas was shorter in the group exposed to radon and then to tobacco smoke compared with the group exposed to radon alone. For an identical tobacco smoke exposure of 350 hours, the mean latency period was inversely related to the cumulative radon dose. All these results showed a clear co-carcinogenic effect of exposure to radon and radon daughters and tobacco smoke in rats.

Combined exposure to radon and mineral fibres and other industrial or environmental airborne pollutants

The experimental protocol described above was used to study the potential co-carcinogenic effects of radon and mineral fibres. Acid leached chrysotile fibres were shown to exhibit less carcinogenic activity *in vivo* than untreated fibres (8). Since mesothelial cells are considered to be the target cells for the induction of tumours by mineral fibres, this experiment was designed to investigate the potential synergistic action of different kinds of unleached or acid leached asbestos fibres and other mineral dusts injected into the pleural cavity of rats after previous inhalation of radon and its daughters. In these experiments, 60 rats exposed to radon were used as controls. Ten groups of 10 rats each were exposed to the same dose of radon and then, 2 weeks later were injected intrapleurally with 2 mg of mineral dust, unleached or leached asbestos fibres, glass fibres and two varieties of quartz. No rats were exposed to mineral fibres alone. The results are summarized in table 1.

Table 1. Combined effects of radon and other environmental or industrial airborne pollutants

Experiments	Results	Conclusion
Radon + Cigarette Smoke	Increased incidence of lung carcinomas up to 4 times greater than in rats exposed to radon alone	**Multiplicative effect**
Radon + Mineral Fibres	1 / 3 Lung carcinomas 1 / 3 Pleural mesotheliomas 1 / 3 Combined pulmonary pleural tumours in excess	**Additive effect**
Radon + Diesel Exhaust	Slight non significant excess of lung carcinomas	**No clear synergistic effect**
Radon + Minerals from Metallic Mine Ores	Slight non significant excess of lung carcinomas	**No clear synergistic effect**
Radon + Methylene chloride	No excess of lung cancer	**No synergistic effect**

The potential carcinogenic or co-carcinogenic role of 4 minerals present in the ores of metallic mines was also investigated. These included, nemalite (a contaminant of Quebec chrysotile), biotite (present in many granites and in the French uranium ore), iron pyrites (present in various iron and gold ores), and finally iron-rich chlorite (present in iron, tungsten and gold ores).

The use of diesel-powered vehicles is steadily increasing worldwide. Among diesel-exhaust exposed populations, only a case control study and a retrospective cohort study in railroad workers (9), showed a significant association between diesel exhaust inhalation and lung cancer, suggesting that occupational exposure to diesel exhausts results in a small but significant excess risk of lung cancer. Experimentally, some evidence of a carcinogenic effect has been previously reported in rats after exposure to diesel exhaust containing high concentrations of diesel soot particles for periods of up to 2 years (10). The potential synergistic effects of diesel exhaust were investigated in rats after previous exposure to radon and radon daughters.

Chlorinated compounds are widely used in a variety of commercial forms for industrial and medical applications. Exposure to methylene chloride induces lung and liver cancers in mice. The aim of this study was to test the potential carcinogenic or cocarcinogenic effect of methylene chloride in rats, acting either alone, as a complete carcinogen, or as a promoter after local pulmonary irradiation by inhalation of radon.

The results of these different studies are summarized in table 1.

CONCLUSION

These results demonstrate the potential co-carcinogenic action of various environmental or industrial airborne pollutants combined with radon exposure, showing either a multiplicative, an additive or nul effect. The strongest co-carcinogenic effect was shown by combined exposure first to radon and then to tobacco smoke and resulted in an increased incidence of lung carcinomas, mainly of the squamous cell type. These results also indicated the possible application of this "radon model" to the investigation of possible interactions between exposure to two occupational and/or environmental pollutants. The importance of such an experimental model for risk assessment should be emphasized since human industrial occupational or environmental exposures are nearly always not single but multiple.

REFERENCES

1. J. M. Samet, R. Pathak, M.V. Morgan, C.R. Key, A.A. Valdivia and J.H. Lubin. Lung cancer mortality and exposure to radon progeny in a cohort of New Mexico underground uranium miners. Health Phys., 61, 745-752, (1991).
2. E.P. Radford and K.G. Renard St Clair. Lung cancer in Swedish iron ore miners exposed to low doses of radon daughters. N. Engl. J. Med., 310, 1485-1494, (1984).
3. A.J. Fox, P. Goldblatt and L.J. Kinlen. A study of the mortality of Cornish tin miners. Br. J. ind. Med., 38, 378-380, (1981).
4. G. Saccomanno, G.C. Huth, O. Auerbach et al. Relationship of radioactive radon daughters and cigarette smoking in the genesis of lung cancer in uranium miners. Cancer, 62, 1402-1408 (1988).
5. I. Berenblum, A re-evaluation of the concept of cocarcinogenesis. In: *Progress in Experimental Tumor Research* , Hamburger, F., Editor, S. Karger, New-York, Vol. 11, pp. 21-30 (1969).
6. G. Monchaux, J.P. Morlier, M. Morin, J. Chameaud, J. Lafuma and R. Masse. Carcinogenic and cocarcinogenic effects of radon and radon daughters in rats. Environ. Health Perspect. 102, 64-73, (1994).
7. J. Chameaud, R. Perraud, J. Chrétien, R. Masse and J. Lafuma, Lung carcinogenesis during in vivo cigarette smoking and radon daughters exposure in rats. In: *Recent Results in Cancer Research*, Springer-Verlag, Berlin-Heidelberg, Vol. 82, pp. 11-20, (1982).
8. G. Monchaux, J. Bignon, M.C. Jaurand, J. Lafuma, P. Sébastien, R. Masse, A. Hirsch, and J. Goni. Mesotheliomas in rats following inoculation with acid-leached chrysotile asbestos and other mineral fibres. Carcinogenesis, 2, 229-236, (1981).
9. E. Garschik, M.B. Schenker, A. Munoz, M. Segal, T.J. Smith, T.J., , S.R., S.K. Hammond and F.E. Speizer. A retrospective cohort study of lung cancer and diesel exhaust exposure in railroad workers. Am. Rev. Respir. Dis., 137 (1988) 820-825.
10. U. Heinrich, H. Muhle, S. Takenaka, H. Ernst, R. Fuhst, U.Mohr, F. Pott and W. Stöber. Chronic effects on the respiratory tract of hamsters, mice and rats after long-term animal inhalation of high concentrations of filtered and unfiltered diesel engine emissions. J. Appl. Toxicol., 6, 383-395, (1986).