INTERNATIONAL AGENCY FOR RESEARCH ON CANCER

ANNUAL REPORT 1977

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INTRODUCTION

This report covers the work of the International Agency for Research on Cancer for the twelve-month period ending 30 June 1977. Like reports of previous years, the first part presents an overall view of the current projects and general developments in environmental carcinogenesis in the Agency's programme. The second part describes individual projects in detail.

The scientific programme

Due to the long-term nature of research, especially that in epidemiology, a certain amount of repetition is bound to occur from year to year. The Director would like to draw attention to the progress that has been made in certain of the projects of the Agency.

Cancer registration

The third volume of *Cancer Incidence in Five Continents* has been published and provides age-standardized incidence rates for 78 populations in 28 countries; work has already begun on a commentary on the first three volumes, which is intended to make the data more accessible to the non-specialist. This publication is the outcome of collaboration between the Agency and the International Association of Cancer Registries (IACR).

Another collaborative activity in which the Cancer Unit, WHO Headquarters, the Agency and IACR are all involved is the preparation of a manual devoted to the techniques of cancer registration. This will be available early in 1978.

Clearing-house for on-going research in cancer epidemiology

The collaboration of the Agency and the German Cancer Research Centre (DKFZ), Heidelberg, Federal Republic of Germany, with the support of the International Cancer Research Data Bank of the National Cancer Institute (USA), has resulted in the publication of the 1977 *Directory of On-going Research in Cancer Epidemiology*. This is the second annual edition and includes abstracts of 905 epidemiological projects undertaken in 70 countries.

Role of alcoholic beverages in human malignancies

With the support of the National Institute on Alcohol Abuse and Alcoholism (USA), the Agency has been able to extend its studies on the role of alcoholic beverages in human cancer.

The study in Ille-et-Vilaine, France has shown that the risk of oesophageal cancer is clearly related to the consumption of alcohol. It is also related to the number of cigarettes smoked. These two agents act independently, but when they are combined the effects are multiplied.

The earlier report of an association between the consumption of beer and cancers of the colon and rectum has prompted studies involving brewery workers renowned for their high daily consumption of beer. A retrospective cohort study has been started in Denmark where more than 14 000 past and present brewery workers are being followed. Causes of death and mortality rates established so far show no significant difference for cancers of the rectum and colon in these workers compared with the general population; however, cancers of the oesophagus and larynx were more frequent. Mortality among the brewery workers from cirrhosis of the liver, gastroduodenal ulcer and traffic accidents was also higher than expected.

A similar study on brewers of another type of beer is being completed in Ireland.

Role of diet in large-bowel cancer

The Agency has begun investigations into what effect diet might have on human cancer. The present studies, which are being carried out mainly in Scandinavia, are designed to test the association between specific dietary factors and the incidence of large-bowel cancer. Three hypotheses are being tested: the first, that a high dietary intake of fat increases the amount of bile acids in the large bowel and that these are subsequently metabolized by bacterial flora to carcinogens; the second, that a high consumption of meat, which is strongly correlated with colon cancer, is involved, although this could be linked to its fat content; and the third, that a deficiency of dietary fibre is a risk factor. So far, dietary fat does not seem to be associated with increased cancer risk; meat consumption was significantly higher in areas in which colon cancer incidence was higher; and the higher intake of dietary fibre and milk, seen in areas of lower cancer incidence, seems to have a protective effect. This study is being extended to other populations.

Health risks from man-made mineral fibres

The Agency is collaborating with the Medical Research Council Pneumoconiosis Unit, Cardiff, UK, and the Institute of Occupational Medicine, Edinburgh, UK, to determine whether there is a carcinogenic hazard for workers exposed to fibres in factories making rock wool, glass wool and continuous fibre. So far, study visits have been made to 46 factories in 11 European countries to ascertain whether the conditions are suitable for historical follow-up or prospective studies. With the cooperation of the industry, data from personnel records, fibre type, method and dates of manufacture and facilities for follow-up have been examined, and 43 out of 72 factories are providing enough continuous data to make studies possible.

The Agency will be responsible for the epidemiological studies and the Institute of Occupational Medicine for estimates of exposure to fibres. Animal studies on the effect of exposure to various fibre sizes and types are being carried out at the MRC Pneumoconiosis Unit.

Role of viruses in human malignancies

In the study on the etiology of Burkitt's lymphoma, the sera from 14 children with the disease have been compared with sera taken from the same children long before the onset of the disease and with sera from other children selected as controls. For 11 of the 14 children, antibody titres against the Epstein–Barr viral capsid antigen were significantly higher than those of the control sera. This interim result is thought to favour the hypothesis that early infection with Epstein–Barr virus, possibly during the perinatal period, represents a determining factor in the later development of Burkitt's lymphoma. If the case-finding programme can continue for another two years, more cases from whom sera were collected before the onset of the disease should appear. This would permit confirmation of the results and conclusions to be drawn from them. The highest praise must go to our Ugandan colleagues for their work in ensuring the success of this programme, in which 45 000 children were bled and followed up for several years.

The role of malaria in the etiology of Burkitt's lymphoma is being studied in Tanzania. Since March 1977, nearly 85% of the children in the North Mara district have been given chloroquine tablets regularly. The effect of this anti-malarial prophylaxis on the incidence of Burkitt's lymphoma will be followed.

The role of the Epstein–Barr virus in the etiology of nasopharyngeal carcinoma was one of the topics reviewed at an international symposium on the etiology and control of nasopharyngeal carcinoma held in April in Kyoto, Japan, in collaboration with the Japanese Society for the Promotion of Science and the National Cancer Institute (USA). The consistent presence of markers of Epstein–Barr virus infection in tumour cells from patients in high-, intermediate- and low-risk areas has confirmed the association between the virus and the tumour; however, the contribution of chronic nasal disease and the effect of life-long consumption of salted fish were considered to merit further study.

Immunogenetic studies in Singapore have identified a specific HLA profile that is an indicator of high risk for nasopharyngeal carcinoma among Southern Chinese, and these findings are being pursued.

Immunology

In the field of carcino-embryonic antigens, the Agency has continued to make its expertise available in the α-fetoprotein (AFP) studies and is currently preparing a reference reagent for radioimmune and immuno-enzyme assays of AFP, in collaboration with the National Institute for Biological Standards and Control, UK.

The Agency is now engaged in the preparation of a reference reagent for the assay of the β-l-specific pregnancy glycoprotein, which is being studied as a marker for trophoblastoid tumours. This work is being carried out in collaboration with the National Institute of Health of The Netherlands.

With the establishment at the IARC Research Centre, London, of a low-temperature bank for the storage and preservation of biological materials—cells and sera—from cancer patients, the Agency has been able to provide materials for research by collaborating laboratories in the UK, the USA, Switzerland and India.

Environmental carcinogens

The importance of precise analytical techniques for identifying and quantifying environmental carcinogens has grown as epidemiological techniques have become more refined. The problem of relating an observed cancer incidence to the level of a given carcinogenic insult remains the key to most etiological studies. At the Agency, efforts have been concentrated on techniques for the analysis of nitroso compounds at the microgram level—and less—and this effort is continuing. Moreover, the breadth of international collaboration in this programme has grown considerably.

Recently, the Agency broadened its scope in this field by calling together experts in analytical techniques for a range of other environmental carcinogens, with the aim of producing a manual of selected analytical methods. The first two volumes will cover *N*-nitroso compounds and vinyl chloride and will be published shortly.

Chemical carcinogenesis

The role of the Agency in coordinating carcinogenicity data is of major importance. The aim is to provide information that can be used in the primary prevention of cancer. Fifteen volumes of monographs evaluating the carcinogenic risk of chemicals to man have now been published, covering 336 chemicals, and monographs will continue to be published at the same rhythm of 2–3 volumes a year. So far, 25 of these compounds have been shown to be associated with cancer in man—16 of the 25 in occupational exposures, eight related to medicinal exposures and one in diet.

Fig. 1 Visit of Dr S. Sheikholeslamzadeh, Iranian Minister of Health and Social Welfare. The Minister is watching the visual display of the computer terminal, watched by his Under-Secretary, Dr G. Soupikian, and members of the Agency.

The world-wide survey of chemicals that are currently being tested for carcinogenicity has continued and provides a valuable communication between scientists working in this field.

Laboratory studies have been largely devoted to the development and improvement of rapid screening tests for environmental chemicals and to the study of the mechanisms of carcinogenesis. These studies, carried out in collaboration with many national institutes, are all directed towards a better assessment of the significance to man of the results of laboratory tests for chemical carcinogens.

Manpower training

The task of increasing research manpower in the fields, especially, of epidemiology and environmental carcinogenesis remains an integral part of the Agency's effort. It was satisfying to be able to increase the number of Research Training Fellowships this year, since it matched the high quality of the majority of the applicants. It is important to note, too, that five out of 25 fellowships awarded are for training in epidemiology.

Epidemiology has been the most important theme in the programme of courses that the Agency has run since 1968; the fact that this year an epidemiology course has been organized in Brasilia represents a deliberate policy of seeking to raise research skills in developing countries. In collaboration with the Regional Office for the Eastern Mediterranean, WHO, the Agency is in the process of organizing another epidemiology course in Karachi.

Linked with its educational role is the publishing activity of the Agency. Since 1971, sixteen volumes have been published in the IARC Scientific Publications series—an activity in which all of the scientific staff of the Agency have been deeply involved.

Headquarters

During this year, installation of the fifth and sixth floors of the Agency's building was completed, and, as a result, a more rational distribution of space in the building has been possible. The biostatistics section, which provides an essential service to all of the research units, has been able to expand and is now equipped with visual display units that permit conversational connexion with the computer at the International Computing Centre, Geneva. Other units have also benefitted from the new space; however, there are still rooms available for visiting scientists.

The auditorium is not only used by the Agency for meetings and specialized courses but has also been made available for national and international meetings organized by the local medical and scientific community.

Personnel

In June 1977, the staff of the Agency totalled 167 and was made up of 36 scientists, 68 technicians and 63 administrative and secretarial staff. Eight visiting scientists spent periods varying from one month to one year, learning new techniques and contributing to the Agency's research programme.

Dr James Huff has joined the Agency from Oak Ridge National Laboratories; he succeeds Dr Claus Agthe, who has returned to WHO to head the Division of Food Additives. Dr Huff is responsible for developing the programme of monographs on the evaluation of the carcinogenic risk of chemicals to man.

In May 1977, Dr Janez Kmet reached retiring age. He joined the Agency from WHO and, since 1968, was largely responsible for the oesophageal cancer study in northern Iran, where he spent most of his time in the field. His stimulating and good-humoured personality was such that his visits to Lyon, although brief and infrequent, were always a pleasure for his colleagues. He will continue to serve the Agency as a consultant to the study in Iran.

Dr D. P. Beri who, as a long-term consultant, headed the Burkitt's lymphoma field station in the West Nile district, Uganda, retired recently to his home in India. His colleagues at the Agency and in Uganda wish him a most happy retirement.

Funding

During 1977, the income of the Agency totalled US $6 811 000. Of this, US $4 640 000 came from the contributions of the Participating States and US $2 171 000 from grants and contracts. The details are given in Table 1.

Table 1. Income and expenditure for 1977 [a]

	Amount (US$)	Percentage of total
Income		
Statutory budget	4 640 000	68.13
Extrabudgetary	2 171 000	31.87
Total	6 811 000	100.—
Expenditure		
Intramural		
Headquarters scientific staff	1 733 000	25.45
Administrative staff and office services	871 000	12.79
Building management	605 000	8.88
Laboratory research (supplies and staff salaries)	670 000	9.84
Publications and information programme	438 000	6.43
Building renovations	186 000	2.73
Other (data processing, library, organizational and scientific meetings)	184 000	2.70
Total	4 687 000	68.82
Extramural		
Contractual and collaborative research	1 645 000	24.15
Fellowships	290 000	4.26
Research centres	97 000	1.42
Duty travel	92 000	1.35
Total	2 124 000	31.18
Grand total	6 811 000	100.—

[a] All figures are estimates. Statutory budget income and expenditure details are extracted from the approved budget, whereas the extrabudgetary figures are based on information available in late July 1977.

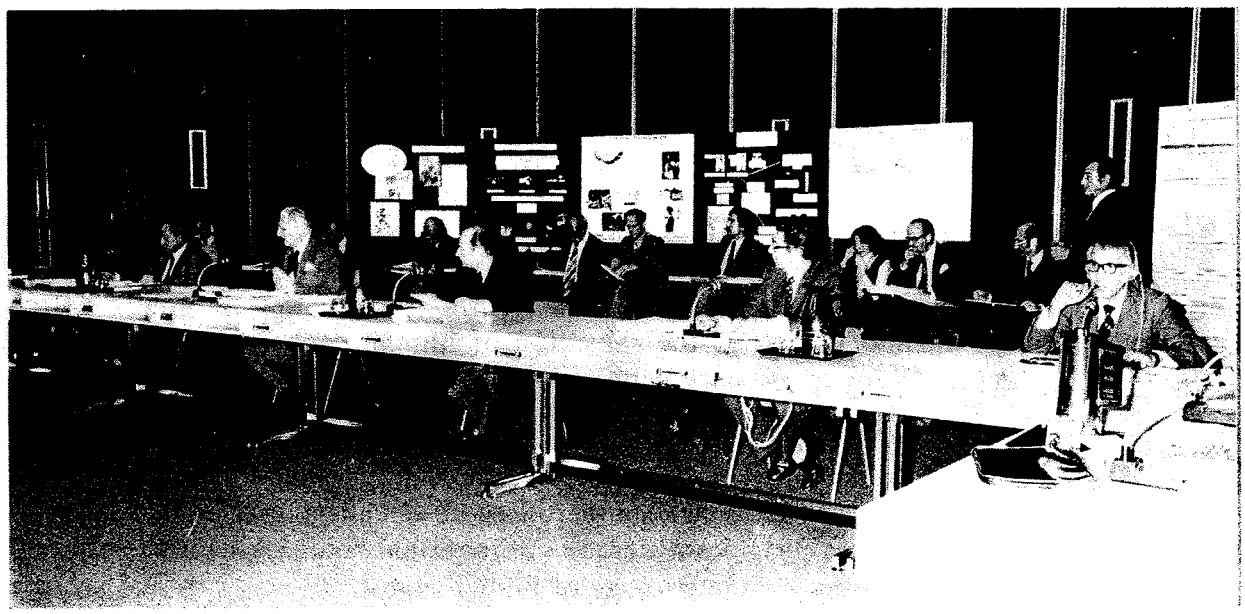
Present and future programme planning

Each year the Scientific Council examines a selected number of projects in depth. Every four years, however, at the request of the Governing Council, the Scientific Council reviews the overall activities of the Agency and the extent of its programmes in the light of recent developments in the field of cancer research; it will meet in February 1978 to make this four-year review.

The decision of the 18th World Health Assembly in 1965 to create the Agency was the culmination of extensive studies and discussions by scientific committees on the need for an international body to stimulate research in cancer. In a series of meetings, held during 1963–1965, leaders in cancer research reviewed many aspects of current scientific developments and formulated certain general concepts that would define the role of an international cancer research organization. A scientific committee nominated by the participating states proposed certain specific programmes and the level of support considered necessary. It was emphasized that the programme should stand on its scientific merit and should concentrate on research particularly suited to an international collaborative approach.

During the early years of the Agency, it was decided to confine operations predominantly to the study of the role of the environment in human cancer. Not only was there increasing concern among both the public and the medical profession about the adverse effects of the environment on human health and about the hazard of cancer in particular, but it was clear also that research in this field was peculiarly well-suited to an international approach and, moreover, was inadequately covered by national efforts.

Fig. 2 The Scientific Council at work. Foreground, left to right: Professor A. Caputo, Professor Sir Richard Doll, Professor S. Eckhardt, Professor J. Miller, Professor C. Mofidi.



This orientation of the Agency's programmes was reaffirmed by the Governing Council in May 1973, and the principles endorsed then are likely to govern the development of the scientific programmes over the next few years, the more so since they are in accord with the International Cancer Plan presented to the World Health Assembly.

Within the Agency's programmes basic research is, inevitably, interrelated with research applied specifically to problems of environmental carcinogenesis—and the research aspect is interrelated with the Agency's role in training scientific manpower.

The development of rapid screening tests for chemical carcinogens, the problems related to the interpretation of such tests and their extrapolation to the human situation require a considerable level of research competence in the Agency's scientific staff and in its Scientific Council. If the scientific staff are to be able to apply new knowledge in basic research to the problems of environmental carcinogenesis, they must be aware of all those developments in different parts of the world that are pertinent to their research projects. They must, moreover, be in a position to enhance the Agency's aim of generating new data by stimulating national collaborative research projects.

New data and new research methods are, in turn, applied to those of the Agency's research activities that are directed towards investigating all those aspects of the human environment that may be related to cancer causation—cultural habits, diet, 'life style', occupation, medicinal drugs, pollution of air, water and food.

An integral part of the Agency's international collaborative research programmes is the training of scientific manpower, especially in the fields of epidemiology and environmental carcinogenesis. Specialized courses and research training fellowships equip young scientists to play a more important role in their national research programmes and often serve to 'ear-mark' them as future scientific collaborators of the Agency.

The publications of the Agency are another arm of its international collaborating activity. They provide governments, industrial managers and scientific workers with authoritative evaluations of cancer hazards in the environment, with standardized national cancer statistics for international comparison and study, and with data on studies in epidemiology and occupational cancer. The manuals of experimental pathology, analytical methods and techniques of cancer registration aim to provide internationally acceptable standards in environmental cancer research.

Before attempting to predict future developments, it is important to summarize what has already been achieved by the Agency and its many scientific collaborators in national and international organizations.

Environmental carcinogenesis

The Agency has played a leading part in establishing the emphasis now generally accorded to environmental factors in the causation of human cancer, thus significantly influencing the development of primary cancer control.

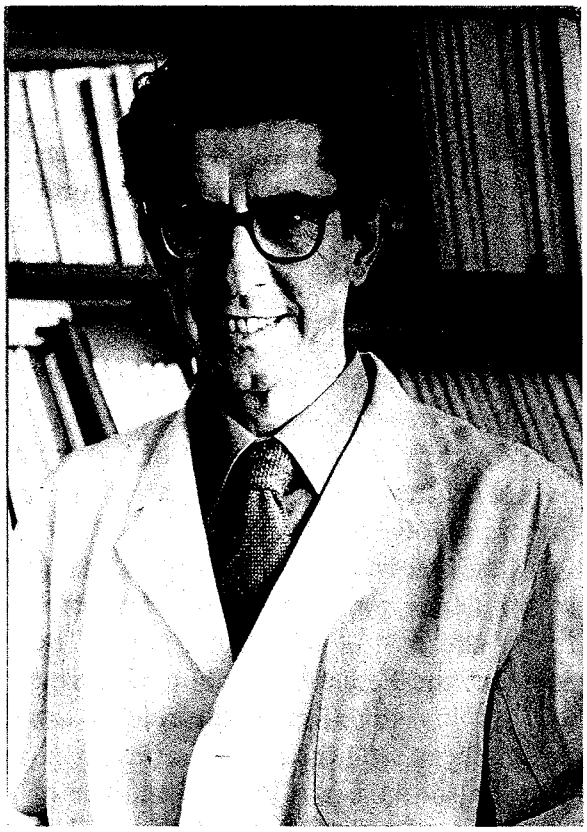
Epidemiology

The collection, computerization and standardization of cancer morbidity data from approximately 80 populations provides a unique international resource which is essential for basic environmental studies on the etiology of human cancer.

A monograph on the purposes and techniques of cancer registration is being completed, in collaboration with the International Association of Cancer Registries, with the aim of improving international comparability of registry data. Advice to and organizational support of cancer registration activities is given in countries where such data are at present unavailable; these include Iran, Indonesia, Papua and New Guinea.

Fig. 3 New members of the Scientific Council

Professor G. L. J. van der Schueren



Professor J. Miller

Professor T. Sugimura

A clearing-house has been organized which collects information on ongoing epidemiological and occupational cancer studies in order to improve international collaboration and to identify areas where research effort is lacking.

A systematic evaluation is being made of the role of alcoholic beverages in cancers of the oesophagus, gastrointestinal tract and liver. The multiplicative effects of cigarette smoking in combination with the consumption of alcoholic beverages are being studied in France, Ireland, Denmark, Belgium and Switzerland.

It has been demonstrated that certain current hypotheses regarding the etiology of cancer of the large bowel and rectum are not consistent with the distribution of these cancers in areas of similar cultural and social background. As yet, no hypothesis that explains their geographical distribution has been unequivocally established, but there is increased concern about the effects of 'life style'. The Agency's work in this area has mainly been in Scandinavia.

A study of familial factors in breast cancer in Iceland—an isolated community—has permitted the elimination of confounding variables and an evaluation of the risk factors involved.

The study of 'latent' carcinoma of the prostate has shown geographical variations of frequency similar to those seen for invasive carcinoma, which in some countries is the major cause of death among men. The study was carried out in Hong Kong, Singapore, Sweden, the Federal Republic of Germany, Jamaica, Uganda and Israel.

An investigation of the role of tobacco habits in carcinoma of the lung and larynx, especially in South-East Asia (Hong Kong and Thailand), showed that the nature of the tobacco smoked may modify the site of the resulting cancer.

A systematic investigation has been made of oesophageal cancer, which is a major cause of death in a belt extending from North China to Mesopotamia. A curious geographical distribution has been found in Iran, where it has been shown that these cancers are not due to cigarettes and alcoholic beverages, as is the case in North America and Europe.

The role of aflatoxins in liver cancer in Africa has been demonstrated in field studies in which aflatoxins were measured in plate-samples and comparisons made of incidences of this cancer.

Special emphasis has been laid on instruction in cancer epidemiology and chemical carcinogenesis by means of courses organized both in Lyon and elsewhere.

Chemical and occupational carcinogenesis

The series of monographs that evaluate the carcinogenic risk to man of environmental chemical stimuli has been published since 1972. The series is now regarded as the standard source for such information both by scientists and public health authorities.

An international survey of chemicals that are undergoing carcinogenicity testing has been maintained.

Laboratory research has been aimed at providing a rational approach to the extrapolation of results of animal studies to man.

A large-scale, multi-generation study was conducted in several laboratories in which DDT was administered to experimental mice and rats. The effect of DDT was shown to be non-cumulative. This study was carried out at the request of WHO, in view of the importance of DDT as an agricultural pesticide and in malaria control.

An international, collaborative study has been set up to evaluate the possible hazard of man-made mineral fibres, in view of their increasing utilization and the fact that they may be used as a substitute for asbestos. The study is being carried out with governmental and industrial collaboration.

A series of monographs has been prepared which is aimed at establishing standard pathological terminology for experimental animal tumours that are produced during biological carcinogenicity testing.

The Agency, in collaboration with The Directorate General for Research, Science and Education of the Commission of the European Communities, organized a workshop to evaluate rapid screening tests for potential chemical carcinogens. It was also demonstrated that *in vitro* tests may prove to be of value in identifying individual susceptibility to external carcinogens.

An international workshop was convened to examine transplacental mechanisms in human carcinogenesis. During the organization of this meeting, the first cases of transplacentral cancer in man were identified.

Viruses and cancer

A study was made of the relationship between infectious mononucleosis and Hodgkin's disease by matching patients who had shown positive Paul-Bunnel tests with data from cancer registries in the UK, Sweden and Norway.

A long-term prospective study was carried out in Uganda to test the hypothesis that the Epstein–Barr virus has an etiological role in Burkitt's lymphoma. This was the first such study of its kind to be organized. It has been extended in Tanzania to determine whether malarial infection is a cofactor in the causation of Burkitt's lymphoma, since infection with the virus alone is insufficient to cause the tumour.

A sero-epidemiological study was carried out to test the association between Epstein–Barr viral infection and nasopharyngeal carcinoma. The study was based in Hong Kong and Singapore because of the high incidence of this disease among the Cantonese Chinese populations in those areas. It was demonstrated for the first time that there was a link between nasopharyngeal carcinoma and certain immunogenetic factors within the HLA system in Chinese.

Miscellaneous

The diagnostic significance of alpha-fetoprotein in liver cancer has been evaluated systematically in Africa and the USSR. An international reference centre for tumour-related antigens has been established at the Agency by WHO.

The future

Attempts to define future developments in the Agency's research areas are beset with difficulties. However, some general lines of strategy may be deduced from past experience, and the following may prove to be of interest.

Feasibility of cancer prevention through environmental control

'Cancer control' applies to any actions aimed at reducing morbidity and mortality from cancer, including primary prevention, which implies control of exposure to environmental carcinogens, whether industrial or cultural. Such actions can also include screening methods for early diagnosis and cancer therapy, which are especially important in those cancers such as carcinoma of the skin in which treatment is highly effective in reducing mortality.

The Agency's contribution to cancer control is in the field of primary prevention. Unfortunately, there are a number of misconceptions regarding the practical possibilities of primary prevention: these are perhaps based in part on earlier successes in the field of communicable diseases, where identification of causal factors led very rapidly to programmes of suppression and eradication. There is also a widely-held idea that exposure to environmental carcinogens implies only exposure to industrial pollution. The view that 80% to 90% of human cancers owe their origins directly or indirectly to environmental factors should not lead to the assumption that 80% to 90% of all cancers could be prevented in the immediate future by the application of available knowledge. Such a misconception gives rise to exaggerated claims and may also prove detrimental to the selection of priorities in cancer research.

Cancers may be classified into four main groups on the basis of current views of etiology: (1) those for which the probable environmental cause is known or strongly suspected; (2) those for which an environmental association can be inferred; (3) those of unknown origin; and (4) those in which genetic factors may be important.

(1) *Cancers for which the probable environmental cause is known or strongly suspected*

The major environmental etiological factors identified so far are personal and cultural habits: cigarette-smoking, which is a causal factor in lung cancer; excess consumption of alcoholic beverages, associated with oesophageal cancer; and excess exposure to sunlight, associated with skin cancer. Other cancers are related to occupational exposure: asbestos and lung cancers with mesothelioma; vinyl chloride with haemangiosarcoma of the liver; beta-naphthylamine with bladder cancer. It has been estimated, however, that in industrialized states cancers due to industrial occupational exposures account for only between 1% and 5% of all human cancers; in contrast, if it were possible to implement measures to control cigarette smoking, approximately 40% to 50% of all cancer deaths in males in the United Kingdom would be prevented.

(2) *Cancers for which an environmental association can be inferred*

The marked geographical variations in incidence of tumours of the digestive tract and of the genitourinary system, and studies of these cancers in migrant populations, have provided evidence of a largely circumstantial nature for an association between these cancers and environmental factors. Recent studies suggest that these factors may not be direct carcinogens but are indirect factors that influence the endocrine or immunological status of the population groups.

Although firm research data is lacking, there is sufficient indication that what is generally referred to as 'life style' may have marked effects on both the endocrine and immunological status. Dietary pattern is probably one of the most important parameters in defining life style, but breast-feeding habits, which are evidently associated, positively or negatively, with breast cancer are also an indication.

Relatively little is known, however, of the effects of low exposures to a large number of carcinogenic factors, such as is implied in the carcinogenic hazard associated with environmental pollution.

(3) *Cancers of unknown origin*

This group includes, particularly, tumours in children and those of the haematopoietic system and of bone and soft tissue. The role of exogenous factors in these cancers and the possible role of genetic and viral factors are as yet unknown; geographical variations of incidence of these cancers tend to be relatively slight.

(4) *Cancers in which genetic factors may be important*

There is probably a complex interrelationship between hereditary susceptibilities and environmental carcinogenic stimuli in the causation of a number of different cancers; this may explain the apparent differences in individual susceptibility to similar carcinogenic exposures. Except for a few rare syndromes, however, in the absence of appropriate environmental stimuli, genetic factors appear to be relatively unimportant, with two major exceptions.

Skin cancer is an example of the cancers in this group. Excessive exposure to sunlight is the environmental stimulus, the effect of which is modified by genetically-determined skin pigmentation. A genetic susceptibility factor has also been identified in a Chinese population at high risk for nasopharyngeal carcinoma, but no unequivocal association with an environmental stimulus has yet been established.

It would appear then, that at present primary prevention is feasible only for human cancers that fall in the first group, for which the probable environmental cause is known. Public health measures could be effective in this area, but, unfortunately, experience indicates that people are reluctant to change their personal habits, and governments are not prepared to take the Draconian measures necessary to enforce such changes. Instead of a ban on cigarette production or on sunbathing, indirect approaches are made, such as the development of 'safer' cigarettes or barrier creams to cut out the ultra-violet irradiation in sunlight. Control of exposure to industrial carcinogens and to carcinogenic medicinal drugs is certainly possible, but it requires a change in social values.

For the second group, there is very little indication that primary prevention that depended on changing the 'life style' of large numbers of people would have much chance of success at present. It is unlikely that the public would accept major modifications in the way that individuals live: obesity and over-nutrition have been known for a long time to reduce lifespan and to carry an increased risk of cancer, yet they remain major health problems in many countries.

For the last two groups, current knowledge of genetic and host susceptibility is limited, and the implications for cancer control remain in any case uncertain.

Unknown risks

Another problem in primary prevention is the estimation of a level of carcinogenic risk. At present, this depends on tests in experimental animals or in *in vitro* systems; in both cases, the results are difficult to extrapolate objectively to man, because of inadequate knowledge of basic mechanisms of carcinogenesis. Nevertheless, there is little option but to continue to utilize the present system of extrapolation of test results, whatever its limitations, in formulating legislation for future control measures.

The problems of developing more objective criteria for extrapolation from animal tests and evaluation of carcinogenic risk will be studied during a meeting at the Agency in October 1977.

National programmes of cancer research should be broad enough not only to ensure that more cancer patients receive effective therapy, but also, through basic research efforts in carcinogenesis, to provide future generations with the protection that could be achieved by better preventive measures. The Agency's programme, with studies in epidemiology of cancer and evaluation of environmental carcinogens, will continue to be complementary to national cancer research programmes.

Environmental carcinogenesis

During the past decade there has been increasing debate about the social responsibility of the biomedical scientist, and in the field of carcinogenic hazards the exchange is at its sharpest. Within the scientific community, the Agency has a responsibility to ensure that its contributions to the debate are based on a rational evaluation of the available data; if the data are inadequate, it may not be possible to draw any conclusion. Today, unfortunately, emotional rather than scientific evaluation is widespread, with a resulting loss of public confidence in experts: scientists may overemphasize hazards in a way that the average man regards as contrary to commonsense. If the scientific community is to maintain the support and confidence of the public, it must avoid 'crying wolf' too often and must limit its pronouncements to those which can be supported by facts.

There is no substitute for a massive research investment for the study of fundamental mechanisms involved in carcinogenesis to provide an eventual basis for rational control. In the absence of such basic knowledge, all available human data must be taken together with the most reliable experimental data to form the basis for regulatory decisions, which may vary from country to country according to local priorities.

The number and amount of synthetic chemicals entering the environment has increased considerably over the years and is likely to continue to do so. Many of these substances are very useful, but utility has sometimes been accompanied by carcinogenicity: PVC is but one example. Even if strict mutagenicity and animal testing are enforced it is likely that other chemicals will nevertheless be manufactured which will eventually prove to be human carcinogens. It is of the utmost importance that highly exposed groups, industrial or otherwise, are closely followed to determine as soon as possible if there is any increase in risk. Given the large number of such groups, the only economical method of following them is to determine the cause of death or, preferably, whether they appear in the files of a cancer registry. This implies the possibility of linking unequivocally the exposure with the effect at the individual level.

A major task of the Agency in the near future will be the fostering of the systematic collection and analysis of data on various exposed cohorts in an attempt to determine the associated health effects. At the same time information on occupational exposures obtained from systematic case-control studies will be assessed.

Hitherto, much of the world's research effort in environmental carcinogenesis has not been coordinated. After suitable feasibility studies, one of the major tasks of the Agency over the next ten years will be the creation of a network to provide standardized, reliable epidemiological information from several centres. These centres will be selected to permit assessment of the effect on cancer risk of changes in life style and industrial and other environmental exposures and will be located in both developed and developing countries. The coordination and integration of their activities should permit rapid identification of the existence and significance to man of carcinogenic agents.

One new aspect of these programmes will, it is hoped, be the development of metabolic epidemiology. This study depends on the development of techniques to investigate variations between population groups at the biochemical level. Within the foreseeable future it may be possible to initiate systematic studies of, for example, variations in enzymatic competence between different population groups, which might influence susceptibility to carcinogenic stimuli.

Information services

In view of the large number of information and abstracting systems—many of them computerized—that are now available, it would appear unnecessary for the Agency to duplicate them. However, an international organization like the Agency needs to evaluate published and abstracted data in its own field of competence. Within its monograph series, in particular, the Agency will continue to make a critical examination of already published information and to distribute this as widely as possible. The clearing-house of epidemiology and occupational cancer and the clearing-house of chemicals under test have both illustrated that there are still many gaps in existing knowledge which must be filled by planned and coordinated research at national and international levels.

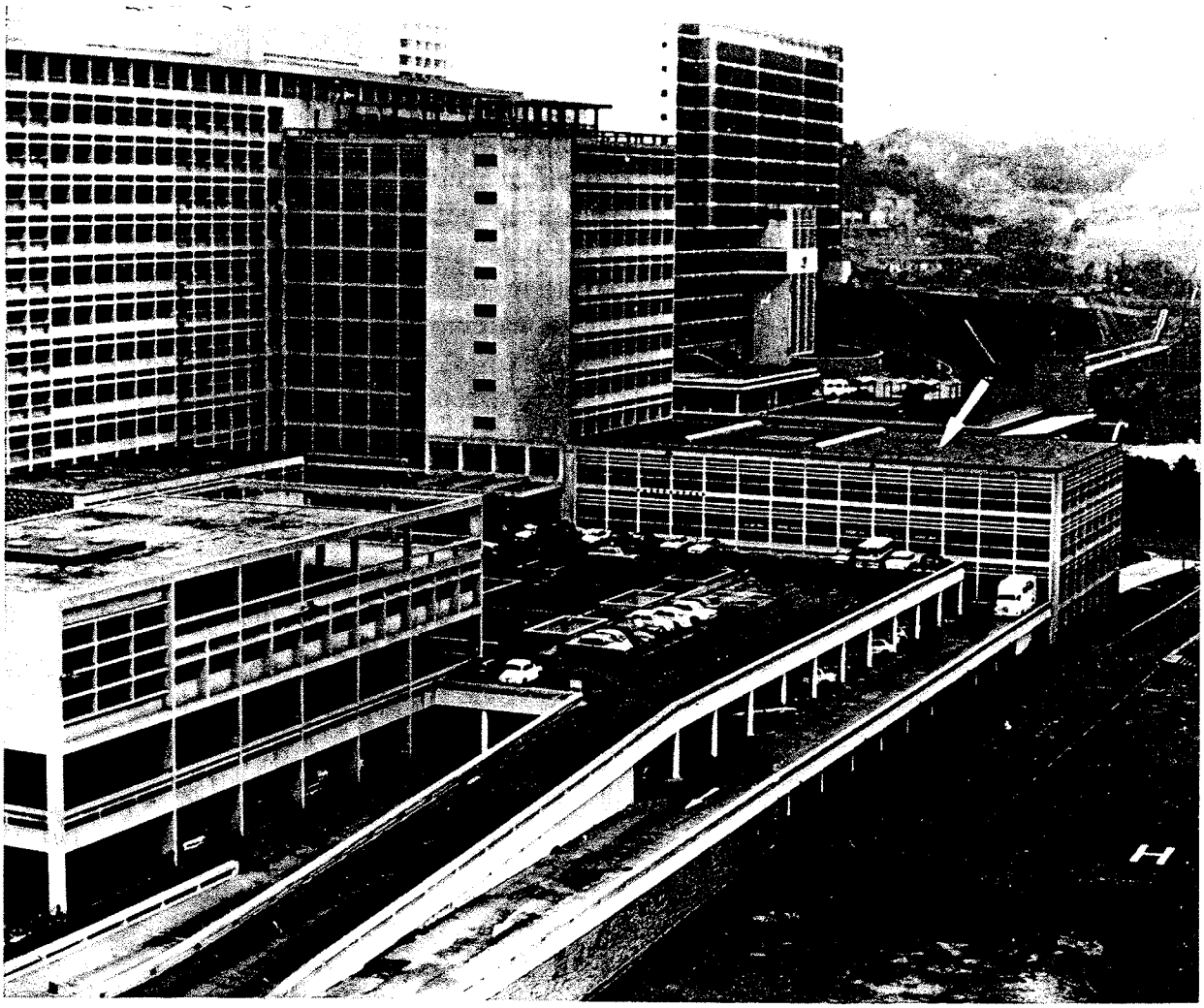
The role of the Agency in relation to cancer problems in developing countries

The Governing Council has required that the Agency develop and execute its programmes where its efforts appear most likely to be successful, irrespective of whether this work is carried out in a member country or not. It has assumed that all countries will benefit equally from the Agency's programme, since cancer is a universal problem, and information obtained in one area will rapidly be diffused to oncologists in others.

Much of cancer research requires major manpower and material resources. Most countries, accordingly, tend to concentrate research on those problems that are most pertinent to their own needs, maintaining just enough general scientific expertise to advise and evaluate research performed elsewhere. Such advice, however, is less readily available in developing states, although few are without some form of expertise in oncology.

The Agency undertakes research to solve problems of particular relevance to individual developing countries, such as the study of liver cancer in Africa, the role of mycotoxins and the carcinogenic properties of anti-parasitic drugs, to provide guidance, either directly or through national resources.

Fig. 4 Regional activities: Medical and Health Department Institute of Radiology and Oncology (indicated by an arrow) at Queen Elizabeth Hospital, Kowloon, Hong Kong where IARC-sponsored research is being carried out.



At a time when many developing countries rely on programmes of industrialization and improved agricultural practices to raise their standards of living, it is important that they have available evaluations of potential cancer hazards from chemicals, e.g., pesticides, drugs, fertilizers, etc. The Agency's current programme in this field was formalized by the Governing Council in Resolution GC/9/R10, which states:

> "By making available to governments all documentation in its (the Agency's) possession on substances on which governments will seek advice, and the conclusions the Agency has been able to draw from this documentation and, if need be, its advice on the methodology of experimental and epidemiological studies."

Manpower training

The Agency has an active training programme in cancer research, emphasizing epidemiology and mechanisms of carcinogenesis. Applications for fellowships and for participation in courses that come from non-member states are treated in the same way as those

from member countries. The candidates are selected *on merit* by an outside committee, which ensures that the training requested is pertinent to the needs of the home laboratory and that the candidate concerned will return to his home country. The committee can only support candidates who have the necessary basic educational level to profit from advanced training.

While most fellows supported by the Agency usually undergo their training in national laboratories, some are also trained in specific research areas within the Agency's laboratories. It is anticipated that the number of such in-house fellows from developing countries will increase as problems of industrialization and environmental cancer control, with their concomitant health and socioeconomic implications, become more prevalent.

The multidisciplinary nature of the Agency's programme, and, especially, its emphasis on epidemiology and environmental carcinogenesis, are of special importance to scientists who wish to maintain a wide knowledge in these problems rather than in a limited field. Thus, an oncologist may be required by the appropriate local authorities to give an opinion on environmental problems which may otherwise lie outside his field of competence. Such requests cannot readily be solved by an examination of primary sources, since they require a critical evaluation by experts of both published and unpublished data. When a fellow has been directly involved in the Agency's programme and has been exposed to several disciplines in coordinated projects he may be better equipped, not only to evaluate problems by using the Agency's resources, but also to have a more extensive appreciation of the expertise required. In addition, close working contact between scientists from developing and from industrialized states within the Agency's own programme will tend to improve technical and personal cooperation.

Conclusions

Clearly, the Agency must become and remain an island of competence and scientific expertise in the field of cancer within the countries of the United Nations and must retain the capacity to comment freely and accurately on the complex problem of environmental carcinogenesis, whether it involves an industrialized state or a developing country.

If it can continue to recruit and retain an expert staff capable of carrying out such tasks, it will fully justify its existence, since all too often national scientists and regulatory bodies are under limiting pressures. The need for such an organization is likely to remain as long as man attempts to improve his standard of living by the production of chemical compounds and as long as carcinogenic mechanisms remain a mystery. It is hoped, furthermore, that the organization can retain the scientific freedom necessary to follow the most recent developments in cancer research and that it will not become isolated from developments at the national level or divorced from the practical problems associated with the solution of day-to-day legislative questions.

1. UNIT OF EPIDEMIOLOGY AND BIOSTATISTICS

Dr C. S. MUIR (Chief)

1. INTRODUCTION

The Agency's role in coordinating descriptive epidemiological data has resulted in the publication of the third volume of the monograph, *Cancer Incidence in Five Continents*. A commentary describing the content of the first three volumes in non-technical terms is in preparation. The Clearing-House for On-going Research in Cancer Epidemiology has produced its second annual directory, and the index for the chapter on neoplasms of the Ninth Revision of the *International Classification of Diseases, Injuries, and Causes of Death* has been extended. Collaboration with the International Association of Cancer Registries in the production of a manual on cancer registration continues.

The search for the cause of oesophageal cancer has continued in the Caspian littoral of Iran and in Normandy. The Agency is involved in an extensive series of international studies to explore the role of alcoholic beverages in neoplasia. Case-control studies in Brittany have shown a multiplicative effect of alcohol and tobacco in the causation of oesophageal cancer. A survey of mortality in a cohort of the Danish brewery workers has been completed, and the results of a similar survey in Ireland are being analysed. Differences in the incidence of large-bowel cancer in urban Denmark and rural Finland have been studied in relation to diet, fibre intake, faecal steroids and microflora, and bowel function. This work is now being extended to other populations. The study of the effects of man-made mineral fibres on health, with particular reference to cancer, continues in Europe. The feasibility of creating an international epidemiological cancer network is being examined.

The statistical section continues to contribute to many of the research programmes of the Agency. Techniques have been devised for the estimation of cumulative cancer risk, and a monograph on modern techniques in the analysis of case-control studies is in preparation.

2. DESCRIPTIVE EPIDEMIOLOGY

The aim in descriptive epidemiology of cancer is to map the occurrence of the disease throughout the world and to improve the comparability of incidence data. The establishing of differences in cancer risk in different populations facilitates the formulation and testing of etiological hypotheses.

2.1 *International Classification of Diseases (ICD)* (Dr C. S. Muir)

A draft of the chapter on neoplasms for the Ninth Revision of the ICD has been prepared in conjunction with the International Classification of Disease Unit at WHO Headquarters (Dr K. Kupka, Mr G. Corbett) and was approved at a revision conference held in Geneva in 1975. The index, however, requires considerable further work, and codes for converting from ICD-O (the ICD adapted for use by oncologists) must now be prepared.

(*a*) *ICD-9 Index* (Mrs J. Nectoux)

When analysing the indexes for the Seventh and Eighth Revisions of the ICD, it became clear that much greater guidance for the coder was required to ensure that a given diagnosis, and its variants, was always coded in the same way. For this reason, particular care was taken to provide a code number for each histological term which has an implied site: thus, nephroblastoma of unstated site of origin must be coded to the kidney. Among other problems tackled was the assignment of likely site to rather vague diagnoses such as 'carcinoma of jaw'. Using the Index of the Eighth Revision of the ICD, such a diagnosis would be coded to malignant neoplasm of bone; in the Ninth Revision Index the proposed assignment is to gum, i.e., ICD 143.

(*b*) *ICD-Oncology (ICD-0)*

A specialized adaptation of the ICD—the ICD-Oncology or ICD-0—was published in December 1976 after extensive multinational field trials [1]. This classification is based on three independent axes: topography, morphology and behaviour, and is thus very flexible. However, for international comparison it is essential that this be collapsible into the neoplasm rubrics of the Ninth Revision of the ICD, and conversion codes are now being elaborated by Dr L. B. Thomas and Mrs C. Percy of the National Cancer Institute (Bethesda, Md., USA).

2.2 *Cancer registries*

(*a*) *International Association of Cancer Registries (IACR)* (Dr C. S. Muir)

In April 1973 an agreement (RA/73/016) was signed with the International Association of Cancer Registries whereby the Agency agreed to provide a secretariat and deputy-secretary for the Association. This agreement was renewed in 1976. The principal collaborative activity has been the publication of Volume III of *Cancer Incidence in Five Continents* (see section 2.3). Two issues of the Association Newsletter were published.

The International Association now has approximately 100 members. Four new registries were admitted to voting membership in 1976: Kuwait, Yorkshire, Scotland, Zaragoza.

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[1] *ICD-0, International Classification of Diseases for Oncology*, 1st ed., Geneva, WHO, 1976.

(*b*) *Caspian Cancer Registry* (see report of the Teheran Research Centre, page 135)

(*c*) *Singapore Cancer Registry* (see report of the Singapore Research Centre, page 127)

(*d*) *Jamaica Cancer Registry* (Professor S. E. H. Brooks: RA/72/014)

The Agency continues to provide minimal financial support to the registry, data from which appeared in the third volume of *Cancer Incidence in Five Continents*.

(*e*) *Dakar Cancer Registry* (Professor C. Quenum)

The cancer registry in Dakar was initiated in 1968 with the assistance of the Agency. Data on 4 766 cancer cases, collected for the years 1969-1974, were put on punch cards at the Agency. This material has now been checked and duplicates eliminated, leaving 4 165 cases for analysis. Of these, 1 894 were residents of the registration area, representing a crude rate of 45.2 per 100 000 per annum in the Cap-Vert province.

(*f*) *Computer teaching exercise*

At the workshop and training course on Cancer Registries and Occupational Cancer held in Lyon in 1975 [1], the practical work included investigation of an imaginary industrial hazard. The purpose of the exercise was to provide 'instant experience'. Each small group of students decided, on the basis of information about a possible industrial hazard in a factory, on appropriate methods of investigation; the computer then supplied the data in the form requested.

This exercise had several drawbacks, and it was decided to extend it to achieve a closer simulation of an actual situation. Such an extention requires considerable programming, and an agreement (RA/76/018) was made with Professor J. A. H. Waterhouse (Birmingham and West Midland Regional Cancer Registry, UK) to undertake this work, which is being supported by the National Cancer Institute (Bethesda, Md., USA). Miss Judith Fewings has reviewed and rewritten the basic analytical sections of the programme and is now directing her attention to the synthetic sections.

(*g*) *Cancer registration in Latin-speaking countries* (Dr A. J. Tuyns)

The Agency was represented at the second meeting held in Milan (Italy), on 19–20 May 1977, of the group for the epidemiology and registration of cancer in Latin-speaking countries. The problems of population-based cancer registries, as opposed to hospital-based registries, were discussed. Reports were presented on the work of various cancer registries operating in Spain, Italy, France and Switzerland. Proposals were put forward for collaborative studies on the reliability of death certificates in these countries and for a collaborative study on cancer of the larynx by several of the registries represented at the meeting.

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[1] International Agency for Research on Cancer (1976) *Annual Report, 1976*, Lyon, p. 29.

(*h*) *Advice to cancer registries and other bodies*

An increasing number of requests for advice and assistance are being received by the Unit, many being referred by the International Union Against Cancer and the Cancer Unit of WHO. Specifically, advice was provided to the following countries:

Algeria: further exploitation of existing data on cancer frequency and improvement of the recording system (Professor A. Yaker, Chief, Laboratory of Pathology, Mustapha Hospital, Algiers)

France: establishment of a cancer registry in the Grenoble area (Dr R. Schaerer, University Hospital Centre of la Tronche, Grenoble)

Spain: establishment of cancer registries (Dr J. M. Francia Viña, Cancer Section, Directorate-General of Public Health, Madrid)

Switzerland: The section of cancer registration and epidemiology of the Swiss League Against Cancer comprises at present six registries operating in various cantons of Switzerland. The League has been assisted in evaluating the functioning of these registries.

In addition, many inquiries about the chapter on neoplasms in the *International Classification of Diseases* and on the availability of information on the geographical distribution of various cancers were answered.

2.3 Cancer Incidence in Five Continents, *Vol. III* (Dr C. S. Muir, Miss S. Whelan)

The third volume of *Cancer Incidence in Five Continents*, containing incidence rates for 78 populations in 28 countries, was published towards the end of 1976 [1]. In addition to age-specific and age-standardized incidence rates by site and sex, further tables giving age-standardized incidence for 49 four-digit rubrics are included.

An extensive discussion is provided on the reliability of data, with tables that give the proportion of registration with histological confirmation of diagnosis, the proportion based on a death certificate only, and correlation with available mortality data. Equivalence tables are given for the chapter on neoplasms of the Seventh and Eighth Revisions of the *International Classification of Diseases*.

A chapter is devoted to a new measure of age-standardized incidence, the cumulative rate, which is both a directly standardized incidence rate and a good approximation to the actuarial or cumulative risk. This rate is the sum for each year of age of the age-specific incidence rates taken from birth to age 74. It can be interpreted either as a directly age-standardized rate with the same population size in each age group or as an approximation to the cumulated risk. The cumulative risk is the risk an individual would have of developing cancer up to a certain age if no other causes of death were in operation. Examples of these rates are given in Table 2. This measure is simple to use and can be combined with a relative risk obtained from analytical studies to obtain measures of risk of particular groups. It is a natural way of expressing the tumour experience of birth cohorts defined by year, and the measure is directly comparable with risks observed in animal experiments when the latter are analysed by the life-table method.

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[1] Waterhouse, J. A. H., Muir, C. S., Correa, P. & Powell, J., eds (1976) *Cancer Incidence in Five Continents*, Vol. 3, Lyon, International Agency for Research on Cancer (*IARC Scientific Publications* No. 15).

Table 2. Cumulative rates for the age-span 0–74 years for selected cancer sites and cancer registries from 1968–1972, expressed as percentages

ICD code	151 Stomach		153–4 Large bowel		162 Bronchus		174 Breast	180 Cervix uteri	185 Prostate	188 Bladder		204–7 Leukaemias		140–209 All sites	
	M	F	M	F	M	F	F	F	M	M	F	M	F	M	F
Nigeria, Ibadan	0.8	0.7	0.3	0.4	0.1	0.1	1.7	2.4	1.4	0.5	0.3	0.4	0.4	8.8	11.4
Colombia, Cali	4.8	3.1	0.7	0.8	2.1	0.6	2.9	6.6	2.0	0.8	0.3	0.6	0.4	18.1	22.8
USA, Alameda County															
White	1.1	0.6	4.9	3.8	7.2	2.3	8.4	1.3	4.6	2.2	0.6	0.9	0.5	32.5	30.2
Black	2.9	0.8	3.8	3.3	8.5	1.8	6.2	2.9	9.9	1.1	0.5	1.1	0.6	39.6	24.8
India, Bombay	1.0	0.6	0.9	0.6	1.6	0.4	2.2	2.4	0.6	0.3	0.1	0.3	0.2	15.0	12.8
Israel, all Jews	2.6	1.5	2.6	2.6	3.7	1.3	6.2	0.5	1.6	2.0	0.4	0.7	0.6	24.9	25.3
Japan, Miyagi	10.2	4.6	1.5	1.2	2.5	0.9	1.3	1.5	0.3	0.4	0.2	0.4	0.3	22.1	14.2
Singapore															
Chinese	5.8	2.3	2.8	1.9	7.6	2.3	2.2	2.0	0.4	0.8	0.3	0.4	0.4	31.6	17.8
Malays	1.2	1.1	0.9	0.6	2.1	1.0	2.0	1.2	0.5	0.5	0.1	0.4	0.2	11.2	10.5
Indians	2.5	2.6	1.2	1.7	1.3	1.3	2.6	2.9	0.7	0.4	0.6	0.5	0.4	15.9	19.9
Norway	2.8	1.4	2.7	2.3	3.0	0.6	4.9	1.8	3.7	1.2	0.4	0.7	0.5	22.5	20.1
UK, Birmingham	2.8	1.2	3.8	2.7	10.3	1.4	5.8	1.3	1.8	2.0	0.5	0.6	0.4	28.7	20.3
New Zealand															
Maoris	3.9	1.9	1.5	1.7	7.4	3.8	5.7	3.2	2.2	0.4	0.7	0.7	0.3	25.8	27.0
Non-Maoris	1.8	0.7	4.4	3.8	6.5	1.0	5.8	1.1	2.7	1.2	0.3	0.8	0.5	25.8	20.9

Note: This table shows, for example, that, in the absence of other causes of death, male Nigerians in Ibadan have a 0.8% chance of developing cancer of the stomach before the age of 75; white females resident in Alameda County, California have an 8.4% chance of developing breast cancer before the same age.

2.4 *Commentary on* Cancer Incidence in Five Continents (Dr M. Stukonis)

The first three volumes of *Cancer Incidence in Five Continents* [1, 2, 3] cover a period of approximately 15 years. The data contained therein consist of a series of tabulations with virtually no commentary, and the significance of the data is not immediately apparent. Work has now started on a monograph which will present the data, including changes over time, in a series of diagrams and charts supported by a brief explanatory text, designed for use by the medical administrator, non-specialist scientist and informed layman. This monograph will subsequently be extended by supplements which will contain analyses of interest to the cancer epidemiologist, including correlation between different cancer sites by sex, analyses of the shape of age-incidence curves, sex ratio by age, etc. As a preliminary step, cumulative rates and risks have been computed from the material contained in the three volumes.

It should be realized, however, that cancer incidence statistics from Africa, Asia and South America are very limited and that there are as yet no data from Australia or USSR. International incidence time trends can currently be studied in only 14 countries (Canada, Colombia, Denmark, the Federal Republic of Germany, Finland, Iceland, Jamaica, Japan, Norway, Singapore, Sweden, the UK, the USA and Yugoslavia).

2.5 *Cancer registration and its techniques* (Dr R. MacLennan)

There is increasing awareness of the contribution that cancer registries can make to many aspects of cancer control, including epidemiological studies, evaluation of survival and health services planning. The Agency has frequently been approached to give advice on the establishment and running of such registries and is now involved in the preparation of a manual on this topic. This manual aims to promote the development of cancer registries, especially in those areas where cancer patterns are as yet poorly described, and to enhance and ensure international comparability of cancer data by providing common nomenclature and descriptions of various aspects of cancer registration. Although the bias of the manual is towards population-based cancer registries, the functions of those based on hospitals [4] are delineated throughout, and the complementary nature of their activities is emphasized. This work has been undertaken in collaboration with the International Association of Cancer Registries (Dr R. Steinitz) and the Cancer Unit, WHO (Dr A. Winkler). Chapters in the manual cover difficulties likely to be encountered and rules to be followed in coding, a review of the input operations in a registry, comments on classification and coding and output operations and reports. Problems of data processing are considered.

2.6 *Ratio studies*

The Agency continues to foster the collection of relative frequency data from cancer institutes and pathology laboratories in regions where cancer registration would be difficult and mortality information unreliable or non-existent.

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[1] Doll, R., Payne, P. & Waterhouse, J., eds (1966) *Cancer Incidence in Five Continents*, Geneva, UICC.

[2] Doll, R., Muir, C. & Waterhouse, J., eds (1970) *Cancer Incidence in Five Continents*, Vol. 2, Geneva, UICC.

[3] Waterhouse, J. A. H., Muir, C. S., Correa, P. & Powell, J., eds (1976) *Cancer Incidence in Five Continents*, Vol. 3, Lyon, International Agency for Research on Cancer (*IARC Scientific Publications* **No. 15**).

[4] *WHO Handbook for Standardized Cancer Registries (Hospital-Based)*, Geneva, WHO (*WHO Offset Series* No. 25), 1975.

(*a*) *Indonesia* (Dr O. M. Jensen, Dr C. S. Muir)

Analysis of material collected by Dr Soeripto of the Pathology Department of the Yogyakarta Medical School has now been completed [1].

(*b*) *Cameroon* (Dr O. M. Jensen, Dr A. J. Tuyns)

The analysis of 2 808 cases of cancer examined histologically by Dr P. Ravisse at the Institut Pasteur of Yaoundé (Cameroon) between 1969 and 1973 has been completed and submitted for publication.

2.7 *Clearing-house for on-going research in cancer epidemiology* (Dr C. S. Muir, Mrs A. Nagy-Tiborcz, Mrs E. Démaret)

The clearing-house for on-going research in cancer epidemiology was created in 1974 by the Agency and the German Cancer Research Centre, Heidelberg, Federal Republic of Germany (Professor G. Wagner, Dr C. O. Köhler, Mr K. Schlaefer: RA/74/003) and operates within the framework of the International Cancer Research Data Bank of the National Cancer Institute (Bethesda, Md., USA).

The goal of the clearing-house is to provide workers in the field and other scientists, governments and public health officials with information on on-going studies in cancer epidemiology through the annual *Directory of On-going Research in Cancer Epidemiology* and by a special searches service.

Although 'epidemiology' is interpreted broadly, the clearing-house does not solicit information on clinical trials, diagnosis or mass-screening programmes, unless these include epidemiological evaluation.

The first annual directory was published in July 1976 [2]. It contained 622 projects reported by 442 principal investigators in 56 countries. Some 2 000 copies of this directory have been distributed to principal investigators, ministries of health, medical journals, cancer research centres and cancer registries, to industry, libraries, medical research boards, research workers and individuals. Copies have also been distributed at meetings and to scientists visiting the Agency and the German Cancer Research Centre. While descriptive epidemiology is undertaken in a fairly wide range of countries, analytical studies tend to be more frequent in North America, Scandinavia and the UK. There are considerable numbers of investigations of industrial and non-industrial high-risk groups and of case-control studies. Studies of breast and respiratory-tract cancer are frequent, but common sites like pancreas and prostate are currently largely ignored.

The clearing-house address list contains at present some 5 000 names. Mailing for the second directory, which began in November 1976, comprised 3 415 letters of invitation. In response, 500 relevant new projects were reported to the clearing-house; 145 persons had no project to report; 235 were not working in the field of cancer epidemiology; and 97 of the projects reported were considered not to fall within the scope of the clearing-house and were forwarded to the current cancer research project analysis centre of the International

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[1] Soeripto, Jensen, O. M. & Muir, C. S. (1977) *Brit. J. Cancer* **36**, 141–148.

[2] Muir, C. S. & Wagner, G., eds (1976) *Directory of On-going Research in Cancer Epidemiology*, 1976, Heidelberg, German Cancer Research Centre.

Cancer Research Data Bank Program, or to the UICC clearing-house for controlled therapeutic trials, so that the information was not lost.

The second annual directory [1] contains information on 905 epidemiological projects undertaken in 70 countries.

3. ANALYTICAL EPIDEMIOLOGY

3.1 *Etiological factors in oesophageal cancer*

(*a*) *France* (Dr A. J. Tuyns, Dr O. M. Jensen). See section 3.2 (*a*)

(*b*) *Caspian littoral* (Dr J. Kmet, Dr N. E. Day). See report of the Teheran Research Centre (page 135).

3.2 *Studies on alcohol and cancer* (Dr A. J. Tuyns)

The Agency, with the support of the National Institute on Alcohol Abuse and Alcoholism (NIAAA) (Rockville, Md., USA), has developed a programme for the study of the role of alcohol in human cancer. An extensive review of pertinent literature has been prepared.

At the fourth annual review meeting (Lyon, 23 March 1977), principal investigators reported on the present status of their studies in Belgium, Denmark, Ireland, France and Switzerland. The laboratory studies carried out in Caen and Lyon were also reported (see page 56).

(*a*) *France* (Dr A. J. Tuyns, Dr O. M. Jensen)

(i) *Methodology of case-control studies: choice of an appropriate control group*

In the study on oesophageal cancer in Ille-et-Vilaine (France), two control groups were interviewed: 600 hospital controls matched for sex and age and a group of 778 persons taken from the general population. These two groups exhibited a certain number of differences, particularly with regard to their consumption of alcohol and tobacco, the main factors being investigated: a higher alcohol consumption was found among hospital controls, mainly due to the inclusion of patients with forms of cancer associated with drinking, and their tobacco consumption was also higher, in relation not only to smoking-related diseases but also to other diseases not hitherto associated with smoking.

The implications of these findings are that hospital controls do not provide an accurate measure of such widespread habits as drinking and smoking and that, for a refined study, population controls should be used. A more detailed discussion of these points has been published [2].

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[1] Muir, C. S. & Wagner, G., eds (1977) *Directory of On-going Research in Cancer Epidemiology*, Lyon, International Agency for Research on Cancer (*IARC Scientific Publications* No. 17).

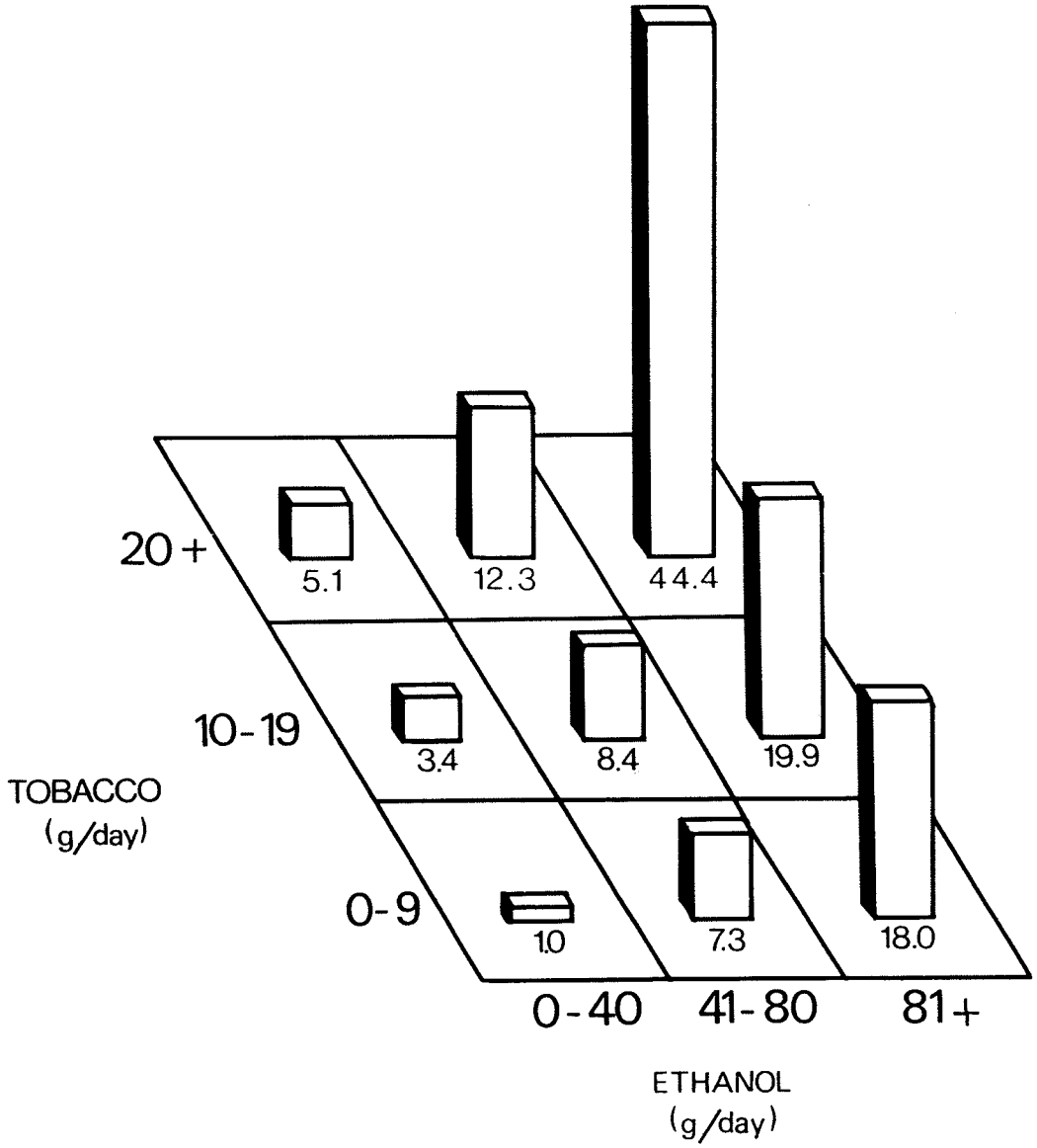
[2] Tuyns, A. J., Jensen, O. M. & Péquignot, G. (1977) *Rev. Epid. Santé publ.*, **25**, 67–84.

(ii) *Alcohol and tobacco*

In Ille-et-Vilaine (France), the risk of oesophageal cancer is clearly related to consumption of alcohol: the logarithm of this risk is a pure linear function of the average daily consumption expressed in grams of pure ethanol. A similar relationship has been found for the consumption of tobacco. These two risks act independently and, when combined, multiply their effects [1] (Fig. 5).

The variations in alcohol and tobacco consumption between males and females and between rural and urban populations are responsible for the differences in risk encountered between these groups—higher in rural areas and very much higher in males; the distribution of oesophageal cancer in Ille-et-Vilaine can be explained in terms of these two factors

Fig. 5 Relative risks for cancer of the oesophagus in relation to daily consumption in grams of alcohol and tobacco.



[1] Tuyns, A. J., Péquignot, G. & Jensen, O. M. (1977) *Bull. Cancer*, **64**, 45–60.

only. This does not exclude the possibility that other factors, whose effect would be negligible in Ille-et-Vilaine, could be much more important in other parts of the world where the risk of oesophageal cancer is also high.

The study in Normandy (France) is being continued and will include cancers of the other segments of the digestive tract and of the larynx. A laboratory has been set up in the locality to study the mutagenicity of various fractions of alcoholic beverages collected in the region (Professor J. Y. Le Talaer, François Baclesse Regional Centre, Caen). These studies are carried out in close collaboration with the Agency (see 56). In the Department of Pathology of the same Centre, work is continuing on the oesophagi collected from patients and from controls (Dr A. M. Mandard) (see page 124).

(iii) *Nutrition and oesophageal cancer*

The nutritional aspects of œsophageal cancer have been examined with Dr G. Péquignot, Nutrition Section of the French National Institute of Health and Medical Research (INSERM) (RA/75/015). Past intake of various foodstuffs by some 200 oesophageal cancer patients was compared with that of 778 population controls. No important difference in nutrients was found, except that intake of retinol and pyridoxine was slightly higher in the group of oesophageal cancer cases than in the controls (Table 3). The distribution of

Table 3. Mean daily intake of selected nutrients among oesophageal cancer patients and population controls in Ille-et-Vilaine, France

	Oesophageal cancer patients [a]	Population controls [b]
Protein	87.7 g	93.2 g
Animal	52.2 g	57.3 g
Vegetable	35.6 g	35.9 g
Carbohydrates	358 g	345 g
From sugar	44 g	46 g
From starch	314 g	299 g
Fats	125.2 g	125.4 g
Vegetable	56.7 g	60.4 g
Animal	68.5 g	65 g
Calories (excl. alcohol) [c]	2910	2882
$\frac{\text{Protein calories}}{\text{Total calories (excl. alcohol)}} \times 100$	12.1 %	13 %
Carotene	457.8 mg	460.7 mg
Retinol	138.3 mg	112.7 mg
Thiamine (Vitamin B_1)	1.48 mg	1.53 mg
$\frac{\text{Thiamine}}{\text{Calories (excl. alcohol)}} \times 1000$	0.051 mg	0.054mg
Riboflavin (Vitamin B_2)	1.57 mg	1.59 mg
Pyridoxine (Vitamin B_6)	41.8 mg	23.0 mg
Vitamin C	126 mg	124 mg
Iron	18.5 mg	16.7 mg
Calcium	719 mg	711 mg
Alcohol (expressed as ethanol)	85.6 g	44.0 g
Total calories [d]	3510	3190

[a] Mean of 200 patients
[b] Mean of 778 controls
[c] Energy (excl. alcohol) 12,180 J 12,063 J
[d] Total energy 14,690 J 13,352 J

cases and controls into consumers and non-consumers of various food items was examined. After appropriate correction for age and urban/rural status—factors that entail variation in diet—differences were found for 13 food items (Table 4). The patients ate meat less often but offal more often; they also ate more dried vegetables, salted butter and potatoes. It is premature to assess the significance of these findings.

No differences were found in the consumption of coffee and tea, but there were fewer water drinkers among oesophageal cancer cases than among controls. There were, however, more consumers of beer, cider, wine and liqueurs among the cancer patients. These observations point to the role of alcohol which has already been described.

Table 4. Dietary items for which differences in consumption were observed between oesophageal cancer patients and population controls in Ille-et-Villaine, France

Daily consumption	Number of oesophageal cancer patients [a]	Expected no. [b]	Significance $P \leqslant$
Meat (> 50 g)	29	42.6	0.05
Offal	30	13.2	0.001
Smoked fish	11	4.6	0.005
Pulses	42	19.9	0.001
Potatoes (> 200 g)	70	53.0	0.025
Salted butter	47	32.9	0.025
Other fats	22	35.6	0.025
Chocolate	11	24.2	0.01
Mineral water	10	19.6	0.05
Beer ($\geqslant$ 500 ml)	7	2.8	0.025
Wine ($\geqslant$ 500 ml)	21	6.7	0.001
Cider ($\geqslant$ 1000 ml)	28	12.1	0.001
Liqueurs ($\geqslant$ 50 ml	15	5.4	0.001

[a] Out of 200 cancer patients
[b] Based on the consumption habits of the controls

(*b*) *Denmark* (Dr O. M. Jensen)

A suggested association between the consumption of beer and cancer of the colon and rectum [1] has been studied by means of a retrospective cohort study of male Danish brewery workers, a group known to have a high daily consumption of beer.

After excluding duplicates and eight persons with incomplete identifying information, 14 313 brewery workers remained for follow-up; for 74 (0.5%), it was not possible to determine whether they were dead or alive by the end of the follow-up period, 1943–1973. Independent examination of a sub-sample of the total cohort confirmed the validity of the follow-up.

Causes of death in the cohort have been determined with the assistance of the Danish National Health Service (Dr J. Mosbech, Mr S. Sørensen: RA/75/017). Expected numbers of deaths were calculated on the basis of general population mortality rates, procured with the help of the Danish Institute of Clinical Epidemiology (Dr O. Horwitz: RA/76/023).

A comparison of observed and expected number of deaths from various diseases among Danish brewery workers showed no significant differences for cancer of the rectum

[1] Breslow, N. E. & Enstrøm, J. E. (1974) *J. nat. Cancer Inst.*, **53**, 631–639.

and cancer of the colon but a significant excess risk for cancer of the oesophagus and larynx. Mortality from cirrhosis of the liver, gastroduodenal ulcer and motor vehicle accidents was also higher than expected. The cohort of brewery workers also showed a slight, but numerically important, excess mortality from all causes (Table 5).

Table 5. Selected causes of death for the period 1974-1973 among male members of the Danish Brewery Workers Union (The expected numbers are based on mortality rates for the male general population after adjustment for age and place of residence).

Cause of death	Observed (O)	Expected (E)	O/E
Malignant neoplasms			
Buccal cavity and pharynx	11	13.55	.81
Oesophagus	36	19.04	1.89 [c]
Stomach	88	95.28	.92
Colon	63	58.18	1.08
Rectum	62	54.56	1.14
Liver and biliary passages	42	28.19	1.49 [b]
Pancreas	44	40.92	1.08
Larynx	25	9.62	2.59 [c]
Lung	280	241.43	1.16 [a]
Urogenital tract	131	119.51	1.10
Leukaemia	25	26.32	.95
Other malignant neoplasms	144	122.56	1.17
Metabolic disorders			
Diabetes mellitus	40	34.45	1.16
Mental disorders			
Psychosis, other mental disorders	7	6.44	1.09
Alcoholism	4	3.61	1.11
Cardiovascular disease			
Stroke	264	240.37	1.10
Chronic rheumatic heart disease	19	22.35	.85
Arteriosclerotic and degenerative heart disease	874	883.08	.99
Other cardiovascular disease	375	356.19	1.05
Respiratory disease			
Pneumonia	72	64.72	1.11
Bronchitis	50	65.68	.76
Gastrointestinal disease			
Gastroduodenal ulcer	67	42.53	1.58 [c]
Appendicitis	9	10.29	.87
Cirrhosis of liver	85	48.12	1.77 [c]
Cholelithiasis, cholecystitis	20	15.04	1.33
Pancreatic disease	8	7.16	1.12
Urogenital disease			
Nephritis	31	22.25	1.39
Violent death			
Motor vehicle accidents	83	62.25	1.33 [b]
Other accidents	104	111.72	.93
Suicide	121	115.61	1.04
Homicide	1	1.81	.55
All causes of death	3185	2942.83	1.08

[a] $P < 0.05$ [b] $P < 0.01$ [c] $P < 0.001$

The cancer morbidity experienced by the cohort has been established in collaboration with the Danish Cancer Registry (Dr J. Clemmesen: RA/73/019). The 1309 cancer cases available for analysis will permit a detailed study of the cancer pattern in sub-groups of the total cohort.

(*c*) *Ireland* (Dr R. MacLennan)

A substantially similar study in Ireland is nearing completion. The health effects on brewery workers of another type of beer are being investigated by the Medico-Social Research Board (Dr G. Dean: RA/76/016).

A parallel study of the cancer experience of whisky distillery workers has now begun.

3.3 *Large-bowel cancer*

Large-bowel cancer, a major cause of death in Europe, North America and Australasia, shows considerable geographical variation. The Agency is coordinating an international collaborative study of this disease in areas where there are contrasts in incidence and where population-based cancer registries exist.

(*a*) *Diet and large-bowel cancer* (Dr R. MacLennan)

The Agency is coordinating a series of studies of three aspects of diet which may explain many of the geographical differences in colon cancer. These are: (i) high fat intake, which may increase the bile acids in the large bowel to be subsequently metabolized by bacterial flora to carcinogens or co-carcinogens; (ii) high meat consumption, although the strong correlation with colon cancer might be related to the fat content; and (iii) deficiency of dietary fibre.

(i) *Intestinal micro-ecology in Denmark and Finland* (Dr R. MacLennan, Dr O. M. Jensen)

A comparison of dietary intake and faecal characteristics has now been completed [1] in population samples from two areas, one in urban Denmark and the other in rural Finland, with a four-fold variation in colon cancer incidence. The study suggests that the etiology of colon cancer may be multifactorial; it is not associated in a simple manner with either dietary fat, neutral steroids, acid steroids or their bacterial metabolites. However, meat consumption was greater in the high-incidence areas. Higher intake of dietary fibre and milk in the low-incidence area suggests the existence of a possible protective effect, unrelated to mouth-anus transit time.

Further dietary and metabolic studies are needed to clarify the relationships between possible carcinogenic and protective effects of diet. Investigations are being initiated in

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[1] IARC Intestinal Micro-ecology Group (1977) *Lancet*, *ii*, 207. Collaborating institutions and members were: Department of Medicine, St. Elizabeth's Hospital, Copenhagen, Denmark (Dr J. Mosbech, Dr K. Buschard, Dr J. Dejgard, Dr H. Bardram, and Dr E. Tvedegard); Department of Community Health, University of Kuopio, Kuopio, Finland (Prof. H. Vuori, Dr S. Kokko, and Dr S. Karjalainen); Institute for Medical Microbiology, Uppsala, Sweden, (Prof. G. Laurell, Dr A. C. Rydén, and Dr A. Schwan); Bacterial Metabolism Research Unit, Colindale, London (Sir Robert Williams, Dr B. S. Drasar, and Dr M. J. Hill); and M. R. C. Dunn Nutrition Unit, Cambridge (Dr W. P. T. James, Dr J. H. Cummings, and Dr D. A. T. Southgate).

two other areas in rural Denmark and in urban Finland. Other studies to examine the same hypothesis under differing conditions will be organized during 1978 in Iceland, Singapore, Australia and New Zealand.

(ii) *Dietary fibre*

Studies of the effects of dietary fibre are at present impeded by the lack of food tables that give values for the various types of dietary fibre. *Ad hoc* laboratory analysis of fibre in food is tedious and expensive. A symposium sponsored jointly by the European Economic Community and the Agency will be held in 1977 in Lyon to recommend methods for measurement of dietary fibre, so that national food tables can be updated with comparable methodology. This will greatly facilitate future international collaborative studies of diet and cancer.

(*b*) *Large-bowel pathology in autopsy series* (Dr R. MacLennan)

The large bowels from a series of autopsies are being examined in a comparable manner in Denmark, Finland, Norway, The Netherlands, Poland and the UK [1].

Collection of material from 280 unselected autopsies in Trömso, Norway (Dr H. Stalsberg, Dr T. J. Eide) is now complete, and collection from Kuopio, Finland, will be finished in 1977. The protocol for this study, which is compatible with that used in studies in North and South America and Japan, is available to interested investigators.

(*c*) *Sub-site distribution of cancer of the large bowel* (Dr O. M. Jensen, Dr R. MacLennan)

It has been suggested that lower rectal cancer differs etiologically from cancers of the upper rectum [2]. In an attempt to generate hypotheses concerning the etiology of this neoplasm, a descriptive study of the distribution of malignant neoplasms within the rectum has been planned. In collaboration with the national cancer registries of Denmark and Norway, whose data show a two-fold difference in rectal cancer incidence and almost identical levels of sigmoid cancer [3], prospective registration of detailed localization of tumours in the sigmoid colon and rectum has begun in Copenhagen and Oslo; the study will be extended to areas with differing rectal cancer incidence.

3.4 *Cancer of the stomach and rectum in Belgium* (Dr A. J. Tuyns)

A review of cancer mortality data in Belgium revealed higher rates for stomach and rectal cancer in the Flemish part of the country than in the Walloon part [4]. More recent data for the years 1972–1975 have now been analysed and show the same pattern. Information on diet available for both parts of the country indicates differences in the consumption of various food items, some of which are consistent with the current hypothesis of the role of dietary fibre. A case-control study in two provinces, one in the Flemish and the other in the Walloon part of the country, has been initiated.

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[1] International Agency for Research on Cancer (1975) *Annual Report, 1975*, Lyon, p. 37.

[2] Berg, J. W. & Howell, M. A. (1974) *Cancer*, **34**, 807.

[3] De Jong, U. W., Day, N. E., Muir, C. S., Barclay, T. H. C., Bras, G., Foster, F. H., Jussawalla, D. J., Kurihara, M., Linden, G., Martinez, I., Payne, P. M., Pedersen, E., Ringertz, N. & Shanmugaratnam, K. (1972) *Int. J. Cancer*, **10**, 463–477.

[4] Ramioul, L. & Tuyns, A. J. (1977) *Acta gastro-ent. belg.* (in press).

3.5 *Smoking, chewing and drinking habits in Northern Thailand* (Dr R. MacLennan)

The case-control study of cancers of the oral cavity, pharynx/larynx and lung in Chiang Mai, Northern Thailand [1, 2] has now been published, jointly with results of analyses of the various types of 'cigar' and cigarette smoked locally [3]. Smokers of cigars with alkaline smoke had increased risks for larynx and lung cancers, whereas those who smoked cigars with an acid smoke had an increased lung cancer risk. These differences in risk were not statistically significant, possibly because of the considerable variation in the type of tobacco and additives in cigars, with consequent imprecision in responses to the fairly broad questions asked in the case-control study.

A medico-anthropological survey of smoking, chewing and alcohol drinking habits was done in Ban Pong, a typical Northern Thai village near Chiang Mai (Miss C. Mougne). The detailed information obtained on these habits will be valuable for future studies in Northern Thailand and Burma.

The daily consumption of cigars in Ban Pong and the results of laboratory analyses of smoke were used to estimate total lifetime tobacco tar exposure in this population. The mean lifetime tar exposure of male and female daily smokers in Ban Pong over 50 years of age is equivalent to that from smoking 30 medium-tar Australian cigarettes per day for 30 years.

3.6 *Lung cancer in Singapore Chinese* (Dr R. MacLennan)

The high incidence of lund cancer in Chinese females in Singapore, especially among those speaking the Cantonese dialect, and the relatively high rates in Chinese males of all dialect groups have been studied by means of interviews of cases and controls. Although a significant dose-response effect for cigarette smoking was found in males and females, smoking cannot explain the high rates of adenocarcinoma of the lung observed in Cantonese females which is apparently unrelated to smoking [4]. No other environmental exposure can be implicated, although persons with a low consumption of green vegetables were at higher risk, perhaps because of an increased risk from smoking in the presence of a relative deficiency of vitamin A [5].

Further studies are now under way, including a case-control study in Hong Kong (Professor M. Colbourne: RA/77/010) of the relation of smoking to lung cancers of different histological type in females and a search for mutagenic activity in vapours produced during Chinese cooking.

3.7 *Lung cancer in Cuban women* (Dr R. MacLennan, Dr O. Joly)

Lung cancer mortality among Cuban women is the highest for females in the Americas. A case-control study is being conducted in Havana in collaboration with the National Institute of Oncology and Radiobiology, Havana (RA/77/016) to test whether this high risk is related to a high cigarette consumption and whether there are differences in risk

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[1] International Agency for Research on Cancer (1975) *Annual Report, 1975*, Lyon, p. 38.
[2] International Agency for Research on Cancer (1976) *Annual Report, 1976*, Lyon, p. 41.
[3] Simarak, S., de Jong, U. W., Breslow, N., Dahl, C. J., Ruckphaopunt, K., Scheelings, P. & MacLennan, R. (1977) *Brit. J. Cancer*, **35**.
[4] International Agency for Research on Cancer (1976) *Annual Report, 1976*, Lyon, p. 41.
[5] Bjelke, E. (1975) *Int. J. Cancer*, **15**, 561–565.

for smokers of cigarettes containing light or dark tobaccos. It is hoped to interview most cases, and completeness of coverage will be checked through the cancer registry. Analytical methods used will be comparable to those of other Agency studies and those of the US smoking and health programme.

3.8 *Latent carcinoma of the prostate*

The international comparative study of the frequency and characteristics of latent carcinoma of the prostate has now been completed. A report has been prepared by Dr H. Tulinius (Iceland), coordinator of the study, and Dr R. A. B. Drury (Uganda), in collaboration with the other participants, Professor G. Dhom (Federal Republic of Germany), Dr C. W. Chan (Hong Kong), Professor B. Gellei (Israel), Dr B. Sparke (Jamaica), Dr Lee Yoke Sun (Singapore), Dr S. Lundberg (Sweden) and Dr N. Breslow (University of Washington, Seattle, Wash., USA), who carried out the statistical analysis for the study, and this has now been published [1].

Standardized methods of microscopic evaluation were used, and each of the 1 327 prostates examined underwent a 'blind' re-evaluation by a pathologist other than the one in the area of origin. The morphological features were examined of 350 latent carcinomas of the prostate found. Statistical analysis of the difference in frequency between areas and age groups was carried out, and likelihood ratio tests showed that both area and age had highly significant effects ($P = < 0.0001$). The statistical significance of the pathologist effect was smaller, although clearly present ($P = < 0.01$).

The study has confirmed that latent carcinoma of the prostate has a high frequency, occurring in some 20% of all males over 44 years of age. The frequency of the smallest latent carcinomas [2] was about 12% in all the areas investigated and did not vary with age, whereas larger latent carcinomas increased sharply with age, exhibiting an area-to-area variation resembling that of clinical carcinoma of the prostate. The 186 medium and large-sized latent carcinomas were found mainly in the two European and two black populations.

3.9 *Industrial exposure*

(*a*) *Health risks from man-made mineral fibres* (Dr R. Saracci, Dr J. E. H. Milne)

(i) *Production industries*

The study is financed by the Joint European Medical Research Board of the Comité International de la Rayonne et des Fibres Synthétiques (CIRFS) and the European Insulating Manufacturers Association (EURIMA) and is directed by the independent scientific and technical committee of the board. The committee includes representatives of the Agency, the Institute of Occupational Medicine, Edinburgh, UK and the Medical Research Council Pneumoconiosis Unit, Cardiff, UK. In response to requests that a representative of organized labour be on the committee, Dr Eric Bolinder, Medical Adviser to the Swedish Trade Union Confederation, was appointed in January 1977, having received a mandate from the International Confederation of Free Trade Unions.

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[1] Tulinius, H. *et al.* (1977) *Int. J. Cancer*, **20**, 680-688.

[2] Five or six slices of each gland were prepared, and each slice was divided into octants; a latent carcinoma was scored as 'small' if it appeared in only one or two of the 40 or 48 octants.

CIRFS and EURIMA have supplied a nominal roll of the 72 factories in their organization, which are situated in 18 countries. Visits have been made systematically to plants in Austria, Denmark, England, the Federal Republic of Germany, Finland, France, Northern Ireland, Norway, Scotland, Spain and Sweden. Data obtained from each plant included numbers of workers, fibre type, method of manufacture, the year production started, availability and quality of personnel records and facilities for follow-up. In some, but not all plants, measurements of mean fibre diameter were available.

On the basis of these data, a preliminary assessment was made for each factory as to its suitability for involvement in either an historical follow-up study or an on-going prospective study. Of the 72 factories, 46 have now been assessed; 13 are judged suitable for historical follow-up (Table 6) and 30 for prospective studies. A retrospective period of 20 years was considered to be a minimum for historical follow-up in view of the long delay for other fibre-induced cancers (e.g., mesothelioma induced by asbestos) between first occupational exposure and the associated neoplasm.

Table 6. Man-made mineral fibre factories[a] judged to be suitable for historical investigation

Fibre type [a]	Present work force	Year records commenced
GW	150	1935
RW	70	1950
Cont.	1100	1950
Cont.	150	1956
RW	130	1942
RW	400	1940
GW	300	1933
RW	450	1943
RW	80	1940
RW	70	1955
GW	1100	1955
GW	140	1941
GW	120	1952

[a] RW, rock wool; GW, glass wool; Cont., continuous glass fibre

Although it would be of great interest to follow workers exposed to different hazards, this study is currently restricted to work forces exposed solely to one type of fibre. Hence previous exposure to asbestos within a factory has meant its exclusion from the study.

There has been close collaboration with the other agencies involved in the study. Following the selection of suitable factories from an epidemiological point of view, the Institute of Occupational Medicine carried out exposure estimations, generally selecting plants on the basis of Agency recommendations.

An industry-sponsored workshop on man-made mineral fibres was held under the auspices of the WHO at the WHO Regional Office for Europe, Copenhagen, 26–27 October 1976. The epidemiological research was discussed and the status of the Agency field studies described.

Visits have been made to cancer institutes and cancer registries in Spain, Switzerland, the Federal Republic of Germany and the Nordic countries, with the object of following up occupational cohorts defined in the man-made mineral fibre industry.

(ii) *User industries*

Higher concentrations of fibres are likely to be associated with some uses of man-made mineral fibre products, e.g., in building (insulation of walls, roofs, etc.), construction and demolition, than occur in the industry itself. An investigation of workers in these occupations would therefore also be desirable, in order to detect possible cancer risks arising from the use of man-made mineral fibres. Several factors, however, could complicate this type of investigation. There may be exposure to other carcinogens, such as asbestos, and the exposure may be discontinuous. Work is organized in small units with high mobility, and follow-up of the exposed people would consequently be difficult. An exploration of such a study is, nevertheless, in progress in Sweden, where relatively wide use of man-made mineral fibres has been made in the building industry for at least two decades, to assess whether an epidemiological investigation of this type would produce meaningful results.

(*b*) *Detection of occupational risk by case-control studies* (Dr R. Saracci, Dr J. Siemiatycki)

Cohort studies, historical or prospective, are generally undertaken when a substance or exposure is suspected to carry risk. A large number of exposed persons are followed to determine which of them develop cancer. In the case-control approach, persons with cancer are asked about previous occupational exposures. Preliminary studies on the feasibility of conducting such investigations in the Lyon area are under way.

(*c*) IARC Monographs on the Evaluation of Carcinogenic Risk of Chemicals to Man (Dr R. Saracci)

Dr Saracci continues to provide epidemiological advice on the preparation of these monographs (see page 89). The lack of epidemiological information for many of the compounds considered indicates the need for a systematic approach to this problem (see section 5).

4. BIOSTATISTICS (Dr N. E. Day)

During the course of the year, Dr L. Muenz returned to his post at the National Cancer Institute, Bethesda, Md., USA, and Dr S. Mandel was appointed as a short-term consultant for six months as from March 1977.

Following the six-month consultantship of Dr E. Schifflers (University of Louvain, Belgium) in 1976, the computing capacity at the Agency was upgraded to make use of new facilities available at the International Computing Centre (ICC) in Geneva and to broaden the service that can be used by the Agency. In particular, conversational connexion with the ICC computer in Geneva has been installed, allowing on-line text editing, data base management and more efficient programme development. Apart from providing a general statistical and computing service to the Agency, the Biostatistics section has been occupied with the projects described below.

4.1 *Collaboration with investigators in Iceland*

(*a*) *Breast cancer—study of familial risk*

The study of familial risk in a well-defined total population, partly financed by the National Cancer Institute, Bethesda, Md., USA, and undertaken with the Icelandic Cancer Registry (Dr H. Tulinius, RA/73/004), is almost complete [1]. Final verification of the matchings between the family files and the breast cancer files is under way. Expanded analysis of the role of reproductive factors in determining breast cancer risk in Iceland is near completion. Two papers are in preparation on the familial risk, one devoted to the Icelandic data and the other treating problems of statistical methodology (see section 4.4 (*a*)).

(*b*) *The Icelandic cervical cancer detection clinic* (in collaboration with Dr G. Johannesson)

Parts of the demographic and reproductive data collected by the clinic were used as control material for the work described in the previous paragraph. The efficacy of the clinic in detecting cancer and reducing mortality has also been examined. Following repeated screening of more than 85% of the female population under 60 years of age, mortality from cervical cancer has decreased more than two-fold over a 10-year period. Prior to the introduction of screening there had been an increasing mortality from the disease. These findings are being examined further.

The feasibility of using the detection clinic as the basis for prospective studies for a variety of risk factors is under consideration.

4.2 *Immunogenetics*

(*a*) *Nasopharyngeal carcinoma* [2]

In collaboration with Dr S. H. Chan and Dr M. J. Simons (University of Singapore), a paper was prepared and presented at the International Symposium on the Etiology and Control of Nasopharyngeal Carcinoma, Kyoto, Japan, 4–6 April 1977 [3].

(*b*) *Methodology*

A presentation was made at the First International Conference on HLA and Disease in Paris in June 1976, and a paper was published in *Tissue Antigens* [4]. A general approach to the problem of estimating genotype-environmental interactions is being developed.

4.3 *Oesophageal cancer in Iran—case-control study*

A sample of previously-interviewed oesophageal cancer cases and the corresponding controls were followed up to ascertain the validity of the diagnosis and to obtain information on the reproducibility of answers received to questionnaires (see page 137).

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[1] International Agency for Research on Cancer (1976) *Annual Report, 1976*, Lyon, p. 45.

[2] International Agency for Research on Cancer (1976) *Annual Report, 1976*, Lyon, p. 44.

[3] Simons, M. J., Wee, G. B., Shanmugaratnam, K., Goh, E. H., Ho, J. H. C., Chan, J. C. W., Darmalingam, S., Prasad, U., Bétuel, H., Day, N. E. & de-Thé, G. B. (1977) In: *Proceedings of the International Symposium on Etiology and Control of Nasopharyngeal Carcinoma (NPC), Kyoto, Japan, April 1977*, Lyon, International Agency for Research on Cancer (in press).

[4] Day, N. E. & Simons, M. J. (1976) *Tissue Antigens*, **8**, 109–119.

4.4 *Analysis of clustering phenomena*

(*a*) *A general method*

A model for clustering, such as occurs in families or households, is being developed, which is more general than those so far proposed and can take account explicitly of the unit ascertainment to give an estimate of the strength of clustering. The method is being applied to the Icelandic familial breast cancer data (see 4.1 (*a*) above) and may be used on data arising from studies of urinary tract tumours associated with endemic Balkan nephropathy.

(*b*) *Time-space clustering of Burkitt's lymphoma* (see page 69)

Fifteen years' data on Burkitt's lymphoma in the West Nile district of Uganda are being analysed with Dr P. Smith, Oxford, UK. The time-space clustering described for the data collected during 1961–65 is clearly not a constant phenomenon: the children involved in the clusters were found to be the older cases. The implication for the latent period of the observed clustering is being studied (see below).

(*c*) *Seasonal variations in Burkitt's lymphoma occurrence*

Seasonal variations in the occurrence of Burkitt's lymphoma could only arise if the variation in the latent period between a final triggering event and clinical diagnosis were not too large. Upper limits on the variability of the latent period have been obtained (Mr B. Lachet, University of Grenoble, France). It appears likely that very few of the tumours can have a latent period of more than two years. An attempt is being made to use the data from the time-space clustering to strengthen this conclusion.

4.5 *Longitudinal serological data*

Methods for analysing data of this type are being developed in order to study the joint evolution of anti-Epstein–Barr virus antibody titres and parameters of malaria infection (see page 78), as well as for the more direct purposes of estimating infection rates and relating them to demographic and environmental variables.

4.6 *Workshop on the analysis of case-control studies in epidemiology*

Methods that have been developed in the last fifteen years for the analysis of case-control studies are scattered throughout the literature and lack a unifying conceptual framework. The time is now ripe to bring together the relevant statistical methods for analyses of this type. A workshop will be held in December 1977, the outcome of which will be a monograph containing a presentation of the methodology together with a critical re-analysis of a selection of published studies. The International Union Against Cancer have agreed to meet part of the cost of bringing participants to the workshop under the International Cancer Research Workshops scheme. A preliminary draft of the first part of the monograph is being written in collaboration with Dr N. Breslow, University of Washington, Seattle, Wash., USA.

5. INTERNATIONAL CANCER NETWORK

The need for an international cancer network to collect data on cancer and the environment in a uniform manner in a small number of contrasting environments was described in the 1976 annual report [1]. Funding has now been obtained for a two-year feasibility study. Dr J. Cooper of the National Cancer Institute, Bethesda, Md., USA, on detachment to the Agency, is preparing working documents and proposals.

6. MISCELLANEOUS

In addition to their involvement in the teaching courses organized by the Agency (see page 112), members of the unit have participated in several educational seminars. Teaching courses in elementary epidemiological and statistical techniques are held for unit secretarial and technical staff, and work proposed or in progress is discussed at weekly staff meetings.

Several members of the staff are on the editorial boards of international cancer journals and are frequently asked to review scientific papers.

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[1] International Agency for Research on Cancer (1976) *Annual Report, 1976*, Lyon, pp. 24–26.

2. UNIT OF ENVIRONMENTAL CARCINOGENS

Dr L. GRICIUTE (Chief)

1. INTRODUCTION

The Unit of Environmental Carcinogens has elaborated methods for the detection and measurement of environmental carcinogens, especially the *N*-nitrosamines. The latest technique for the detection and measurement of *N*-nitrosamines employs the Thermal Energy Analyzer coupled with gas chromatography: by coupling the Analyzer with high-pressure liquid chromatography, both volatile and non-volatile *N*-nitroso compounds can be detected and measured.

The cooperative studies on methods of analysis of volatile *N*-nitrosamines have been extended to include bread and methods of detection of non-volatile *N*-nitrosoamino acids.

Work on a manual of selected methods of analysis for environmental carcinogens is in progress. Volume 1, devoted to volatile *N*-nitrosamines, is in an advanced stage of preparation.

In coordination with the epidemiological studies of oesophageal cancer in Normandy, France, alcoholic drinks collected in that region have been analysed for known carcinogens. These studies are being carried out in collaboration with the François Baclesse Regional Centre, Caen, and include the testing of apple brandy for mutagenicity and in animal experiments.

Studies on the relationship between asbestos and cancer have been continued in collaboration with the Pneumoconiosis Unit of the Medical Research Council (Penarth, UK) and the Institute of Experimental and Clinical Medicine of the Estonian SSR.

2. QUANTITATIVE DATA ON ENVIRONMENTAL CARCINOGENS

2.1 *Analysis of* N-*nitrosamines* (Mr E. A. Walker)

(*a*) *Thermal Energy Analyzer*

During the year, considerable experience has been gained in the use of the Thermal Energy Analyzer (TEA) for the analysis of volatile *N*-nitrosamines. Although the detector has generally proved to be highly selective for *N*-nitrosamines, a large initial peak corresponding to a solvent peak was normally recorded, which was probably a spurious response to untrapped pyrolysis products. To eliminate this, a short, Tenax gas chromatography column (60–80 mesh) was interposed between the pyrolyser and the cold trap to improve

trapping efficiency. This virtually eliminated the 'solvent peak', revealing a new peak that corresponded to a *N*-nitrosamine which emerged before *N*-nitrosodimethylamine (NDMA) in the analysis of some samples of apple brandy. Few *N*-nitrosamines have a shorter retention time than NDMA; if the peak was real, it could have been consistent with an unsaturated alkyl grouping, and one such possibility was *N*-nitrosomethylvinylamine (NMVA). The peak was subsequently found to have the same retention time as NMVA and to exhibit a similar instability to temperature. In mass spectrometry (MS) investigation, the peaks corresponding to NO^+ and to the parent ion of NMVA were detected. Samples have been analysed by Dr D. Fine (Thermo Electron Research Center, Waltham, Mass., USA) using the TEA coupled with high-pressure liquid chromatography (HPLC), and his results confirmed the presence of NMVA. This compound is unstable, decomposes during gas chromatography at temperatures normally employed for analysis of the volatile *N*-nitrosamines and may polymerize. A TEA/HPLC interface, a gift from the Thermo Electron Corporation, is being installed to permit continued investigation.

(*b*) *Mass spectrometry (MS)*

Specific ion monitoring by MS coupled with gas chromatography (GC) has been used for identification of *N*-nitrosamines. Very good correspondence was found between the TEA and MS; however, even with a high resolution (8 000–10 000) it has been found necessary to monitor several ions to obtain reliable confirmation by MS. The general conclusion was that for absolute confirmation of an *N*-nitrosamine, more than one method of analysis, preferably including MS, should be employed.

(*c*) *Collaborative analytical studies*

(i) *Volatile* N*-nitrosamines in processed meat*

Another collaborative study on *N*-nitrosamines in processed meat has been conducted. In this study, a specially prepared, highly spiced meat, typical of many poor quality products and representing a severe test of analytical cleanup procedures, was prepared for the Agency in the laboratories of the British Food Manufacturers Research Association (BFMIRA) (Dr C. L. Walters). Participants were allowed a choice of method and were given a list of possible *N*-nitrosamines for which to analyse, with no indication of those that had actually been added. The survey demonstrated that adequate methods exist for the detection and determination of a range of volatile *N*-nitrosamines in processed meat (Tables 7, 8 and 9). The presence of NDMA and *N*-nitrosopiperidine (NPip) in the blank cans demonstrated that the addition of spices can introduce *N*-nitrosamines into processed meat. It was particularly gratifying to observe that GC screening methods (Table 3), without the use of MS or TEA, although somewhat lengthy, can provide reliable results in laboratories which have experience in the methods. A statistical evaluation of the results, calculated on the basis of added *N*-nitrosamines, showed a general increase in the accuracy of the results obtained in comparison with previous studies, in spite of the added difficulties imposed in this study.

Table 7. Corrected results from the third collaborative study on analysis of *N*-nitrosamines in a heavily spiced luncheon meat

Lab. no.		NDMA: Amount added — 18 μg/kg			Blank	NDEA: Amount added — 4.8 μg/kg			Blank	NDBA: Amount added — 9.1 μg/kg			Blank	NPip: Amount added — 0 μg/kg			Blank	NPy: Amount added — 13.2 μg/kg			Blank
Thermal Energy Analyzer																					
3	GC	19.7	22.0	19.5	3.0	5.0	6.0	4.9	0.9	6.8	8.8	8.4	ND	6.8	7.2	6.6	6.6	11.1	11.9	10.4	1.9
4	GC	18.0	20.0	18.0	3.3	4.9	5.0	5.1	0.3	6.4	6.6	6.2	0.1	5.4	6.1	5.7	4.1	7.8	8.3	7.3	0.8
5	GC	6.9	11.7	14.1	[a]	4.4	3.3	8.1	ND	7.4	7.4	7.0	0.3	7.0	6.1	7.7	7.0	10.5	10.2	11.4	0.7
15	GC	23	24	22	7	6	6	6	ND					7	8	8	6	12	18	13	4
16 [b]	GC	16.7	15.0	16.3	3.0	4.3	3.7	4.7	2.5	8.7	7.3	9.3	ND	4.7	4.7	4.7	5.0	13.3	13.3	14.0	3.0
20	GC	16.9	16.8	22.6	4.5	6.7	6.1	5.7	0.5	9.2	7.6	9.1	ND	9.4	7.6	8.4	8.4	15.0	11.0	14.2	2.0
		20.0				5.4				10.0				7.8				11.0			
21	GC	21.5	18.7	21.6	4.2	5.6	4.3	5.0	1.4	7.4	6.4	8.6	ND	5.7	6.0	4.7	6.5	10.4	9.5	7.4	1.8
		20.5	24.9	22.6	8.7	4.5	5.4	5.1	0.2	7.5	7.6	7.2	ND	5.6	6.7	5.5	6.1	8.1	9.3	10.4	0.7
21	HPLC	19.8	20.2	19.8	3.8	5.2	4.8	4.6	1.3	7.3	7.6	8.1	ND	5.6	6.0	4.5	5.8	9.5	8.3	8.1	1.5
		20.6	22.7	20.3	8.0	4.1	4.9	4.5	ND	6.9	7.2	7.5	ND	4.7	6.8	5.5	6.3	7.6	8.4	9.0	0.7
22	GC	29	24	23	5	9	5	3	ND	8	6	6	ND	11	9	8	9	18	16	14	6
25 [b]	GC	18.3	18.2	19.7	5.1	5.2	5.4	5.5	ND	17.4	17.0	15.4	8.8	10.5	8.7	9.8	10.5	15.8	14.0	15.1	5.3
Chemiluminescence																					
4		19.0	14.0	12.0		4.4	3.6	2.8		7.3	8.4	5.2		5.1	7.1	5.1		5.7	9.2	6.3	
31		76	95	93	89	7	8	11	4	12	16	19	ND	10	13	13	6	16	17	21	ND

NDMA — *N*-nitrosodimethylamine; NDEA — *N*-nitrosodiethylamine; NDBA — *N*-nitrosodi-*n*-butylamine; NPip — *N*-nitrosopiperidine; NPy — *N*-nitrosopyrrolidine; GC — gas chromatography; HPLC — high-pressure liquid chromatography; ND — not detectable.

[a] NDMA could not be quantitated in this case because of complete overlap of the NDMA peak by a large interfering peak of unknown origin.

[b] Mean of several determinations.

Table 8. Corrected results from the third collaborative study on analysis of *N*-nitrosamines in a heavily spiced luncheon meat (mass spectrometry)

Lab. no.	NDMA: Amount added — 18 µg/kg			Blank	NDEA: Amount added — 4.8 µg/kg			Blank	NDBA: Amount added — 9.1 µg/kg			Blank	NPip: Amount added — 0 µg/kg			Blank	NPy: Amount added — 13.2 µg/kg			Blank
1	25	25	22	3	6	8	6	<1	<1	<1	7	<1	6	8	10	5	12	14	19	2
	20				6				7				13				23			
4	23.0	15.0	16.0	3.6	4.5	6.5	4.6	ND	ND	9.1	ND	ND	4.3	6.8	6.6	4.1	6.1	11.0	7.5	ND
5	13.1	13.3	16.7	3.9	4.7	3.8	6.0	0.4	7.0	7.9	6.2	0.3	7.8	6.6	11.0	7.1	8.1	9.3	12.4	0.7
18	17.2	15.4	16.5	2.4	3.7	4.1	4.3	<0.3	4.6	3.4	4.1	ND	5.7	6.2	5.6	5.5	7.5	8.6	8.7	ND
18	15.2	15.6	15.2	2.9	4.7	5.9	5.8	<0.3	4.3	4.3	4.7	ND	6.2	6.1	7.4	8.0	10.5	12.3	10.7	ND
24	21.3	29.6	31.9	8.6	2.1	5.9	7.1	0.6	7.2	10.2	12.0	1.4	10.9	10.6	11.7	14.9	19.8	16.0	21.8	ND

NDMA — *N*-nitrosodimethylamine; NDEA — *N*-nitrosodiethylamine; NDBA — *N*-nitrosodi-*n*-butylamine; NPip — *N*-nitrosopiperidine; NPy — *N*-nitrosopyrrolidine; ND — not detected

Table 9. Corrected results from the third collaborative study on analysis of *N*-nitrosamines in a heavily spiced luncheon meat (gas chromatography)

Lab. no.	NDMA: Amount added — 18 µg/kg			Blank	NDEA: Amount added — 4.8 µg/kg			Blank	NDBA: Amount added — 9.1 µg/kg			Blank	NPip: Amount added — 0 µg/kg			Blank	NPy: Amount added — 13.2 µg/kg			Blank
3 (Hall)	26.5	25.6	28.0	3.6	4.0	3.8	4.0	0.4	5.8	6.9	6.8	ND	7.7	7.6	7.4	8.1	12.0	10.2	9.6	ND
3 (HFB)	10.5	19.9	11.7	4.6	4.4	7.0	5.2	ND	6.2	7.3	5.5	1.9	9.4	8.5	10.4	10.8	6.3	8.3	5.6	2.7
15	19	22	19	5	5	5	6	ND					7	8	8	5	21	22	21	ND
16[a]	15.3	14.0	15.3	3.0	4.5	4.5	4.7	3.0	7.0	5.7	9.7	ND	4.7	5.0	5.0	5.0	12.0	12.0	12.7	2.5
20	19.6	23.6	17.0	5.5	7.0	8.3	4.9	1.1	9.5	8.2	9.2	ND	9.8	10.5	9.4	8.9	14.6	14.3	13.5	1.5
26	17.8	16.3	20.3	3.6	4.9	4.9	3.5	ND	7.9	8.3	8.4	1.4	7.3	7.0	6.0	7.0	10.1	10.5	10.1	1.7
27	ND	2.5	37.0	ND	0.2	ND	10.8	ND	ND	10.8	ND	ND					ND	11.2	9.1	ND
29	16.7	21.0	6.5	ND	27.3	25.2	2.7	ND	det.	det.	det.	det.	34.7	33.0	44.0	ND	ND	ND	ND	ND

NDMA — *N*-nitrosodimethylamine; NDEA — *N*-nitrosodiethylamine; NDBA — *N*-nitrosodi-*n*-butylamine; NPip — *N*-nitrosopiperidine; NPy — *N*-nitrosopyrrolidine; Hall — Hall electrolytic conductor; HFB — heptafluorobutyramide formation; ND — not detected; det. — detected but impossible to quantify.
[a] Mean of several determinations.

(ii) *Volatile* N-*nitrosamines in bread*

Bread samples (*tanoori*) collected during the oesophageal cancer study in Iran have been distributed to five participating laboratories of the European Sub-committee for analysis of volatile *N*-nitrosamines. The results indicated an accuracy comparable with that for determination of *N*-nitrosamines in processed meat.

(iii) *Non-volatile* N-*nitrosamines*

The estimation of standards of three non-volatile *N*-nitrosamines, *N*- nitrososarcosine, *N*-nitrosoproline and *N*-nitrosohydroxyproline, is currently being undertaken in a number of laboratories. The results so far show that satisfactory estimation could be made using a silylation technique; this has been used in the Agency laboratory, and consistent results were obtained using flame ionization detection and TEA detection in both GC and HPLC modes. It is anticipated that this study will eventually be extended to the determination of these compounds in food products.

(*d*) *Studies on* N-*nitrosamine formation* (Mr E. A. Walker, Mrs B. Pignatelli, Dr M. Castegnaro)

(i) *General kinetic studies*

The kinetics of *N*-nitrosamine formation have so far been measured at 20°C to avoid problems caused by decomposition of nitrous acid at higher temperatures; however, in order to simulate *in vivo* formation, such studies should be carried out at 37°C. At 20°C, the formation reaction was arrested by addition of a large excess of sodium hydroxide solution to give a strongly alkaline medium, before extraction of the *N*-nitrosamines with methylene chloride. At 37°C, however, this method gave variable results, which appear to be due to continued formation of *N*-nitrosamines in the organic solvent. It is believed that decomposition products of nitrous acid nitrosate the co-extracted amine. Sulphamic acid was therefore added as an alternative method of destroying nitrite, and when it was added in a 100-fold excess rapid destruction of the nitrite was achieved. Reproducible kinetics were then obtained.

A study on the effect of ethanol on *N*-nitrosamine formation showed no increase either at 20°C or 37°C with the sulphamic acid method. Earlier studies using excess caustic soda to arrest reaction showed an apparent increase, which may also be explained by formation in the extracting solvent. These effects will be the subject of further investigation.

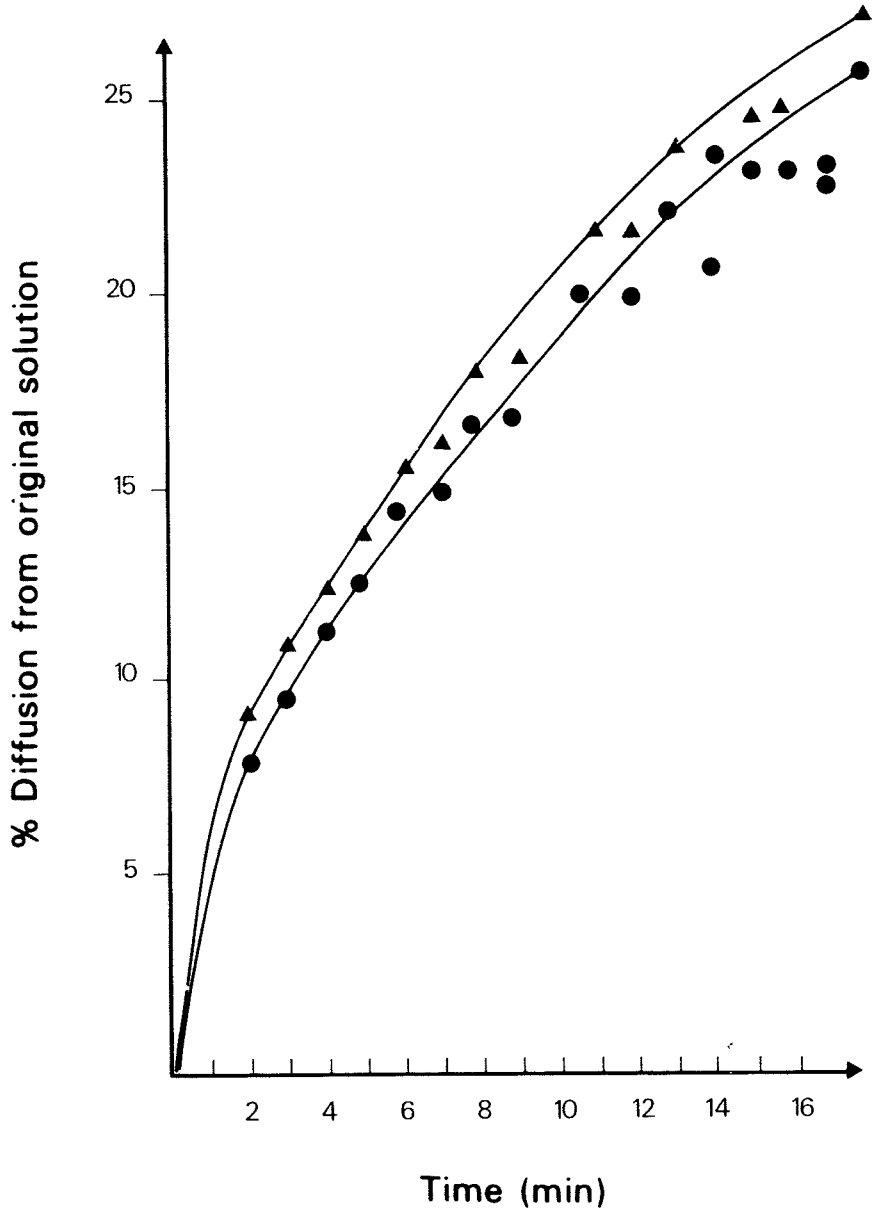
(ii) *Effect of phenols and* p-*nitrosophenol*

Model studies of the effect of phenols on *N*-nitrosamine formation have been continued. It was shown that a concentration of 10^{-2} mol l^{-1} of phenol doubled the amount of *N*-nitrosodiethylamine (NDEA) formed at 20°C; a concentration of only 4×10^{-4} mol l^{-1} of *p*-nitrosophenol was sufficient to effect the same increase in NDEA formation. Similar results have been obtained in a collaborating laboratory (Dr M. Knowles, Ministry of Food, Agriculture and Fisheries, UK) on the formation of *N*-nitrosopyrrolidine (NPy) in the presence of *p*-nitroso-*o*-cresol.

(*e*) *Safety in handling* N-*nitrosamines and their solutions*

There is widespread use of rubber gloves as a protective measure in handling chemical carcinogens in the laboratory, but its validity does not seem to have been checked. Since *N*-nitrosamines and solutions of *N*-nitrosamines in organic solvents, particularly methylene chloride, are frequently handled in the analytical laboratory, the efficiency of gloves as a protection has been investigated.

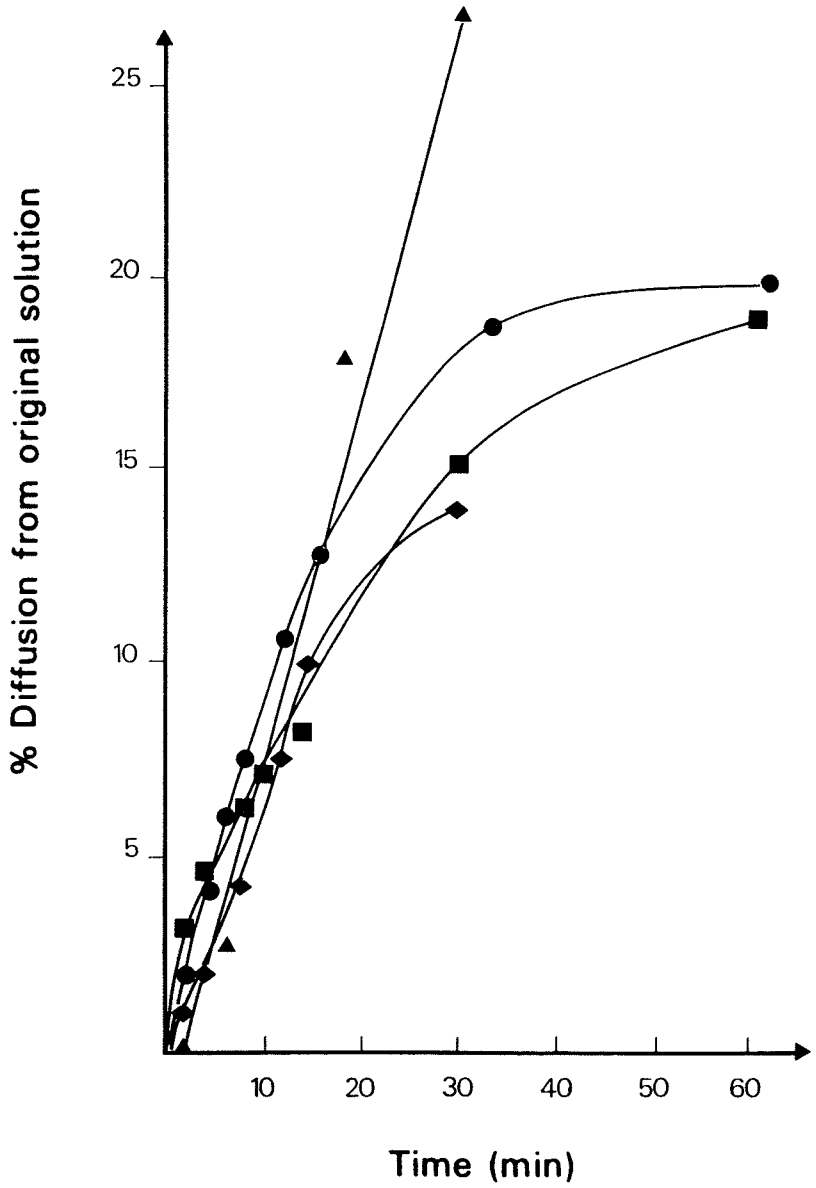
Fig. 6 Diffusion of *N*-nitrosodimethylamine (▲) and *N*-nitrosodiethylamine (●) in methylene chloride to the interior of protective gloves containing methylene chloride.



A simple test was devised, in which a glove filled with methylene chloride or a physiological saline solution was suspended in a beaker containing a solution of *N*-nitrosamine in methylene chloride. The rate of diffusion through the rubber was determined by analysing the solvent contained in the glove at intervals. Four gloves were tested: two types of thin

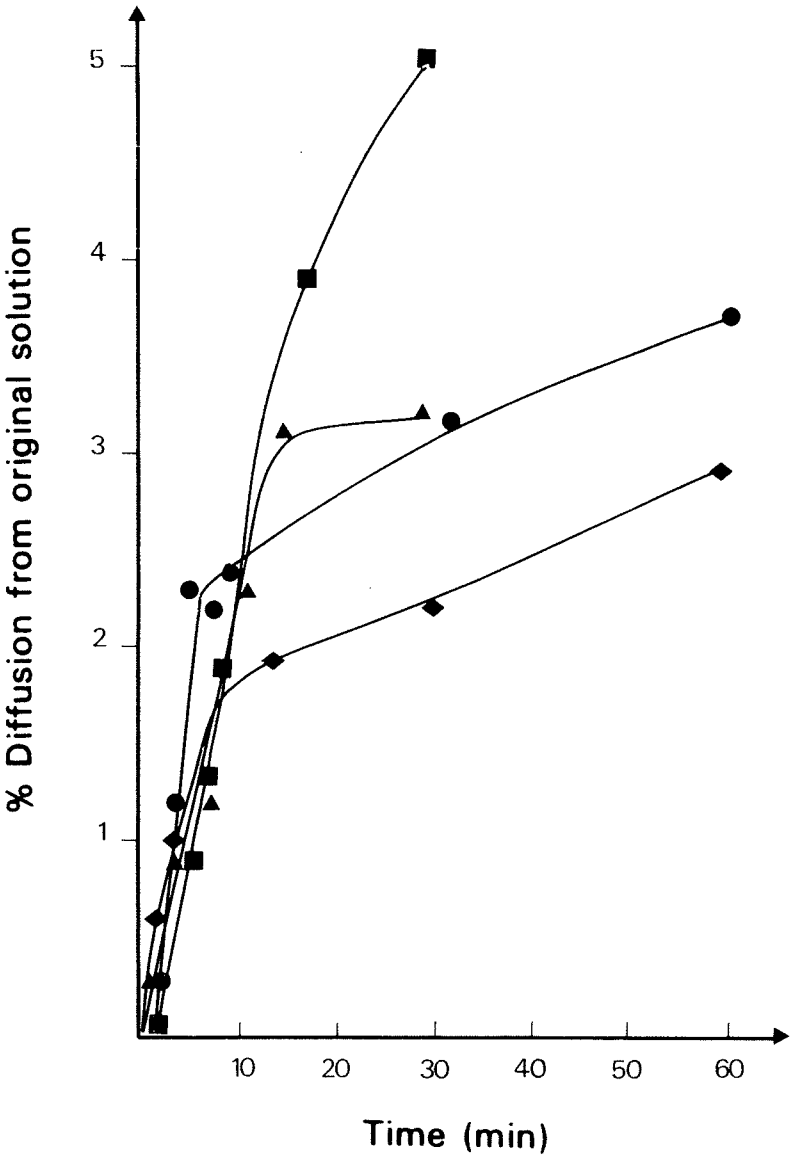
surgical gloves and two types commonly used in the household and industry; several *N*-nitrosamines were used in the test. All of the gloves were permeable to *N*-nitrosamines (Figs 6, 7 and 8). Permeability was reduced when the transfer was from organic solvent to aqueous solution but was still significant.

Fig. 7 Diffusion of *N*-nitrosodimethylamine in methylene chloride to the interior of protective gloves containing physiological saline: ■ — Latex glove A; ◆ — Latex glove A coated with hairspray; ● — Latex glove B; ▲ — Latex glove C (thick).



The thin surgical gloves were also tested one inside the other but separated by a thin layer of saline, adsorbing talc or a barrier cream. The results showed that when two gloves are worn and the inner glove is coated in any one of the three ways, penetration is very considerably reduced. A coating of hairspray applied to the surface of gloves had no effect.

Fig. 8 Diffusion of *N*-nitrosodiethylamine in methylene chloride to the interior of protective gloves containing physiological saline: ■ — Latex glove A; ◆ — Latex glove A coated with hairspray; ● — Latex glove B; ▲ — Latex glove C (thick).



When 10 mg NDMA were applied to the surface of a glove filled with methylene chloride, it was rapidly detected in the interior. This simulated the result of opening of bottles whose caps were contaminated.

The rate of penetration of different *N*-nitrosamines varied considerably (Figs 7 and 8). Of the compounds tested, NDMA and NPy penetrated the most rapidly, and the rate of penetration decreased in the homologous series of *N*-nitrosamines, e.g., dimethyl > diethyl > dipropyl > dibutyl. No penetration at all was detected with *N*-nitrosodi-*n*-butylamine.

The experiments show that rubber gloves do not provide complete protection, that penetration by *N*-nitrosamines is rapid and, therefore, that gloves should be discarded immediately after handling *N*-nitrosamines or their solutions.

2.2 *Manual of selected methods of analysis of environmental carcinogens* (Dr L. Griciute)

The second meeting of the editorial board for the manual of selected methods of analysis for environmental carcinogens (Chairman: Professor H. Egan, Laboratory of the Government Chemist, London) was held in Lyon on 19–20 January 1977 [1].

The material for Volume 1 of the manual, dealing with the analysis of volatile *N*-nitrosamines, was presented by the review board (Chairman: Professor R. Preussmann, German Cancer Research Centre, Heidelberg, Federal Republic of Germany).

Specialists on the analysis of vinyl chloride and related compounds (Mr W. Thain, Mr D. Squirrel, Dr W. R. Eckert) reviewed the analytical methods for these substances, and it was decided that the next volume of the manual should be devoted to their analysis. The other priorities remained unchanged.

2.3 *Carcinogens in alcoholic beverages* (Mr E. A. Walker, Dr M. Castegnaro, Mr G. Toussaint, in collaboration with Professor J. Y. Le Talaer, François Baclesse Regional Centre, Caen, France; Dr J. F. Drilleau, Cider Industry Research Centre, Le Rheu, France; Mrs J. Guérain, National Union of Alcohol Distillers, Paris)

(*a*) N-*Nitrosamines in different beverages*

Examination of over 100 different apple brandies from Normandy, France, with the TEA detector has shown the presence of trace amounts of *N*-nitrosamines in the majority of samples. Average levels found were: NDMA–0.5 µg/l, NDEA–0.1 µg/l, and *N*-nitrosodipropylamine (NDPA)–0.1 µg/l. Similar levels of NDMA and NDEA have been detected in the 10 cider samples so far examined, but, so far, no NDPA has been found. Analyses of a number of apple brandies in which NDMA had previously been detected by a GC method [2] have been confirmed by TEA analysis. However, the greater sensitivity and selectivity of the TEA has allowed effective analysis down to the 0.01 µg/l level and revealed the presence of other *N*-nitrosamines. A number of whiskies, rums, armagnacs and cognacs have also been examined in order to obtain a comparative background on *N*-nitrosamine levels in alcoholic drinks in general. Level of NDMA, NDEA and NDPA similar to those in apple brandy were detected. Analysis of beers and wines from different regions has been started, and NDMA was found at an average level of about 2 µg/l in 27 samples of beer. Out of 21 samples of different wines, only one was positive, containing NDMA at a level of 0.05 µg/l (see Table 10).

N-Nitrosomethylvinylamine (NMVA), NDEA and NDPA have all been shown experimentally to be oesophageal carcinogens [3, 4], but it remains to be proved whether the small traces found could be etiological factors in the occurrence of oesophageal cancer in northern France. Other alternatives should be considered: firstly, the traces found could be an indication that nitrosatable material is present and that larger amounts of these *N*-nitrosamines are formed *in vivo* after consumption of the alcoholic beverage; and secondly, undetected carcinogens may be present. Studies are being pursued along both lines.

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[1] IARC Internal Technical Report No. 77/001.
[2] Castegnaro, M., Pignatelli, B. & Walker, E. A. (1974) *Analyst*, **99**, 156–162.
[3] Druckrey, H., Preussmann, R., Ivankovic, S. & Schmähl, D. (1967) *Z. Krebsforsch.*, **69**, 103.
[4] Magee, P. N., Montesano, R. & Preussmann, R. (1976) *ACS Monograph Series No. 173*, 491–625.

Table 10. Levels of volatile nitrosamines in various alcoholic beverages

Sample type	Nitrosamines found	Level		
		Min.	Max.	Average
Apple brandy (122 samples)	NDMA	ND	10	0.5
	NDEA	ND	1.30	0.1
	NDPA	ND	3.60	0.1
Cider (10 samples)	NDMA	ND	1.60	0.3
	NDEA	ND	1.10	0.1
Cognac (8 samples)	NDMA	ND	0.3	0.1
	NDEA	ND	0.2	0.05
	NDPA	ND	0.2	0.02
Armagnac (4 samples)	NDMA	0.1	0.9	0.4
	NDEA	0.2	0.3	0.2
Rum (8 samples)	NDMA	ND	0.3	0.1
	NDEA	ND	0.2	0.1
	NDPA	ND	0.2	0.05
Whisky (7 samples)	NDMA	ND	0.5	0.1
	NDEA	ND	0.2	0.02
	NDPA	ND	0.2	0.02
Beer (27 samples)	NDMA	ND	7	2
Wine (21 samples)	NDMA	ND	0.05	—

NDMA — *N*-nitrosodimethylamine; NDEA — *N*-nitrosodiethylamine; NDPA — *N*-nitrosodipropylamine; ND — not detected

With regard to the possibility of *in vivo* formation, the absence of any promoting effect by ethanol and the marked promotion by phenol constituents has already been discussed (see sections 2.1 (*d*) (i) and 2.1 (*d*) (ii)). Furthermore, it has been found that addition of nitrite to apple brandy which has been acidified (pH 3.5) can lead to a 50-fold increase in the concentration of NDMA and NDEA. Several unknown peaks were also shown by TEA detection in the 1 μg/kg range, but the concentration was insufficient for MS identification.

(*b*) *Studies on the biological activity of apple brandy*

A number of samples of apple brandy have been tested for mutagenicity with and without metabolic activation. Sixty-seven farm samples, 139 commercial samples and 35 samples made by experimental distillation have been tested. None showed mutagenic activity.

A random selection of samples were submitted to a simple distillation below 95°C, and the distillate and residue were tested separately. A weak but positive response was obtained for some samples from both the alcoholic distillate and the residual aqueous fraction.

Nine out of 25 farm samples showed mutagenicity without metabolic activation, and five out of the nine also gave a positive response with activation. Two out of 18 commercial samples gave a positive response without metabolic activation, and none with activation. The aqueous residues were freeze-dried, and the dry residues so obtained were

also tested; however, only one sample of farm brandy showed any mutagenic activity. *N*-Nitrosamine determination on both distillate and aqueous residue showed approximately equal concentrations. *N*-Nitrosamine was not concentrated in either fraction and showed no correlation with mutagenic activity.

In an attempt to explain these results, three different farm samples were each spiked at the 5, 20 and 50 µg/kg levels with NDMA and NDEA, treated as already described, and the distillates and aqueous residues examined for mutagenicity. Of the alcoholic distillates obtained from samples spiked at the 5 µg/kg level, two out of three showed positive mutagenicity only with metabolic activation; at the 20 µg/kg level, all three were positive only with activation; and at the 50 µg/kg level, two out of three were positive only with activation. The residue from one out of three samples spiked at the 50 µg/kg level was positive with activation. The *N*-nitrosamines were found to be approximately equally distributed between the distillate and aqueous residue.

No definite conclusion could be drawn from these results other than that evidence of mutagenicity was more frequent in farm samples than in commercial samples. The observed mutagenic effect without activation in the unspiked samples suggested that this was not due to *N*-nitrosamines, which show mutagenicity only after metabolic activation, nor to polycyclic aromatic hydrocarbons (PAH), which all remain in the dry residue. However, the results from the spiked samples are confusing, since mutagenicity should have been expected both with and without activation; spiking induced mutagenicity with activation, but seemed to eliminate the mutagenicity without activation. It is concluded from these results that a more efficient separation is required to concentrate any mutagens into smaller fractions, and it is planned to use a spinning band distillation column for this purpose.

(*c*) *Analysis of experimental distillation products*

Experimental distillation of cider, designed to compare the products from farm and commercial fermentation and distillation, has been carried out by Dr Drilleau. The products of fermentation were separated by centrifuging, and both the supernatant liquor and the wet residue (lees) were distilled. The fractions were then analysed in the Agency laboratory for *N*-nitrosamines and hydrocarbons and tested for mutagenicity (Dr Le Talaer). The analytical data suggest that *N*-nitrosamines found in apple brandy originate in the cider. So far, there is no evidence that PAH are produced during distillation. There is still no explanation for the presence of PAH in farm apple brandy. No evidence of mutagenicity was found in any fraction of the distillate.

(*d*) *Patulin in cider*

Although patulin was reported to have been detected in ciders [1, 2], both the methods of thin-layer chromatography and HPLC used for these studies have been suspected. More recent investigation was made using a semi-preparative HPLC method of clean-up developed for determination of aflatoxins in crude olive oil [3]. It was then found that although patulin could be recovered quantitatively from apple juice, it was not possible to do so from cider, in which patulin was apparently unstable.

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[1] Drilleau, J. F. & Bohuon, J. (1973) *C.R. Hebd. Séances Acad. Agric. Fr.*, **59**, 1031–1037.
[2] International Agency for Research on Cancer (1975) *Annual Report, 1975*, Lyon, p. 51.
[3] International Agency for Research on Cancer (1976) *Annual Report, 1976*, Lyon, p. 54.

3. *N*-NITROSAMINES IN OTHER ENVIRONMENTAL SAMPLES

3.1 *Nitrite in saliva—possible relevance to* in vivo *formation of* N-*nitroso compounds*

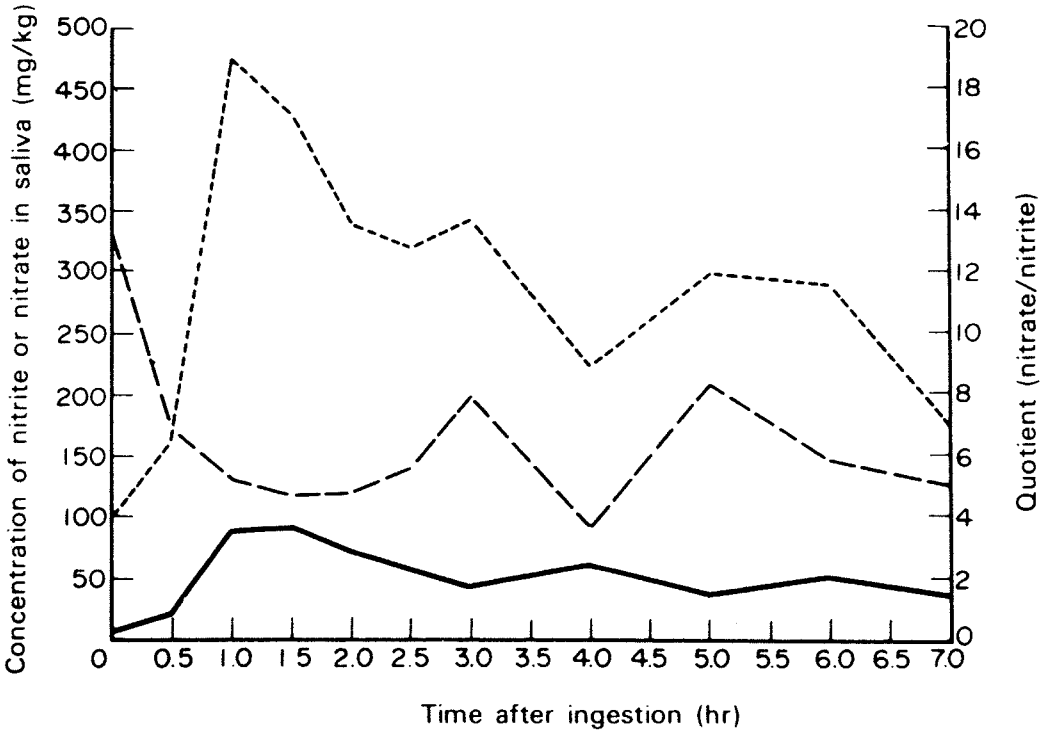
(Mr E. A. Walker, Dr M. Castegnaro, in collaboration with Dr R. Preussmann and Dr G. Eisenbrand, Institute of Toxicology and Chemotherapy, Heidelberg, Federal Republic of Germany)

(*a*) *Salivary concentrations of nitrite and nitrate after ingestion of nitrate*

The influence of dietary nitrate on the nitrite content of human saliva was investigated in 11 volunteers in 30 experiments. Concentrations of nitrate and nitrite in saliva were measured after ingestion of nitrate-containing vegetable juices or spinach.

Average concentrations in saliva before the start of the experiments were 74 ± 50 mg/l of nitrate and 9 ± 5 mg/l of nitrite. These concentrations began to increase 30 min after ingestion of nitrate, the maximum concentrations of nitrate being reached after 1 hr and of nitrite after 1.2 hr (Fig. 9).

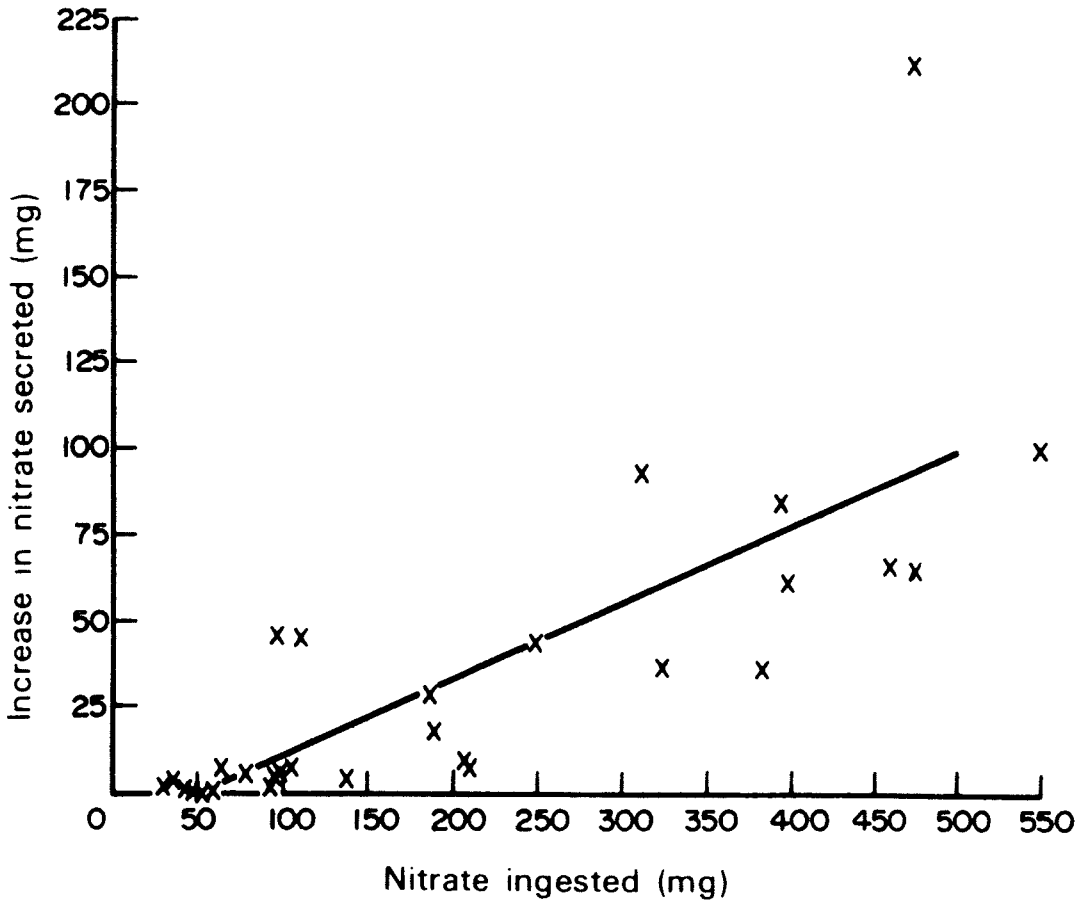
Fig. 9 Concentrations (mg/l) of nitrate (------) and nitrite (———) in saliva after ingestion of 450 mg nitrate-containing red beetroot juice (250 ml). Quotient of nitrate/nitrite (– – – –).



For comparison of the data, the absolute amounts of nitrate and nitrite were calculated on the assumption of a constant salivary flow throughout the total time period. The relationship between secreted nitrate and nitrite and ingested nitrate was demonstrated (Figs 10 and 11). There was also a direct correlation between the amount of secreted nitrate and the nitrite produced in saliva (Fig. 12).

The average increase in concentration was estimated to be 1.1 mg/l of nitrate and 0.2 mg/l of nitrite per mg of nitrate ingested. Ingestion of about 500 mg nitrate (in 200 g

Fig. 10 Relationship between amount of nitrate ingested and nitrite secreted (n = 31).



of radishes, for example) produced an average salivary nitrite concentration of about 100 mg/l; thus, assuming a salivary flow of 50 ml/hr, about 20 mg nitrite could enter the stomach within 5 hr.

(*b*) *The gastro-oral circulation of nitrate*

After ingestion, the absorbed nitrate caused an increased nitrate concentration in the blood, dependent on the rate of absorption from the gastrointestinal tract. The secretion of nitrate through the salivary glands obviously depended on the blood level of nitrate. In the oral cavity, about 20% of the nitrate was reduced to nitrite; both nitrate and nitrite were swallowed and reached the stomach, where nitrite quickly disappeared by reaction with the stomach contents, by absorption and by other processes (Fig. 13). Nitrate, however, could be reabsorbed and recirculated, and this is a very likely explanation for the observed second and third maxima (Fig. 9).

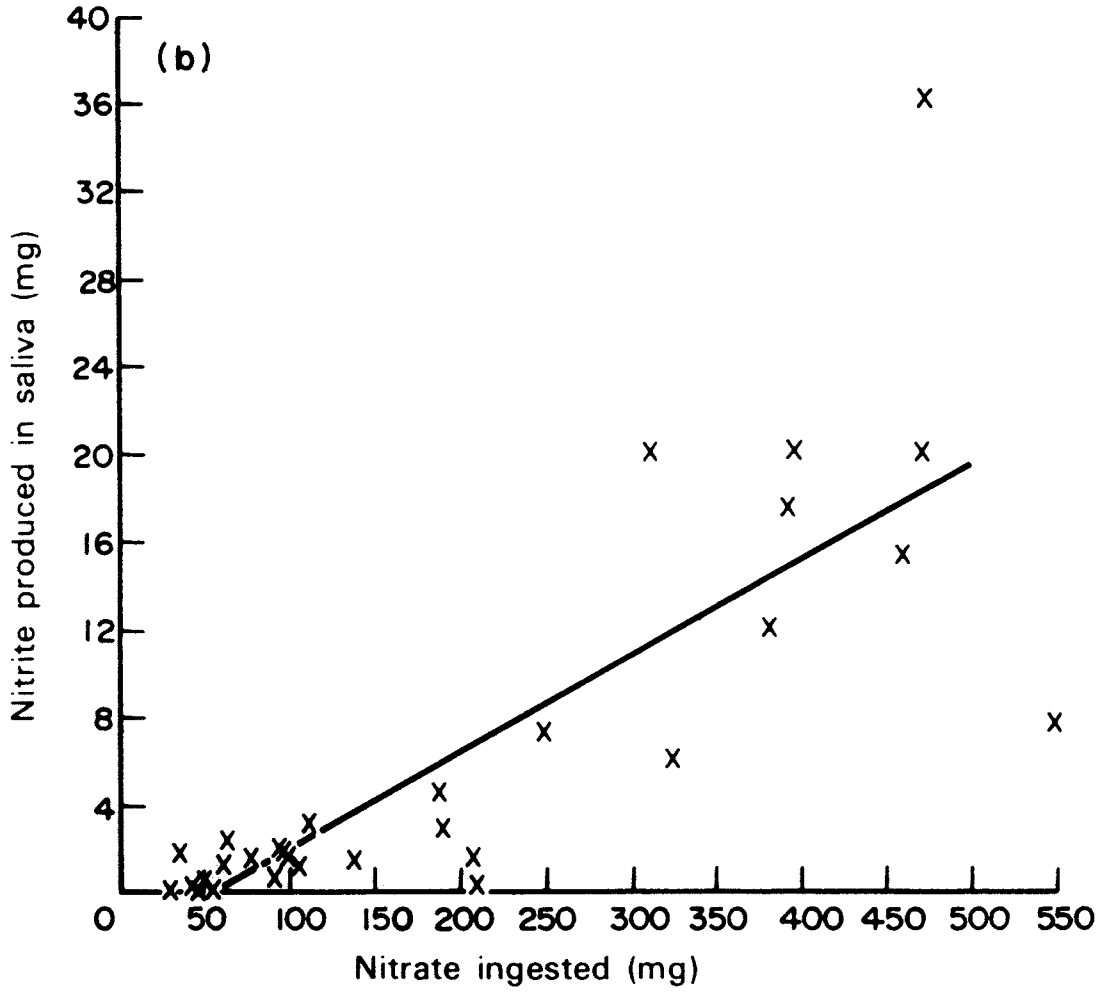
(*c*) *Possible influence of salivary nitrite on the endogenous formation of* N-*nitroso compounds*

The results show that the nitrate content of the diet was the major factor governing the nitrite content of saliva. When vegetable food with a high nitrate content was consumed, the nitrite concentration in saliva reached much higher levels than were previously thought possible.

The stability of nitrate and nitrite in saliva has been tested. Without addition of preservatives the nitrate was reduced very quickly to nitrite and ammonia, but this could be prevented by storage at −18°C. A less satisfactory method of stabilization was the addition of 0.1 normal caustic soda (1: 1, v/v), which permitted the storage of the samples for several days at room temperature or for several weeks at 4°C.

The individual factors that influence the ability to form nitrite are being studied. Saliva samples are incubated with known amounts of nitrate, and the yields of nitrite formed in saliva give an indication of inter-individual differences in bacterial flora of the oral cavity.

Fig. 11 Relationship between amount of nitrate ingested and nitrite produced in saliva (n = 31).



Collaborative pilot studies, in which the Agency (Dr M. Castegnaro) and the Institute of Toxicology and Chemotherapy, Heidelberg, Federal Republic of Germany (Dr G. Eisenbrand), are involved, have been carried out on techniques of analysis and sample preservation to be used in a projected field study on nitrite in saliva samples to be collected in Iran. The collection, which will be made in areas of both high and low oesophageal cancer incidence, is planned for Autumn 1977.

Fig. 12 Relationship between amount of nitrate secreted and nitrite produced in saliva.

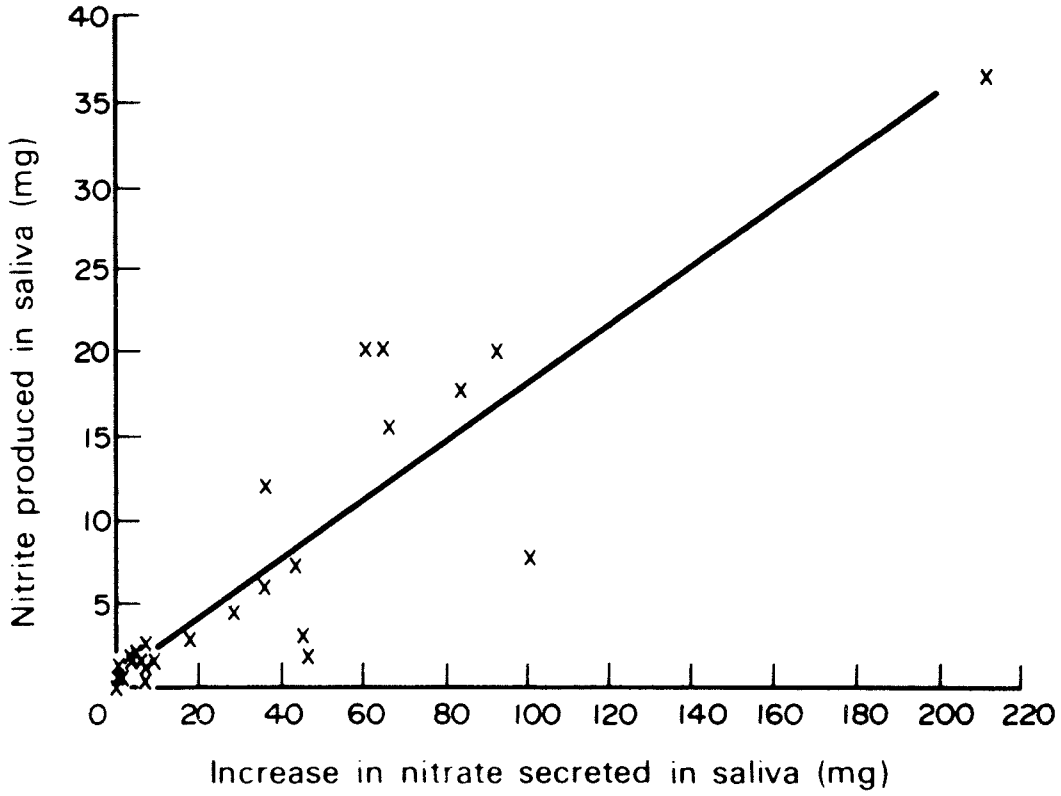
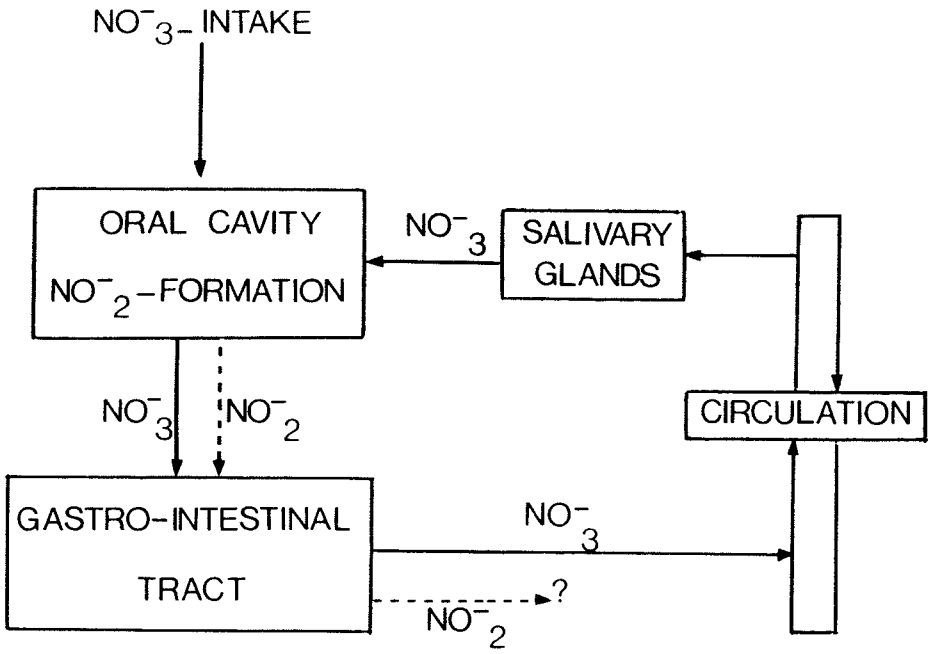


Fig. 13 Flow chart of the gastrointestinal circulation of nitrate.



3.2 N-*Nitrosamines in miscellaneous samples* (Mr E. A. Walker, Dr M. Castegnaro)

(*a*) *Urine*

In the presence of nitrate and nitrate-reducing bacteria, nitrosatable amines may be converted to *N*-nitrosamines. A pilot study is being carried out on urine samples from bilharzial bladder cancer patients in collaboration with Dr R. M. Hicks of the School of Pathology, Middlesex Hospital Medical School, London, and Dr I. El-Sebai, Dr A. A. El-Aaser and Dr M. M. El-Merzabani of the Cancer Institute, Cairo University, Cairo.

Fourteen 24-hour samples were taken, made alkaline, distilled and extracted with methylene chloride. After reducing the volume of methylene chloride, the samples were dispatched to the Agency for *N*-nitrosamine analysis. Although all of the samples were found to contain some volatile *N*-nitrosamines, it is felt that these results cannot be regarded as completely reliable, since *N*-nitrosamines may have been formed in the methylene chloride solution (see page 52). Alternative sampling methods are to be tested, including the addition of a bacteriocide such as merthiolate.

(*b*) *Animal feed*

'Spontaneous' tumours which are observed in control animals in biological experiments may result from contamination of the animal feed with chemical carcinogens. Regular analyses for *N*-nitrosamines and polycyclic aromatic hydrocarbons in the diet used in the Agency and in collaborating laboratories has been planned. Many animal feeds contain fried fish meal in their formulation, and this may be a source of *N*-nitrosamines. Nearly 50 samples have been examined for volatile *N*-nitrosamines (Table 11); considerable variations were found within a single feed batch.

Table 11. Volatile *N*-nitrosamines found in animal feeds

	Min. µg/kg	Max. µg/kg	Average µg/kg
NDMA	ND	42	4.8
NDEA	ND	17	1.6
NDPA	ND	1.5	0.16
NDBA	ND	4.1	0.22
NPip	ND	210	7.61
NPy	ND	4.4	0.3

NDMA — *N*-nitrosodimethylamine; NDEA — *N*-nitrosodiethylamine; NDPA — *N*-nitrosodipropylamine; NDBA — *N*-nitrosodi-*n*-butylamide; NPip — *N*-nitrosopiperidine; NPy — *N*-nitrosopyrrolidine; ND — not detected : limit of detection 0.1 µg/kg

(*c*) *Soya sauce from Singapore*

Nineteen samples representing a wide range of varieties of soya sauce have been analysed for volatile *N*-nitrosamines. Barely detectable traces (< 0.05 µg/l) were found.

(*d*) N-*Nitrosamines in tobacco*

Samples of smoke condensate from French tobaccos (provided by Professor R. Truhaut, Laboratory of Toxicology and Industrial Hygiene, Paris) were found to contain NDMA, NDEA, *N*-nitrosomethylethylamine (NMEA) and NPy.

Analysis has also been carried out for volatile *N*-nitrosamines in pipe scrapings which, it is reported, are chewed by inhabitants of the Transkei (southern Africa), a region of high oesophageal cancer incidence. The samples were taken during epidemiological studies (Dr E. Rose, National Research Institute for Nutritional Diseases, Tygerberg, South Africa, and Dr N. E. Day), and all were found to contain a number of volatile *N*-nitrosamines (Table 12). *N*-Nitrosonornicotine was not included in the analysis since no standard was available.

Table 12. Levels of *N*-nitrosamines found in the scrapings of pipes used for smoking tobacco in the Transkei

Lab. sample no.	NDMA µg/kg	NPy µg/kg
154	240	115
155	45	130
156	100	180
157	340	910
158	95	315
159	140	300
160	100	320
161	270	805
162	285	470
163	270	1600

NDMA — *N*-nitrosodimethylamine; NPy — *N*-nitrosopyrrolidine

4. STUDIES ON THE RELATIONSHIP BETWEEN ASBESTOS AND CANCER

4.1 *Medical Research Council Pneumoconiosis Unit, Llandough Hospital, Penarth, UK* (RA/70/014)

Principal investigator: Dr J. C. Wagner

(*a*) *Physics and chemistry*

(i) *Standardization of methods for monitoring dust counts in the asbestos industry* (Mr W. H. Walton, Institute of Occupational Medicine, Edinburgh, UK)

A considerable amount of information on the monitoring methods used in different countries has been collected, and trials on standardized methods involving laboratories in various countries have been conducted. Government organizations are now defining standardized methods for monitoring dust counts in the asbestos industry. A working party to consider the measurement of physical characteristics of fibres in relation to their biological importance was held at the Agency, 30 June - 1 July 1977.

(ii) *Asbestos in tissue* (Dr F. D. Pooley, Department of Mineral Exploitation, University College, Cardiff, UK)

The material from the mesothelioma and asbestos surveys (see section 4.1 (*c*)) has been examined. The methods for the preparation of tissue, examination of fibrous materials and recording of data have now been standardized. Information so far obtained was presented at the meeting on fibrous dusts at the end of June 1977.

(iii) *UICC standard reference samples* (Dr V. Timbrell, Medical Research Council Pneumoconiosis Unit, Llandough Hospital, Penarth, UK)

Since the preparation of the UICC Standard Reference Samples of asbestos dust in 1966, more than 700 sets have been dispatched to laboratories throughout the world. Detailed reports on their usage and results obtained are currently being accumulated. A review of these studies will be prepared.

(*b*) *Experimental pathology*

(i) *Animal studies*

Experiments have been completed in which the effects of a Grade 7 Canadian chrysotile from a commercial consignment were compared with Italian talc and two previously-studied chrysotile samples — UICC Canadian chrysotile and superfine chrysotile (SFA). The dusts were administered intrapleurally and by inhalation. In the intrapleural inoculation experiment, mesotheliomas occurred with all of the samples of chrysotile but not with the talc. Out of 48 rats injected intrapleurally, five developed mesotheliomas with UICC Canadian chrysotile, 13 with Grade 7 and 18 with the SFA. In the inhalation study, in which there were three durations of exposure, it is of interest that ten carcinomas of the lung developed in the animals exposed to the UICC Canadian chrysotile, three lung carcinomas and one mesothelioma in those exposed to the SFA, and only one in those inhaling the Grade 7 chrysotile.

A method has been devised for the preliminary isolation and concentration of polyvinyl chloride-paste polymer in human and animal tissue (Dr R. Davies). Techniques are now being established for the *in vitro* study of the release of lysosomal enzymes from rat macrophages following their exposure to various mineral dusts.

(ii) *Immunology*

In the study of workers at high risk after exposure to crocidolite abestos in a dockyard in the UK, 140 men have been studied on five occasions, using a range of tests for cellular immunology. Preliminary results indicate that the combination of cigarette smoking and asbestos exposure produces an excess of leucocytes, which include lymphocytes and their sub-sets.

(*c*) *Pathology*

An attempt was made to collect all of the mesotheliomas of the pleura that occurred in the UK during 1976. Lung and tumour tissues from 110 cases have been submitted, and the tumours were studied by the British Mesothelioma Panel under the chairmanship of Dr J. S. P. Jones, University of Nottingham, UK. The lung tissue from these cases and the controls have been prepared for analysis for asbestos and other minerals (Dr F. D. Pooley, Dr V. Timbrell). In 1977, lung tissue from cases in which there is evidence of asbestosis, pleural plaques and carcinoma of the lung, as well as those with mesothelioma, will be studied and compared with those from selected control populations.

A meeting was held in Cardiff, UK, in December 1976 (under the aegis of the European Economic Community) to discuss the problems of analysis of tissue for fibrous dust, the standardization of the diagnosis of asbestosis and the histological criteria for grading the severity of the disease.

Professor I. Baris, from the University of Hacettepe, Ankara, Turkey, visited the MRC Pneumoconiosis Unit to discuss the occurrence of mesotheliomas of the pleura in two villages in Turkey. He brought biopsy material from a number of patients, nine of which showed the histological features of mesothelioma. A mineral analysis of all available pulmonary material is being undertaken (Dr F. D. Pooley).

(*d*) *Epidemiology*

With regard to the detailed study of men exposed to asbestos in dockyards in the UK (Dr P. G. Harries), the main report on the survey of all employees in the four Royal Naval Dockyards and the proportional cancer registration study have been published [1, 2]. The three follow-up studies—of the original group of laggers and sprayers, of the one-in-ten sample first seen in 1966–1968 and of the sample of men aged 50–59 in 1972–1973—are nearing completion.

The mortality and repeat morbidity studies of asbestos cement workers in New Orleans, USA (Professor H. Weill) are continuing.

4.2 *Institute of Experimental and Clinical Medicine, Tallinn, Estonian SSR* (RA/74/011)
Principal investigator: Dr A. Vôsamäe

An investigation was begun on the synergistic effect of phenols extracted from Estonian shale oil on lung carcinogenesis induced by asbestos dust and by benzo(*a*)pyrene in Wistar rats.

Benzo(*a*)pyrene was administered at a dose of 5 mg/0.5 ml of polyglucin; Canadian chrysotile asbestos dust (UICC sample) at a dose of 1 mg/0.5 ml of polyglucin; and phenols from shale oil at 0.5 ml of a 1 % solution in polyglucin. All animals received intratracheal instillations at fortnightly intervals.

Group I (60 animals) received 5 instillations of benzo(*a*)pyrene.
Group II (60 animals) received 6 instillations of Canadian chrysotile asbestos dust.
Group III (60 animals) received 5 instillations of phenols from shale oil.
Group IV (80 animals) received 5 instillations of benzo(*a*)pyrene and phenols.
Group V (80 animals) received 5 instillations of asbestos dust and phenols.
Group VI (controls):

(*a*) 40 animals received 5 instillations of 0.5 ml of polyglucin.

(*b*) 40 animals received no intratracheal treatment.

The intratracheal instillations were started in April 1977. To date, some animals in Groups I and IV have died.

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[1] Harries, P. G., Rossiter, C. E. & Coles, R. M. (1976) *RN Clinical Research Working Party Report 1/76*, Alverstoke, Gosport, Institute of Naval Medicine.
[2] Lumley, K. P. S. (1976) *Brit. J. industr. Med.*, **33**, 108–114.

5. CARCINOGENICITY TESTING OF DAPSONE (Dr L. Griciute)

In the experiments with the anti-leprosy drug dapsone [1], treatment of the animals is finished, but many are still alive.

Several tumours (mammary adenoma, fibrosarcoma of the mediastinum, suprarenal carcinoma and reticulosarcoma) were detected in dead BD IV rats treated with dapsone during the second year of the experiment, but these tumours are common in old, untreated rats. In the control group of rats treated intratracheally with benzo(*a*)pyrene instillations, several bronchogenic carcinomas and mediastinal sarcomas have been detected so far.

Tumours were rare in dapsone-treated C57BL mice; but pulmonary adenomas were common in control mice treated with urethane.

At this stage of the study, the results do not suggest that dapsone is a potential carcinogen, since the rate of survival is high, the tumours were detected mainly during the second year of the experiment, and there was no predominant site for the tumours.

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[1] International Agency for Research on Cancer (1976) *Annual Report, 1976*, Lyon, p. 60.

3. UNIT OF BIOLOGICAL CARCINOGENESIS

Dr G. BLAUDIN DE-THÉ (Chief)

1. INTRODUCTION

The main emphasis of the programme has been studies directed towards the etiology of Burkitt's lymphoma and nasopharyngeal carcinoma.

In the prospective study of Burkitt's lymphoma in the West Nile District of Uganda, 1976 was another year with a low incidence of the disease. A total of 14 Burkitt's lymphoma cases have been detected since 1973 within the cohort of children bled in the initial serum survey. Pairs of pre- and post-Burkitt's lymphoma sera from these 14 cases, together with selected controls from neighbours and families, were tested for Epstein–Barr virus reactivities. The results show that anti-viral capsid antigen titres were significantly higher in the sera of children who subsequently developed Burkitt's lymphoma than in control sera, and there was a very high relative risk associated with anti-viral capsid antigen antibody titres, which are more than two dilutions higher than the mean, standardized for age, sex and locality. This favoured the recently proposed hypothesis [1] that an early Epstein–Barr virus infection, possibly during the perinatal period of life, could represent a critical risk factor for subsequent development of Burkitt's lymphoma.

The other biological factor suspected of playing a role in the etiology of Burkitt's lymphoma, hyperendemic malaria, is being investigated directly in the malaria suppression scheme, which was launched in the Mara Region of Tanzania in March 1977. All children up to 10 years of age (aproximately 70 000) in North Mara will be given chloroquine tablets twice monthly for a period of five years, to see whether the suppression of malaria to hypoendemic levels will eliminate Burkitt's lymphoma in this area.

In the study of nasopharyngeal carcinoma, the main results have been the finding of an Epstein–Barr virus-specific immunoglobulin in the saliva of patients. This immunoglobulin was restricted to patients with nasopharyngeal carcinoma and was not found in the other conditions asscciated with the virus—infectious mononucleosis and Burkitt's lymphoma—nor in other tumours. Furthermore, it was found that these secretory immunoglobulins were formed in the tumour itself and might, therefore, become a diagnostic tool for early or differential detection.

A symposium was organized in Kyoto, Japan, on the etiology and control of nasopharyngeal carcinoma, to review the progress made since the previous symposium in 1964 and to make recommendations for future studies in this field. The results of immunogenetic studies in Singapore (see report of the Singapore Research Centre, page 127) have con-

[1] de-Thé, G. (1977) *Lancet*, *i*, 335–338.

firmed that certain HLA profiles are associated with a risk for development of nasopharyngeal carcinoma, at least in Cantonese Chinese. The consistent presence of markers of Epstein–Barr virus infection in the tumour cells in high-, intermediate- and low-risk areas has confirmed the association between the virus and the tumour, although in nasopharyngeal carcinoma patients the humoral response to the virus varied somewhat between different geographical areas and racial groups. The roles of chronic nasal disease and the consumption of salted fish at an early age were factors which merited further study; however, epidemiological findings failed to indicate any important etiological role for environmental factors in the development of the tumour.

Two newsletters were published in the international reference programme for oncogenic herpesviruses, in which technical sheets were included on the immunological and virological characteristics of the most widely-used lymphoblastoid cell line (Raji).

The third international symposium on oncogenesis and herpesviruses was organized in Boston, Mass., USA, in collaboration with Dr W. Henle (Children's Hospital of Philadelphia, Pa., USA) and Dr F. Rapp (Hershey Medical School, Hershey, Pa., USA).

2. ETIOLOGICAL STUDIES OF BURKITT'S LYMPHOMA (Dr A. Geser, Dr G. de-Thé)

2.1 *Prospective study of Burkitt's lymphoma in the West Nile District, Uganda*

East African Virus Research Institute, Entebbe, Uganda

Principal investigator: Dr P. M. Tukei

Burkitt's Lymphoma Field Station, Arua, Uganda

Principal investigator: Dr D. P. Beri

Kuluva Hospital, Arua, Uganda

Principal investigator: Dr E. H. Williams

(*a*) *Case detection*

Ten new cases of Burkitt's lymphoma were detected in the entire West Nile District in 1976, two of which were from the cohort of children that had been bled between 1972–1974, bringing the total of 'pre-bled' cases to 14. The sera from these two children have been tested for anti-Epstein–Barr virus antibody activities (see section 2.1 (*b*)).

A new analysis of time-space clustering of the Burkitt's lymphoma cases detected in the West Nile District up to the end of 1975 showed that, firstly, the incidence of Burkitt's lymphoma varied over a period of years in different localities and that, secondly, the kind of clustering previously described [1], which involves a much shorter time scale, still occurred (Fig. 14).

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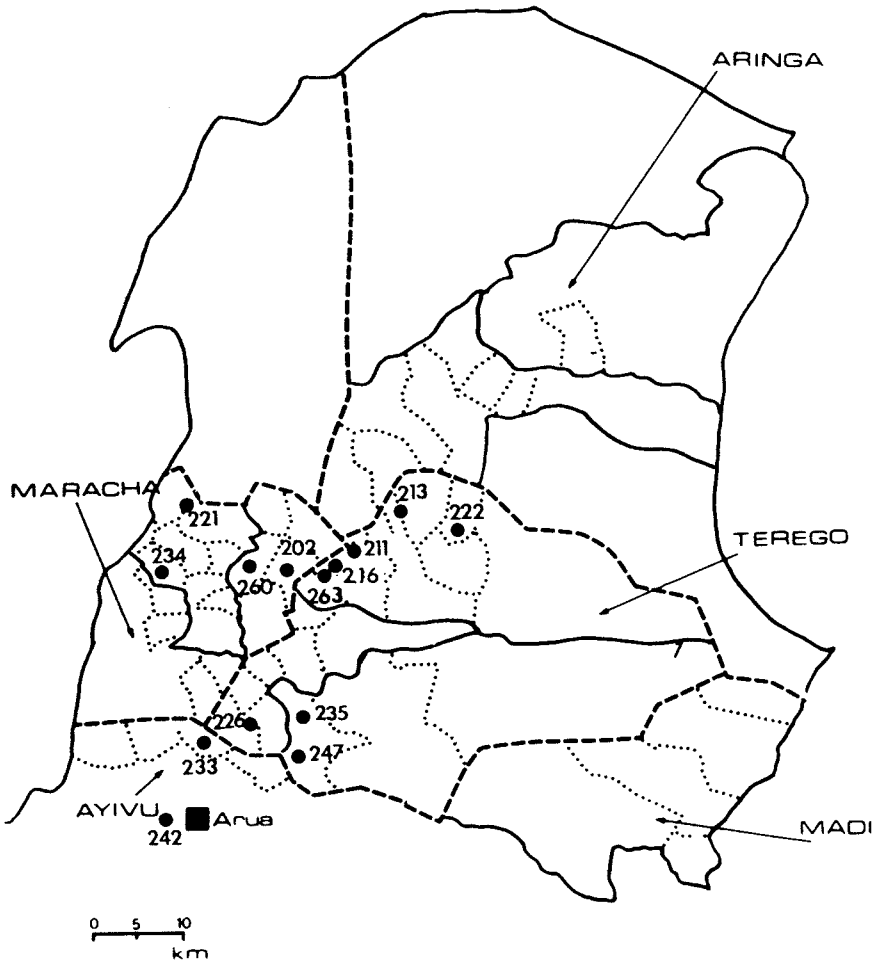
[1] Pike, M. C., Williams, E. H. & Wright, B. (1967) *Brit. med. J., ii*, 395.

Fig. 14 Boundaries of the study area for Burkitt's lymphoma in the West Nile District of Uganda, with localization of the cases previously bled in the main study (●).



An analysis has been started of the data collected in the repeated surveys of sample groups of the cohort from which sera were collected initially. It may show whether variations in intensity of malaria infection (and of Epstein–Barr virus infection) in different areas at different times could explain the observed time-space clustering of Burkitt's lymphoma in the study area.

The field work in the in-depth study ended in June 1977. The numbers of sample groups and of 0–8-year-old children that were examined in each round are given in Table 13. The collected sera have been stored and are being tested for antibodies against Epstein–Barr viral capsid antigen, early antigen and complement-fixing antigen. They are also being tested for antibodies against cytomegalovirus.

(*b*) *Testing of 'pre-Burkitt's lymphoma' sera* (Dr G. de-Thé, Mrs M. F. Lavoué)

Since 1973, 14 cases of Burkitt's lymphoma have been detected in the northern West-Nile district, Uganda, from the cohort of children bled between 1972–1974 (Fig. 14). The sera that were collected from these children before the onset of the disease ('pre-bled') have been tested, together with sera from neighbours and families, as controls, for anti-

Table 13. Numbers of sample groups and of 0-8-year-old children in the in-depth study of sera collected in the West Nile District, Uganda

Examination round no.	No. of groups	No. of 0-8-year-old children
1	22	1518
2	16	1186
3	13	652
4	8	468

bodies against anti-Epstein–Barr viral capsid antigen, early antigen, nuclear antigen and complement-fixing antigen. The tests were carried out in parallel in the Agency and by Dr W. Henle (Children's Hospital of Philadelphia, Pa., USA) (Table 14).

Table 14. Levels of antibodies against Epstein-Barr viral antigens in sera collected from Burkitt's lymphoma patients (BL) before the onset of the disease ('pre') and after the onset of the disease ('post') and from controls (C) and neighbours (N)

Case registry number	Pre or post sera	Time interval (months)	VCA	EA	EA-R	EA-D	EBNA	CF/s	IF/EBNA smears	V-DNA gene equivalents
BL 202	Pre	8	320	10	< 10	< 10	640	40		
	Post		320	1280	160	< 10	1280	160	NSA	NBA
C 202	Pre	14	80/160	< 10	< 10	< 10	640	80		
	Post		80	< 10	< 10	< 10	320	80		
N 202	Pre		320	< 10	< 10	< 10	160	40		
	Pre		160/320	< 10	< 10	< 10	< 5	10		
	Pre		80	< 10	< 10	< 10	640	40		
	Pre		10	< 10	< 10	< 10	20	10/20		
BL 211	Pre	17	80	< 10	< 10	< 10	640	40/80		
	Post		2560	80	—	40	640	40/80	NSA	NBA
C 211	Pre	18	80/160	< 10	< 10	< 10	640	80		
	Post		80	< 10	< 10	< 10	640	80		
N 211	Pre		160	20	< 10	< 10	160	20		
	Pre		80	< 10	< 10	< 10	160	20		
	Pre		320	< 10	< 10	< 10	1280	80		
	Pre		160	20	≦ 10	< 10	40	5		
BL 213	Pre	17	1280	< 10	< 10	< 10	640	80		
	Post		1280	10	10	< 10	2560	80	NSA	NBA
C 213	Pre	17	320	10	< 10	< 10	160/320	40		
	Post		640	10/20	< 10	< 10	640	40		
N 213	Pre		40/80	< 10	< 10	< 10	40	20		
	Pre		640	160	40	< 10	10	5		
	Pre		80	< 10	< 10	< 10	160	40		
	Pre		160	20	< 10	< 10	80	10/20		
BL 216	Pre	18	320	< 10	< 10	< 10	320	20/40		
	Post		160	< 10	10	< 10	1280	80	NSA	65
C 216	Pre	21	80	< 10	< 10	< 10	10	< 10		
	Post		80	< 10	< 10	< 10	640	80		
N 216	Pre		80	< 10	< 10	< 10	2560	320		
	Pre		40	< 10	< 10	< 10	2560	160/320		
	Pre		320	< 10	< 10	< 10	640	160		
	Pre		160	< 10	< 10	< 10	80	40		

Table 14 (continued)

Case registry number	Pre or post sera	Time interval (months)	VCA	EA	EA-R	EA-D	EBNA	CF/s	IF/EBNA smears	V-DNA gene equivalents
BL 221	Pre	7	640	10	20/40	< 10	640	40/80		
	Post		640	80	40	< 10	640	20/40	NSA	54
C 221	Pre	6	40	< 10	< 10	10	640	40		
	Post		40	< 10	< 10	10	160	40		
N 221	Pre		320	< 10	< 10	< 10	640	40/80		
	Pre		160	< 10	< 10	< 10	640	20		
	Pre		320	< 10	< 10	< 10	320	80		
	Pre		80	< 10	< 10	< 10	160	40		
BL 222	Pre	20	640	< 10	< 10	< 10	40	20		
	Post		1280	10	10/20	< 10	2560	80	NSA	116
C 222	Pre	20	80	< 10	< 10	< 10	160	80		
	Post		160	< 10	< 10	< 10	320	80		
N 222	Pre		320	< 10	< 10	< 10	1280	160		
	Pre		80	< 10	< 10	< 10	640	80/160		
	Pre		320	< 10	< 10	< 10	1280	160		
	Pre		320	20	< 10	< 10	320	80		
BL 226	Pre	20	≧2560	20	< 10	20	20	10		
	Post		1280	80/160	160	< 10	< 5	10	Positive	49
C 226	Pre	20	80	< 10	< 10	< 10	640	160		
	Post		80	< 10	< 10	< 10	640	160		
N 226	Pre		640	< 10	< 10	< 10	160	40		
	Pre		640	< 10	< 10	< 10	1280	160		
	Pre		160	< 10	< 10	< 10	320	80		
	Pre		160	< 10	< 10	< 10	320	80		
BL 233	Pre	13	1280	< 10	< 10	< 10	320	160		
	Post		640	< 10	< 10	< 10	160	80	NSA	NBA
C 233	Pre	13	20	< 10	< 10	< 10	40	< 10		
	Post		20	< 10	< 10	< 10	160	40		
N 233	Pre		160	< 10	< 10	< 10	160	20		
	Pre		80	< 10	< 10	< 10	320	160		
	Pre		640	< 10	< 10	< 10	160	40		
	Pre		40	< 10	< 10	< 10	20	AC		
BL 234	Pre	11	160	< 10	< 10	< 10	640	160		
	Post		80	< 10	< 10	< 10	160	160	No cells	Negative
C 234	Pre	12	160	< 10	< 10	< 10	320	80		
	Post		160	< 10	< 10	< 10	160	80		
N 234	Pre		40	< 10	< 10	< 10	640	160/320		
	Pre		640	< 10	< 10	< 10	320	160/320		
	Pre		80	< 10	< 10	< 10	160	40		
	Pre		40	< 10	< 10	< 10	640	80/160		
BL 235	Pre	14	160	< 10	< 10	< 10	40	AC		
	Post		640	20	40	< 10	40	10	Positive	38
C 235	Pre	14	80	< 10	< 10	< 10	160	80		
	Post		80	< 10	< 10	< 10	160	80		
N 235	Pre		40	< 10	< 10	< 10	640	80		
	Pre		160	< 10	< 10	< 10	40	20		
	Pre		80	< 10	< 10	< 10	320	160		
	Pre		80	< 10	< 10	< 10	640	80		

Table 14 (continued)

Case registry number	Pre or post sera	Time interval (months)	VCA	EA	EA-R	EA-D	EBNA	CF/s	IF/EBNA smears	V-DNA gene equivalents
BL 242	Pre	33	1280	< 10	< 10	< 10	320	AC		
	Post		≥ 2560	80	20/40	< 10	320	160	NSA	NBA
C 242	Pre	30	40	< 10	< 10	< 10	160	AC		
	Post		80	< 10	< 10	< 10	160	40		
N 242	Pre		80	< 10	< 10	< 10	20	20		
	Pre		320	< 10	< 10	< 10	40	80		
	Pre		80	< 10	< 10	< 10	40	10		
	Pre		160	< 10	< 10	< 10	40	40		
BL 247	Pre	18	40	< 10	< 10	< 10	160	40		
	Post		40	< 10	< 10	< 10	40	20	Negative	Negative
C 247	Pre	18	1280	40	10/20	< 10	160	80/160		
	Post		640	10/20	< 10	< 10	160	40/80		
N 247	Pre		320	< 10	< 10	< 10	160	20/40		
	Pre		160	< 10	< 10	< 10	640	160/320		
	Pre		320	< 10	< 10	< 10	160	80		
	Pre		160	< 10	< 10	< 10	80	40		
BL 260	Pre	51	320	< 10	< 10	< 10	320	80		
	Post		640	< 10	< 10	< 10	2560	≥ 320	Negative	Negative
C 260	Pre	50	640	20	< 10	< 10	640	40		
	Post		160	< 10	< 10	< 10	320	40/80		
N 260	Pre		160	10	< 10	< 10	640	40		
	Pre		320	< 10	< 10	< 10	80	≤ 10		
	Pre		320	80	40	< 10	640	160		
	Pre		320	< 10	< 10	< 10	1280	AC		
BL 263	Pre	54	≥ 2560	2560	—	≤ 640	640	AC		
	Post		≥ 2560	2560	—	≤ 320	2560	40	Positive	40
C 263	Pre	54	*	*	*	*	*	*		
	Post		10	< 10	< 10	< 10	320	40		
N 263	Pre		80	< 10	< 10	< 10	640	40/80		
	Pre		10	< 10	< 10	< 10	160	40		
	Pre		80	< 10	< 10	< 10	160	40		
	Pre		80	< 10	< 10	< 10	640	80		

VCA — viral capsid antigen; EA — early antigen; EA-R — early antigen, restricted; EA-D — early antigen, diffuse; EBNA — nuclear antigen; CF/s — complement-fixing antigen; IF — immunofluorescence; V-DNA — Epstein–Barr viral DNA; NSA — no serum available; NBA — no biopsy available; AC — anti-complementary activity

The controls used were of three types:

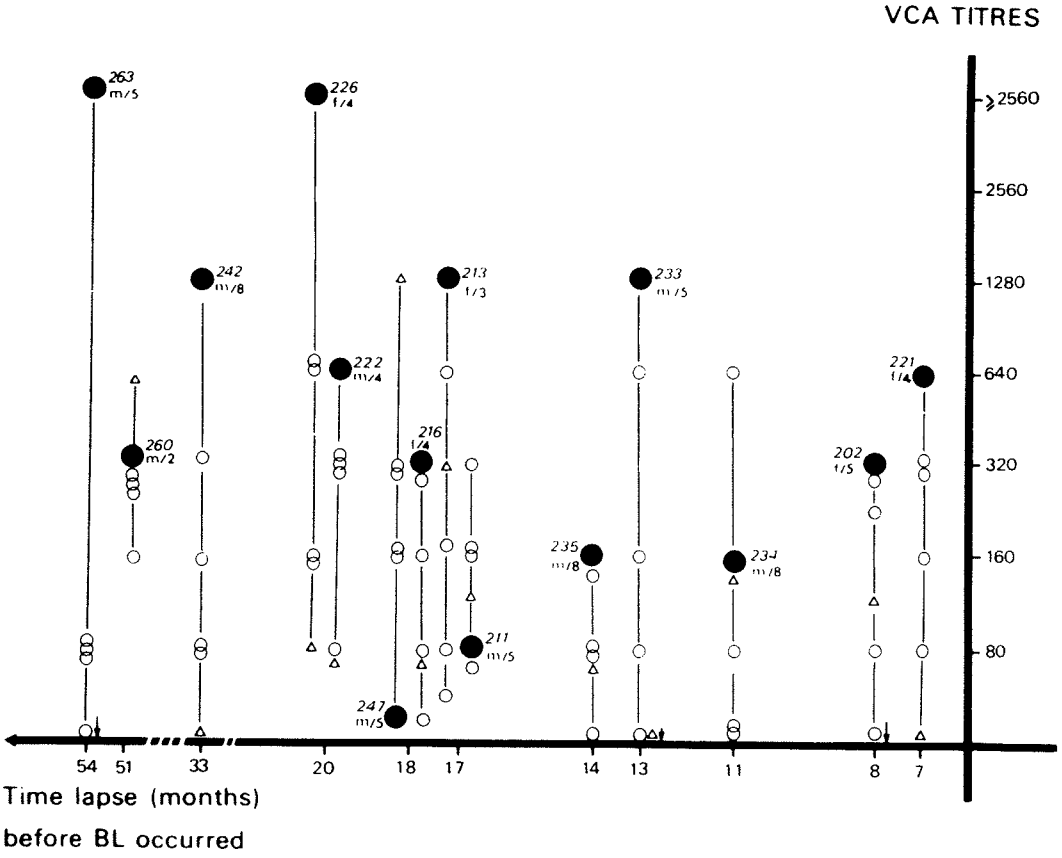
1. A randomly selected control chosen by the statistical clerk at the time that a Burkitt's lymphoma case occurred. This control was a neighbour of the same sex and age and was bled at the same time as the Burkitt's lymphoma case during the main study ('pre-bled' serum). The families of both the case and the control were bled again at the time the Burkitt's lymphoma was diagnosed ('post-bled' serum).

2. Four neighbours selected from the serum bank and, as far as possible, of the same age and sex and from the same locality as the case, and also bled in the main study ('pre-bled' serum)

3. A sample group of the general population of the same age-group as the case, whose serological profile was determined in the sero-epidemiological sub-study

All of the 'pre-bled' sera from the cases, with the exception of two, had either the highest, or close to highest, antibody titres against viral capsid antigen as compared to the randomly selected controls and neighbours (Fig. 15). However, the levels of the other Epstein–Barr virus reactivities — early antigen, nuclear antigen, and complement-fixing antigen—showed no significant differences between cases and controls (Fig. 16).

Fig. 15 Antibody titres against Epstein–Barr viral capsid antigen in 'pre-bled' sera from Burkitt's lymphoma cases (●), randomly-selected controls (△) and neighbours (○).

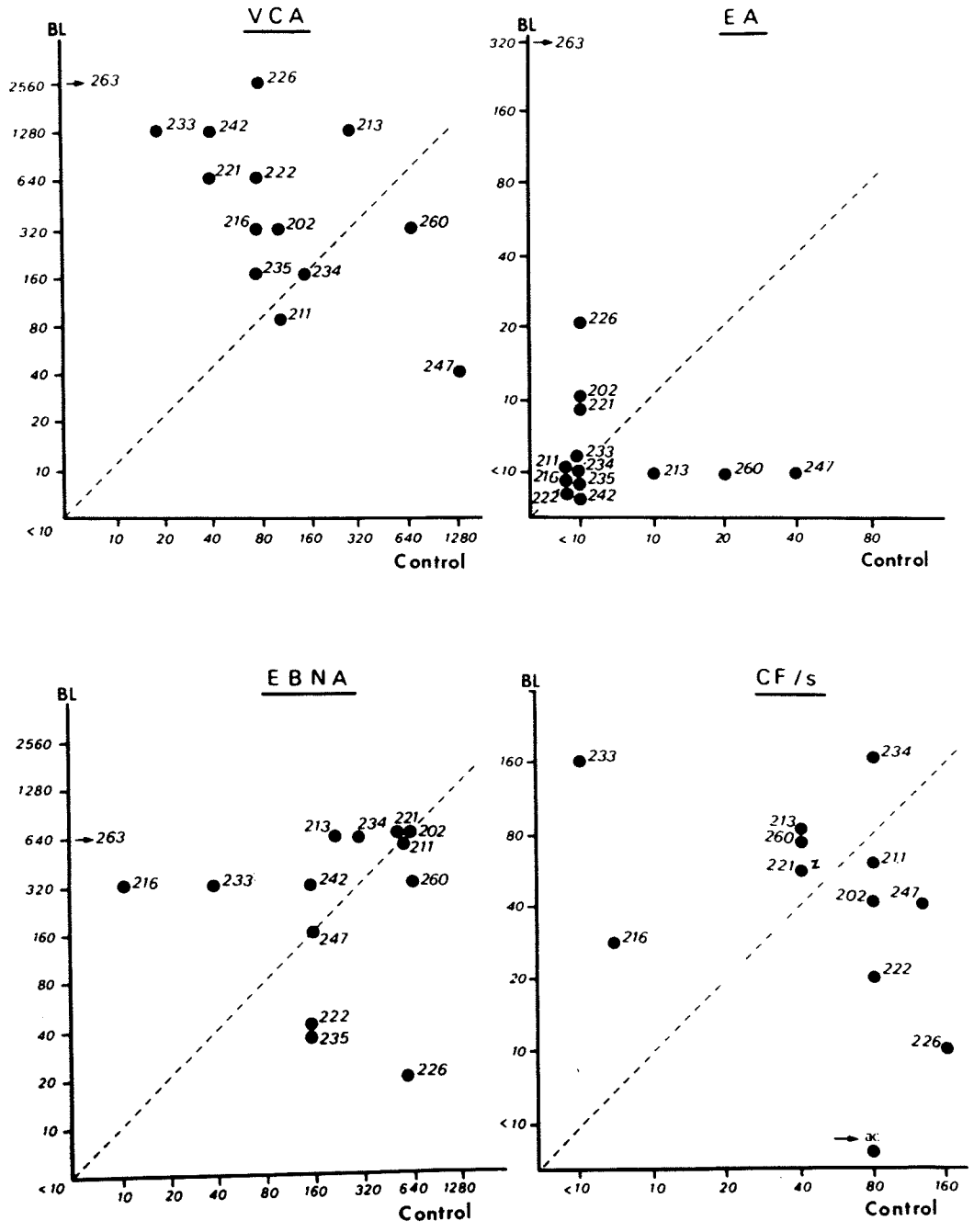


Comparison of the 'pre-bled' sera from the Burkitt's lymphoma patients with sera from the general population showed, with the same two exceptions, that patients had much higher anti-viral capsid antigen antibody titres (Fig. 17).

When the 'pre-bled' sera from the patients, which had been collected 7–54 months before the onset of the disease, were compared with sera collected from them after the onset of the disease and with sera from controls, the observed titres showed unexpected stability, with the exception of early antigen reactivities which showed seroconversion in eight cases (Fig. 18).

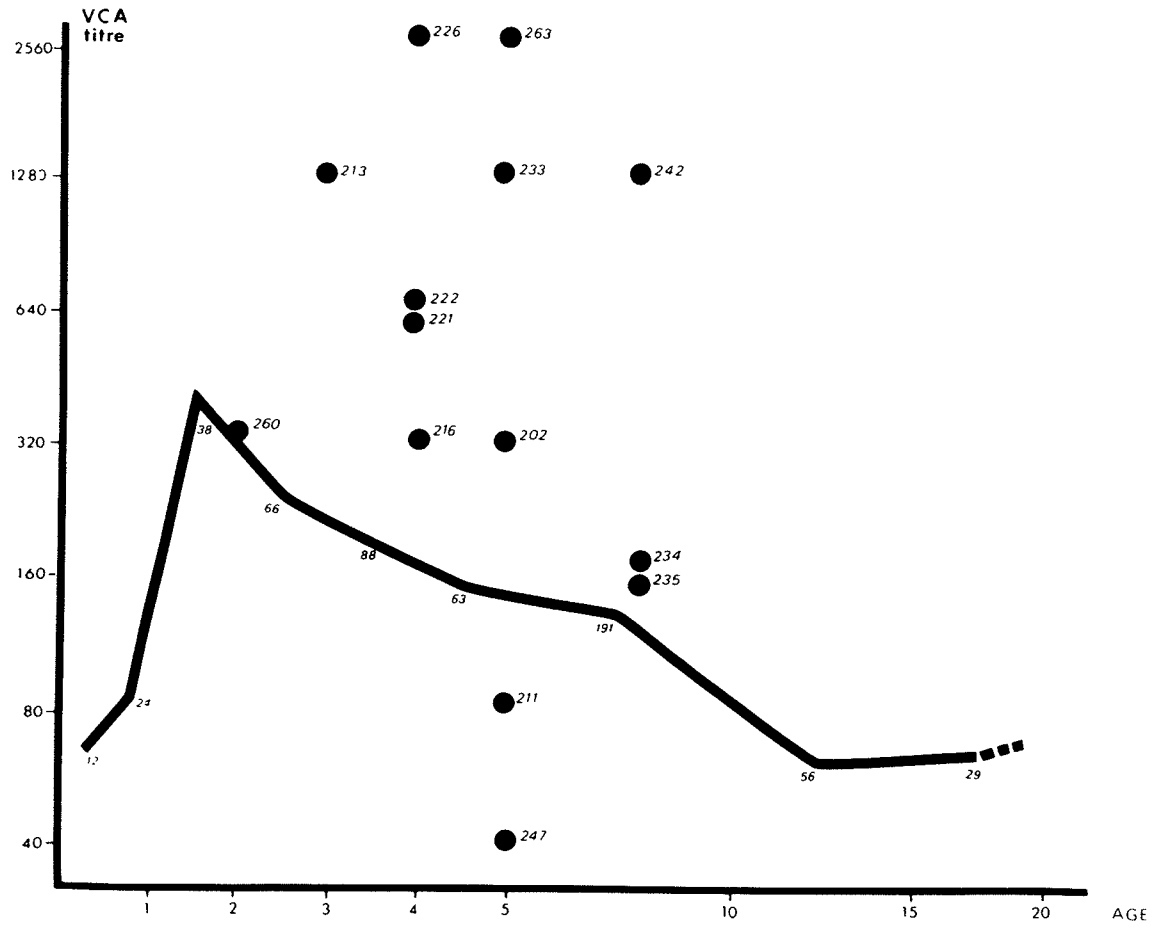
Statistical analysis of the data on the antibody titres against viral capsid antigen (Dr N. E. Day) in cases and controls showed a highly significant difference between the two groups ($P = 0.005$).

Fig. 16 Antibody titres against Epstein–Barr viral antigens — VCA — viral capsid antigen; EA — early antigen; EBNA — nuclear antigen; CF/s — complement-fixing antigen — before tumour development in 14 pairs of Burkitt's lymphoma patients and controls.



In summary, it may be concluded that a high level of antibodies to viral capsid antigen a long time prior to development of Burkitt's lymphoma, far from giving any protection against development of the disease (as is the situation in infectious mononucleosis), rather seems to signify a high risk of developing Burkitt's lymphoma. These conclusions favour

Fig. 17 Antibody titres against Epstein–Barr viral capsid antigen in 'pre-bled' sera from Burkitt's lymphoma cases (●) and from the general population (——).



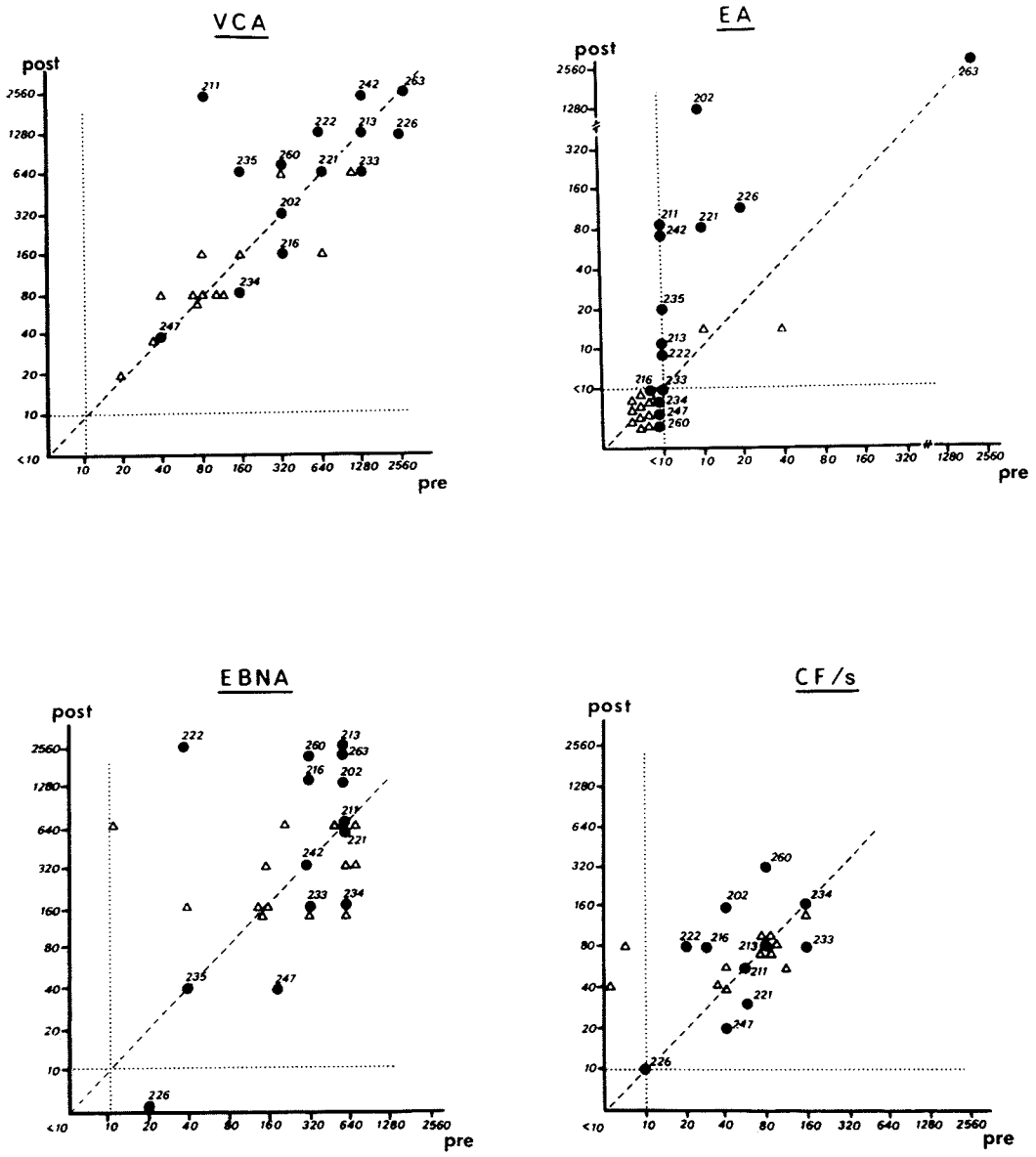
the hypothesis recently proposed [1] that an early infection with Epstein–Barr virus, possibly during the perinatal period of life, could represent a determining factor for the later development of Burkitt's lymphoma.

One of the two cases that presented exceptions to the general conclusions was examined in depth. Although from the clinical and pathological point of view this case was consistent with a diagnosis of Burkitt's lymphoma, no Epstein–Barr viral DNA could be detected in the tumour cells; the anti-viral capsid antigen antibody titre 18 months prior to tumour development was exceedingly low and showed no increase at the time of tumour development. This case, however, had some clinical and cytological peculiarities, and it is possible that there is a Burkitt's-type lymphoma in tropical areas that is not related to the Epstein–Barr virus. The case is, in fact, similar to that of the rare Burkitt's-type lymphomas seen in temperate climates, in which neither Epstein–Barr viral markers nor antibodies are associated with the tumour.

The two exceptional cases do not necessarily imply that the Epstein–Barr virus is not an etiological factor in Burkitt's lymphoma, but rather that there is a low incidence of a lymphoma everywhere in the world that is unrelated to the virus. The Burkitt's lymphoma

[1] de-Thé, G. (1977), *Lancet*, *i*, 335–338.

Fig. 18 Antibody titres against Epstein–Barr viral antigens — VCA — viral capsid antigen; EA — early antigen; EBNA — nuclear antigen; CF/s — complement-fixing antigen — in sera collected from Burkitt's lymphoma patients (●) before the onset of the disease ('pre') and after the onset of the disease ('post') and from controls (△).



which has a high incidence in the tropical belt and for which the Epstein–Barr virus appears to be a causal factor is superimposed on the rarer non-virus-related lymphoma.

Two more years of case detection should yield sufficient 'pre-bled' cases to permit an exact investigation of the etiological role of the virus in Burkitt's lymphoma. The Epstein–Barr virus is, however, only one of the causal factors of Burkitt's lymphoma and is probably associated with a specific age and mode of infection, and the search for other cofactors is important.

Very early and probably massive Epstein–Barr virus infection, separated so far apart in time from the onset of the disease, could only represent an initiating agent. The promot-

ing factor appears to be within the intermediate environment of the case, and this may be malaria infection itself. Alternatively, malaria may be an epidemiological marker for an as yet unknown precipitating factor.

2.2 *Cofactors in the etiology of Burkitt's lymphoma* (Dr A. Geser)
Shirati Mission Hospital, Musoma, Tanzania

Principal investigator: Dr G. Brubaker

The close geographical association of hyperendemic malaria and high incidence of Burkitt's lymphoma was further confirmed in continuing epidemiological studies in East Africa during the present reporting period. Malaria thus remains the environmental factor most likely to be involved in the etiology of this tumour, and the proposed malaria suppression trial [1] has now been launched in North Mara, Tanzania.

This project offers at the same time excellent prospects for studying the operational and immunological aspects of a malaria control programme integrated into the new community structure which has evolved in Tanzania. For this reason, the division of malaria and other parasitic diseases and the special programme for research and training in tropical diseases, both of WHO, have agreed to participate in this trial. With their support, chloroquine distribution to children in North Mara was initiated in March 1977. Excellent cooperation has been received from local medical and administrative authorities. The population is cooperative and willing to assist the programme; so far, nearly 85% of the eligible children report regularly for drug distribution, and the prevalence of malaria parasitaemia has declined from about 40% to about 5% in the child population covered by the scheme. A system is now being established to compare child mortality and morbidity in North Mara with that in South Mara, where no explicit malaria suppression is yet being carried out.

The detection and registration of Burkitt's lymphoma cases will continue in North and South Mara in order to evaluate the effect of the anti-malaria measures on the incidence of Burkitt's lymphoma.

A mathematical model is being constructed to see whether variations in the amount of rainfall in the West Nile, Uganda is in any way synchronized with the considerable variation in Burkitt's lymphoma incidence that has been observed from season to season and from year to year.

3. ETIOLOGICAL STUDIES ON NASOPHARYNGEAL CARCINOMA

3.1 *Symposium*

An international symposium on the etiology and control of nasopharyngeal carcinoma was organized jointly by the Japanese Society for Promotion of Science, the Virus Cancer Program of the National Cancer Institute (USA) and the Agency and was held in Kyoto, Japan from 4–6 April 1977.

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[1] International Agency for Research on Cancer (1976) *Annual Report, 1976*, p. 70.

The progress made since the first UICC symposium on this topic, organized in 1964 by Professor K. Shanmugaratnam in Singapore, was reviewed in the fields of both clinical and fundamental research. Recent research results were also presented.

The participants separated into disciplinary-defined subgroups to discuss and make recommendations for future research; they covered histopathological classification, clinical staging, epidemiology, genetics, virology and immunology.

3.2 *Characterization of Epstein–Barr virus antigens—development of field-adaptable serological tests* (Dr G. Lenoir)

In order to develop simple, highly specific serological tests that can be applied to the testing of a large number of specimens, a programme on the characterization and purification of the Epstein–Barr virus antigens has been set up:

(*a*) *Characterization of the Epstein–Barr virus antigens* (in collaboration with Professor J. Daillie, Department of Biology, Claude-Bernard University, Lyon, France)

Two antigenic systems are presently under investigation. The first is the Epstein–Barr virus nuclear antigen, which is the first antigen detectable in a cell after Epstein–Barr virus infection. This antigen is expressed in all Epstein–Barr virus genome-carrying cells, either *in vitro* in lymphoblastoid cells (whether they are producing viral particles or not) or *in vivo* in tumour cells, i.e., Burkitt's lymphoblasts or epithelial cells of nasopharyngeal carcinomas. The presence of this antigen in a cell is a reflection of its malignant state.

Nuclear antigen is presently purified using gel filtration techniques as well as various chromatography columns, in particular those with affinity for DNA. Under dissociating conditions, such as a 6 M concentration of guanidine chloride, the antigenic molecule, which has a molecular weight of 180 000 daltons in its native form [1], is dissociated into smaller components with molecular weights of about 70 000. A radioimmunoassay for the detection of this antigen in tumour cells will be developed as soon as its purification reaches a satisfactory level.

The second system under study is that of the early antigens. Their characterization is important since high levels of antibodies directed against them are found preferentially in patients with Epstein–Barr virus-related diseases, such as Burkitt's lymphoma, nasopharyngeal carcinoma and infectious mononucleosis, and not in the general population. They are being purified by techniques similar to those employed for the nuclear antigen. Furthermore, we have been able to show that the early antigens contain a molecule with an Epstein–Barr virus-specific DNA polymerase activity [2].

(*b*) *Development of field adaptable Epstein–Barr virus serological tests* (in collaboration with Dr A. Voller, Zoological Society of London)

The micro-Elisa test, a new serological test using alkaline phosphatase-labelled anti-human immunoglobulins, has been applied to the detection of antibodies directed against Epstein–Barr virus antigens. Using crude preparations of P3 HR1 cells as a source of

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[1] Lenoir, G., Berthelon, M. C., Favre, M. C. & de-Thé, G. (1976) *J. Virol.*, **17**, 672–674.

[2] Ooka, T., Daillie, J., Costa, O. & Lenoir, G. (1978) In: *Proceedings of the 3rd International Symposium on Oncogenesis and Herpesviruses, Boston, 1977* (in press).

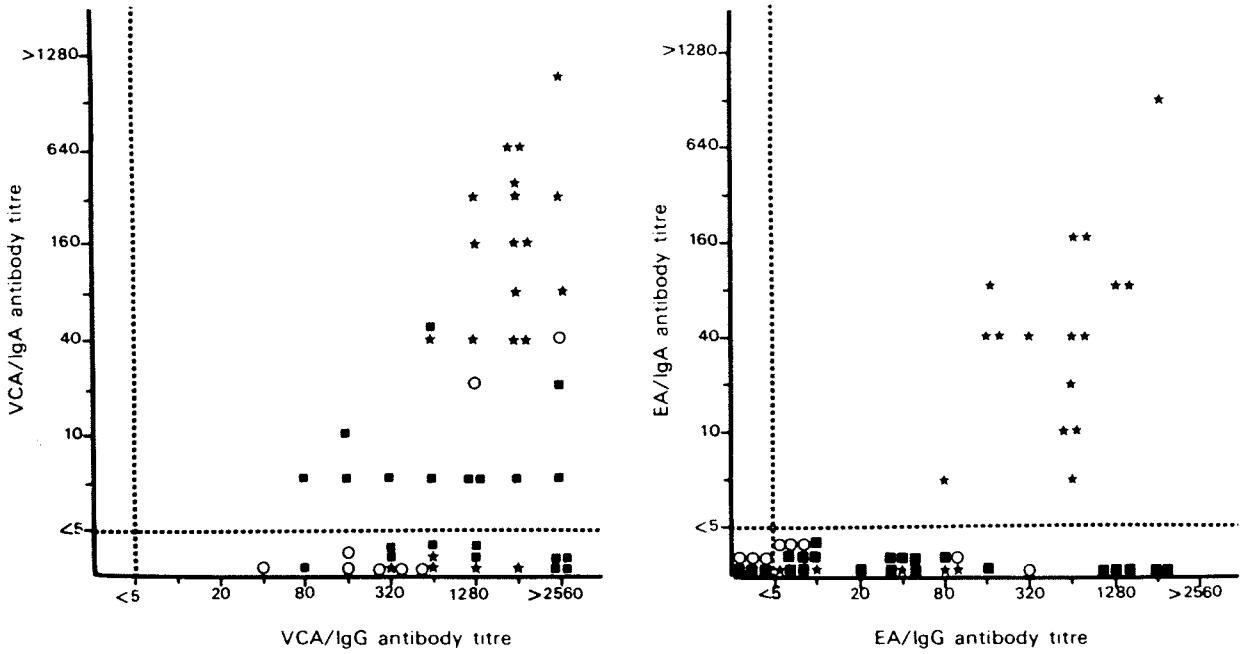
antigens, the testing of a large number of sera of various origins (nasopharyngeal carcinoma, Burkitt's lymphoma and infectious monocucleosis) has shown that this test could be applied to our field needs. However, purified antigen preparations are required for specific detection of viral capsid antigen and early antigen reactivities.

Using a partially purified early antigen preparation (obtained from Raji cells treated with 5-iodo-2'-deoxyuridine), detection of the specific early antigen antibodies correlates very well with the data obtained by immunofluorescence. Presently, the major problem is the detection of the viral capsid antibodies in low-titre sera. For this purpose, we are preparing viral capsid antigen extracts with an increased specificity activity. In addition, detection of antibodies of the IgA and IgM class of immunoglobulins is also under investigation, and this test will be used directly in the nasopharyngeal carcinoma and Burkitt's lymphoma programmes.

3.3 *Immunoglobulins* (Mrs C. Desgranges-Blanc)

Patients with nasopharyngeal carcinoma and Burkitt's lymphoma are known to have high serum antibody titres against Epstein–Barr virus antigens [1, 2]. These antibodies are mainly of the IgG class of immunoglobulins. A recent study by Wara *et al.*[3] showed an increase in total immunoglobulin A (IgA) in the sera of patients with nasopharyngeal carcinoma. These sera were tested for Epstein–Barr virus-specific IgA, anti-viral capsid antigen and early antigen. Immunofluorescence tests were used to compare this group of

Fig. 19 Immunoglobulin A (IgA) and immunoglobulin G (IgG) antibody titres against Epstein–Barr viral antigens — VCA — viral capsid antigen; EA — early antigen — in patients with nasopharyngeal carcinoma (★), Burkitt's lymphoma (■) or other cancers of the head and neck (○).



[1] Henle, W., Henle, G., Burtin, P., Cachin, Y., Clifford, P., de Schryver, A., de-Thé, G., Diehl, V., Ho, H. C. & Klein, G. (1970) *J. nat. Cancer Inst.*, **44**, 225–231.

[2] de-Thé, G., Ho, J. H. C., Ablashi, D. V., Day, N. E., Maracio, A. J. L., Pearson, G., Sohier, R. & Martin-Berthelon, M. C. (1975) *Int. J. Cancer*, **16**, 713–721.

[3] Wara, W. M., Wara, D. W., Phillips, T. L. & Ammann, A. J. (1975) *Cancer*, **35**, 1313–1315.

sera with those of Burkitt's lymphoma patients and of patients with other cancers of the head and neck. It was found that nasopharyngeal carcinoma patients had levels of anti-viral capsid antigens equal to or higher than 1/40; other patients consistently had lower titres. The anti-early antigen IgA titres were positive, that is, greater than 5, only in nasopharyngeal carcinoma patients (Fig. 19). Thus, the presence of these Epstein–Barr virus-specific immunoglobulins—and especially of the anti-early antigen IgA—could be used as a diagnostic test for nasopharyngeal carcinoma.

The IgA levels in sera from nasopharyngeal carcinoma patients were compared with those in their families and in the normal population (Table 15). The levels found in the families of nasopharyngeal carcinoma patients—a high risk group—were significantly higher than those in the normal population.

Table 15. Presence of Epstein–Barr virus specific IgA in sera from nasopharyngeal carcinoma (NPC) patients, family members and the normal population

	VCA/IgA	%	EA/IgA	%
NPC patients	127/145	87.6 %	106/145	73.1 %
NPC family members	88/278 *	31.7 %	2/278	0.8 %
Normal population	93/522 *	17.9 %	0/522	0 %

* $\chi^2 = 19.84$ $P < 0.001$
VCA = viral capsid antigen;
EA = early antigen

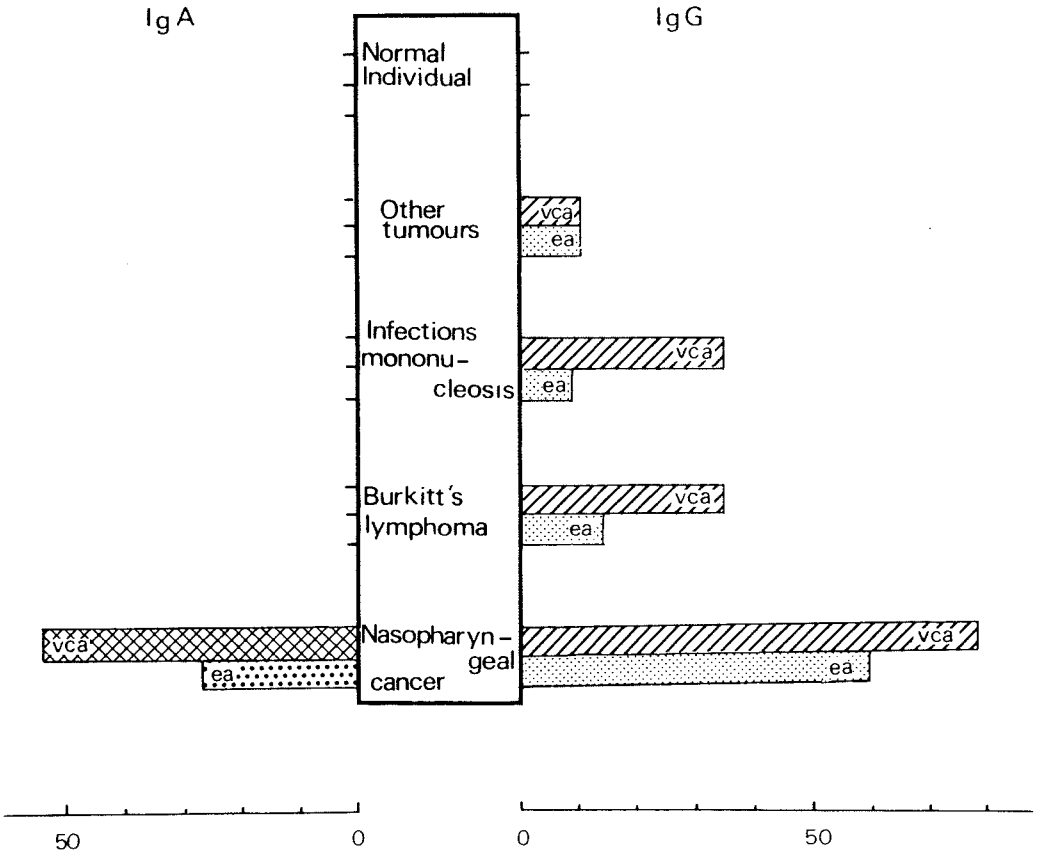
A case has been reported from Hong Kong [1] in which a relative of a nasopharyngeal cancer patient was found to have a positive Epstein–Barr virus-specific IgA titre in his serum. Seven months later, this relative also developed a nasopharyngeal carcinoma; a second serological test at that time showed an increase in the IgA titre, although the IgG titre, which had previously been high, remained constant. A prospective serological study in a high-risk population, testing for the Epstein–Barr virus-specific IgA, could provide a means of early detection of those most susceptible to the development of nasopharyngeal carcinoma.

Saliva was also collected from patients with nasopharyngeal carcinoma and tested for immunoglobulins. As in the sera, IgA was found, and this contrasted with the situation in all other persons tested—those with Burkitt's lymphoma or infectious mononucleosis, patients with other head and neck tumours and 'normal' controls—in whom no Epstein–Barr virus-specific IgG was found in the saliva (Fig. 20).

When secretory tissue from nasopharyngeal carcinoma patients was tested with a specific antiserum, immunofluorescence revealed the secretory origins of the IgA. This was confirmed by chromatographic analysis of the saliva after filtration, in which the separated fractions sedimented with a coefficient of 11s for secretory IgA and 7s for IgA(α) and IgG (Fig. 21). The Epstein–Barr virus-specific IgA seemed to originate from a local, immune secretory reaction, and this also might be used as a rapid, complementary diagnostic aid for nasopharyngeal carcinoma.

[1] Ho, H. C., personal communication.

Fig. 20 Presence of the immunoglobulins (IgA, IgG) specific to Epstein–Barr viral antigens — VCA — viral capsid antigen; EA — early antigen — in saliva from different groups of persons.



The saliva samples were also examined for the presence of 'complete' Epstein–Barr virus, by their ability to transform human cord-blood leucocytes, and compared with saliva samples from patients with Burkitt's lymphoma and infectious mononucleosis (Table 16). The percentage of positive samples in the saliva from nasopharyngeal carcinoma patients was even lower than in the samples from healthy controls and much lower than the levels in the other two Epstein–Barr virus-associated diseases—Burkitt's lymphoma and infectious mononucleosis. Nevertheless, electron microscopic examination of the samples that were negative in the leucocyte-transformation test still showed the presence of clusters of Epstein–Barr viruses (Fig. 22).

Examination by immunofluorescence of total immunoglobulins extracted from a nasopharyngeal tumour showed Epstein–Barr virus-specific IgA(α) and secretory pieces. When fixed sections of the localized tumour were examined, (α)-chain IgA were localized in the plasmocytes surrounding the tumoral tissue (Fig. 23A), and the secretory pieces were present in mucous gland cells (Fig. 23B) and in the epithelial tumour cells (Fig. 23C).

3.4 *Genetic studies* (Dr J. P. Lamelin)

The association between nasopharyngeal carcinoma and HLA antigens of the major histocompatibility complex has been further explored among newly-diagnosed Chinese

patients from Singapore. An extensive survey of HLA antigens, both their characterization and distribution, is now in progress in sera collected in Tunisia, using a panel of local antisera.

Fig. 21 Presence of immunoglobulins (IgAα, IgG) specific to Epstein–Barr viral capsid antigen (VCA) in secretory tissue from nasopharyngeal carcinoma patients, as shown by immunofluorescence (IF) and confirmed by chromatography. SP — secretory IgA.

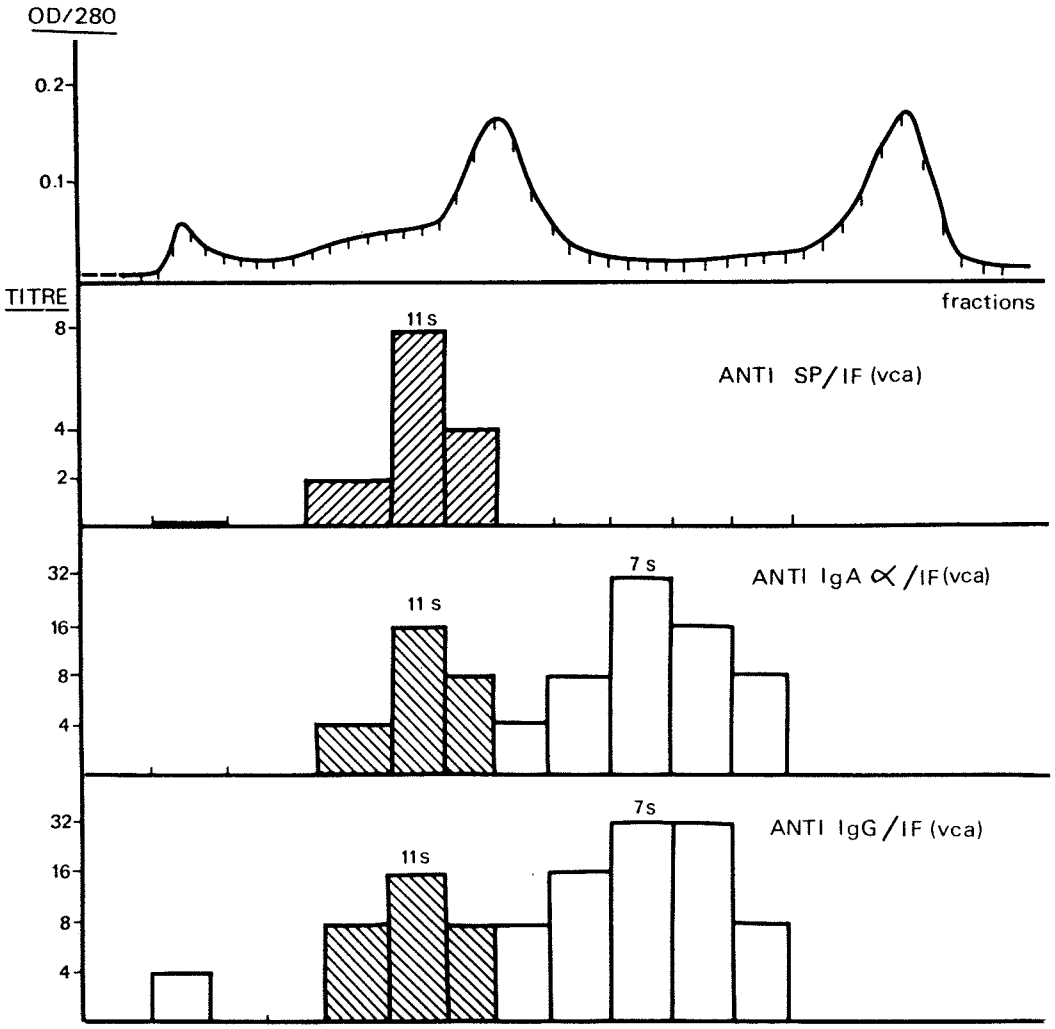
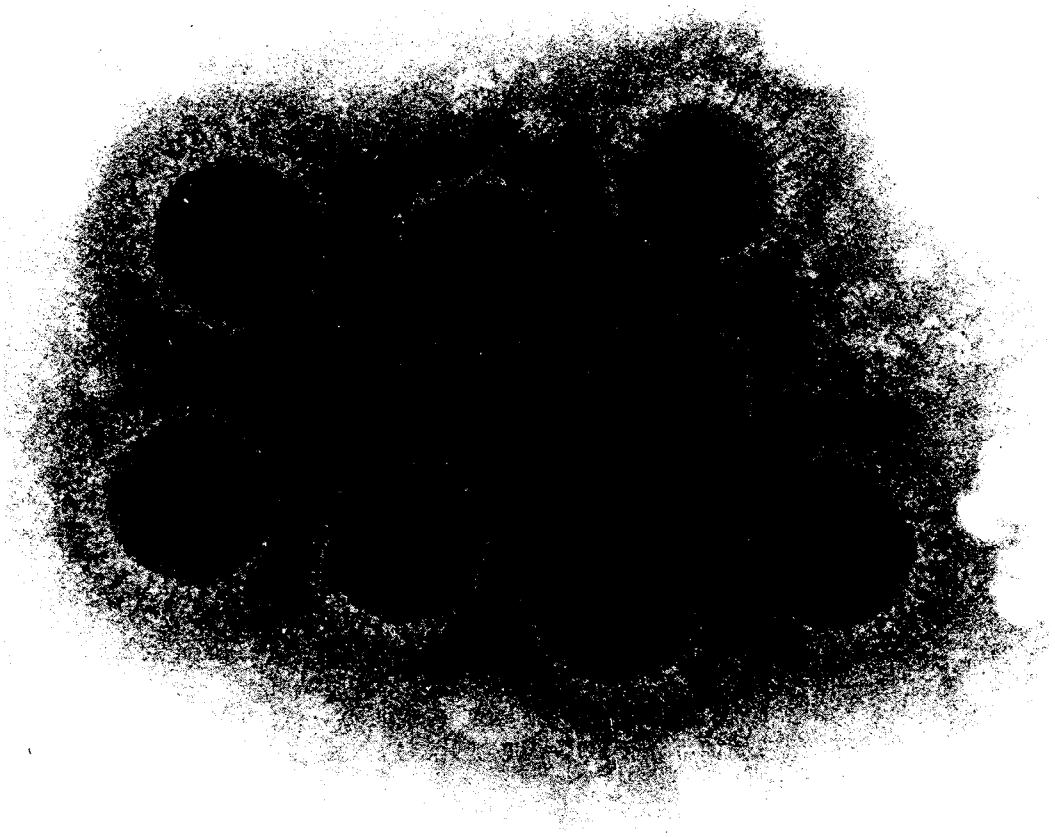


Table 16. Transformation of human umbilical cord leucocytes by saliva samples from patients with nasopharyngeal carcinoma (NPC), infectious mononucleosis (IM) and Burkitt's lymphoma (BL) and from matched controls

Source of saliva	No. of samples tested	No. positive for transforming EBV	Percentage positive
NPC patients	23	2	8.7
IM patients	12	8	66.7
BL patients	24	12	50
Healthy subjects	29	6	20.7

EBV-Epstein–Barr virus

Fig. 22 Presence of clusters of Epstein–Barr viruses which did not transform human cord blood leucocytes in samples of saliva from patients with nasopharyngeal carcinoma.



(*a*) *Singapore*

WHO Immunology Research and Training Centre, Singapore

Principal investigator: Dr S. H. Chan (See report of the Singapore Research Centre, page 127)

(*b*) *Tunis–Lyon*

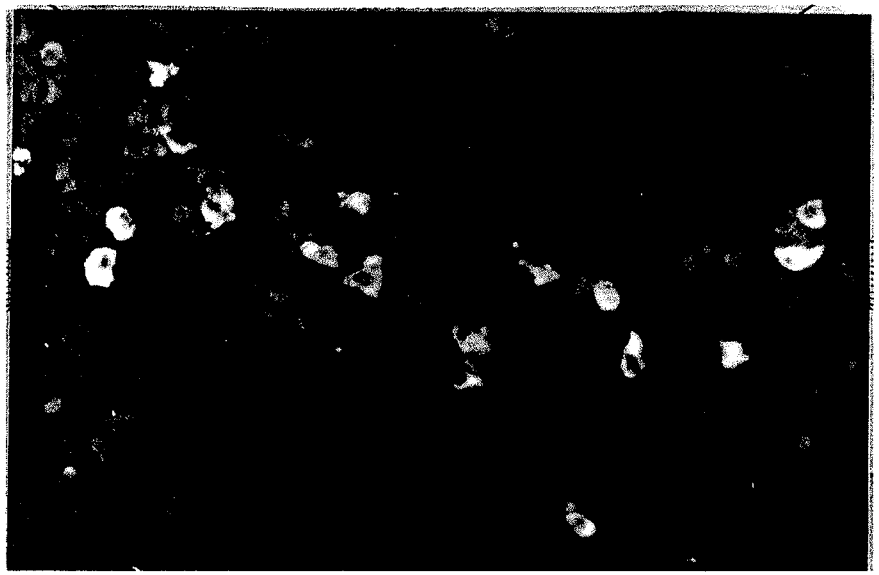
Salah-Azaiz Institute, Tunis (Professor N. Mourali, Professor R. Sohier)

Blood Transfusion Centre, Beynost, France (Dr H. Bétuel)

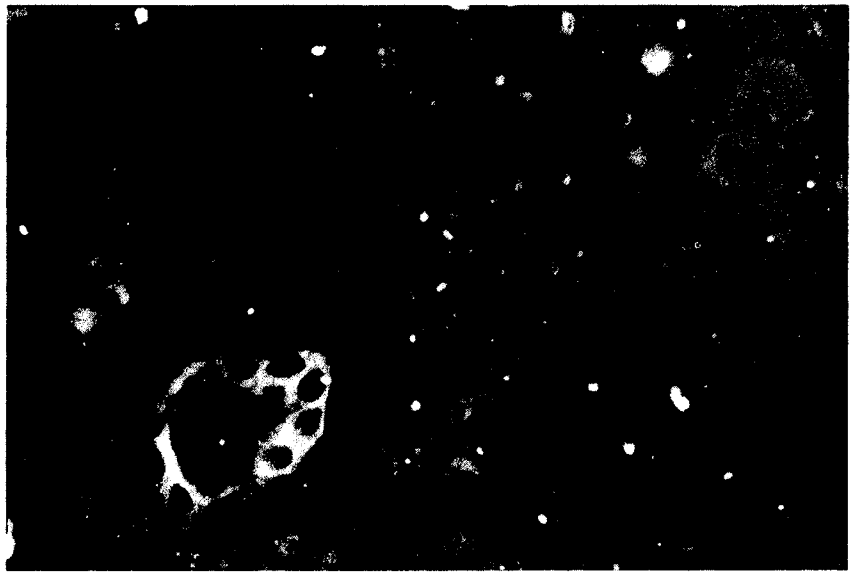
A total of 1030 sera from multiparous women were collected, and, of these, 323 were screened for HLA antibodies on 80 different lymphocyte suspensions (mostly from blood donors), in order to establish the HLA profile of the general population. This will provide the base-line for comparison with nasopharyngeal cancer patients, similar to the investigation in Singapore that led to the discovery of the HLA haplotype A-2 B-Sin 2 among the population at high risk for nasopharyngeal carcinoma there.

Anti-HLA antibody activity was found in 137 sera (42%), which were classified as follows: polyspecific, 37 (11%); relatively monospecific, 26 (8%); and antigens that were present in less than 10% of the population, 74 (23%). The data will be analysed to establish the actual distribution of HLA antigenic specificities among the Tunisian population.

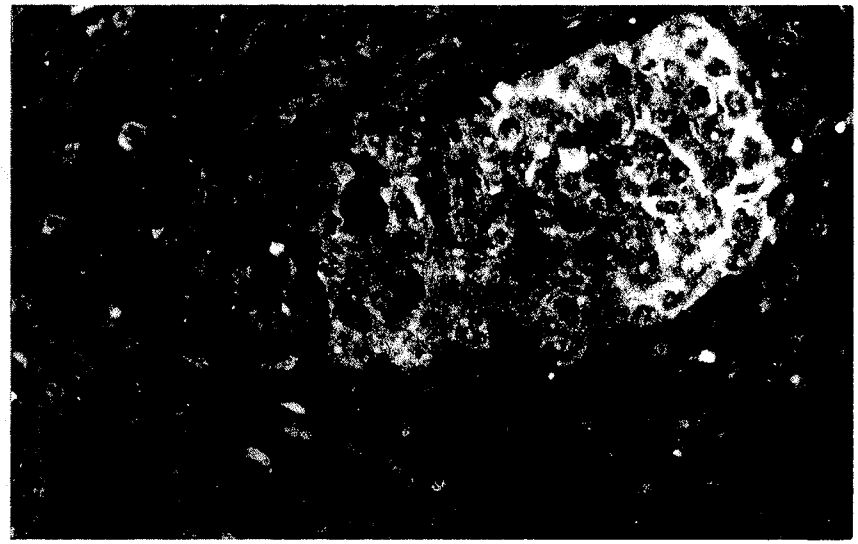
Fig. 23 Sections from a nasopharyngeal tumour, showing (A) (α)-chain IgA in the plasmocytes surrounding the tumoral tissue; (B) secretory pieces in mucous gland cells; and (C) secretory pieces in epithelial tumour cells.



A



B



C

Further typing of nasopharyngeal carcinoma patients in Tunisia will depend upon whether the sera now collected show reactions with specificities characteristic of this population.

3.5 *Immunology* (Dr J. P. Lamelin)

The concept of immune surveillance and its failure as a major factor in oncogenesis is the origin of a broad interest in different aspects of cell-mediated immune reactions of cancer patients. For nasopharyngeal cancer patients, there are two additional and specific reasons for this interest. Firstly, among at least the 'high-risk' group of Cantonese Chinese, 'susceptibility' to nasopharyngeal carcinoma was found to be associated with the HLA haplotype A-2 B-Sin2, although the underlying mechanism was not known. In view of the link between the major histocompatibility complex and the genes that control T-cell functions, 'susceptibility' may operate within these T-lymphocytes. Secondly, since the usual treatment given to nasopharyngeal carcinoma patients is radiotherapy, this might provoke an impairment of the patient's immune reaction which might supervene on pre-existing abnormalities.

Three aspects of cell-mediated immunity have been studied:

(i) *Non-specific cell-mediated immune response*

(*a*) *WHO Immunology Research and Training Centre, Singapore*

Principal investigator: Dr S. H. Chan

Newly-diagnosed nasopharyngeal carcinoma patients were tested for their *in vivo* skin reactivity to purified protein derivative—the Mantoux test—and for the *in vitro* responsiveness of their blood lymphocytes to phytohaemagglutinin. Although in some of the patients the results of these tests were within the normal limits, a significant depression was found in the group as a whole compared with the control group. After more than 3½ years' follow-up, the survival rate of nasopharyngeal carcinoma patients with normal responses to the Mantoux or phytohaemagglutinin tests appears to be better than that of those with lower responses. These results indicate that these two non-specific tests for cell-mediated immunity are of prognostic value for patients with nasopharyngeal carcinoma.

(*b*) *University of Lyon, Lyon, France*

Principal investigator: Dr J. P. Revillard

Peripheral blood lymphocytes from 18 Tunisian nasopharyngeal carcinoma patients were compared with those from matched controls, before and after cobaltotherapy, for their ability to form E- and EAC-rosettes and to mount a proliferative response with the mitogens, phytohaemagglutinin or concanavalin A, and anti-lymphocytic globulins. A slight decrease in the percentage of E-rosettes formed and a moderate hyporesponsiveness to phytohaemagglutinin and concanavalin A were observed in the lymphocytes taken before treatment, but the results were not statistically significant. Within the group of treated patients, a much greater depression was found, including the response to anti-lymphocytic globulins, although in a few long-term survivors the response to mitogens was equal to that in the controls. These findings underline the difficulty of interpreting the results of longitudinal studies of cell-mediated immunity, specific or non-specific, in nasopharyngeal carcinoma patients.

(ii) *Specific cell-mediated immune response towards Epstein–Barr virus- and nasopharyngeal carcinoma-related antigens*

University of Hong Kong, Hong Kong

Principal investigator: Dr M. H. Ng

The blastogenic response has been compared between lymphocytes from nasopharyngeal carcinoma patients and those from patients with other tumours. First, the response of the two groups of lymphocytes to a 'neutral' environmental antigen, purified protein derivative, was shown to be 'positive' and identical. Then, the responses induced by membrane extracts from Epstein–Barr virus-transformed lymphoblastoid cell lines and from pooled biopsy specimens from nasopharyngeal carcinoma patients were compared using a tritiated thymidine incorporation technique. A significantly higher frequency of 'positive' responses was obtained with the lymphocytes from the nasopharyngeal carcinoma patients.

In collaboration with Dr D. A. Stevens (visiting scientist, Santa Clara Valley Medical Centre, San José, Calif., USA), peripheral leucocytes from Tunisian nasopharyngeal carcinoma patients were compared with those from matched controls by measuring their capacity to attach to plastic in the presence of Epstein–Barr virus-related antigens. The results of a pilot study, involving only ten nasopharyngeal carcinoma patients and six individuals with other tumours, did not indicate any disease specificity with this technique.

(iii) *Soluble immune complexes*

Circulating, soluble immune complexes were found to be more frequent in the sera from nasopharyngeal carcinoma patients than in those from matched controls. These results, obtained by the Clq binding test, were confirmed in the comparative study undertaken by the WHO Immunology Research Laboratory, Geneva, Switzerland to assess and compare various techniques (see page 120).

A larger series of sera from nasopharyngeal carcinoma patients and controls were examined by Dr J. P. Revillard (Department of Immunology, Claude Bernard University, Lyon, France), also using the Clq binding test. The level of circulating immune complexes was shown to be higher in sera from nasopharyngeal carcinoma patients from Hong Kong, irrespective of the stage of their disease, than in normal, matched controls.

The situation was similar among Tunisian nasopharyngeal carcinoma patients, in whom two additional, unexpected observations were made. In a series of controls consisting of family members of the selected patients, the level of circulating immune complexes was found to be intermediate between that of the patients themselves and that of unrelated 'normal' Tunisian individuals. Moreover, in individual sera from both the patients and family controls, a correlation was found between the level of binding and the level of antibodies against Epstein–Barr viral capsid antigen, but no correlation was found between the level of binding and the level of antibodies against either Epstein–Barr nuclear antigen or early antigen. This finding raised important questions as to the nature of the antigen present in the complexes. It is being studied further.

4. UNIT OF CHEMICAL CARCINOGENESIS

Dr L. TOMATIS (Chief)

1. INTRODUCTION

The identification of potentially carcinogenic chemicals in the environment and the evaluation of their carcinogenic risk to man are the main activities of the unit. The goal is to provide information that can be used in primary prevention of cancer. Up to July 1977, fifteen volumes in the series *Evaluation of Carcinogenic Risk of Chemicals to Man* have been published, containing monographs on 336 chemicals. Evidence or a strong suspicion of an association with the occurrence of cancer in man was found for 25 of the chemicals considered. For 16 of these 25 chemicals, the human hazard arose in an occupational exposure, for eight it was related to therapeutic measures, and there was one example of dietary exposure. There is good evidence that the general population can also be exposed to several of the occupational and iatrogenic carcinogens.

Among the difficulties encountered in the compilation of the monographs and, specifically, in making the final assessment of the existence of a risk for man were the predominance of experimental over epidemiological data and the inadequacies of a number of experimental reports, which rendered some of the published data unusable.

A worldwide survey on chemicals being tested for carcinogenicity has continued in an attempt to increase communication between scientists working in this field and, thereby, to reduce unnecessary duplication of carcinogenicity testing. The information from the survey has been disseminated in periodical bulletins.

A limited programme of long-term carcinogenicity testing has been carried out, but a large part of laboratory activities has been devoted to the development and improvement of rapid screening tests for environmental chemicals and to the study of the mechanisms of carcinogenesis that may permit the development of criteria for a better assessment of the significance to man of results obtained at the laboratory level. A particular effort has been made to identify a group of tests to be carried out in parallel that would reduce false-negative results to a minimum. Through the laboratory programmes, liaison has been established with a number of national institutes, which has served towards the development of growing international collaboration in this field.

The investigation of the possible role of prenatal events in the occurrence of cancer in children (see page 121) has continued. In collaboration with the National Institute for the Study and Treatment of Tumours, Milan, Italy, a cancer registry in the Lombardy region of Italy is being established.

2. COORDINATION OF CARCINOGENICITY DATA

2.1 *Monographs on* The Evaluation of the Carcinogenic Risk of Chemicals to Man (Dr L. Tomatis, Dr J. E. Huff and Mr J. D. Wilbourn, in collaboration with Dr C. Agthe, Division of Food Additives, WHO, Geneva, Switzerland)

The 1976 IARC Annual Report [1] listed the substances considered in Volumes 10 and 11 of the IARC monographs. Volumes 1–9 of the monographs were described in previous Annual Reports. Since July 1976 four more volumes of the monographs have been published.

Volume 12, which considered a number of carbamates, thiocarbamates and carbazides (Table 17), was prepared by a working group which met in June 1976 and was published in November 1976. In October 1976, another working group considered some miscellaneous pharmaceutical substances (Table 17), and the monographs were published as Volume 13 in March 1977.

Table 17. Substances included in *IARC Monographs on the Evaluation of the Carcinogenic Risk of Chemicals to Man*, Volumes 12, 13, 14, and 15

Substance	Volume	Substance	Volume
Acriflavinium chloride	13	Ledate	12
Asbestos	14	Maneb	12
Aurothioglucose	13	Methyl carbamate	12
1,2-Bis (chloromethoxy)ethane	15	Methyl iodide	15
1,4-Bis (chloromethoxymethyl)benzene	15	Methyl selenac	12
Carbaryl	12	Metronidazole	13
Chlorinated dibenzodioxins	15	Monuron	12
Chloropropham	12	Niridazole	13
Chloroquine	13	Oxazepam	13
Copper 8-hydroxyquinoline	15	Oxymetholone	13
2,4-D and esters	15	Oxyphenbutazone	13
Diallate	12	Phenacetin	13
Diazepam	13	Phenicarbazide	12
1,2-Dibromo-3-chloropropane	15	Phenobarbital	13
trans-1,4-Dichlorobutene	15	Phenobarbital sodium	13
Dihydroxybenzenes	15	Phenylbutazone	13
Dimethoxane	15	Phenytoin	13
Dimethylcarbamoyl chloride	12	Phenytoin sodium	13
Disulfiram	12	Potassium bis (2-hydroxyethyl)-dithiocarbamate	12
Dithranol	13	Pronetalol hydrochloride	13
Dulcin	12	Propham	12
Eosin	15	*n*-Propyl carbamate	12
Eosin disodium salt	15	Pyrimethamine	13
Ethionamide	13	*para*-Quinone	15
Ethylene dibromide	15	Semicarbazide (hydrochloride)	12
Ethyl selenac	12	Sodium diethyldithiocarbamate	12
Ethyl tellurac	12	Succinic anhydride	15
Ferbam	12	2,4,5-T and esters	15
Hexamethylphosphoramide	15	Thiram	12
Hycanthone	13	1,2,3-Tris (chloromethoxy)propane	15
Hycanthone mesylate	13	Zectran	12
8-Hydroxyquinoline	13	Zineb	12
Isopropyl alcohol	15	Ziram	12
Isopropyl oils	15		

[1] International Agency for Research on Cancer (1976) *Annual Report, 1976*, Lyon, p. 85.

A first monograph on asbestos was published in 1972 [1], and an updated evaluation of the data was drafted by a working group in December 1975. The publication of this monograph was delayed, however, pending the results of further studies known to be nearing completion and because several members of the Working Group considered that the chemical, physical and geological characteristics of asbestos fibres had not been fully covered. Therefore, in December 1976 a special working group was convened to reconsider asbestos, including chrysotile, actinolite, amosite, anthophyllite, crocidolite and tremolite fibres. The group made an evaluation of all available data published or accepted for publication up to December 1976, and a new section on the physical and chemical properties of asbestos fibres was prepared. This was published as Volume 14 in April 1977.

A working group met in February 1977 to consider some fumigants, the herbicides 2,4-D and 2,4,5-T, chlorinated dibenzodioxins and some miscellaneous industrial chemicals. These monographs were published in Volume 15 (Table 17).

The evaluations in each volume of the IARC monographs are made by an international group of experts in the main disciplines of cancer research; 134 experts from 21 countries have attended as members of the working groups which prepared the evaluations published in the first 15 volumes of the monographs. Representatives from the Stanford Research Institute, USA, the National Cancer Institute, USA, and other observers from various countries have also attended these meetings. The members and observers who attended the working groups which resulted in Volumes 12, 13, 14 and 15 are listed in Table 18. The lists of experts involved in the preparation of Volumes 1–7 [2] and Volumes 8–11 [3] have been published previously.

Table 18. Members of the working groups who contributed to the preparation of monographs published in Volumes 12, 13, 14, and 15 of the IARC Monographs on the Evaluation of the Carcinogenic Risk of Chemicals to Man

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Dr A. Abbondandolo, Laboratory for Mutagenesis and Differentiation, C.N.R., Pisa, Italy
Dr L. Bahna, Cancer Research Institute, Slovak Academy of Sciences, Bratislava, Czechoslovakia
Dr R. R. Bates, US Food and Drug Administration, Washington, D.C.
Dr F. Berrino, National Institute for the Study and Treatment of Tumours, Milan, Italy
Dr G. Berry, Medical Research Council Pneumoconiosis Unit, Llandough Hospital, Penarth, Glamorgan, UK
Professor J. Bignon, Intercommunal Hospital Centre, Créteil, France
Professor E. Boyland, London School of Hygiene and Tropical Medicine, London
Dr I. Chernozemsky, Institute of Oncology, Medical Academy, Sofia
Dr G. J. van Esch, National Institute of Public Health, Bilthoven, The Netherlands
Professor L. Fiore-Donati, Institute of Anatomy and Pathological Histology, Borgo Roma Polyclinic, Verona, Italy
Dr L. Fishbein, National Cancer for Toxicological Research, Jefferson, Ark., USA
Dr R. Gingell, The Eppley Institute for Research in Cancer, Omaha, Nebr., USA
Dr P. Grasso, The British Industrial Biological Research Association, Carlshalton, Surrey, UK
Dr H. C. Grice, Health and Welfare Canada, Tunney's Pasture, Ottawa

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[1] International Agency for Research on Cancer (1973) *IARC Monographs on the Evaluation of Carcinogenic Risk of Chemicals to Man, Vol. 2, Some Inorganic and Organometallic Compounds*, Lyon, pp. 17–47.
[2] International Agency for Research on Cancer (1974) *Annual Report, 1974*, Lyon, pp. 70–73.
[3] International Agency for Research on Cancer (1976) *Annual Report, 1976*, Lyon, pp. 86–88.

Table 18. (continued)

Dr D. E. Hathway, Imperial Chemical Industries Ltd, Alderley Park, Cheshire, UK
Dr T. Hirayama, National Cancer Center Research Institute, Tokyo
Dr J. E. Huff[1], Oak Ridge National Laboratory, Oak Ridge, Tenn., USA
Dr P. F. Infante, National Institute for Occupational Safety and Health, Cincinnati, Ohio, USA
Professor A. M. Langer, Mount Sinai School of Medicine, New York, N.Y., USA
Dr R. A. Lemen, School of Public Health, University of Illinois, Chicago, Ill., USA
Dr W. Lijinsky, Frederick Cancer Research Center, Frederick, Md., USA
Professor D. B. Ludlum, Albany Medical College, Albany, N.Y., USA
Dr L. Massé, National School of Public Health, Rennes, France
Dr J. McCann, Department of Biochemistry, University of California, Berkeley, Cal., USA
Professor U. Mohr, Department of Experimental Pathology, Medical School, Hanover, Federal Republic of Germany
Dr S. D. Murphy, School of Public Health, Department of Physiology, Harvard University, Boston, Mass., USA
Professor D. Neubert, Institute of Toxicology and Embryonal-Pharmacology, The Free University of Berlin, Federal Republic of Germany
Dr V. B. Okulov, N. N. Petrov Research Institute of Oncology, Leningrad, USSR
Dr F. D. Pooley, Department of Mineral Exploitation, University College, Cardiff, UK
Professor R. Preussmann, Institute of Toxicology and Chemotherapy, German Cancer Research Centre, Heidelberg, Federal Republic of Germany
Professor G. Prodi, Institute of Cancerology, University of Bologna, Bologna, Italy
Dr D. P. Rall, National Institute of Environmental Health Sciences, Research Triangle Park, N.C., USA
Dr C. Ramel, University of Stockholm, Wallenberg Laboratory, Stockholm
Dr G. Saint-Ruf, C.N.R.S., Marcel Delépine Centre, Department of Organic-Biological Chemistry, Orléans, France
Professor C. Schlatter, Institute of Toxicology, Confederated Technical School and University of Zurich, Schwerzenbach, Switzerland
Professor H. W. Schlipköter, Medical Institute for Air Hygiene and Silicosis Research, University of Dusseldorf, Dusseldorf, Federal Republic of Germany
Professor I. J. Selikoff, Mount Sinai School of Medicine, New York, N.Y., USA
Dr V. F. Simmon, Stanford Research Institute, Menlo Park, Cal., USA
Professor A. Somogyi, Federal Health Agency, Berlin, Federal Republic of Germany
Dr B. Teichmann, Central Institute for Cancer Research, Academy of Sciences of the GDR, Berlin-Buch, German Democratic Republic
Dr B. Terracini, Institute of Anatomy and Pathological Histology, University of Turin, Turin, Italy
Dr V. S. Turusov, Cancer Research Center, USSR Academy of Medical Sciences, Moscow
Professor V. Vojvodic, Bulevar Reovlucije 257/III, Belgrade
Dr J. C. Wagner, Medical Research Council Pneumoconiosis Unit, Llandough Hospital, Penarth, Glamorgan, UK
Dr J. K. Wagoner, National Institute for Occupational Safety and Health, Cincinnati, Ohio, USA
Professor J. A. H. Waterhouse, Regional Cancer Registry, Birmingham, UK
Dr P. Westerholm, National Board of Occupational Safety and Health, Stockholm
Dr G. M. Williams, Naylor Dana Institute for Disease Prevention, Valhalla, N.Y., USA

[1] Present address: International Agency for Research on Cancer, Lyon, France.

Table 18. (continued)

Representatives from the National Cancer Institute

Dr J. A. Cooper[1], Deputy Associate Director, Carcinogenesis Program, Division of Cancer Cause and Prevention, National Cancer Institute, Bethesda, Md., USA
Dr S. Siegel, Coordinator, Technical Information Activities, Bioassay and Carcinogenesis Program, Division of Cancer Cause and Prevention, National Cancer Institute, Bethesda, Md., USA

Representatives from the Stanford Research Institute

Dr O. H. Johnson, Senior Industrial Economist, Chemical-Environmental Program, Stanford Research Institute, Menlo Park, Cal., USA
Dr K. E. McCaleb, Director, Chemical-Environmental Program, Stanford Research Institute, Menlo Park, Cal., USA
Dr V. von Schuller-Götzburg, Manager, Chemical Information Services — Europe, Stanford Research Institute, Zurich, Switzerland

Representatives from the World Health Organization

Dr C. Agthe, Chief, Food Additives Unit, WHO, Geneva, Switzerland
Dr J. F. Bertaux, Medical Officer, Pharmaceuticals, WHO, Geneva, Switzerland
Dr A. Davis, Chief, Schistosomiasis and Other Helminthic Infections, WHO, Geneva, Switzerland
Dr M. Vandekar, Medical Officer/Toxicologist, Division of Vector Biology and Control, WHO, Geneva, Switzerland

Representatives from the Commission of the European Communities

Dr W. J. Hunter, Commission of the European Communities, Health and Safety Directorate, Kirchberg/Luxembourg
Mrs M. T. van der Venne, Commission of the European Communities, Health and Safety Directorate, Kirchberg/Luxembourg

Observers

Dr B. Hildebrand, Toxicology Department, BASF Aktiengesellschaft, Ludwigshafen, Federal Republic of Germany
Dr R. J. Levine, Senior Medical Scientist, Center for Occupational and Environmental Safety and Health, Stanford Research Institute, Menlo Park, Cal., USA
Dr C. Levinson, International Federation of Chemical and General Workers' Unions, Geneva, Switzerland
Dr H. Ott, Commission of the European Communities, Environmental Research Programme, Brussels
Dr S. Villa-Trevino, Centre for Investigation and Advanced Studies, National Polytechnical Institute, Department of Cellular Biology, Mexico City

[1] Present address: International Agency for Research on Cancer, Lyon, France.

Three working groups have been planned for 1977–1978. Some aromatic amines which were not included in Volumes 1 or 4 will be considered in June 1977. In October 1977 some *N*-nitroso compounds and polychlorinated biphenyls that are known to occur in the environment will be evaluated or re-evaluated. In February 1978 a working group will consider some plastics and synthetic rubber compounds, including their monomers, polymers and copolymers. In June 1978 it is tentatively proposed to consider some organochlorine compounds and to up-date certain monographs already published in Volume 5 of the series.

The first 15 volumes of the series comprise 294 monographs, in which evaluations or re-evaluations were made of 336 compounds. Twenty-six chemicals or industrial processes have been associated with, or are highly suspected of being associated with, the induction of human cancer (Table 19). Although exposure to these chemicals is mainly occupational or medicinal, environmental exposure is also known to occur. Nine of the 26 compounds are or have until recently been used as drugs. For 157 of the 336 compounds, definite evidence exists of carcinogenicity in experimental animals, but no adequate epidemiological studies were available; however, for most of these chemicals, human exposure is known to occur. For the remaining 154 compounds, no evaluation could be made (Table 20). A review of the monograph programme was presented at the symposium on origins of human cancer which was held in Cold Spring Harbor, N.Y., USA in September 1976 [1].

The criteria used to evaluate the carcinogenic risk of chemicals to man were established in 1971 and have been applied in essence by all the working groups whose deliberations resulted in the 15 volumes so far published. In October 1977, a joint IARC/WHO *ad hoc* working group will meet to review these criteria and to revise the preamble to the IARC monographs, in which the criteria are set out as guidelines. The new preamble will represent the basic outline on which monographs will be prepared. The working group will also decide whether the content and format of the monographs should be changed as a consequence of the modifications to the preamble.

2.2 *Survey of chemicals being tested for carcinogenicity* (Mrs M. J. Ghess, Dr J. E. Huff, Dr H. Bartsch)

The aims of this project are to avoid unnecessary duplication of research, to increase communication between scientists working in this field, to make a census of available research facilities throughout the world and to ascertain the number of chemicals which are being tested.

Three surveys have been carried out by questionnaire, and six information bulletins have been issued since the project began. A fourth survey is in progress.

The first three surveys were taken and the six bulletins issued at intervals of 6–9 months, to obtain quickly and to catalogue worldwide information on long-term carcinogenicity testing. Now that the majority of on-going investigations are known, and since the time needed to complete long-term tests may be three years or more, future surveys will be carried out at intervals of 18–24 months.

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[1] Tomatis, L. (1977) In: *The Origins of Human Cancer*, Cold Spring Harbor (*Cold Spring Harbor Cell Proliferation Series* Vol. 4) (in press).

Table 19. Chemicals or industrial processes associated with cancer induction in man — target organs and main routes of exposure

Chemical or Industrial Process	Main Type of Exposure [1]	Target Organs Man	Main Route of Exposure [2]
1. Aflatoxins	Environmental, occupational [3]	Liver	Oral, inhalation [3]
2. 4-Aminobiphenyl	Occupational	Bladder	Inhalation, skin, oral
3. Arsenic compounds	Occupational, medicinal and environmental	Skin, lung, liver [3]	Inhalation, oral, skin
4. Asbestos	Occupational	Lung, pleural cavity, gastrointestinal tract	Inhalation, oral
5. Auramine (manufacture of)	Occupational	Bladder	Inhalation, skin, oral
6. Benzene	Occupational	Haemopoietic system	Inhalation, skin
7. Benzidine	Occupational	Bladder	Inhalation, skin, oral
8. Bis(chloromethyl)ether	Occupational	Lung	Inhalation
9. Cadmium using industries (possibly cadmium oxide)	Occupational	Prostate, lung [3]	Inhalation, oral
10. Chloramphenicol	Medicinal	Haemopoietic system	Oral, injection
11. Chloromethyl methyl ether (possibly associated with bis(chloromethyl)ether)	Occupational	Lung	Inhalation
12. Chromium (chromate producing industries)	Occupational	Lung, nasal cavities [3]	Inhalation
13. Cyclophosphamide	Medicinal	Bladder	Oral, injection
14. Diethylstilboestrol	Medicinal	Uterus, vagina	Oral
15. Haematite mining (?radon)	Occupational	Lung	Inhalation
16. Isopropyl oil	Occupational	Nasal cavity, larynx	Inhalation
17. Melphalan	Medicinal	Haemopoietic system	Oral, injection
18. Mustard gas	Occupational	Lung, larynx	Inhalation
19. 2-Naphthylamine	Occupational	Bladder	Inhalation, skin, oral
20. Nickel (nickel refining)	Occupational	Nasal cavity, lung	Inhalation
21. N,N-Bis(2-chloroethyl)-2-naphthylamine	Medicinal	Bladder	Oral
22. Oxymetholone	Medicinal	Liver	Oral
23. Phenacetin	Medicinal	Kidney	Oral
24. Phenytoin	Medicinal	Lympho-reticular tissues	Oral, injection
25. Soot, tars & oils	Occupational, environmental	Lung, skin (scrotum)	Inhalation, skin
26. Vinyl chloride	Occupational	Liver, brain [3], lung [3]	Inhalation, skin

[1] The main types of exposures mentioned are those by which the association has been demonstrated and exposures other than those mentioned may also occur
[2] The main routes of exposure given may not be the only ones by which such effects could occur
[3] Denotes indicative evidence

Table 20. Evaluations made by Working Groups for substances included in *IARC Monographs on the Evaluation of Carcinogenic Risk of Chemicals to Man*

Number of chemicals evaluated	336		
Chemicals carcinogenic to man		26	
Chemicals carcinogenic to experimental animals only		156	
Chemicals for which a final evaluation of carcinogenicity could not be made		154	
including: chemicals for which a suspicion of carcinogenicity was found			53
chemicals for which the available data were inadequate for evaluation			78
chemicals for which the available data did not indicate a carcinogenic effect			23

Bulletin No. 6 included up-dated information from the first five bulletins as well as more information on investigations being carried out in the USA.

As reported in the Annual Report 1976 [1], data gathered from the Autumn 1975 survey—reported in Bulletin No. 6 and issued in March 1976—gave information from 89 institutes (mainly non-commercial) in 19 countries on a total of 828 chemicals. For the first time, chemicals listed in this bulletin were crosslinked with studies reported in the *Directory of On-going Research in Cancer Epidemiology* [2].

Of the compounds being tested for carcinogenicity in experimental animals, 126 have also been considered by the working groups whose evaluations were published in Volumes 1–15 of the monograph series. For 49 of these chemicals under test, the data were insufficient to permit a final evaluation of carcinogenicity to be made by the working groups. Publication of the results of the studies in progress may permit future working groups to evaluate the carcinogenic risk of these chemicals.

Approximately 75% of the 828 chemicals under test are currently being manufactured or are known to occur naturally. The remaining 25% comprise laboratory curiosities, chemicals produced for experimental purposes only and chemical analogues.

Bulletin No. 7, which will result from the present survey, will be finalized early in 1978. Particular emphasis will be placed on up-dating information provided in Bulletin No. 6, with special attention to studies that have been completed and published.

2.3 *Carcinogenicity testing*

(*a*) *DDT*

(i) *Department of Experimental Oncology, University of Genoa, Italy*

Principal investigators: Professor L. Santi, Dr L. Rossi

The lifelong administration to rats of DDT mixed in the diet (dose–500 mg/kg body weight) results in the late appearance of liver tumours [3].

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[1] International Agency for Research on Cancer (1976) *Annual Report, 1976*, Lyon, p. 88.

[2] Muir, C. S. & Wagner, G., eds (1976) *Directory of On-going Research in Cancer Epidemiology*, 1976, Heidelberg, German Cancer Research Centre.

[3] Rossi, L., Ravera, M., Repetti, G. & Santi, L. (1977) *Int. J. Cancer*, **19**, 179–185.

(ii) *Department of Occupational Health, Hadassah Medical School, Jerusalem*

Principal investigator: Professor M. Wassermann

Following the results obtained in previous investigations on the levels of chlorinated pesticides and polychlorinated biphenyls in human tissues, the feasibility of an investigation on the possible long-term effect of PCBs in occupationally-exposed people is being considered.

(*b*) *Maleic hydrazide* (Dr V. Ponomarkov)

(i) Two experimental groups of C57BL mice were used. Maleic hydrazide was given either subcutaneously on days 1, 7, 14 and 21 at a total dose of 55 mg or orally at a weekly dose of 510 mg/kg body weight. The experiments are still in progress.

(ii) *National Institute of Public Health, Utrecht, The Netherlands*

Principal investigators: Dr G. van Esch, Dr R. Kroes

The long-term administration of maleic hydrazide to rats did not result in an increased incidence of tumours compared with that in the untreated controls.

(*c*) *Styrene* (Dr V. Ponomarkov)

Styrene was given to 0_{20} mice orally during their lifetime at weekly intervals (dose—1350 mg/kg body weight). The experiment is finished and the data are being analysed; preliminary evaluation indicates an increased incidence and earlier appearance of lung tumours compared with the controls. In a second experiment, weekly doses (500 mg/kg body weight) were given orally to BD-IV rats. This experiment is nearing completion. A third group, of C57BL mice, received weekly doses (300 mg/kg body weight) of styrene. The experiment is still in progress.

(*d*) *Styrene oxide* (Dr V. Ponomarkov)

Styrene oxide is given orally at weekly doses of 600 mg/kg body weight to 0_{20} mice. This experiment is nearing completion. Another experimental group was started recently in which BD-IV rats were given styrene oxide orally at weekly doses of 100 mg/kg body weight.

(*e*) *Vinylidene chloride* (Dr V. Ponomarkov)

Vinylidene chloride is given orally to BD-IV rats and C57BL mice at weekly doses of 150 mg/kg body weight and 70 mg/kg body weight, respectively. The experiment is still in progress.

(*f*) *2-Chlorobutadiene* (Dr V. Ponomarkov)

2-Chlorobutadiene is administered orally to BD-IV rats at weekly doses of 50 mg/kg body weight. The experiment is still in progress.

(*g*) *Phenobarbital* (Dr V. Ponomarkov)

The lifelong administration of phenobarbital to rats at doses of 500 ppm in drinking-water has resulted in the late appearance of liver tumours [1].

(*h*) *5-Bromodeoxyuridine* (Dr A. Likhachev, Dr V. Ponomarkov)

5-Bromodeoxyuridine is given intraperitoneally to BD-IV rats on days 1, 3, 7 and 21. The experiment is still in progress.

(*i*) *Magenta*

School of Medicine, Hanover, Federal Republic of Germany

Principal investigator: Professor U. Mohr

Syrian golden hamsters and Sprague-Dawley rats are given twice weekly oral doses of either commercial magenta or *p*-rosaniline for lifespan. The experiments are still in progress.

(*j*) N-*Phenyl-2-naphthylamine*

School of Medicine, Hanover, Federal Republic of Germany

Principal investigator: Professor U. Mohr

Syrian golden hamsters and Sprague-Dawley rats are given twice weekly oral doses of *N*-phenyl-2-naphthylamine for lifespan. The experiments are still in progress.

(*k*) *2-Chloroethylene oxide and 2-chloracetaldehyde*

National Institute of Health and Medical Research, Orsay, France

Principal investigator: Dr F. Zajdela

Repeated subcutaneous administration of chloroethylene oxide (the suspected ultimate carcinogen of vinyl chloride) in mice induced local tumours with a frequency comparable to bis(chloromethyl)ether when both compounds were tested at maximal tolerated toxic doses. 2-Chloroacetaldehyde was tested in a two-stage experiment by skin painting in mice. Subcutaneous administration proved to be impracticable due to strong necrotic activity. Both experiments are terminated.

(*l*) *Some pesticides*

National Institute of Public Health, Budapest

Principal investigator: Dr M. Börzsönyi

The long-term carcinogenicity testing of the following pesticides has been initiated: Benlate (butylcarbamoylmethyl-2-benzimidazol carbamate) alone or in combination with sodium nitrite; Ficam (2,2-dimethyl-1,3-benzodioxol-4-yl-*N*-methyl carbamate); Formetanate (3-dimethylaminomethyleneiminophenyl-*N*-methyl carbamate); Dodine (*N*-

[1] Rossi, L., Ravera, M., Repetti, G. & Santi, L. (1977) *Int. J. Cancer*, **19**, 179–185.

cyclododecyl-2,6-dimethyl morpholine); Tirofine (*N,N'*-1,4-piperazinediyl-bis-2,2,2-trichloroethylene-bis-formide) alone or in combination with sodium nitrite; Carbendazim (methyl-2-benzimidazol carbamate) alone or in combination with sodium nitrite. The experiments are still in progress.

3. EVALUATION OF THE SIGNIFICANCE FOR MAN OF DATA ON EXPERIMENTAL CHEMICAL CARCINOGENESIS AND OF THE DEVELOPMENT OF RAPID SCREENING TESTS

3.1 *Metabolism of carcinogens and DNA-repair studies*

(*a*) *The role of DNA-repair processes in the carcinogenicity of alkylating agents*

(Dr R. Montesano, Dr G. Margison, Mrs J. Margison, Dr A. Likhachev, Miss H. Brésil)

Data on the degree of alkylation and the persistence of various alkylated bases in the DNA of various organs of hamsters treated with [^{14}C]-*N*-nitrosodimethylamine (NDMA) have been published [1], as well as data on the levels of 0^6-methylguanine (0^6-meGua), 7-methylguanine (7-meGua) and other alkylated bases in liver DNA at various times after ^{3}H-*N*-nitrosomethylurea administration to rats pretreated or not with NDMA [2, 3]. Four main topics that continue this work have been investigated and are reported here.

(i) *DNA-excision repair capacity of hamster liver*

In certain experiments, assessment of DNA-excision repair capacity is partly based on the assumption that the relative amounts of alkylated bases produced *in vivo* are dependent only on the proximal alkylating species and not on the source of the DNA. Thus, reaction of *N*-nitrosomethylurea (NMU) with DNA *in vitro* produced an 0^6-meGua: 7-meGua ratio of 0.107. In tissues in which excision repair is low (e.g., rat brain), this ratio is achieved. For chemical reasons, the same ratio would be expected for NDMA, and again, in organs or under conditions where excision repair is slow, such a ratio is obtained. In other cases in which excision of 0^6-meGua is rapid (e.g., rat liver), the ratio of 0^6-meGua:7-meGua is less than 0.107, at even very short times after administration of NDMA or NMU, indicating that excision of 0^6-meGua is almost as rapid as its production. This is not so for 7-meGua which is not subject to an active excision process and can therefore be used as an indirect measure of the absolute amount of 0^6-meGua generated in the DNA (since the 0^6-meGua:7-meGua ratio is always 0.107). A dose of NDMA or NMU that produces the same amounts of 7-meGua thus generates the same total amount of 0^6-meGua. It is therefore interesting that two hours after administration of NMU (30 mg/kg), more than 80% of the 0^6-meGua produced had been excised from hamster liver DNA, whereas even seven hours after NDMA application (2.5 mg/kg producing approximately the same initial level of 7-meGua) only 10% of the 0^6-meGua had been excised. This suggests that identical DNA damage produced by different agents can be repaired at different rates, and further experiments are under way to explore this problem.

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[1] Margison, G. P., Margison, J. M. & Montesano, R. (1976) *Biochem. J.*, **157**, 627–634.

[2] Margison, G. P., Brésil, H., Margison, J. M. & Montesano, R. (1976) *Cancer Lett.*, **2**, 79–86.

[3] Montesano, R., Margison, G. P. & Likhachev, A. (1977) In: Farber, E., *et al.*, eds, *Physiopathology of Carcinogenesis in the Digestive Organs*, Tokyo, University of Tokyo Press and Baltimore, University Park Press (in press).

(ii) *Accumulation of 0^6-meGua in target and non-target organ DNA* [1]

Chronic administration of NDMA to BD-IV rats induces tumours of the liver but not of the kidney or lung. BD-IV rats were treated with ^{14}C-NDMA (2 mg/kg/day) by gavage on weekdays and sacrificed at 2, 4, 8, 16 and 24 weeks after the beginning of the treatment and in each case 72 hours after the last NDMA administration. The amounts of 7-meGua and 0^6-meGua in liver, lung and kidney DNA were then determined.

Although a large amount of 7-meGua was present in liver DNA, no 0^6-meGua was detected throughout the period of the experiment, indicating that, at least under present experimental conditions, chronic treatment with NDMA does not lead to a specific accumulation of 0^6-meGua in target organ DNA. However, 0^6-meGua did accumulate in the DNA of kidney and lung, organs in which tumours are not induced during chronic treatment with NDMA. This is clearly seen (Fig. 24) in the 0^6-meGua:7-meGua ratios, which had increased in kidney and lung DNA after only four weeks of treatment. The greatest increase in this ratio was between 16 and 24 weeks, when values five to six times greater than those detected at four weeks were obtained.

After a single dose of 2.5 mg/kg NDMA, 0^6-meGua was lost from kidney DNA at a rate similar to that observed with liver (half-life, slightly less than 20 hours). However, following chronic treatment, 0^6-meGua was detected in the DNA of the kidney (and lung) indicating that, in contrast to the liver, accumulation of this base had occurred. This may have been due to a reduction in the level of the 0^6-meGua excision enzymes or a loss of their activity by exhaustion or overloading.

These results are consistent with the previous observations that chronic administration of NDMA has no effect on the rate of removal of 0^6-meGua from liver DNA [2].

(iii) *Methylation of DNA in various tissues of rats by 1,2-dimethylhydrazine (1,2-DMH)* [3]

1,2-DMH induces tumours predominantly of the intestinal tract in rats and other animal species. Single subcutaneous doses of 1,2-DMH to rats resulted in tumours of the colon and kidney. Within the intestinal tract, the tumours developed mostly in the colon and, less frequently, in the duodenum or rectum. In the present studies, we have examined the formation of 7-meGua and the degree of alkylation at sites other than the 7-position in the DNA of various tissues of rats following a single subcutaneous administration of 300 mg/kg of ^{3}H-1,2-DMH.

The highest level of 7-meGua was found in the DNA of the liver three hours after administration of the carcinogen. 7-meGua was also detected in the DNA of the mucosa of the colon (14% of the liver value), the kidney (3.2%) and the mucosa of the ileum (1.7%). By 72 hours, the level of 7-meGua had decreased in the DNA of all the organs. At 3 hours, 0^6-meGua was detected in the DNA of the liver and, to a lesser extent, in the mucosal DNA of the colon. However, at 72 hours, 0^6-meGua was found only in the mucosal DNA of the colon and at the same level as that observed at 3 hours.

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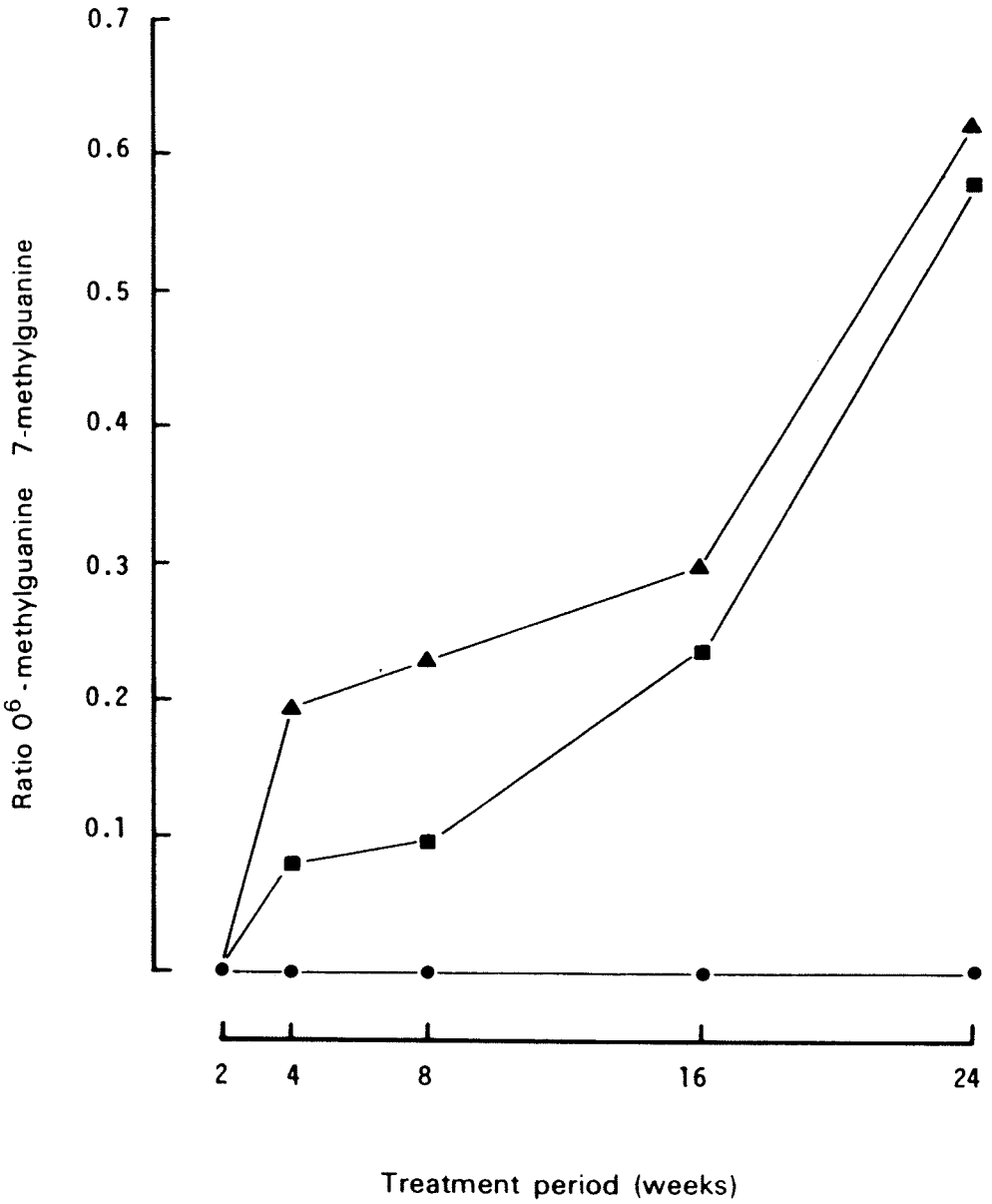
[1] Margison, G. P., Margison, J. M. & Montesano, R. (1977) *Biochem. J.*, **165**, 463–468.
[2] Margison, G. P., Brésil, H., Margison, J. M. & Montesano, R. (1976) *Cancer Lett.*, **2**, 79–86.
[3] Likhachev, A. J., Margison, G. P. & Montesano, R. (1977) *Chem.-biol. Interactions*, **18**, 235–240.

Fig. 24 O^6-meGua: 7-meGua ratio in rat tissues DNA during chronic oral administration of ^{14}C-NDMA (0.492 mCi/mol; 2 mg/kg/day on weekdays). Liver (●), lung (■), kidney (▲).



The large differences in the relative persistence of O^6-meGua in these tissues is reflected in the O^6-meGua:7-meGua ratios (see Table 21). These ratios are probably more accurate than the individual measurement of alkylated bases, since they do not depend on the actual

Table 21. 7-meGua and O^6-meGua in the DNA of the liver and colon 3 hr after subcutaneous injection of ^{3}H-1,2-dimethylhydrazine (300 mg/kg)

Tissue	*dpm/μmol guanine* 7-meGua	O^6-meGua	O^6-meGua / 7-meGua
Liver	4224	57	0.014
Colon	586	33	0.056

amount of the parent base present in DNA nor on the possible variations occurring during the extraction and/or analysis of the DNA samples.

These results confirm and extend the earlier results of Hawks & Magee [1] that demonstrated methylation of nucleic acids by 1,2-DMH in the intestinal tract of rats and show, in addition, that 0^6-meGua seems to persist preferentially in the DNA of organs such as the colon, in which tumours are induced.

(*b*) *Metabolism of vinyl chloride* (Dr H. Bartsch, Mr C. Malaveille, Dr R. Montesano, Dr T. Kuroki, Mr A. Barbin and Miss H. Brésil, in collaboration with Professor N. Loprieno, Laboratory of Genetics, University of Pisa, Pisa, Italy; Professor H. F. Stich, Cancer Research Centre, University of British Columbia, Vancouver, Canada; Dr F. Zajdela, Radium Institute, Faculty of Science, Orsay, France; Dr A. Croisy and Dr P. Jacquignon, Institute of Chemistry of Natural Substances, Gif-sur-Yvette, France; and Dr E. Huberman, Oak Ridge National Laboratory, Oak Ridge, Tenn., USA)

The various adverse biological effects of vinyl chloride, a recognized carcinogen in animals and man, appear to be dependent upon its enzymic conversion to chemically reactive metabolites [2, 3]. Since liver microsome-mediated formation from vinyl chloride *in vitro* of two chemically reactive products, chloroethylene oxide and 2-chloroacetaldehyde, has been reported, the biological activity of these two compounds and of monochloroacetic acid, an identified urinary metabolite in rats and in man, and of a putative metabolic intermediate, 2-chloroethanol, were compared in the following short-term tests for the detection of potential carcinogens:

(i) assay for electrophilic reactivity with 4-(4-nitrobenzyl)pyridine;

(ii) tissue-mediated mutagenicity tests with *Salmonella typhimurium* strains;

(iii) tissue-mediated mutagenicity assay with *Schizosaccharomyces pombe* and *Saccharomyces cerevisiae*;

(iv) induction of ouabain- and azaguanine-resistant mutations in Chinese hamster V79 cells;

(v) induction of DNA-repair synthesis, measured by unscheduled incorporation of tritiated thymidine into cultured human skin fibroblasts; and

(vi) host-mediated assay in mice with *S. pombe*.

Chloroethylene oxide was a most active compound in the assay systems (i)–(vi), and it also induced local tumours in mice upon repeated subcutaneous administration (Zajdela & Croisy, unpublished data). These results strongly support the hypothesis that this epoxide is one of the principal mutagenic and carcinogenic intermediates formed from vinyl chloride by mammalian metabolism. This study also permitted a comparison of the sensitivities of the various assays in the detection of chloroethylene oxide [4, 5, 6].

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[1] Hawks, A. & Magee, P. N. (1974) *Brit. J. Cancer*, **30**, 440–447.

[2] Bartsch, H. & Montesano, R. (1975) *Mutation Res.*, **32**, 93–114.

[3] Bartsch, H., Malaveille, C., Barbin, A., Brésil, H., Tomatis, L. & Montesano, R. (1977) *Environm. Hlth Persp.*, **17**, 193–198.

[4] Bartsch, H. & Loprieno, N. (1977) In: Böhme, H. & Schöneich, J., eds *Proceedings of the 6th EEMS meeting, Gernrode, German Democratic Republic*, Abhandlungen der Akademie der Wissenschaften der DDR, No. 9, pp. 161–165.

[5] Loprieno, N., Barale, R., Baroncelli, S., Bartsch, H., Bronzetti, G., Cammellini, A., Corsi, C., Frezza, D., Nieri, R., Leporini, C., Rossellini, D. & Rossi, A. M. (1976) *Cancer Res.*, **36**, 253–257.

[6] Huberman, E., Bartsch, H. & Sachs, L. (1975) *Int. J. Cancer*, **16**, 639–644.

(*c*) *Studies on mechanisms of carcinogen activation and on factors involved in the organ-specific carcinogenesis of chemicals* (Mr C. Malaveille, Dr H. Bartsch, Dr T. Kuroki, Dr G. P. Margison, Mr A. Barbin, Mrs G. Brun, Miss A. M. Camus, Mrs J. M. Margison, in collaboration with Dr P. L. Grover and Dr P. Sims, Chester Beatty Research Institute, London; Dr G. Kolar and Dr M. Wiessler, German Cancer Research Centre, Heidelberg, Federal Republic of Germany)

The role of organ-specific, enzymic release of alkylating intermediates in determining in which tissue a tumour develops in response to some *N*-nitrosamines or to 3,3-dimethyl-1-phenyltriazene has been investigated, using synthetic putative or identified metabolites and *in vitro* mutagenicity tests and studies of the binding of radioisotope-labelled metabolites to nucleic acids in target and non-target tissues in rats. As shown with these systemically-acting carcinogens, specific tissue activation to ultimate reactive intermediates seems to be a prerequisite of, but not always a final determinant of, the organ-specific carcinogenicity of the parent compound. For instance, 3,3-dimethyl-1-phenyltriazene appears to require metabolic activation in the liver to form a transported alkylating intermediate, but different rates of removal of miscoding bases generated in DNA of target and non-target organs are apparently among the factors that determine organ specificity [1, 2].

Tissue-mediated mutagenicity tests with *S. typhimurium* or Chinese hamster V79 cells have also been used in studies on the mechanism of metabolic activation of polycyclic aromatic hydrocarbons, to help pin-point which particular dihydro-diol derivatives are biologically reactive precursors of vicinal diol-epoxides [3, 4]. The results obtained on a series of dihydro-diols derived from 7-methylbenz(*a*)anthracene strongly support the hypothesis that a vicinal diol-epoxide, 3,4-diol-1,2-oxide, of 7-methylbenz(*a*)anthracene may be one biologically important metabolite of the parent hydrocarbon *in vivo* [5].

(*d*) *Carcinogen metabolism by human tissues* [6] (Mrs N. Sabadie, Dr H. Bartsch, Mr C. Malaveille, Miss A. M. Camus, Mrs G. Brun, in collaboration with Dr H. B. Richter-Reichhelm and Professor U. Mohr, School of Medicine, Hanover, Federal Republic of Germany, Professor W. Schindler, Lung Clinic, Hanover, Federal Republic of Germany and Dr A. H. Conney, Department of Biochemistry and Drug Metabolism, Hoffmann-La-Roche, Nutley, N.J., USA)

It has been suggested that the differences in susceptibility between individuals exposed to the same level of environmental carcinogens may arise in part from differences in metabolic activation in the processes of detoxification of chemical carcinogens. To test this hypothesis, the metabolism of vinyl chloride, *N*-nitrosomorpholine, *N*-nitroso-*N'*-methylpiperazine and benzo(*a*)pyrene were measured in adult liver and lung tissue, using mutagenicity tests with *S. typhimurium* strains or spectrofluorometric benzo(*a*)pyrene-hydroxylase determinations. When hepatic benzo(*a*)pyrene-hydroxylase activity in biopsies from

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[1] Bartsch, H., Margison, G. P., Malaveille, C., Camus, A. M., Brun, G., Margison, J. M., Kolar, G. F. & Wiessler, M. (1977) *Arch. Toxicol.* (in press).

[2] Camus, A. M., Wiessler, M., Malaveille, C. & Bartsch, H. (1977) *Mutation Res.* (in press).

[3] Malaveille, C., Kuroki, T., Sims, P., Grover, P. L. & Bartsch, H. (1977) *Mutation Res.*, **44**, 313–326.

[4] Kuroki, T. (1978) In: Tso, P. O. P. & Gelboin, H. V., eds, *Polycyclic Hydrocarbons and Cancer*, New York, Academic Press (in press).

[5] Malaveille, C., Tierney, B., Grover, P. L., Sims, P. & Bartsch, H. (1977) *Biochem. biophys. Res. Commun.*, **75**, 427–433.

[6] The tissues were obtained from biopsy or surgical samples taken for diagnostic or therapeutic purposes.

adult human subjects was plotted against the liver microsome-mediated mutagenicity, a statistically significant positive correlation was obtained between the rate of oxidative benzo(*a*)pyrene metabolism and mutagenic effects of *N*-nitrosomorpholine, *N*-nitroso-*N'*-methylpiperazine and vinyl chloride[1].

Benzo(*a*)pyrene-hydroxylase activity was measured in normal and neoplastic lung tissue from more than forty patients. Assay conditions were such that the increase of phenolic metabolites was proportional to the protein concentration and the incubation time. More than ten-fold differences between individuals were found in the ability of liver and lung tissues to metabolize carcinogens. The results suggest that *N*-nitrosomorpholine, *N*-nitroso-*N'*-methylpiperazine and benzo(*a*)pyrene are metabolized by enzyme systems that are under similar regulatory control.

3.2 *Chemical carcinogenesis and mutagenicity in cultured cells* (Dr R. Montesano, Dr T. Kuroki, Miss C. Drevon and Mrs L. Saint Vincent)

(*a*) *Rat liver epithelial cells*

Various criteria have been used to differentiate normal cells from cells transformed neoplastically *in vitro*. These criteria have been developed mainly from studies with cells of mesenchymal origin, and it is questionable to what extent they are also applicable to epithelial cells. In collaboration with Dr K. Sanford (National Cancer Institute, Bethesda, Md., USA) and Dr B. Weinstein (Columbia University College of Physicians and Surgeons, New York, N.Y., USA), various epithelial cell lines, developed in the Agency laboratory, have been evaluated for their capacity to form colonies in soft agar, for their production of plasminogen activator and for morphological changes *in vitro*[2]. The results have been correlated with the tumorigenic capacity of the same cell lines after injection into syngeneic hosts. The data given in Table 22 show that growth in soft agar and some cytological changes correlated well with tumorigenicity *in vivo*. However, the production of plasminogen activator showed little correlation with tumorigenicity. The present data also provide evidence that plasminogen activator production is neither a necessary nor a sufficient property for growth in agar. It is possible that protease production is linked to the mechanism of metastasis formation; however, the cell cultures IAR6-1, IAR6-1-RT7 and IAR-27, which produced a high yield of metastatic lesions in the lungs when injected into rats, have a low *in vitro* production of plasminogen activator (Table 22).

(*b*) *Chinese hamster V79 cells*

Microsome-mediated mammalian cell mutagenesis has been established using V79 Chinese hamster cells[3]. The cells, grown in monolayer, were treated with the chemicals in the presence of a postmitochondrial fraction (S15) from rat liver and a NADPH-generating system for 1–15 hr, depending on the chemical. The cells were then washed and incubated in fresh culture medium for 2–3 hr and plated for cytotoxicity and mutagenicity tests.

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[1] Sabadie, N., Camus, A., Malaveille, C., Richter-Reichhelm, H. B., Mohr, U. & Bartsch, H. (1977) *Proc. Amer. Assoc. Cancer Res.*, **18**, 37.

[2] Montesano, R., Drevon, C., Kuroki, T., Saint Vincent, L., Handleman, S., Sanford, K. K., DeFeo, D. & Weinstein, I. B. (1977) *J. nat. Cancer Inst.* (in press).

[3] Kuroki, T., Drevon, C. & Montesano, R. (1977) *Cancer Res.*, **37**, 1044–1050.

Table 22. Properties of tumorigenic and non-tumorigenic rat liver epithelial cell lines [a]

Cell culture (week in culture when tested)	Initial cytological diagnosis	Growth in agar	Fibrinolytic activity [b]	
			6 hr	18 hr
Tumorigenic [c]				
IAR6 (36 wks)	+	ND	ND	
(49-52 wks)	+	+ [e]	4	18
IAR6-1 (52-63 wks)	+	+	0	26
IAR6-1-RT7 (80 wks)	+	+	11	34
IAR2-28 (48-49 wks	+	+	23	61
(91-103 wks)	+	+	17	44
IAR20-PC1-3 (30-46 wks)	+	+	30	47
IAR22 (46 wks)	—	+	ND	
(50-66 wks)	+	+	19	50
IAR27 (53-74 wks)	+	+	5	28
W8 (~17 wks)	+	+	0	0
HTC (>12 yrs)	+	+	34	70
Non-tumorigenic				
IAR6 (20 wks)	—	—	3	26
IAR20 (20 wks)	ND	—	25	42
(32-43 wks)	—	—	25	47
IAR20-PC1 (25-31 wks)	—	—	10	29
IAR20-PC1-3 (22-25 wks)	—	—	ND	
IAR27 (28-34 wks)	± [d]	—	23	47
K16 (19 wks)	—	—	0	0
Undetermined state				
K22 (15 wks)	—	—	ND	
W22 (16 wks)	—	—	19	50
W22-AAF (16 wks)	—	—	ND	

[a] Montesano, R., Drevon, C., Kuroki, T., Saint Vincent, L., Handleman, S., Sanford, K. K., Deteo, D. & Weinstein, I. B. (1977) *J. nat. Cancer Inst.* (in press).

[b] % lysis ^{125}I-fibrin by intact cells. In all cases assays were also done in the presence of cells but in the absence of plasminogen, and negligeable fibrinolysis was obtained, indicating that the cells are elaborating a plasminogen activator. Cell line IAR20-PC1 MNNG (40 wks) was the only exception and it appears to be elaborating a plasminogen-independent protease.

[c] The cell cultures were considered tumorigenic from the time they produced tumors after inoculation into syngeneic rats.

[d] On first examination, this coverslip preparation was diagnosed (+) but on reexamination was diagnosed (—).

[e] This cell culture gives positive result in Lyon but negative in New York; for the other cultures the same results were obtained in both places. ND-not determined.

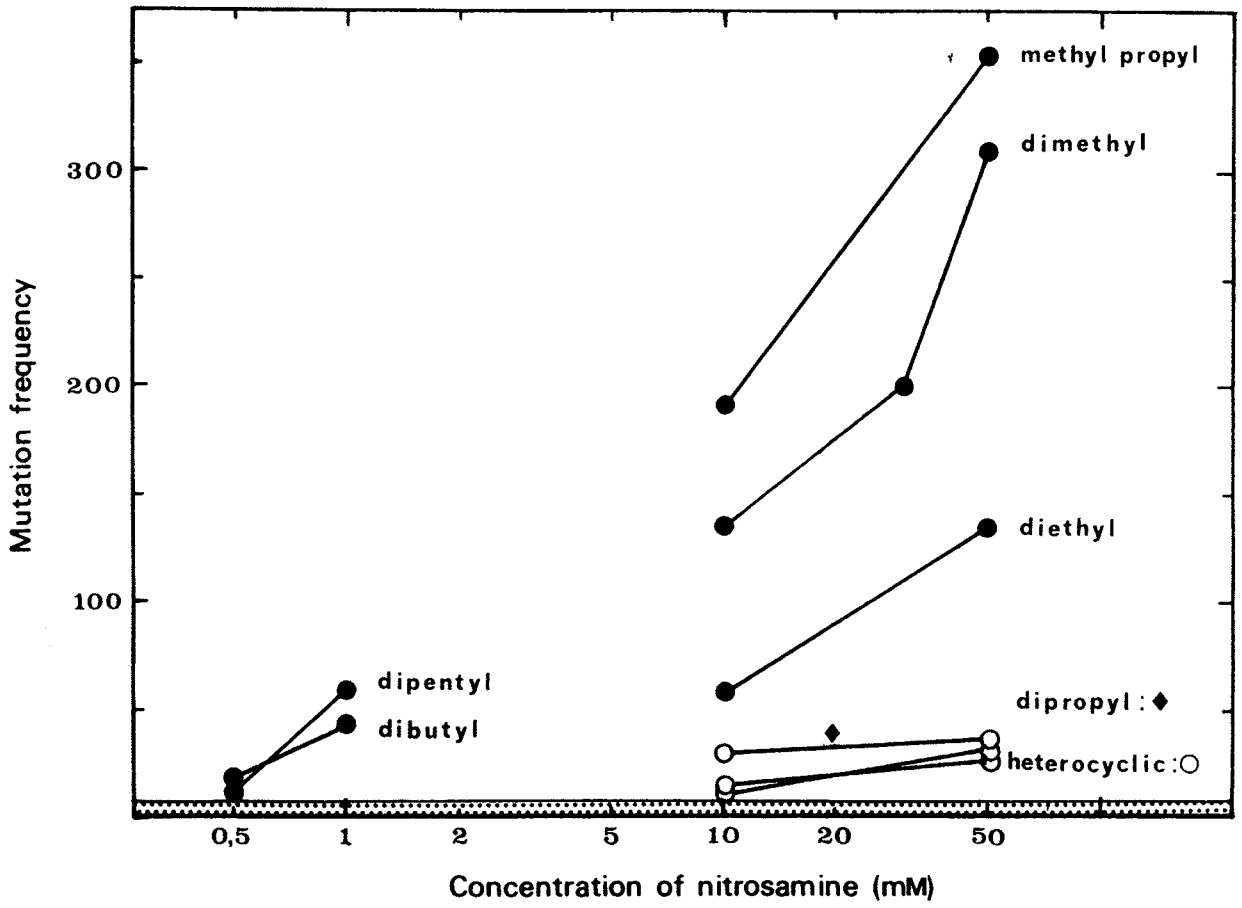
Mutagenesis was determined both by resistance to 8-azaguanine, a purine analogue, and by resistance to ouabain, a specific inhibitor of the Na^+-K^+-activated ATPase of plasma membrane. In this assay system, the S15 fraction and cofactors alone were not toxic to the V79 cells after up to 15 hr of incubation.

N-Nitrosodimethylamine (NDMA) could induce mutagenicity and toxicity in a dose-related fashion in the presence of a microsomal fraction and cofactors; this effect was not observed when cofactors necessary for the mixed-function oxidases were omitted from the incubation mixture. Modulators of drug-metabolizing enzymes affected the microsome-mediated mutagenicity of NDMA. Phenobarbital pretreatment of the rats from which the liver fraction was prepared led to an approximately two-fold increase in the mutation rate compared with tissues from untreated rats with concentrations of NDMA from 2-50 mM, while methylcholanthrene pretreatment resulted in an increase in the mutation frequency with a higher concentration of NDMA (50 mM). The reduction in the muta-

genicity of NDMA observed with liver fraction from animals pretreated with aminoacetonitrile was consistent with the protective effect of this drug against the acute liver toxicity and carcinogenicity of NDMA observed in rats.

Various other nitrosamines were assayed for their mutagenicity in the microsome-mediated system. The results are shown in Fig. 25 and Table 23, in which mutagenicity in the V79 cells is compared with data obtained in *S. typhimurium*. Of the nine carcinogenic nitrosamines tested, all were mutagenic to the V79 Chinese hamster cells, with the exception of *N*-nitrosomethylphenylamine; this compound was not mutagenic in *S. typhimurium* (Table 23) or *E. coli*. Two non-carcinogenic nitrosamines, *N*-nitrosodiphenylamine and *N*-nitrosomethyl-*tert*-butylamine, had no mutagenic effect, either with V79 cells or with *S. typhimurium* (Table 23).

Fig. 25 Mutagenicity of various *N*-nitrosamines for Chinese hamster V79 cells in the presence of post-mitochondrial fraction from rat liver. Mutation frequency is expressed as the number of 8-azaguanine-resistant mutants per 10^5 survivors. Heterocyclic *N*-nitrosamines include the derivatives of morpholine, pyrrolidine and methyl-piperazine.



A series of chemicals are being tested in the microsome-mediated mutagenesis system [1]. Vinyl chloride monomer, a human carcinogen, was found to be mutagenic to the V79 cells in the presence of microsomal fraction and cofactors, while polycyclic hydrocarbons,

[1] Drevon, C., Kuroki, T. & Montesano, R. (1977) In: *Proceedings of the International Mutagenesis Conference, July 1977* (in press).

such as benzo(*a*)pyrene, were only slightly mutagenic in the microsome-mediated system. These chemicals were, however, moderately mutagenic when tested in a cell-mediated system, in which the V79 cells were co-cultured with metabolically competent but lethally-irradiated rat embryo cells. This suggested that, at least in the case of hydrocarbons, mutagenicity assay systems incorporating a liver microsomal fraction may not simulate the metabolic pathway that occurs in intact cells. A series of chemicals are now being tested in both microsome- and cell-mediated assay systems, and the use of liver parenchymal cells for cell-mediated systems is being considered.

Table 23. Mutagenicity of various nitrosamines in the presence of liver fraction from phenobarbital-pretreated rats

N-Nitrosamine	Mutagenic effect in:	
	S. typhimurium	Chinese hamster cells
Carcinogenic		
N-Nitrosodimethylamine	+	+
N-Nitrosodiethylamine	+	+
N-Nitrosodi-*n*-propylamine	+	+
N-Nitrosomethyl-*n*-propylamine	+	+
N-Nitrosodi-*n*-butylamine	+	+
N-Nitrosodi-*n*-pentylamine	+	+
N-Nitrosomethylphenylamine	—	—
N-Nitrosomorpholine	+	+
N-Nitrosopyrrolidine	+	+
N-Nitroso-*N'*-methylpiperazine	+	+
Noncarcinogenic		
N-Nitrosodiphenylamine	—	—
N-Nitrosomethyl-*tert*-butylamine	—	—

(*c*) *Karyotyping of tumour cells*

First Institute of Pathology, Semmelweis Medical University, Budapest

Principal investigator: Professor K. Lapis

Several primary cultures of nervous tissue tumours, in particular of double primary tumours, induced by *N*-nitrosoethylurea in BD-VI rats have been initiated, with the purpose of verifying the karyotype of tumour cells. Various cell lines from human embryos and from mouse embryo lung were established in culture, and the following characteristics were examined in relation to the tumorigenicity of these cells: growth pattern *in vitro*; changes in morphology, karyotype and chromatin patterns; agglutinability by concanavalin A; and immunological changes.

3.3 *Detection of potential chemical carcinogens in short-term tests* (Dr H. Bartsch, Mr C. Malaveille, Dr T. Kuroki, Miss A. M. Camus, Miss G. Planche, Mr A. Barbin, Mrs G. Brun)

(*a*) *Mutagenicity testing*

Pure chemicals or complex mixtures have been tested in an *in vitro* assay system using *S. typhimurium* strains developed by Professor B. N. Ames (University of California, Berkeley, Calif., USA), in the presence of a microsomal fraction from liver or other organs

from rodents and from human biopsy specimens [1]. Three types of assays were used: plate incorporation assays [2], a liquid incubation system [3] and the plate incorporation assay adapted to test volatile compounds [4].

Special attention has been given (i) to improving the efficiency and reproducibility of the *Salmonella*/microsome mutagenicity test [5, 6, 7]; (ii) to characterizing ultimate reactive metabolites which appear to be involved in carcinogenesis and mutagenesis, to identify *in vivo* possible biological reactive intermediates which cannot be isolated [8]; and (iii) to testing chemicals of socio-economic importance, which have not yet been assayed for their carcinogenic activity. Several halo-olefins have been found to be mutagenic [9, 10].

The 150 compounds that have been tested for mutagenic activity in the *Salmonella*/microsome mutagenicity test with its adapted procedures include drugs, industrial chemicals, mycotoxins, pesticides and halo-olefins. Sixty-six compounds for which evidence of carcinogenicity exists in the literature have been found to be mutagenic. Fourteen compounds which are described as carcinogenic were not detected as mutagens. Only one false-positive result was obtained—a 'non-carcinogen' which showed mutagenic activity. Four chemicals for which there was negative evidence of carcinogenicity were also not mutagenic. Of 84 compounds on which no data on carcinogenicity testing have been published, 35 were non-mutagenic and 49 mutagenic.

Mutagenicity tests were also carried out on complex mixtures, e.g., residues obtained after extracting bread, wheat and tea samples and extracts of the carbonized residue from opium pipes collected in the oesophageal cancer study in Iran (see page 137). Among these mixtures, only extracts of the residues formed in the interior of opium smokers' pipes gave consistently positive results in the mutagenicity tests when assayed in the presence of a rat liver activation system and *S. typhimurium* strains TA100 and TA98.

(*b*) *Comparative electrophilicity, mutagenicity, DNA repair induction activity and carcinogenicity of some* N- *and* O-*acyl derivatives of* N-*hydroxy-2-aminofluorene* (Dr H. Bartsch, Mr C. Malaveille, Mr A. Barbin, Dr T. Kuroki, in collaboration with Professor H. F. Stich, Cancer Research Centre, University of British Columbia, Vancouver, Canada; Professor E. C. Miller and Professor J. A. Miller, McArdle Laboratory for Cancer Research, University of Wisconsin Medical Center, Madison, Wis., USA)

The activity of carcinogenic, chemically-reactive esters derived from *N*-hydroxy-2-aminofluorene, namely *N*-myristoyloxy-*N*-acetyl-2-amino-fluorene, *N*-acetoxy-*N*-myristoyl-2-aminofluorene, *N*-myristoyloxy-*N*-myristoyl-2-aminofluorene and *N*-acetoxy-*N*-

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[1] The tissues were obtained from biopsy or surgical samples taken for diagnostic or therapeutic purposes.
[2] Ames, B. N., Durston, W. E., Yamasaki, E. & Lee, F. D. (1973) *Proc. nat. Acad. Sci. (Wash.)*, **70**, 2281–2285.
[3] Bartsch, H., Malaveille, C. & Montesano, R. (1975) *Cancer Res.*, **35**, 644–651.
[4] Bartsch, H., Malaveille, C. & Montesano, R. (1975) *Int. J. Cancer*, **15**, 429–437.
[5] Malaveille, C., Planche, G. & Bartsch, H. (1977) *Chem.-biol. Interactions*, **17**, 129–136.
[6] Bartsch, H., Camus, A. M. & Malaveille, C. (1976) *Mutation Res.*, **37**, 149–162.
[7] Bartsch, H. (1977) In: *Proceedings of the Meeting of the Scientific Committee, Carlo Erba Foundation, Occupational and Environmental Health Section*, pp. 105–116.
[8] Planche, G., Croisy, A., Malaveille, C. & Bartsch, H. (submitted for publication).
[9] Barbin, A., Planche, G., Croisy, A., Malaveille, C. & Bartsch, H. (1977) In: *Abstracts, Second International Conference on Environmental Mutagens, Edinburgh.*
[10] Bartsch, H., Malaveille, C., Barbin, A., Planche, G. & Montesano, R. (1976) *Proc. Amer. Assoc. Cancer Res.*, **17**, 17.

acetyl-2-aminofluorene were compared qualitatively and quantitatively in three short-term tests that are currently used for the detection of potential carcinogens: assay for electrophilicity, mutagenicity in bacteria or V79 Chinese hamster cells and induction of unscheduled DNA repair synthesis in cultured fibroblasts.

The data showed a general qualitative correspondence between the induction of DNA repair synthesis, electrophilicity and carcinogenic activity of these esters, although quantitative differences were evident. However, correlation of these activities with mutagenicity (tested with *S. typhimurium* TA1538 and TA98) was poor; no mutagenicity was demonstrable for the two *N*-myristoyloxy derivatives. These findings underline the need for carrying out multiple short-term assays in predicting the potential carcinogenic activity of chemicals [1].

(*c*) *Screening for adverse biological effects of pesticides and of an antischistosomal drug within a WHO evaluation and testing programme* (Dr H. Bartsch, Mr C. Malaveille, Mrs G. Brun, Dr E. de la Peña, Dr T. Kuroki, in collaboration with Professor N. Loprieno and Dr A. Abondandolo, Laboratory of Genetics, Pisa, Italy; Dr E. Vogel, Department of Radiation Genetics and Chemical Mutagenesis, State University of Leiden, The Netherlands; Dr M. Vandekar, Division of Vector Biology and Control and Dr A. Davis, Division of Malaria and other Parasitic Diseases, WHO, Geneva, Switzerland)

Several pesticides are being tested in the *Salmonella*/microsome mutagenicity assay, and Embay 8440, a new compound with antischistosomal activity, is being evaluated for its possible adverse biological effects in the following systems: *Salmonella*/microsome mutagenicity assay; gene conversions and forward mutations in yeast; host-mediated assay in mice, using yeast as genetic indicator; mutation induction in Chinese hamster V79 cells, scoring for 6-thioguanine, 8-azaguanine or ouabain resistance; induction of unscheduled DNA repair synthesis in cultured human cells; and induction of X-linked recessive lethal mutations in germ cells of male *Drosophila melanogaster*.

3.4 *Research training*

(*a*) *In the Agency*

Dr E. de la Peña, Institute of Soil Science, Section for Growth Protection, C.S.I.C., Madrid, received training on the *Salmonella*/microsome mutagenicity test system during a four-month period under a fellowship from the Ministry of Education and Science, Madrid. Subsequently he was involved in the testing of several pesticides for their mutagenic activity within the framework of a WHO insecticide evaluation and testing programme.

Dr A. Y. Likhachev, from the Professor N. N. Petrov Research Institute of Oncology, Leningrad, USSR, was a recipient of an IARC Research Training Fellowship, and, during the year spent at the Agency, received training in the techniques of isolation of nucleic acids and of alkylated DNA bases. The studies in which he was involved are described on pages 98.

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[1] Bartsch, H., Malaveille, C., Stich, H. F., Miller, F. C. & Miller, J. A. (1977) *Cancer Res.*, 37, 1461–1467.

Dr E. Matos, from the Angel H. Roffo Oncology Institute, University of Buenos Aires, and Dr J. A. Castro, from the Laboratory of Biotoxicological Chemistry, Ministry of Defence, Buenos Aires, received an International Cancer Research Technology Transfer fellowship from the UICC and spent a period of one month in the Unit in order to become acquainted with the mutagenesis assay in Chinese hamster cells and the procedures of separation of DNA alkylated bases by liquid chromatography.

(*b*) *Training course on* in vitro *mutagenicity testing*

A training course on the *Salmonella*/microsome mutagenicity test was organized in April 1977 in conjunction with Dr M. Roberfroid, Dr M. Mercier and Dr F. Poncelet from the University of Louvain, Laboratory of Medical Chemistry, Brussels.

3.5 *Workshop on* in vitro *mutagenicity testing*

A workshop was organized by Professor H. Greim and Dr I. E. Mattern in March 1977 at the Institute for Radiation and Environmental Research at Neuherberg, Munich, Federal Republic of Germany, and co-sponsored by the Agency, the European Environmental Mutagen Society and the Commission of the European Communities. The main aim of this workshop, which was attended by 30 participants from eight European countries and Canada, was to formulate the standard protocol for carrying out bacterial *in vitro* mutagenicity tests.

4. PRENATAL CARCINOGENESIS

4.1 *Experimental studies*

The administration of *N*-nitrosoethylurea to pregnant BD-VI rats resulted in a high incidence of nervous tissue tumours and the occurrence of kidney tumours in F_1 descendants. Tumours of the nervous tissue were also observed in F_3 descendants, thus confirming the persistence of an increased cancer risk in subsequent generations [1]. This experiment is being repeated (Dr A. Likhachev, Dr V. Ponomarkov).

School of Medicine, Hanover, Federal Republic of Germany

Principal investigator: Professor U. Mohr

The study of the effects of administration of 7,12-dimethylbenzanthracene to pregnant C57BL mice on three subsequent generations is still in progress.

Cancer Research Center, Academy of Medical Sciences of the USSR, Moscow

Principal investigator: Dr V. S. Turusov

The study of the effect of the administration of *N*-nitrosomethylurea to pregnant rats on four subsequent generations is in progress.

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[1] Tomatis, L., Ponomarkov, V. & Turusov, V. (1977) *Int. J. Cancer*, **19**, 240–248.

4.2 *Prenatal events in childhood cancer*

N. N. Petrov Research Institute of Oncology, Leningrad, USSR

Principal investigator: Professor N. P. Napalkov

Prenatal exposure of BALB/c mice to *N*-nitrosoethylurea resulted in a high incidence of tumours in F_1 descendants, and an increased and earlier appearance of tumours was observed in F_2 descendants. Postnatal X-ray irradiation (150 roentgens) increased the number of lung tumours, mainly adenocarcinomas (in more than 80% of females); on the other hand, X-ray irradiation without transplacental pretreatment with *N*-nitroscethylurea decreased the lung tumour incidence as compared with controls. At the same time, transplacental treatment with the nitrosamine prevented development of ovarian tumours in mice subsequently irradiated in postnatal life. It was also found that F_2 descendants, i.e., those exposed to *N*-nitrosoethylurea only before conception, had no enhanced sensitivity to repeated exposure to the carcinogen, contrary to the effect that is usually observed in transplacentally treated F_1 animals. A dose-response relationship was observed in combined prenatal treatment with *N*-nitrosoethylurea and postnatal irradiation[1, 2, 3].

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[1] Napalkov, N. P., Tomatis, L., Likhachev, A. Y. & Kolodin, V. I. (1977) *Vop. Onkol.*, **23**, 3, 41–51.
[2] Napalkov, N. P., Tomatis, L., Likhachev, A. Y. & Kolodin, V. I. (1977) *Vop. Onkol.*, **23**, 4, 49–57.
[3] Napalkov, N. P., Tomatis, L., Likhachev, A. Y. & Kolodin, V. I. (1977) *Vop. Onkol.*, **23**, 5, 66–70.

5. UNIT OF RESEARCH TRAINING AND LIAISON

Dr W. DAVIS (Chief)

1. INTRODUCTION

As announced in last year's Annual Report, the programme of travel fellowships has been suspended. In contrast, however, an increased number of research training fellowships have been awarded. Two courses have been organized, and five new titles have been added to the *IARC Scientific Publications* series.

2. THE FELLOWSHIPS SELECTION COMMITTEE

The annual meeting of the Fellowships Selection Committee took place in Lyon from 2–3 May 1977. Following the decision of the Committee at its meeting in 1976, the closing date for receipt of applications was brought forward to 31 January 1977, which greatly facilitated the work of the unit in preparing the applications for review and in arranging interviews of as many candidates as possible.

The members of the Committee were unanimous in their opinion that the quality of the applications this year was higher than ever before and were gratified to see the increasing number of applications in the fields of priority accorded by the Agency.

The members of the Committee were:

Professor C. Heidelberger, Director for Basic Research, Los Angeles County Comprehensive Cancer Center, Los Angeles, Calif., USA (*Chemical Carcinogenesis*)

Professor N. P. Napalkov, Director, N. N. Petrov Research Institute of Oncology, Leningrad, USSR (*Chemical Carcinogenesis*)

Professor N. F. Stanley, Department of Microbiology, University of Western Australia, Perth, Australia (*Microbiology*)

Professor H. Sugano, Director, Cancer Institute (Japanese Foundation for Cancer Research), Tokyo (*Experimental Pathology*)

Professor M. Bagshaw was unable to attend, due to recent illness.

3. RESEARCH TRAINING FELLOWSHIPS

As a result of savings made in the Agency's budget, an extra US$115 000 was made available to the fellowships programme, which meant that the Committee was able to recommend the award of 25 fellowships this year. Sixty applications were reviewed by the Committee. The distribution by scientific discipline is given in Table 24.

Table 24. Distribution of Research Training Fellowships by scientific discipline, 1977

Scientific discipline	No. of fellowships
Biochemistry	3
Cell biology	1
Cytogenetics	1
Environmental carcinogenesis	3
Epidemiology	5
Experimental carcinogenesis	1
Immunology	4
Immunopathology	1
Molecular biology	3
Virology	3

4. CORVISSIANO FELLOWSHIPS

The second Corvissiano fellow, Dr T. Kuroki, from Tokyo, has been working at the Agency since August 1976. His work, in the Unit of Chemical Carcinogenesis, has been of great importance in the development of rapid screening tests for environmental carcinogens and in the establishment of an international network of collaborating centres in this field.

Dr Francesca Repetto, of the Department of Health of the Lombardy Region, Italy, will come to Lyon as a Corvissiano fellow for one year as of October 1977. She will work under the supervision of Dr R. Saracci, and one of the aims of her stay will be the preparation of an atlas of cancer mortality in the countries of the European Economic Community.

5. SPECIALIZED COURSES

5.1 *Epidemiology of cancer*

Thirty-seven participants from 23 different countries attended the Agency's sixth cancer epidemiology course held in Lyon from 30 August to 10 September 1976. Dr P. Cole (Department of Epidemiology, Harvard University, Cambridge, Mass., USA) was responsible for the coordination of the programme, and was most ably assisted by Professor E. D. Acheson (University of Southampton, UK) and the late Professor D. D. Reid (London School of Hygiene and Tropical Medicine, London). Important contributions were also made by Dr L. J. Kinlen, University of Oxford, UK, and Professor C. Zippin, University of California, San Francisco, Calif., USA. Members of the Agency's Unit of Epidemiology and Biostatistics also participated actively in the teaching programme.

Professor Cole's notes on 'history and conceptual problems in cancer epidemiology'. which were distributed to all the participants, greatly enhanced the effectiveness of the afternoon lectures on methodology.

The first course organized by the Agency in Latin America took place in Brasilia from 29 November to 4 December 1976, in collaboration with the National Cancer Division of Brazil (Director, Dr H. Torloni). Thirty-two participants (22 from Brazil and 10 from other Latin American countries) took part in the course, which was coordinated by Dr P. Correa (Louisiana State University Medical Center, New Orleans, La., USA). The course,

which was conducted in Spanish, included lectures by Dr Nubia Muñoz (Interdisciplinary Programme and International Liaison Unit) and by a former IARC staff member, Dr N. Breslow, University of Washington, Seattle, Wash., USA (Fig. 26).

Fig. 26 Participants at the Latin-American course on the epidemiology of cancer, Brasilia, 29 November–4 December 1976.

Another regional epidemiology course will be organized this year in collaboration with the Jinnah Postgraduate Medical Centre, Karachi, Pakistan (Director, Professor N. A. Jafarey) and with the WHO Regional Office for the Eastern Mediterranean. The course, entitled 'Epidemiology of Chronic Diseases with Emphasis on Cancer', will take place from 22 October to 3 November in Karachi. Professor I. Kessler, Department of Epidemiology, The John Hopkins University, Baltimore, Md., USA, will be course coordinator, and Dr H. Tunstall-Pedoe, Department of Epidemiology, St Mary's Hospital, London, will lecture on the epidemiology of cardiovascular diseases, which are of particular interest in the Eastern Mediterranean Region. Dr C. S. Muir, Dr R. MacLennan and Dr J. Kmet will complete the teaching faculty. It is hoped that the participants will be representative of as many countries as possible in the region.

5.2 *Chemical carcinogenesis*

Professors Elizabeth and J. Miller (McArdle Laboratory for Cancer Research, University of Wisconsin, Madison, Wisc., USA) will be responsible for the coordination of the programme of the Agency's first course on chemical carcinogenesis to be held in Lyon from 14–19 November 1977.

5.3 *Use of nonhuman primates in cancer research*

In collaboration with Academician B. A. Lapin, Director of the Institute of Experimental Pathology and Therapy, Suchumi, Georgian SSR, the Agency is planning to organize a course on the utilization of nonhuman primates in cancer research, to be held in Suchumi in 1978.

6. PUBLICATIONS

Five new titles have appeared in the *IARC Scientific Publications* series: *Environmental Pollution and Carcinogenic Risks* (joint publication with the French National Institute of Health and Medical Research); *Pathology of Tumours in Laboratory Animals*, Volume I, *The Rat*, Part 2; *Environmental* N-*Nitroso Compounds - Analysis and Formation; Cancer Incidence in Five Continents*, Volume III; and *Air Pollution and Cancer in Man*. Four more are in press—*Directory of On-Going Research in Cancer Epidemiology*, Volume II (joint publication with the German Cancer Research Centre); *Cancer Registration and its Techniques; Proceedings of the Symposium on Nasopharyngeal Carcinoma, Kyoto, 1977*; and *Selected Methods of Analysis for Environmental Carcinogens*, Volume I, *Nitrosamines*. The list of publications in the series is given in Table 25.

Table 25. List of IARC Scientific Publications

No.	Title	Year of publication
1	*Liver Cancer*	1971
2	*Oncogenesis and Herpesviruses*	1972
3	N-*Nitroso Compounds — Analysis and Formation*	1972
4	*Transplacental Carcinogenesis*	1973
5	*Pathology of Tumours in Laboratory Animals*, Vol. 1: *The Rat*, Part 1	1973
6	*Pathology of Tumours in Laboratory Animals*, Vol. 1: *The Rat*, Part 2	1976
7	*Host Environment Interactions in the Etiology of Cancer in Man*	1973
8	*Biological Effects of Asbestos*	1973
9	N-*Nitroso Compounds in the Environment*	1974
10	*Chemical Carcinogenesis Essays*	1974
11	*Oncogenesis and Herpesviruses II*, Parts 1 and 2	1975
12	*Screening Tests in Chemical Carcinogenesis*	1976
13	*Environmental Pollution and Carcinogenic Risks*	1976 [a]
14	*Environmental* N-*Nitroso Compounds — Analysis and Formation*	1976
15	*Cancer Incidence in Five Continents*, Vol. III	1976
16	*Air Pollution and Cancer in Man*	1977
17	*Directory of Ongoing Research in Cancer Epidemiology*, Vol. II	1977 [b]
	Cancer Registration and its Techniques	1977 [c]
	Proceedings of Symposium on Nasopharyngeal Carcinoma, Kyoto, 1977	1977 [c]
	Selected Methods of Analysis for Environmental Carcinogens, Vol. 1: *Nitrosamines*	1977 [c]
	Proceedings of the Third International Symposium on Oncogenesis and Herpesviruses	1977 [d]
	Proceedings of the Fifth Meeting on the Analysis and Formation of N-*Nitroso Compounds*	1978 [d]
	Proceedings of the Symposium on Carcinogenic Risks — Strategies for Intervention	1978 [a, d]
	Pathology of Tumours in Laboratory Animals, Vol. 2, *The Mouse*	1978 [d]
	Pathology of Tumours in Laboratory Animals, Vol. 3, *The Hamster*	1978 [d]

[a] joint publication with INSERM
[b] joint publication with DKFZ
[c] in press
[d] in preparation

Table 26 shows the up-to-date figures for the distribution and sales of the scientific publications and monographs.

Table 26. Distribution of *IARC Scientific Publications* and *Monographs on the Evaluation of Carcinogenic Risk of Chemicals to Man*

	Official distribution	Sales
Scientific Publications		
No. 1	673	810
2	780	1347
3	933	813
4	905	786
5	1008	1056
6	866	750
7	1004	652
8	1003	931
9	961	831
10	983	897
11 — Part 1	1067	572
11 — Part 2	1068	577
12	1107	922
13	866	600
14	920	556
15	842	796
16	946	569
17	1950	525
Monographs Series		
Vol. 1	2640	2035
2	1837	1824
3	1903	1740
4	1661	1559
5	1888	1393
6	1663	1426
7	1977	1294
8	1878	1091
9	1881	1055
10	1902	1221
11	2027	766
12	1919	1068
13	1834	663
14	1900	1110
15	1737	613

7. SYMPOSIA

One of the conclusions of the symposium on environmental pollution and carcinogenic risks held at the Agency from 3–5 November 1975 was the need for definition of criteria on which to base decisions for regulatory action against carcinogenic hazard. In an attempt to fulfill this need, the Agency is organizing a second symposium, again in collaboration with the French National Institute for Health and Medical Research, entitled 'Carcinogenic Risks—Strategies for Intervention', which will take place at the Agency from 30 November to 2 December 1977, under the patronage of Madame Simone Veil, French Minister of Health.

8. COORDINATING COMMITTEE FOR HUMAN TUMOUR INVESTIGATION

The Seventh International Symposium on the Biological Characterization of Human Tumours took place in Budapest from 13–15 April 1977. Dr G. Aczel, Hungarian Deputy Minister of Health, and Professor I. Zoltan, President of the Federation of Hungarian Medical Societies, opened the proceedings.

At its meeting immediately after the symposium, the Coordinating Committee decided unanimously to accept the invitation from the Hellenic Society for Chemotherapy and the National Hellenic Research Foundation to hold the eighth symposium in Athens in 1979.

6. INTERDISCIPLINARY PROGRAMME AND INTERNATIONAL LIAISON UNIT

Dr C. A. LINSELL (Chief)

1. INTRODUCTION

The collection phase of the programme for banking material for cell-mediated immunity studies has been concluded, although specimens from those patients whose clinical course is being followed in East Africa and India will continue to be banked. The bank will be used mainly for studies to assess whether host factors in populations with differing cancer patterns and from widely different environments can be compared.

The preservation techniques used in Africa have been verified, and tests of parameters of immunity are being undertaken in laboratories in Switzerland, the USA and the UK, as well as in the collaborating centres in Kenya and India.

Techniques in the field of cell-mediated immunology are developing rapidly, and their correlation with the clinical condition of the patient needs to be established. The preserved material which is banked and clinical data relating to it will permit early application of the latest techniques.

The liver cancer programme, in which the association with the ingestion of aflatoxin was demonstrated, has progressed to the stage of active intervention. It is now proposed, with the assistance of the Food and Agriculture Organization of the United Nations (FAO) and the United Nations Environment Programme (UNEP), to carry out a field intervention study to provide the methodology for global preventive measures. Attempts will be made to reduce aflatoxin levels in food both by improving agricultural methods and by better storage. To investigate another possible factor in the etiology of liver cancer, a cohort of hepatitis B-virus carriers is being established to assess their risk for that cancer.

Liaison activities with Headquarters and regional offices of WHO, and with the International Union Against Cancer continue. Official liaison has been established with UNEP, which has indicated that environmental carcinogenesis is one of its major priorities.

2. IMMUNOLOGY

2.1 *Tumour-associated antigens* (Dr P. Sizaret)

(*a*) *Experimental model: tumour-specific antigens in rat liver cells transformed* in vitro *by chemical carcinogens* (Dr T. Yokota, in collaboration with the Unit of Chemical Carcinogenesis)

A neo-antigen was shown to be present in a liver-cell line of BD rat origin transformed by *N*-nitrosodimethylamine, and three other neo-antigens were present in BD rat liver-cell lines transformed spontaneously or by *N*-nitrosodimethylamine or *N*-nitrosomethylgua-

nidine. None of these antigens was present on normal BD adult rat liver or spleen cells nor on Wistar rat liver or intestinal carcinoma cells. The antigens were not found in 10-, 15- and 19-day-old rat fetuses, nor were corresponding antibodies found in sera of multiparous pregnant rats; this confirms that these are not fetal antigens.

(*b*) *β1-Specific pregnancy glycoprotein (B1 SP1): comparison with other tumour markers for the diagnosis and management of trophoblastic tumours* (Miss N. Martel)

Preliminary results of the collaborative retrospective study confirmed that B1 SP1 is indeed associated with trophoblastic tumours. Although there were several cases in which chorionic gonadotrophin hormone (HCG) was elevated and the B1 SP1 normal, there were also rare cases in which HCG was normal and B1 SP1 elevated.

An international prospective study of B1 SP1, HCG and the pregnancy-associated α2 glycoprotein (PAAG), for which an association with certain types of cancer has also been reported, has been initiated in collaboration with Professor K. D. Bagshawe (Charing Cross Hospital, London), Dr H. Bohn (Behringwerke Institute, Marburg, Federal Republic of Germany), Dr R. M. Lequin (Catholic University, Nijmegen, The Netherlands) and Professor Y. S. Tatarinov (2nd Moscow Medical Institute, Moscow). The assays include 133 serial plasma specimens provided by Professor Bagshawe and 80 specimens from Professor N. N. Trapeznikov (Cancer Research Center, Moscow). Reagents for the assay of the B1 SP1 and the PAAG were provided by Behringwerke AG.

(*c*) *Preparation of a reference material for the assay of the B1 SP1*

Official liaison was established with the standardization committee of the International Union of Immunological Societies, and Dr Sizaret was elected chairman of the committee for the standardization of pregnancy-associated proteins.

The criteria for the preparation of reference materials for the B1 SP1 and PAAG were determined after consultation with eight experts: Professor K. D. Bagshawe, Dr B. H. Berne (Meloy Laboratories, Springfield, USA), Dr H. Bohn, Dr M. G. Damber (Umeå University, Umeå, Sweden), Dr R. M. Lequin, Dr W. H. Stimson (University of Strathclyde, Glasgow, UK), Dr G. N. Than (Obstetrical and Gynaecological Clinic of the University, Pécs, Hungary) and Professor Y. S. Tatarinov.

To provide material for a standard preparation, six litres of sera from pregnant women were collected in collaboration with Dr A. Mestrallet (Minguettes Polyclinic, Vénissieux, France), Dr J. C. Couturier (Rillieux Polyclinic, Rillieux, France), Dr J. Reboul d'Aubigny (Saint Augustin Clinic, Lyon, France), Dr A. Schmid (Tonkin Clinic, Villeurbanne, France) and Professor A. Notter and Dr R. Garmier (Hôtel-Dieu, Lyon, France). The National Institute of Health of The Netherlands is assisting the Agency in the preparation of the standard. A pilot study to check the stability of the standard is under way with the help of Dr J. H. de Bruijn of the National Institute of Health of The Netherlands, Bilthoven.

(d) Reference material for the assay of α-fetoprotein (AFP) by sensitive techniques

Since the association of AFP with liver-cell cancer was first established by Professor Y. S. Tatarinov in 1963, methods for the detection of this oncofetal protein have become more sensitive, and radioimmunoassay or immunoenzymoassay are now commonly used. Therefore, a reference reagent, assayed against the WHO AFP standard, which is more suitable for these techniques is being prepared in collaboration with Dr T. W. G. Smith and Dr S. G. Anderson of the National Institute for Biological Standards and Control, UK. The cord sera for this reference material were provided by Professor P. Magnin and Miss J. Chabre, Edouard Herriot Hospital, Lyon, France.

2.2 *Storage and preservation of biological material and host factor studies* (Dr A. G. Levin, IARC Research Centre, London)

Emphasis has been placed on the collection and preservation of serial specimens from cancer patients. In order to relate *in vitro* immunological tests, which are subject to many variables, to a patient's clinical status and prognosis, it is useful to be able to examine serial specimens under the same experimental conditions. The serial specimens of lymphocytes (each of which is usually divided into four aliquots) for some cancers that are currently preserved and available in the bank are shown in Table 27.

Table 27. Cryopreserved lymphocyte specimens available from patients with selected diagnoses

	Total no. of patients	Length of time followed 0-3 mths	4-6 mths	7-12 mths	1-2 years	2 or more years
Nasopharyngeal carcinoma	258	203	25	14	14	2
Burkitt's lymphoma	132	86	24	16	5	1
Breast carcinoma	66	63	1	2	—	—
Cervical carcinoma	64	43	12	8	1	—
Oesophageal carcinoma	61	60	1	—	—	—
Malignant melanoma	20	20	—	—	—	—
Sarcoma	20	19	—	—	1	—
Hodgkins' disease	17	16	—	—	1	—
Hepatocellular carcinoma	10	10	—	—	—	—

Among the individuals whose cells and sera are banked are 159 patients from whom tumour tissue is also preserved. Specimens have been collected from seven families in the Shirati area of Tanzania in which one or more Burkitt's lymphoma patients have been detected and from six control families in the same area. Similar specimens are available from six families of nasopharyngeal carcinoma patients in Kenya.

Clinical and epidemiological data are computer-stored and linked to data on the biological specimens, mainly cryopreserved lymphocytes and sera, in collaboration with Professor M. Healy and Mr F. Rich (Computing and Statistics Division of the Clinical Research Centre, London) and Dr N. E. Day (Unit of Epidemiology and Biostatistics).

(*a*) *Lymphocyte stimulation and HLA testing* (in collaboration with Dr S. Knight and Mr P. Hall, Division of Surgical Sciences, Clinical Research Centre, London and Dr C. M. Steel, Medical Research Council Clinical and Population Cytogenetics Unit, Edinburgh, UK)

Laboratory testing of cryopreserved cells has stressed lymphocyte stimulation and HLA typing. Satisfactory responses to both mitogens and allogenic cells were obtained with cryopreserved lymphocytes which had been stored for more than three years [1]. Micro techniques have been employed which will allow stimulation of an aliquot of cryopreserved lymphocytes by several cell lines or mitogens in the same experiment, as well as a determination of the number of T-cells by rosette testing. These studies are primarily designed to test lymphocyte reactivity to cellular antigens, and for this purpose lymphoblastoid cell lines which can be characterized immunogenetically are employed. A fluorochromasia technique has been utilized so that cryopreserved cells can be reacted against standard HLA antisera and serum from parous females from East Africa, particularly from the Shirati area of Tanzania. This tissue-types individuals, whose cells will be studied by other techniques to investigate possible new immunogenetic specificities.

(*b*) *HLA testing* (Drs W. & J. Bodmer, Department of Biochemistry, University of Oxford, UK, and Dr C. Entwistle, National Tissue Typing Centre, Bristol, UK)

Sera from parous females in an area endemic for Burkitt's lymphoma were tested against lymphocytes of known HLA type from Caucasian donors. The reactions are currently being compared with tests of these sera against cells from East African subjects with and without Burkitt's lymphoma, in order to detect new immunogenetic specificities.

(*c*) *Antibody-dependent lymphocytotoxicity to Epstein–Barr virus antibodies* (Dr G. Pearson, Department of Microbiology, Mayo Clinic, Rochester, Minn., USA)

Preliminary studies in individuals whose cells and sera are preserved in the bank have demonstrated antibody-dependent lymphocytotoxicity titres in a group of nasopharyngeal carcinoma and Burkitt's lymphoma patients. It has also been confirmed that the IgA fraction of anti-viral capsid antigen is higher in nasopharyngeal carcinoma patients, as compared with Burkitt's lymphoma and other cancer patients. The relation of these findings to clinical status and prognosis is under study.

(*d*) *Antigen-antibody complexes* (Dr P. H. Lambert, WHO Immunology Research Laboratory, University of Geneva, Geneva, Switzerland)

The presence of antigen-antibody complexes may be relevant to the prognosis of patients with leukaemia, and complexes of this type were found in patients with nasopharyngeal carcinoma from East Africa, using the Clq binding technique. Sera from these patients were contributed to a WHO-sponsored study to evaluate techniques for assaying antigen-antibody complexes. Although the Clq binding technique may be suitable, it seems likely that more than one technique will be necessary to determine the nature of complexes present.

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[1] Steel, C. M., Ennis, M., Levin, A. G. & Wasunna, A. (1977) *Cytobios* (in press).

(*e*) *Carcinoembryonic antigen, hepatitis B virus and α-fetoprotein studies* (Professor A. M. Neville, Chester Beatty Research Institute, Institute of Cancer Research, London; Dr D. Dane, Department of Virology, Middlesex Hospital Medical School, London; and Dr P. Sizaret)

The study included a group of rural subjects, representative of the East African population, and Nairobi blood donors, a high socio-economic group. Both groups showed significantly increased levels of carcinoembryonic antigens and α-fetoprotein, as well as increased frequency of hepatitis B virus antigen and antibody when compared with European reference levels.

(*f*) *Comparative cellular and serum studies on malignant melanoma patients in East Africa and the USA* (Dr S. Golub, Department of Surgery, University of California, Los Angeles, Calif., USA)

Cryopreserved material from melanoma patients from East Africa will be tested using melanoma-specific antigenic material. This study will compare the immunological responsiveness of East African patients with those in the USA.

(*g*) *Macrophage function in an animal tumour system* (in collaboration with Dr W. Butler, St George's Hospital Medical School, London, and Dr J. Soothill, Institute of Child Health, London)

The relation of macrophage function to carcinogenesis is being studied by measuring the clearance of iodinated polyvinyl pyrrolidone from *N*-nitrosamine-induced rat tumours.

2.3 *Immunological studies on breast cancer* (Dr D. Jussawalla, Director, Tata Memorial Centre, Parel, Bombay, India)

Cryopreserved lymphocytes collected serially from breast cancer patients, over 20 of whom have been followed for more than one year, are being tested by stimulation with mitogens and T- and B-cell rosettes in an effort to relate *in vitro* reactivity to the patient's clinical condition and treatment.

In a second study with cryopreserved material, the HLA pattern of Parsee breast cancer patients and their families is being investigated. Twenty-eight families of such patients have been interviewed, and tissue-typing studies are in progress in collaboration with the Indian Council for Medical Research Blood Group Laboratory, Bombay.

3. PRENATAL EVENTS AND CHILDHOOD CANCER (Dr N. Muñoz, Dr L. Tomatis, Dr N. E. Day, in collaboration with Professor K. H. Degenhardt, Institute for Human Genetics, Frankfurt/Main, Federal Republic of Germany; Dr C. Rumeau-Rouquette, National Centre for Scientific Research, Villejuif, France; Dr N. Wald, Department of the Regius Professor of Medicine, Radcliffe Infirmary, Oxford, UK; Dr J. F. Bithell, Childhood Cancer Research Group, University of Oxford, Oxford, UK; Professor E. V. Kuenssberg, Royal College of General Practitioners, Edinburgh, UK; Professor L. Saxén, Department of Pathology, University of Helsinki, Helsinki; Dr G. Fara, Department of Hygiene, Faculty of Medicine and Surgery, University of Milan, Milan, Italy; and Dr A. Pacsa, Institute of Microbiology, University Medical School, Pécs, Hungary)

This project uses material from established prospective studies designed to assess the role of prenatal events on the incidence of congenital malformations. The pregnancy records of the 14 mothers whose children developed cancer were reviewed. Five of the 14 mothers received oestrogen-progesterone preparations during the first trimester; two were treated with valium during the third and fourth months of pregnancy for periods of 1–2 months. Although serum samples had been collected from 7 of the 14 mothers, only one sample now remained. The record linkage to identify more children with cancer continues only in the UK, the Federal Republic of Germany and Finland. A new cohort of 5 000 pregnancies, each with 2–4 prenatal serum samples, from Pécs, Hungary, has been added to this collaborative study, which when complete, will have investigated more than 10 000 pregnancies.

Although prenatal exposure to drugs can be studied in all cases, the role of prenatal viral infections on childhood cancer can only be studied in the cases from Oxford and Pécs, where adequate prenatal serum samples have been collected.

3.1 *Congenital cancer* (Dr N. Muñoz, in collaboration with Dr L. M. Kinnier-Wilson, Marie Curie Memorial Foundation, The Chart, Oxted, Surrey, UK)

A total of 126 malignant tumours that caused death at birth or during the first 28 days of life have been abstracted from the Oxford Childhood Cancer Registry for the period 1953 to 1972. Neuroblastoma, leukaemia, Wilms' tumour, sarcoma and teratoma accounted for 84% of these tumours. The prenatal records were analysed of the mothers of these children and of the mothers of control children matched by sex and date of birth. Influenza, rubella, toxaemia, epilepsy, use of sedatives and anticonvulsants, and exposure to diagnostic irradiation during pregnancy were reported more often by the mothers of the children with cancer than by the control mothers. Congenital defects were also reported more often among children with cancer and among their siblings than among control children and their siblings.

4. LIVER CANCER (Dr C. A. Linsell, Dr N. Muñoz)

4.1 *Aflatoxin intervention study* (in collaboration with the Food Additives Unit, WHO, Geneva, Switzerland, the Food Policy and Nutrition Division, FAO, Rome, and UNEP)

A correlation between liver cancer risk and aflatoxin exposure has been demonstrated in some countries of Africa and Asia. Since individual exposure to aflatoxin cannot be measured, the ultimate test of a causal association between aflatoxin and liver cancer has to be derived from intervention studies. Swaziland was selected for a feasibility study in March 1977 since:

(*a*) Previous IARC studies have established baselines of aflatoxin contamination in different regions of the country.

(*b*) It is a relatively small country with approximately half a million inhabitants, a good infrastructure of health and agricultural services, and limited imports of basic staple foods.

(*c*) National harvest improvement schemes have been established.

(*d*) Cancer registration on a country-wide basis is possible.

The immediate objective of this study will be to reduce the aflatoxin level observed in the lowlands of Swaziland, where a high frequency of liver cancer has been reported, to the levels found in the highlands, where there is a lower liver cancer frequency. The long-term effect on the trend of liver cancer rates will be followed. To prove the specificity of the effect of aflatoxin contamination, monitoring of other factors also associated with liver cancer, such as hepatitis B viral infection, will be required. A joint proposal by WHO/FAO/IARC/UNEP has been made to the Swaziland authorities.

4.2 *Cohort study on hepatitis B virus and liver cancer* (Professor Phoon Wai On, Department of Social Medicine and Public Health, University of Singapore, Singapore)

A cohort study to determine the risk of developing liver cancer among chronic carriers of hepatitis B virus has been planned. A Chinese population in Singapore has been selected because of the high prevalence of hepatitis B viral infection, the high incidence of liver cancer and the existence of a cancer registry which will allow the determination of the liver cancer risk by record linkage after an appropriate follow-up period. This will avoid the need for individual and direct follow-up. A profile will be established of hepatitis B viral infection by measuring the hepatitis B surface, core and 'e' antigens in 5 000 Chinese males over 45 years of age. Twenty-two liver cancer cases are to be expected at the end of a five-year period of study of this population. The Department of Social Medicine and Public Health, University of Singapore, provides medical surveillance of institutions that care for old people in Singapore and welcomes the support and information derived from this project.

4.3 *Experimental studies*

The combined effect *in vitro* of chronic persistent infection with hepatitis B virus and exposure to aflatoxin is being studied to complement the field studies. Collaboration has been established between the gastroenterology departments of the All-India Institute of Medical Sciences, New Delhi, and of Athens University, Athens, and the WHO Reference Laboratory for Viral Hepatitis, London School of Hygiene and Tropical Medicine, London.

4.4 *Hepatitis B markers in liver cancer* (Professor D. Trichopoulos, Department of Hygiene and Epidemiology, University of Athens Medical School, Athens; Dr E. Tabor, Bureau of Biologics, Food and Drug Administration, Bethesda, Md., USA; and Dr P. Sizaret)

Sera from 46 patients with primary liver cancer and from 82 sex- and age-matched controls from Athens have been analysed for various markers of hepatitis B virus. A higher frequency of hepatitis Bs antigen (HBsAg) and anti-hepatitis Bc (markers of chronic persistent infection) and a lower frequency of anti-HBsAg were found among patients with liver cancer than among controls (Table 28). The IARC laboratory has assessed AFP levels in all sera of this study. Sera from 50 more patients with primary liver cancer, from 40 patients with secondary liver cancer, and from sex- and age-matched controls are being tested.

Table 28. Prevalence of hepatitis B markers in hepatoma patients and matched controls, Athens, 1976-77

Groups	Total no.	HBsAg and anti-HBc %	HBsAg only %	Anti-HBs and anti-HBc %	Anti-HBs only %	All markers negative %
Cases	46	31	2	17	24	26
Controls	82	5	12	12	42	35

5. OESOPHAGEAL CANCER (Dr N. Muñoz)

A histological study is planned of oesophagi from patients who died of diseases other than cancer of the oesophagus and on whom autopsy was performed within 12 hours after death. The following areas with varying risks for oesophageal cancer have been selected: high-risk areas: Gonbad (Iran), Alma Ata (Kazakh SSR) [1] and Transkei (South Africa); intermediate-risk areas: Caen (France), Singapore, Teheran, Nairobi and La Plata (Argentina); low-risk areas: Loma Linda (Calif., USA), Sao Paulo (Brazil) and Cali (Colombia).

The oesophagi are being processed and the histology evaluated by a standard protocol. Material will be collected by the Agency and then circulated among the participants, who will meet subsequently to assess the results of the study.

The study has already started in Iran (in collaboration with Dr A. Cor, Gonbad, Dr K. Ladan, Rasht, and Professor A. Armin, University of Teheran), where 16 oesophagi from medico-legal autopsies in Gonbad (a high-risk area), Rasht (a low-risk area) and Teheran (a low-risk area) have been examined; severe atrophic changes and some dysplastic changes have been observed, mainly in those from the high-risk area. A total of 100 oesophagi from high- and low-risk areas will be collected in the course of the study.

In parallel with the human study, oesophagi have been collected from dogs, goats and sheep in both Gonbad and Rasht (in collaboration with Mr P. Ghadirian, Babol, Iran). A parasite, *Gongylonema pulchrum*, has been identified in 22 out of 36 oesophagi from goats and sheep in Gonbad. This parasite was found in only one out of 20 cases collected in Rasht. Antibodies to this parasite, which is also known to affect man, will be investigated in oesophageal cancer patients and controls in the study areas. Other gongylonemata, *G. neoplasticum* and *G. orientalis*, have been reported to produce malignant tumours of the oesophagus in rats and mice. Ten out of 14 dog oesophagi showed nodular lesions produced by *Spirocerca lupi*.

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[1] Kusakina, G. K., Kolycheva, N. I., Kadiakina, A. N. (1974) *Arch. Pathol.*, **36**, 28-31

7. IARC RESEARCH CENTRE, NAIROBI

Professor A. WASUNNA (Head)

The Nairobi Research Centre has been moved to the Department of Surgery, Faculty of Medicine, University of Nairobi. The IARC laboratory and much of the equipment, particularly that used for assay of aflatoxin in foodstuffs, has been transferred to the Department of Public Health Laboratory Services, Ministry of Health, Kenya. This has facilitated the establishment of an FAO/UNEP collaborative project with the Kenya Government, designed to monitor mycotoxins in East Africa.

Fig. 27 Professor Ambrose Wasunna, Head of the IARC Research Centre, Nairobi.

1. EPIDEMIOLOGY OF OESOPHAGEAL CARCINOMA

The epidemiology of oesophageal carcinoma in Kenya is being investigated in collaboration with the Departments of Surgery and Pathology, University of Nairobi. Epidemiological data on all oesophageal cancer patients in Kenyatta National Hospital, Nairobi, and in the provincial hospital, Kisumu, Western Kenya, where the disease is especially frequent, have been collected, using a standard protocol.

2. COLLECTION OF BLOOD SPECIMENS

More than 100 patients from whom blood specimens had previously been obtained for cryopreservation have been followed, and further specimens are being collected in collaboration with the Departments of Surgery, Head and Neck Surgery, Radiation Therapy and Pathology of the Faculty of Medicine, University of Nairobi.

3. IMMUNOLOGICAL RESPONSE IN CANCER PATIENTS

Laboratory studies carried out in Nairobi have included a quality control investigation of laboratory procedures, such as differential counts and rosette counting, which form the basis of various immunological tests. Total lymphocyte counts and T-cell rosettes were used to monitor the immunological response of patients with Kaposi's sarcoma at different stages of the disease. This tumour is relatively common in East Africa, and the clinical course is often much more aggressive than elsewhere.

4. EFFECT OF NUTRITIONAL STATUS (in collaboration with Dr A. Butterworth, Wellcome Trust/WHO Research Laboratory, Nairobi)

A study has been started to compare the response to stimulation with mitogens of lymphocytes and sera from well-nourished with that from poorly-nourished Kenyans. Preliminary results indicate that the sera of the latter inhibit the response of their lymphocytes to phytohaemagglutinin stimulation.

5. BURKITT'S LYMPHOMA (in collaboration with Dr G. Brubaker, Shirati Hospital, Tanzania)

Surveillance of families of Burkitt's lymphoma patients has continued. A panel of antisera from the Shirati area has been established for immunogenetic studies.

8. IARC RESEARCH CENTRE, SINGAPORE

Professor K. SHANMUGARATNAM (Head)

1. THE SINGAPORE CANCER REGISTRY (RA/67/009)

Principal investigator: Professor K. Shanmugaratnam
Supporting staff: 3

Registration of all cancers occurring in the Republic of Singapore has continued. The results of registration over a seven-year period (1968–1974) have been analysed with respect to age, sex and major ethnic groups.

The registry has provided epidemiological and survival data used in the following research projects:

(*a*) Immunogenetics and immunology of nasopharyngeal carcinoma
(*b*) Nasopharyngeal cancer incidence among specific Chinese communities.

On-going research includes: (i) histopathological-specific rates of cancers of the breast and gonads (in collaboration with the Department of Pathology, University of Singapore), (ii) histopathology of nasopharyngeal cancer and correlation with survival rates and other biological variables (in collaboration with the Departments of Pathology, Otorhinolaryngology and Radiotherapy, University of Singapore, and the WHO Immunology Research and Training Centre, Singapore), and (iii) variations in cancer incidence by place of birth (in collaboration with IARC Unit of Epidemiology and Biostatistics).

2. IMMUNOGENETIC AND IMMUNOLOGICAL STUDIES ON NASOPHARYNGEAL CARCINOMA

Principal investigator: Dr S. H. Chan
Supporting staff: 7

2.1 *Immunogenetics*

(*a*) *HLA locus A and B typing*

Typing of newly-diagnosed and long-term surviving nasopharyngeal cancer patients and family members is being continued. A register of all patients that have been HLA typed has been begun and contains information such as age at diagnosis, sex, ethnic and dialect group, date of diagnosis, histology, HLA profile, cell-mediated immune functions, Epstein–Barr virus serology, response to radiotherapy, tumour recurrence and date of death, as well as reference numbers of cancer registration, biopsy and radiotherapy. This will permit definition of newly-diagnosed patients and survivors and determination of the

relationship between HLA profiles and response to therapy, recurrences, survival and immune functions. So far, 350 patients have been entered. A more restricted register has also been started of patients who were histologically negative although suspected of having nasopharyngeal cancer. This will permit correlation of HLA profile and immune functions with subsequent emergence of nasopharyngeal carcinoma.

(i) *Normal subjects*

Cord blood from 238 normal Chinese (100 Cantonese, 95 Hokkien and 43 Teochews) has been collected and HLA-typed. In addition, 103 samples of cord blood from normal Malays have been typed. The HLA profiles of these 341 subjects will serve as the 'normal' HLA profile in this part of the world. Certain locus A antigens are strongly associated with certain locus B antigens—a linkage disequilibrium. HLA-A2 and B Sin2, HLA-AW19 and HLA-BW17, HLA-A blank and HLA-BW17 are pairs of such antigens that show the strongest linkage among the normal Chinese population. These same antigen pairs show an even stronger association among Chinese patients with nasopharyngeal cancer.

(ii) *Newly-diagnosed nasopharyngeal carcinoma patients*

Since the identification of the antigen B Sin2, 141 newly diagnosed nasopharyngeal carcinoma patients have been HLA-typed. For locus A antigens, the excess of A2 among the new patients was less than that found previously, a reflection probably of the improved survival associated with A2 (see Section 2.1 (*a*) (iii)). The highly significant deficit of HLA-A11 among the nasopharyngeal cancer cases was even more striking than observed previously.

At locus B, the relative risk associated with B-Sin2 was slightly less than that observed previously but still highly significant. There was an emergence of a new association, the increase in risk associated with BW17, which was even higher than B Sin 2. That this was not seen in earlier studies was mainly due to two reasons. First, patients reported earlier comprised a mixture of newly-diagnosed patients and survivors, and the frequency of BW17 among survivors was very low (see section 2.1 (*a*) (iii)). Second, among the 91 comparison group patients suspected of having nasopharyngeal cancer but found to be histologically negative, 11 were subsequently found to have nasopharyngeal cancer or metastases to the neck lymph nodes, and the frequency of BW17 was high among these patients.

Eighty-one (57%) of the 141 newly-diagnosed patients had joint occurrence of A2 and B Sin2, compared with 81 (34%) of 238 normal subjects ($\chi^2 = 19.8$; $P = 0.00001$). BW17 has been found to be strongly associated with AW19 or an A blank. The joint occurrence of BW17 and either AW19 or A blank was seen in 36 (26%) of 141 nasopharyngeal carcinoma patients and in 26 (11%) of 238 normal subjects ($\chi^2 = 13.8$; $P = 0.00002$).

(iii) *Long-term survivors (more than 5 years)*

The frequency of BW17 was significantly lower among the long-term survivors (8%) when compared with newly-diagnosed patients (28%). B Sin2 has also a lower frequency in long-term survivors, but the effect is less marked and does not reach statistical significance. On the other hand, the occurrence of A2 without BW17 or B Sin2 was higher

(44%) among the long-term survivors than among the newly-diagnosed patients (24%). These data suggest that BW17 and, to a lesser extent, B Sin2 are poor survival factors, whereas A2 without BW17 or B Sin2 is associated with good survival.

(iv) *Age at onset*

In newly-diagnosed patients below 30 years of age, there is a significantly lower frequency of A2-B Sin2 than in older age groups and a higher frequency of A blank-BW17.

(*b*) *Locus D typing*

The interpretative problems of detecting a MLR typing response have now been resolved, and a greater restriction of locus D genes has been found among nasopharyngeal cancer patients than controls.

(*c*) *Ia locus*

There is strong evidence to suggest that the Ia locus is located very closely to, if not identical with, the D locus. Furthermore, its gene product on the lymphocyte surface is also very close to that of the D locus. In contrast to locus A and B cell surface gene products, which are expressed in all nucleated cells, the products of the Ia locus are only expressed on B lymphocytes, monocytes/macrophages, sperm cells and epithelial/endothelial cells. Since Ia typing is a serological assay, it is more amenable to large-scale screening than is D locus typing.

Over the last 12 months, a total of 1420 maternal sera were screened against unselected and B-enriched lymphocytes from nasopharyngeal cancer patients and controls; 254 of these had B-cell (Ia-like) reactivities. Some of the 254 sera also contained weak anti-locus A and B activities. These are being systematically absorbed with pooled platelets (no Ia antigen) from 40 blood donor packs (500 ml each).

The IARC Research Centre, Singapore, participated in the Seventh Histocompatibility Workshop, in which lymphocytes from nasopharyngeal cancer patients and families were tested against a panel of well-defined locus A, B and C antisera. B-Enriched lymphocytes from patients and controls were screened against a panel of workshop Ia sera as well as against the Research Centre's own Ia sera. It is hoped that Ia workshop numbers will be assigned to the reactivities of some of the Research Centre's sera.

(*d*) *Primed lymphocyte transformation (PLT)*

Because of the difficulty in obtaining nasopharyngeal carcinoma-associated D locus homozygous cells for D locus typing, a new method of typing, the PLT assay, was started. Briefly, if lymphocyte A is cultured with lymphocyte B which is treated with mitomycin C, a clone of A cells with specificities for the D locus antigen of B will appear. This is known as 'priming' of A cells against locus D antigen of B. Other lymphocytes which do not share common D locus antigens with B will not stimulate the 'primed' A lymphocytes. This phenomenon is analogous to the primary and secondary antigen responses *in vitro*. Therefore, if A and B share a common locus D antigen (family members), the 'primed' A

lymphocyte will now only recognize and respond to the non-shared locus D antigen of B. This situation can be achieved by using family members of NPC patients in which haplotypes could be designated by HLA locus A and B typing.

In preliminary experiments we have shown consistent specificities, and the ability to detect specificity is supported in a family study, shown in Table 29. It can be seen that lymphocyte 3, primed by the locus D antigen on haplotype (*a*) of lymphocyte 4, responded to lymphocytes from other members of the family who carried the (*a*) haplotype.

Table 29. Secondary response of lymphocyte 3 primed by lymphocyte 4

Stimulated by		cpm	Net cpm	SI [a]
Nasopharyngeal carcinoma patient	a/b	2019	1562	4.4
Spouse	c/d	609	152	1.33
Child	c/b	457	0	1
Child	c/a	1800	1343	3.9
Child	a/d	1660	1203	3.63

[a] Stimulation index

The advantages of the PLT assay for locus D typing over the conventional assay include ready availability of appropriate cells, a positive response and relatively short period of incubation. However, it also suffers from the disadvantage of requiring large quantities of cells. Therefore, if the Ia locus is closely associated with the D locus, genetic typing of the former will have a distinct advantage, both logistically and technically.

(*e*) *Other genetic markers*

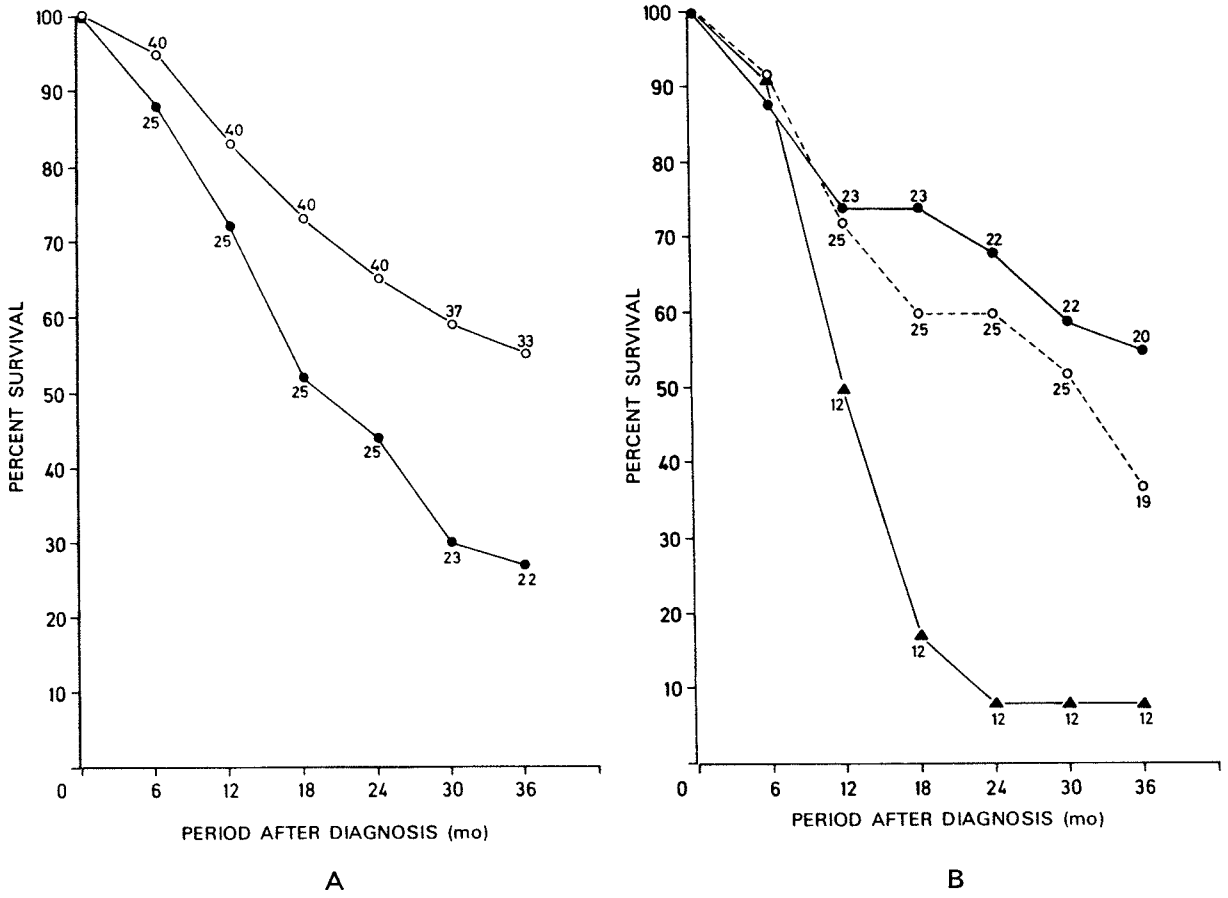
Blood samples from over 200 nasopharyngeal carcinoma patients have been typed for 25 genetically controlled red-cell enzymes and five serum protein systems by Professor R. Kirk (Canberra, Australia); comparative groups consisted of non-nasopharyngeal carcinoma patients and normal controls. The gene frequencies of nasopharyngeal carcinoma patients and controls differed significantly; the non-nasopharyngeal carcinoma patients occupied an intermediate position.

2.2 *Immunology*

(*a*) *Prognostic value of non-specific cell-mediated immune response*

Newly-diagnosed nasopharyngeal carcinoma cases and suitable controls were tested for their skin reactivity to purified protein derivative and the *in vitro* responsiveness of their blood lymphocytes to phytohaemagglutinin. Although these tests were within normal limits in some of the patients, a significant lowering of response was found in the group as a whole as compared with the control group. Analysis of survival up to 3½ years indicates that patients with normal responses survived longer than those with poor responses (Fig. 28). Furthermore, it was shown that hyporesponsiveness to phytohaemagglutinin is not related to stage of the disease. These results indicate that non-specific cell-mediated immunity tests both *in vivo* and *in vitro* are of prognostic value in patients with nasopharyngeal carcinoma.

Fig. 28 Relationship between cell-mediated immune responses (Mantoux response) at time of diagnosis and survival of nasopharyngeal carcinoma patients.
A to purified protein derivatives (○ — normal response; ● — lower response).
B—to phytohaemagglutinin (● — normal response; ○ — lower response; △ — very low response).



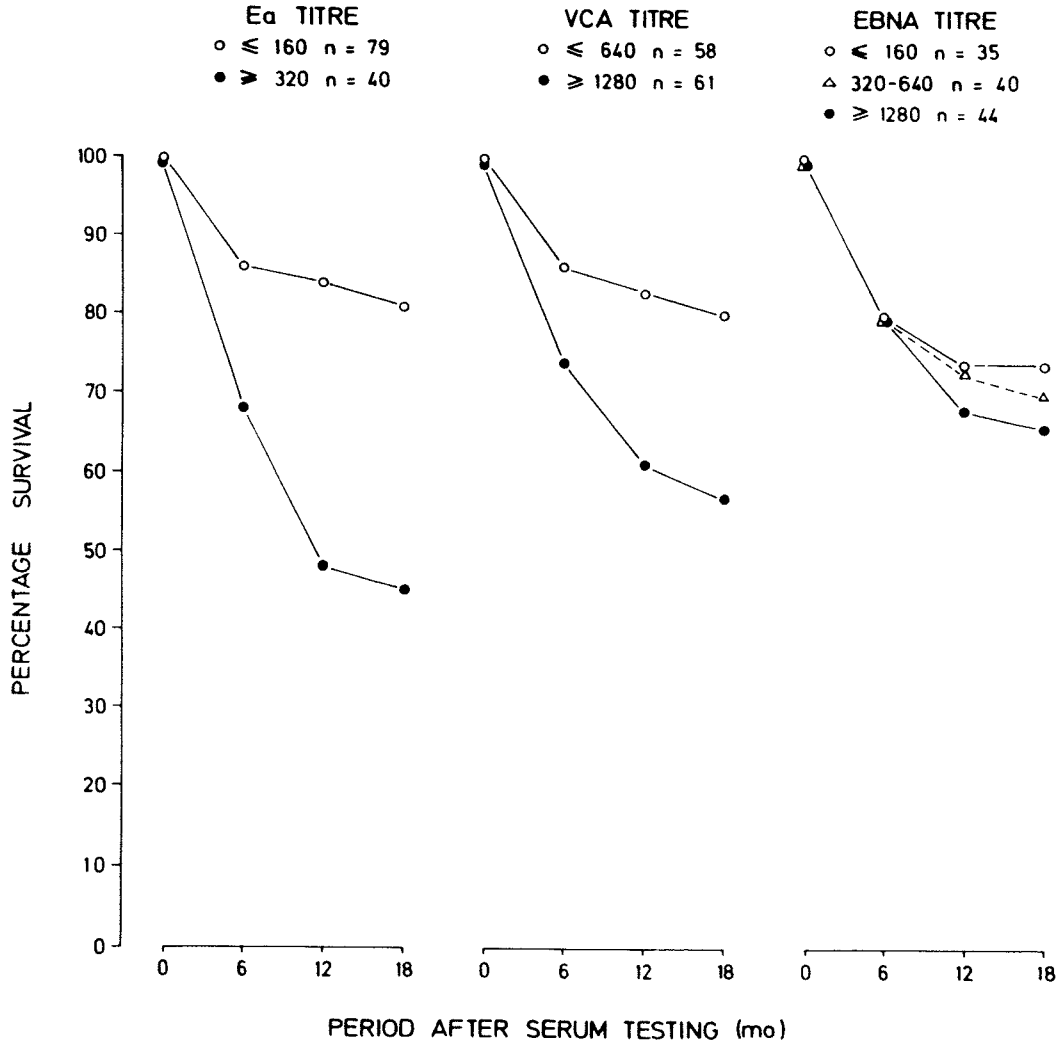
(*b*) *Specific immunity to Epstein–Barr-virus-related antigens*

(i) *Serology*

Anti-viral capsid antigen, early antigen and nuclear antigen antibody titres were measured in the Unit of Biological Carcinogenesis, using sera or plasma from nasopharyngeal carcinoma patients. These patients have now been followed for more than two years, and the study of survival rates indicates that high titres of antibodies against viral capsid antigen and early antigen are associated with poor prognosis (Fig. 29).

In a retrospective study, anti-viral capsid antigen, early antigen and nuclear antigen antibodies, as well as antibody-dependent lymphocytotoxicity (performed by Dr P. Levine, National Cancer Institute, Bethesda, Md., USA) were assessed in three groups of nasopharyngeal carcinoma patients, those that were dead within two years, those that survived 2–4 years and long-term survivors (more than five years), and in non-nasopharyngeal carcinoma patients (Table 30). These results indicate that high titres to the Epstein–Barr virus-associated antibodies are associated with a poor prognosis, whereas high titres to antibody-dependent lymphocytotoxicity are associated with good prognosis.

Fig. 29 Relationship of antibody titres to Epstein-Barr virus-associated antigens at time of diagnosis and survival of nasopharyngeal carcinoma patients. EA — early antigen; VCA — viral capsid antigen; EBNA — nuclear antigen.



(ii) *Cell-mediated immunity*

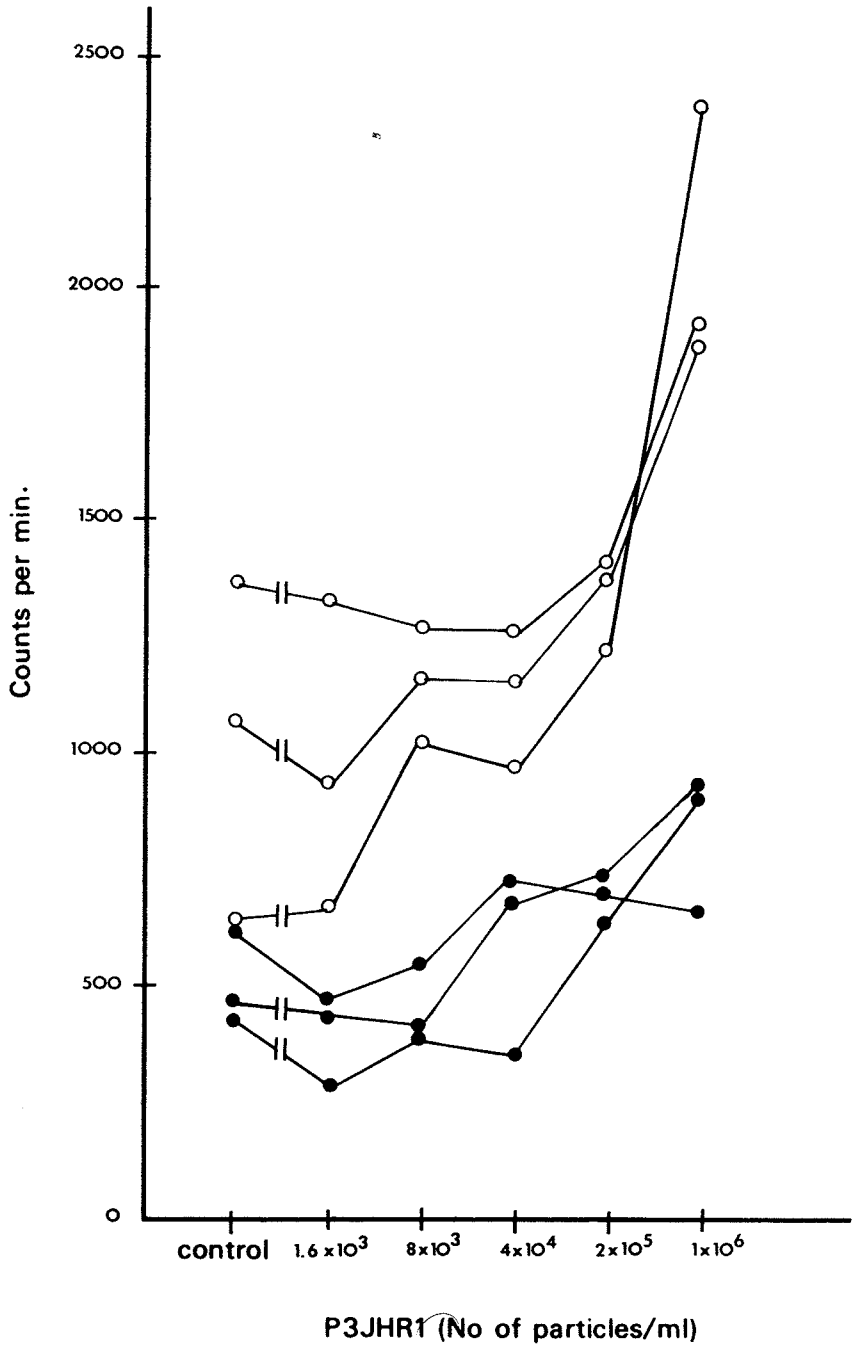
Blast transformation

Dose responses in nasopharyngeal carcinoma patients and control subjects were observed for inactivated 'purified' virion particles from P_3JHR1, B95 and HSV I and for Raji soluble antigen. Nasopharyngeal carcinoma patients have lower responses to P_3JHR1 than controls (Fig. 30). The individual responses to P_3JHR1 were similar to that to B95 but were quite different from that to Raji antigen. In addition, supernatants from QIMR/WIL cell lines, another source of EBV antigen, were also used. Assessment of blast transformation using these antigens is in progress.

Leucocyte adherence inhibition Cr^{51} assay

A leucocyte adherence inhibition Cr^{51} assay has been developed. Mononuclear cells were incubated with Cr^{51} and added to various concentrations of antigens. The response to antigen was expressed as percent inhibition of counts in cells from control tubes.

Fig. 30 Dose-response of nasopharyngeal carcinoma patients (●) and controls (○) to P_3JHR1 virion antigen.



In initial experiments we have observed that the leucocytes of anti-viral capsid antigen-positive, normal subjects had significantly lower counts in assays containing the P_3JHRI virion antigen than control assays. The range of inhibition was between 20%–70%. The specificity has yet to be checked by studying EBV-negative individuals, but in Singapore such subjects are extremely rare.

Table 30. Geometric mean titres of Epstein-Barr virus-associated antibodies and of antibody-dependent lymphocytotoxicity (ADLC) in different groups of Singapore Chinese nasopharyngeal carcinoma (NPC) positive and negative patients

	NPC +ve (newly-diagnosed)			NPC +ve > 5 years	NPC –ve
	Total	Dead (< 2 yrs)	Alive (2-4 yrs)	Survivors	
EA	212.1 (39.8–1131.4)[a] n = 125	261.0 (49.1–1387.8) n = 53	181.2 (34.1–963.8) n = 72	35.2 (10–124) n = 27	14.1 (3.2–62.3) n = 64
VCA	1019.2 (348.5–2983.9) n = 125	1214.8 (452.2–3263.4) n = 53	896.4 (291.9–2752.5) n = 72	311.9 (136.5–712.7) n = 27	118.9 (51.1–276.5) n = 64
EBNA	549.7 (150.7–2004.5) n = 124	529.1 (135–2073.7) n = 52	564.7 (161.9–1969.7) n = 72	247.6 (65.2–940) n = 27	174.7 (61.4–497) n = 63
ADLC	513.9 (131.2–2013.2) n = 60	346.6 (81.7–1470.5) n = 26	694.4 (201.0–2398.4) n = 34	1185.1 (504.8–2782.6) n = 18	511.4 (189.9–1377.6) n = 34

[a] ± 1 SD limits
EA — early antigen; VCA — viral capsid antigen; EBNA — nuclear antigen

We have also used the assay system to test two patients with tumours and two normal subjects. There were differential responses in these patients and normal subjects to the two tumours extracts, but the number of patients tested is too small to show whether there are any specificities in the responses.

3. RISK FACTORS FOR LUNG CANCER IN SINGAPORE CHINESE

Co-investigators: Dr J. L. Da Costa, Dr Y. K. Ng
Coordinating officer: Dr R. MacLennan

See report of the Unit of Epidemiology and Biostatistics, Section 3.6.

9. IARC RESEARCH CENTRE, TEHERAN

Dr B. ARAMESH (Head)
Dr J. KMET (IARC Team Leader)
Mr P. GHADIRIAN (Head, Babol Research Station)
Professor T. HEWER, University of Bristol, UK (Consultant)
Professor D. MCLAREN University of Edinburgh, UK (Consultant)
Mrs L. BANISADRE (Administrative Assistant)

Collaborating Institute:

Institute of Public Health Research, University of Teheran (Director, Dr A. Nadim)

1. CASPIAN CANCER REGISTRY

The work of the Caspian Cancer Registry has continued (RA/70/024), and data up to June 1976 have now been received. The calculation of incidence rates and an analysis of time trends awaits the detailed results of the 1976 census.

2. OESOPHAGEAL CANCER STUDIES

2.1 *Initial observations*

In the first stages of this study, food intake, smoking and drinking patterns, other personal habits, occupation, economic and agricultural practices, methods of food storage, preservation and preparation, etc., were examined in zones of contrasting incidence of oesophageal carcinoma in the Caspian littoral of Iran. Typical dietary items were also analysed for polycyclic aromatic hydrocarbons, volatile *N*-nitrosamines and aflatoxins as well as for nitrate and nitrite. The results of this preliminary survey are now being published in summary form [1].

2.2 *Study of cases, controls and their households*

The analysis is now almost complete, and an article describing the results is in preparation. The finding of lower economic status among the oesophageal cancer patients than among controls, as reported last year, can now be defined in greater detail:

(*a*) The lower socio-economic status is not confined to the present; status in the past shows at least as strong an effect. For men, the number of years at school is strongly and negatively associated with risk (Table 31); and for women, the number of children born alive but who later died is a strong risk factor (Table 32). These two variables are among the most objective that were studied.

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[1] Joint Iran-IARC Study Group (1977) *J. nat. Cancer Inst.* (in press).

Table 31. Relationship of number of years at school (as an indication of socio-economic status) to risk for oesophageal cancer among Iranian men [a]

	Never at school	Attended school at least 1 year	Relative risk [c]
Oesophageal cancer patients [b]	132	9	3.08(1.38-7.11)
Controls	236	47	
Other cancer patients	40	9	1.56(0.55-4.50)
Controls	75	24	

[a] Numbers are only from regions in which at least some positive and negative cases or controls were available.
[b] On the basis of histological or radiological diagnosis
[c] Adjusted for region and age

Table 32. Relationship of number of children who had died [a] (as an indication of socio-economic status) to risk for oesophageal cancer among Iranian women [b]

	Fewer than 4 children died	Four or more children died	Relative risk [a]
Oesophageal cancer patients [c]	40	49	0.48(0.27-0.86)
Controls	99	60	
Other cancer patients	64	11	2.08(0.92-4.76)
Controls	111	38	

[a] The total number of children did not differ between oesophageal cancer patients and controls
[b] Numbers are only from regions in which at least some positive and negative cases or controls were available
[c] On the basis of histological or radiological diagnosis
[d] Adjusted for region and age

(*b*) When the analysis is confined to tumours diagnosed on either histological or radiological grounds, the gradient of risk with socio-economic status is, if anything, increased.

(*c*) The socio-economic effect is stronger for oesophageal cancer patients than for those with other tumours.

The interpretation of these findings would be that poor socio-economic status of long duration is specifically associated with risk for oesophageal cancer and could reflect poor nutritional status (see section 2.4) or opium addiction (see section 2.3), or a combination of the two.

Further findings in the study areas which, because of their consistency with findings in other areas of the world, give support to the overall results are as follows:

(*a*) In the lower-incidence areas of Gilan and Babol, tobacco smoking is positively associated with risk. In the high-incidence area, little tobacco smoking is practised.

(*b*) Cancers of the lung, larynx and hypopharynx are associated with cigarette smoking; the numbers are too small to distinguish regional effects.

(*c*) Late first pregnancy increases the risk for breast cancer, and high parity decreases the risk.

To investigate the accuracy of replies to the first questionnaire, patients were re-interviewed, using a greatly shortened questionnaire. Eighty-five out of 98 patients with oesophageal cancer had died in the 18 months' (average) interval between the two visits; this added support to the diagnosis. The results of the re-interviewing indicated an acceptable level of repeatability.

2.3 *Search for environmental carcinogens*

As previously noted [1], the levels of volatile *N*-nitrosamines, polycyclic aromatic hydrocarbons and aflatoxins in diet were found to be low and substantially similar in high- and low-incidence areas. As no other food item appeared to be incriminated, attention has been focused on bread, the staple food in the high incidence area, and on the wheat used in its preparation. Contaminants, in particular, fungi and their associated toxins and extraneous seeds, have been investigated at the Commonwealth Mycology Unit, London (Dr C. Booth) and at Rothamstead Experimental Station, Harpenden, UK (Professor J. Lacey). The significance of the fungal contaminants found is now being assessed by Dr P. Austwick (Institute of Comparative Medicine, Zoological Society, London), but it appears that these are unlikely to be important.

Twenty samples of wheat, taken from storage pits in the high-incidence area, have been analysed for aflatoxins, sterigmatocystin and ochratoxin by the multi-detection method [2]. In one sample only, a trace of aflatoxin was detected; the other mycotoxins were not detected at all.

Over 50 adventitious species of seeds were found in the wheat samples examined at the Official Seed Testing Station for England and Wales, Cambridge, UK (Mr R. Flood). Among the five species that occurred very frequently was *Lolia temulentum*, consumption of which has been associated with acute illness and hepatotoxicity. The full significance of these findings cannot be assessed until a field botanical survey in the region has been completed (Professor T. Hewer, University of Bristol, UK).

The feasibility of conducting long-term animal feeding experiments using traditional Turkoman bread is now being explored (in collaboration with Dr R. Preussmann, German Cancer Research Centre, Heidelberg, Federal Republic of Germany).

The longer the investigating teams have worked in the area, the more they have gained the confidence of the inhabitants, and over the past year it has been possible to substantiate reports about the widespread consumption of opium and opium derivatives in the region. The dottle is frequently scraped from the opium smoker's pipe and, after reprocessing, eaten; it is often given to young children as a polyvalent medicine. Another female population with a high incidence of oesophageal cancer, the Xhosa in the Southern Transkei, also eat pipe scrapings. Samples from both areas have been obtained for mutagenicity testing. The extracts from the opium dottle have been shown to be strongly mutagenic (Dr H. Bartsch, personal communication), and further enquiries are underway (see page 107).

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[1] International Agency for Research on Cancer (1976) *Annual Report, 1976*, Lyon, p. 127.
[2] Stoloff, L., Neshein, S., Yin, L., Rodericks, J. V., Stack, N. C. Campbell, A. D. (1971) *J. AOAC*, S4-1, 91-97.

2.4 *Nutrition*

Previous work has shown that the Turkoman diet is unusually restricted. A series of preliminary studies were made to determine whether intake, as estimated by dietary surveys, indicated malnutrition which could be confirmed by clinical evidence. In September 1976, eight locations were visited in the high-incidence area (seven inhabited by Turkomans and one by Zabolis) and one village in the low-incidence province of Gilan. There was clear evidence of extensive riboflavin deficiency but no evidence of deficiencies of either vitamin A or C (Table 33).

Table 33. Clinical nutritional assessment of children and pregnant and lactating women in zones of contrasting oesophageal cancer risk in the Caspian littoral of Iran

	High-incidence areas			*Low-incidence area*
	Gomishan	*Pishkamar*	*Moravah Tappeh*	*Gilan*
No. of children	528	56	408	114
% with protein/calorie malnutrition	6	5	5	3
% with riboflavin deficiency	9	32	15	14
No. of pregnant and lactating women	240	21	160	28
Average time of breast feeding (months)	20	17	23	7
% with riboflavin deficiency	5	24	14	7

These studies were confirmed in March-April 1977 by clinical (Professor D. McLaren, University of Edinburgh, UK) and biochemical (Dr F. Siassi, Institute of Public Health Research, Teheran) assessments of riboflavin and other deficiencies. This work was undertaken in the region of Korand, a high-incidence locality. As before, there was no evidence of vitamin A or C deficiencies. Iron-deficiency anaemia was frequent in lactating women. Riboflavin deficiency, as established by erythrocyte glutathione reductase estimation and determination of the urinary riboflavin:creatinine ratio, was found to be very widespread. Some 66 subjects with established riboflavin deficiency were given 60 mg riboflavin daily, and their blood and urine were examined some three weeks after treatment. Fifty-five of these subjects completed the treatment, and in 53 there was definite improvement. Some 56 subjects with no clinical evidence of riboflavin deficiency, matched by age and sex, constituted a control group. It is not known whether riboflavin deficiency favours neoplasia, but this measure would appear to be a useful index of nutritional deficiency in the region.

2.5 *Genetic factors*

The incidence rates for cancer of the oesophagus in the regions of Southern Gonbad and Southern Gorgan are second only to those found in Turkomans. Hitherto, this population has been regarded as being of Persian rather than of Turkic stock, and thus a rather lower incidence of oesophageal cancer would have been expected. Further enquiries have shown that this region was settled by Turkic-speaking Gzylbashi, who moved into this region from Azerbaijan in the 17th century. The genetic similarity of this group to other

Turkic people remains to be confirmed; however, Professor R. Kirk (National University, Canberra) who undertook an analysis of material collected during the period of initial observation in 1972–1973 in 38 villages of contrasting oesophageal cancer incidence and sex ratio, has found pronounced genetic similarities between Turkomans and Azeri Turks and between both groups and Iranian Kurds [1].

Preliminary results from a case-control study of HLA profiles among oesophageal cancer cases and matched controls indicate that there is probably a greater risk among those with the BW40 haplotype, which is particularly common among a Turko-Mongolian group. This work has been continued as a case and case-family study. To date, some 30 histologically confirmed cases and their families have been examined [2].

2.6 *Precancerous lesions* (Dr N. Muñoz)

See report of Interdisciplinary Programme and International Liaison Unit, p. 124.

2.7 *Review of progress*

A progress review meeting was held at the Teheran Research Centre in October 1976 and was attended by Iranian and IARC investigators and by representatives of the National Cancer Institute, USA (Dr M. Schneiderman, Dr F. Cohen), which partially supports this work. It was considered that the study was particularly suitable for assessment of the role and relation of environmental and host factors and that exploration of both aspects should continue.

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[1] Kirk, R. L., Keats, B., Blake, N. J., McDermid, E. J., Ala, F., Karimi, M., Nickbin, B., Shabazi, H. & Kmet, J. (1976) *Amer. J. phys. Anthrop.* (in press).

[2] Mohagheghpour, N. (1976) In: *Proceedings of the First International Symposium on HLA and Disease, Paris, 1976* (in press).

Annex 1

PARTICIPATING STATES AND REPRESENTATIVES AT THE SIXTEENTH SESSION OF THE IARC GOVERNING COUNCIL 28–29 APRIL 1977

Australia

Dr D. S. M. GRAHAM
Regional Medical Director
Australian Embassy
Rome

Dr J. RAVENSCROFT
Director, Australian Treasury Office
Australian Permanent Mission
Geneva, Switzerland

Belgium

Professor S. HALTER *(Chairman)*
Secretary-General
Ministry of Public Health and the Family
Brussels

Dr G. CLAUS
Director-General
Ministry of Public Health and the Family
Brussels

France

Professor E. J. AUJALEU
Honorary Director-General
National Institute of Health and Medical Research
Counsellor of State
Paris

Dr J. F. DUPLAN
Counsellor
National Institute of Health and Medical Research
Bordeaux

Miss M. A. MARTIN-SANÉ
Counsellor
Ministry of Foreign Affairs
Paris

Federal Republic of Germany

Mr H. VOIGTLÄNDER *(Rapporteur)*
International Relations Section
Federal Ministry for Youth, Family Affairs and Health
Bonn

Dr H. KAISER
Ministerial Council
Ministry of Finance
Bonn

Italy

Professor L. SANTI
Director, Institute of Oncology, University of Genoa
Genoa

Professor U. VERONESI
Director-General
National Institute for the Study and Treatment of Tumours
Milan

Japan

Dr A. TANAKA *(Vice-Chairman)*
Ministry of Health and Welfare
Tokyo

Mr T. ONISHI
First Secretary
Permanent Delegation of Japan to the International Organizations in Geneva
Geneva, Switzerland

The Netherlands

Dr J. SPAANDER
Director-General
National Institute of Public Health
Bilthoven

Mr W. J. KAKEBEEKE
Deputy Director International Affairs
Ministry of Health and Environmental Protection
Leidschendam

Union of Soviet Socialist Republics

Dr D. D. VENEDIKTOV
Deputy Minister of Health
Ministry of Health of the USSR
Moscow

Dr N. N. BLOKHIN
Director, Cancer Research Center
Academy of Medical Sciences
Moscow

Professor V. P. DEMIDOV
Chief of Cancer Department
Ministry of Public Health of the USSR
Moscow

Dr O. L. BRATKOV
Senior Inspector
Ministry of Public Health of the USSR
Moscow

Dr Y. I. PUCHKOV
Chief, Department of International Scientific Relations
Cancer Research Center
Academy of Medical Sciences
Moscow

United Kingdom

Dr J. LEARMONTH GOWANS
Secretary
Medical Research Council
London

Dr R. C. NORTON
Senior Principal Medical Officer
Medical Research Council
London

United States of America

Dr G. T. O'CONOR
Associate Director for International Affairs
National Cancer Institute
National Institutes of Health
Bethesda, Md.

Mr R. F. ANDREW
Director, Agency for Health and Drug Control
Department of State
Washington, D.C.

World Health Organization

Dr T. A. LAMBO
Deputy Director-General

Mr A. GROENENDIJK
Director, Division of Budget and Finance

Mr F. GUTTERIDGE
Director, Legal Division

Observers

Dr J. F. DELAFRESNAYE
Executive Director
International Union Against Cancer
Geneva, Switzerland

Professor S. ECKHARDT
Incoming Chairman of the IARC Scientific Council

Professor A. C. UPTON
Outgoing Chairman of the IARC Scientific Council

Annex 2

MEMBERS OF THE SCIENTIFIC COUNCIL AT ITS THIRTEENTH SESSION, 5–7 JANUARY 1977

Professor A. CAPUTO
Director
Regina Elena Institute for Cancer Research
Rome

Sir RICHARD DOLL
Regius Professor of Medicine,
Oxford University
Radcliffe Infirmary
Oxford, UK

Dr J. F. DUPLAN *(Vice-Chairman)*
Director of Research
National Institute of Health and Medical Research
Bordeaux, France

Dr S. ECKHARDT
Director, National Institute of Oncology
Budapest

Professor J. MILLER
Head, Experimental Pathology Unit
Walter and Eliza Hall Institute of Medical Research
Royal Melbourne Hospital PO
Melbourne, Australia

Professor C. MOFIDI
Professor of Human Ecology, University of Teheran
Secretary General, Central Council of Universities and Academic Institutions of Iran
Teheran

Professor K. MUNK
Director, Institute of Virology
German Cancer Research Centre
Heidelberg, Federal Republic of Germany

Professor T. Sugimura
Director, National Cancer Center Research Institute
Tokyo

Professor N. N. TRAPEZNIKOV
Deputy Director, Cancer Research Center
Academy of Medical Sciences of the USSR
Moscow

Professor A. C. UPTON *(Chairman)*
Dean, School of Basic Health Sciences
State University of New York at Stony Brook
Stony Brook, N.Y., USA

Professor T. G. VAN RIJSSEL *(Rapporteur)*
Pathology Laboratory, Faculty of Medicine,
National University of Leiden,
Leiden, The Netherlands

Professor G. L. J. VAN DER SCHUEREN
Director, Tumor Centre
St. Rafael Academic Hospital
Leuven, Belgium

Annex 3

RESEARCH AGREEMENTS IN OPERATION BETWEEN IARC AND VARIOUS INSTITUTIONS JUNE 1976 – JUNE 1977

Support of IARC Research Centres

RA/68/002	University of Singapore (Contribution to the maintenance of an IARC Research Centre at the University of Singapore)
RA/75/020	University of Nairobi (Contribution to the maintenance of an IARC Research Centre at the University of Nairobi)

Reference centres/Serums banks

RA/73/029	Institute of Experimental Oncology, University of Genoa, Genoa, Italy (IARC Reference Centre for environmental carcinogenesis)
RA/73/033	Medical College, Hanover, Federal Republic of Germany (Creation of an IARC Reference Centre for environmental carcinogenesis)
RA/74/003	Institute for Documentation, Information and Statistics, German Cancer Research Centre, Heidelberg, Federal Republic of Germany (Clearing-house for on-going research in cancer epidemiology)
RA/75/013	Cancer Research Center, Academy of Medical Sciences of the USSR, Moscow (Creation of an IARC Reference Centre for environmental carcinogenesis)
RA/75/014	National Institute of Hygiene, Budapest (Creation of an IARC Reference Centre for environmental carcinogenesis)
RA/76/005	Department of Obstetrics and Gynaecology, Hôtel Dieu, Lyon, France (Supply of serum samples for the preparation of an international standard of pregnancy proteins)
RA/76/006	Minguettes Polyclinic, Vénissieux, France (Supply of serum samples for the preparation of an international standard of pregnancy proteins)
RA/76/007	Tonkin Clinic, Villeurbanne, France (Supply of serum samples for the preparation of an international standard of pregnancy proteins)

RA/76/009 Saint Augustin Clinic, Lyon, France
(Supply of serum samples for the preparation of an international standard of pregnancy proteins)

RA/76/010 Rillieux Polyclinic, Rillieux, France
(Supply of serum samples for the preparation of an international standard of pregnancy proteins)

RA/76/019 Angel H. Roffo Oncological Institute, Buenos Aires
(Creation of an IARC Reference Centre for environmental carcinogenesis)

RA/76/022 Department of Obstetrics and Gynaecology, Hôtel Dieu, Lyon, France
(Supply of serum samples for the preparation of an international standard of pregnancy proteins)

RA/77/004 Cairo Cancer Institute, Cairo
(Agreement for collection and extraction by dichloromethane of urinary samples from twenty patients with bilharzia and twenty patients with urinary bladder cancer)

RA/77/005 British Food Manufacturing Industries Research Association, Leatherhead, Surrey, UK
(Extraction and examination of urine samples from twenty patients with bladder cancer and twenty paraplegic patients)

Cancer registries/Incidence studies

RA/67/009 IARC Research Centre, University of Singapore
(Cancer registry at Singapore)

RA/70/024 Institute of Public Health Research, University of Teheran, Teheran
(Study on the incidence of cancer in the Caspian littoral of Iran)

RA/72/014 Department of Pathology, University of the West Indies, Kingston, Jamaica
(Partial support of the Jamaica Cancer Registry)

RA/73/016 International Association of Cancer Registries
(Provision of a secretariat and other supporting services)

RA/75/022 Medico-Social Research Board, Dublin
(Study of causes of death among Guinness brewery workers in Dublin)

RA/76/003 Royal College of General Practitioners, Birmingham, UK
(Record linkage from an outcome of pregnancy studies carried out in Birmingham and Dundee to the cancer registry records in Oxford)

RA/76/004 University of Helsinki, Helsinki
(Record linkage from a study of congenital malformation to cancer registry records in Finland)

RA/76/016 Medico-Social Research Board, Dublin
(Study of causes of death among Guinness brewery workers in Dublin)

RA/76/018 Birmingham Cancer Registry, Birmingham, UK
(Development and completion of a computer-based exercise on a hypothetical occupational cancer risk)

RA/76/023 Danish Institute of Clinical Epidemiology, Copenhagen
(Provision of mortality tapes to be used in the analysis of the study of mortality among Danish brewery workers)

Oesophageal cancer studies

RA/75/015 National Institute of Health and Medical Research, Division of Medico-Social Research, Le Vésinet, France
(Study of cases of œsophageal cancer and their controls in the Calvados region of France)

RA/77/002 Department of Physiology, University of Edinburgh, Edinburgh, Scotland
(Study of nutritional status among a population at high risk for œsophageal cancer from the Gonbad area of Iran)

RA/77/003 Institute of Public Health Research, Teheran
(Joint Iran/IARC study of characteristics of selected population groups in areas of differing œsophageal cancer incidence in the Caspian littoral of Iran — population and nutritional studies in the Atrak river area)

Studies on cancers linked with herpesviruses

RA/70/013 Hong Kong Anti-Cancer Society, Hong Kong
(Studies on the relationship between herpes-type infection and nasopharyngeal carcinoma)

RA/70/017 Department of Pathology, University of Singapore
(Studies on the relationship between herpes-type infection and nasopharyngeal carcinoma)

RA/71/020 East African Virus Research Institute, Entebbe, Uganda
(Follow-up study on Burkitt's lymphoma and Epstein-Barr herpesvirus infection in the West Nile District of Uganda)

RA/72/030 Netherlands Cancer Institute, Amsterdam
(Collaborative studies on immunoserology of Burkitt's lymphoma and nasopharyngeal carcinoma)

RA/73/009 Shirati Hospital, Tarime, Tanzania
(Studies on the epidemiology of Burkitt's lymphoma in the North Mara District of Tanzania)

RA/73/017 Sainte Marie-Thérèse Clinic, Lyon, France
(Study of the role of the herpesvirus type Epstein-Barr in the establishment of permanent cell lines from cord blood specimens)

RA/74/001 Association for the Development of Cancer Research, Primatology Laboratory, National Centre for Scientific Research, Villejuif, France
(Studies on the induction of lympho-epithelial tumours in the marmoset with Epstein-Barr virus)

RA/74/018 University of Hong Kong, Queen Mary Hospital Compound, Hong Kong
(Isolation and purification of Epstein-Barr virus specific antigens)

RA/75/002 Ross Institute, London School of Hygiene and Tropical Medicine, London
(Malaria antibody testing to be carried out by the Institute on sera from Burkitt's lymphoma studies in the West Nile District of Uganda and the Mara Region of Tanzania)

RA/75/005 Gustave Roussy Institute, Villejuif, France
(Study of the role of herpesvirus type Epstein-Barr in nasopharyngeal cancer)

RA/75/006 Lyon Blood Transfusion Centre, Plasma Desiccation Department, Histocompatibility Department, Beynost, Miribel, France
(HLA typing of blood from families with nasopharyngeal carcinoma in Tunisia)

RA/75/008 Edouard Herriot Hospital, Centre for Study and Research on Metabolic and Renal Diseases, Lyon, France
(Study on cellular immunology on the blood of nasopharyngeal carcinoma patients and their families from Tunis, Paris, Lyon and Marseille)

RA/75/009 Institute of Scientific Cancer Research, National Centre for Scientific Research, Villejuif, France
(Study of the biological activity of DNA of herpes simplex virus and Epstein-Barr virus on human lymphoblastoid lines)

RA/76/002 Zoological Society of London, London
(Development of micro-elisa test for Epstein-Barr virus serology)

RA/76/020 National Institute of Cancerology, Tunis
(Collaboration project on nasopharyngeal carcinoma in Tunisia)

RA/77/008 Shirati Mission Hospital, Musoma, Tanzania
(Studies on the effect of partial malaria suppression on incidence of Burkitt's lymphoma in North Mara)

RA/77/015 Department of General and Applied Biology, Claude Bernard University, Villeurbanne, France
(Characterization and purification of viral antigens for the development of serological tests)

Laboratory studies

RA/75/018 François Baclesse Regional Centre, Caen, France
(Animal experiments on the possible carcinogenic effects of cider alcohols)

Liver cancer studies

RA/75/025 Department of Hygiene and Epidemiology, School of Medicine, University of Athens, Athens
(Contribution to the collection of sera from patients with liver disease and controls to assess alpha-fetoprotein levels)

RA/76/012 Geneva Tumour Registry, Geneva, Switzerland
(Study of liver disease, including primary liver cancer, in the canton of Geneva)

RA/77/007 School of Pathology, Middlesex Hospital Medical School, London
(Estimation of hepatitis B antigen and antibody and other viral antibodies in serum specimens from East African cancer patients)

Studies on chemical carcinogens

RA/69/005 Department of Occupational Health, Hebrew University-Hadassah Medical School, Jerusalem
(Analysis of fat and other tissues for the presence of chlorinated hydrocarbons)

RA/70/002 Medical College, Hanover, Federal Republic of Germany
(Investigation of the effects of chemical carcinogens administered transplacentally on the fetal reproductive organs)

RA/70/003 Institute of Pathology, Medical University, Budapest
(Investigation of the effects of minute doses of chemical carcinogens on cells cultured *in vitro*)

RA/72/011 Institute of Experimental Oncology, University of Genoa, Genoa, Italy
(Investigation on effects of long-term administration of DDT and phenobarbital to rats)

RA/72/031 Institute for Experimental Toxicology and Chemotherapy, German Cancer Research Centre, Heidelberg, Federal Republic of Germany
(Study of elaboration of analytical methods for the identification and quantification of *N*-nitroso compounds in various environmental media)

RA/74/007 National Institute of Public Health, Bilthoven, The Netherlands
(Study on the potential carcinogenicity of maleic hydrazide)

RA/74/011 Ministry of Health, Institute of Experimental and Clinical Medicine, Tallinn, Estonian SSR
(Investigations on the combined carcinogenic action of asbestos dust and *N*-nitroso compounds in the hamster)

RA/74/034 P. Jacquignon Laboratory, Institute of Chemistry of Natural Substances, National Centre for Scientific Research, Gif-sur-Yvette, France
(Synthesis of reactive intermediates of some pesticides and organochlorine compounds)

RA/75/023 The Plant Pathology Department, Rothamsted Experimental Station, Harpenden, UK
(Study of microorganisms and toxins in Iranian wheat)

RA/75/024 Mycotoxin Research and Training Centre, Department of Biochemistry and Nutrition, University of Isfahan, Isfahan, Iran
(Study of mycotoxins in Iranian wheat)

RA/76/001 Free University of Brussels, Brussels
(Investigation of an *in vitro* biochemical assay for somatic mutagenesis by chemical mutagens/carcinogens)

RA/76/017 Cancer Research Center, Academy of Medical Sciences of the USSR, Moscow
(Investigation on the effect of prenatal exposure to a chemical on successive untreated generations)

RA/76/027 Institute for Experimental Toxicology and Chemotherapy, German Cancer Research Centre, Heidelberg, Federal Republic of Germany
(Study of elaboration of analytical methods for identification and quantification of *N*-nitroso compounds in various environmental media)

Studies on carcinogens other than chemicals

RA/72/034 Medical Research Council Pneumoconiosis Unit, Penarth, UK
(Research study programme on asbestos cancers)

RA/76/008 Joint European Medical Research Board, Liverpool, UK
(International epidemiological research programme to examine health effects, particularly as these concern cancer, of exposure to man-made mineral fibres in man)

RA/76/026 Institute of Oncology of the University of Genoa, Genoa, Italy
(Study on interaction between asbestos and chromosomal proteins from different human tissues)

Studies of various other cancer forms

RA/73/004 Department of Pathology, University of Iceland, Reykjavik
(Investigations on familiality of carcinoma of the breast)

RA/75/021 Department of Internal Medicine, Copenhagen County Hospital, Copenhagen
(Analysis of data from a study of intestinal microecology and colon cancer in Scandinavia)

RA/76/014 Cancer Research Center, Academy of Medical Sciences of the USSR, Moscow
(Editorial activities for the production of *Pathology of Tumours in Laboratory Animals*)

RA/77/010 Department of Community Medicine, University of Hong Kong, Hong Kong
(Case-control study of lung cancer in Chinese in Hong Kong)

RA/77/013 Tata Memorial Centre, Parel, Bombay, India
(*In vitro* immunological studies on cancer patients in Bombay)

Support of meetings

RA/76/025 Institute for Radiation and Environmental Research, Neuherberg, Federal Republic of Germany
(Support of a workshop on Bacterial *in vitro* Mutagenicity Test Systems, 6–9 March 1977, in Neuherberg)

RA/77/001 International Epidemiological Association, Johns Hopkins University, Baltimore, Md., USA
(Partial support of attendance costs of selected participants to a symposium and sessions relating to cancer at 8th International Scientific Meeting of IEA in Puerto Rico, 17–23 September 1977)

RA/77/006 Faculty of Medicine, Department of Microbiology, University of Kyoto, Kyoto, Japan
(Support of International Symposium on Nasopharyngeal Carcinoma, Kyoto, 4–6 April 1977)

RA/77/009 Faculty of Medicine, Department of Microbiology, University of Kyoto, Kyoto, Japan
(IARC contribution towards the International Symposium on Nasopharyngeal Carcinoma, Kyoto, 4–6 April 1977)

RA/77/011 Protein Laboratory, University of Copenhagen, Copenhagen
(Support of a workshop of the 5th Meeting of the International Research Group for Carcino-embryonic Proteins)

RA/77/014 Department of Microbiology, Milton S. Hershey Medical Center, Pennsylvania University, Hershey, Pa., USA
(Contribution to the Third International Symposium on Oncogenesis and Herpesviruses, Cambridge, Mass., USA, 25–30 July 1977)

Annex 4

MEETINGS AND WORKSHOPS ORGANIZED BY IARC, 1976–77

Course on epidemiology and biostatistics in cancer research	Lyon, 30 August – 10 September 1976
Evaluation of carcinogenic risk of chemicals to man: some miscellaneous pharmaceutical substances	Lyon, 18–25 October 1976
Study of risk of man-made mineral fibres	Copenhagen, 25–29 October 1976
Course on cancer epidemiology	Brasilia, 29 November – 4 December 1976
Evaluation of carcinogenic risk of chemicals to man: asbestos	Lyon, 14–17 December 1976
The biological action of alcohol	Lyon, 11 January 1977
Editorial board on manual of selected methods of analysis of *N*-nitroso compounds	Lyon, 19–20 January 1977
Evaluation of the carcinogenic risk of chemicals to man: some fumigants, the herbicides 2,4-D and 2,4,5-T, chlorinated dibenzodioxins and miscellaneous industrial chemicals	Lyon, 8–15 February 1977
Study of risk of man-made mineral fibres	Lyon, 3–4 March 1977
Alcohol and cancer meeting	Lyon, 23–24 March 1977
International symposium on etiology and control of nasopharyngeal carcinoma	Kyoto, Japan, 13–16 April 1977
European sub-committee — selection of manuscripts for presentation at fifth meeting on the analysis and formation of *N*-nitroso compounds, Durham, N.H., USA, August 1977	Lyon, 14 April 1977
Evaluation of the carcinogenic risk of chemicals to man: some aromatic amines and related nitro compounds: hair dyes, colouring agents and miscellaneous industrial chemicals	Lyon, 7–14 June 1977
Study of risk of man-made mineral fibres	Lyon, 30 June – 1 July 1977

Annex 5

VISITORS TO IARC, JULY 1976 TO JUNE 1977

Dr J. ABBAT
Director General, Laboratory Centre for Disease Control, Department of Health and Welfare, Ottawa

Mr K. ABRAMOWITZ
Robert S. First, Inc., Management Consultants, New York, N.Y., USA

Mr P. ADAMSON
New Internationalist Publications Limited, Benson, Oxfordshire, UK

Professor M. R. ALDERSON
Division of Epidemiology, Institute of Cancer Research, Royal Cancer Hospital, Sutton, Surrey, UK

Mr Y. ALEXANDRE
Asbestos Trade Union Office, Paris

Dr O. U. ALOZIE
Senior Programme Officer, Division of Geophysics, Global Pollution and Health, United Nations Environment Programme, Nairobi

Dr B. ARMSTRONG
University of Western Australia, Department of Medicine, Perth Medical Centre, Nedlands, W.A., Australia

Dr E. ARRHENIUS
Division of Cellular Toxicology, Environmental Toxicology Unit, University of Stockholm, The Wallenberg Laboratory, Stockholm

Dr A. AZORDEGAN
Department of Epidemiology and Biostatistics, Institute of Public Health Research, University of Teheran, Teheran

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INTERNAL TECHNICAL REPORTS, 1976–77

IARC Internal Technical Report No.

77/001 Report of the second meeting of the editorial board for the manual of selected methods of analysis for environmental carcinogens (Lyon, 19–20 January 1977)

Annex 7

PAPERS PUBLISHED OR SUBMITTED FOR PUBLICATION BY IARC STAFF

Bartsch, H. (1976) *The predictive value of mutagenicity assays to assess the possible carcinogenic risk of chemicals.* In: Rosenfeld, C. & Davis, W., eds, *Environmental Pollution and Carcinogenic Risks*, Lyon, International Agency for Research on Cancer (*IARC Scientific Publications* **No. 13**), pp. 229–240

Bartsch, H. (1976) *Detection of potential carcinogens as mutagens: factors for efficiency and limitations of the* Salmonella *test system.* In: *Proceedings of the Meeting of the Scientific Committee, 12 December 1975, Carlo Erba Foundation*, pp. 105–116

Bartsch, H. (1977) *The use of microbial mutagenicity assays for the detection of chemical carcinogens —a critique.* In: *Méchanismes d'Altération et de Réparation du DNA, Relations avec la Mutagenèse et la Cancérogenèse chimique*, Paris, Editions du CNRS, pp. 255–272

Bartsch, H., Camus, A. & Malaveille, C. (1976) Comparative mutagenicity of *N*-nitrosamines in semi-solid and in a liquid incubation system in the presence of rat or human tissue fractions. *Mutation Res.*, **37**, 149–162

Bartsch, H. & Loprieno, N. (1977) *Vinyl chloride (VCM): an example for evaluating adverse biological effects in short-term tests.* In: Böhme, H. & Schoneich, J., eds, *Proceedings of the 6th EEMS Meeting, Gernrode, German Democratic Republic*, Abhandlungen der Akademie der Wissenschaften der DDR, **No. 9**, pp. 161–165

Bartsch, H., Malaveille, C., Barbin, A., Brésil, H., Tomatis, L. & Montesano, R. (1976) Mutagenicity and metabolism of vinyl chloride and related compounds. *Environm. Hlth Persp.*, **17**, 193–198

Bartsch, H., Malaveille, C., Stich, H. F., Miller, E. C. & Miller, J. A. (1977) Comparative electrophilicity, mutagenicity, DNA repair induction activity, and carcinogenicity of some *N*- and *O*-acyl derivatives of *N*-hydroxy-2-aminofluorene. *Cancer Res.*, **37**, 1461–1467

Bartsch, H., Margison, G. P., Malaveille, C., Camus, A. M., Brun, G., Margison, J. M., Kolar, G. F. & Wiessler, M. (1977) Some aspects of metabolic activation of chemical carcinogens in relation to their organ specificity. *Arch. Toxicol.* (in press)

Bogovski, P. & Day, N. (1977) Accelerating action of tea on mouse skin carcinogens. *Cancer Lett.*, **3**, 9–13

Bourgeaux, C., Martel, N., Sizaret, P. & Guerring, J. (1976) Prognostic value of alpha-fœtoprotein radioimmunoassay in surgically treated patients with embryonal cell carcinoma of the testis. *Cancer*, **38**, 1658–1660

Camus, A., Bertram, B., Kruger, F. W., Malaveille, C. & Bartsch, H. (1976) Mutagenicity of β-oxidized *N,N*,di-*n*-propylnitrosamine derivatives in *S. typhimurium* mediated by rat and hamster tissues. *Z. Krebsforsch.*, **86**, 293–302

Camus, A. M., Wiessler, M., Malaveille, C. & Bartsch, H. (1977) High mutagenicity of *N*-(α-acyloxyl)alkyl-*N*-alkyl nitrosamines in *S. typhimurium*: model compounds for metabolically activated *N,N*-dialkylnitrosamines. *Mutation Res.* (in press)

Castegnaro, M. & Walker, E. A. (1976) *Developments in nitrosamine analysis.* In: Tinbergen, B. J. & Krol, B., eds, *Proceedings of the 2nd International Symposium on Nitrite in Meat Products, Zeist, 1976*, Wageningen, Centre for Agricultural Publishing and Documentation, pp. 187–190

Chan, S. H., Chew, T. S., Goh, E. H., Simons, M. J. & Shanmugaratnam, K. (1976) Impaired general cell mediated immune function *in vivo* and *in vitro* in patients with nasopharyngeal carcinoma. *Int. J. Cancer*, **18**, 139–144

Chan, S. H., de-Thé, G. & Goh, E. H. (1977) Prospective study of EBV antibody titres and survival in patients with nasopharyngeal carcinoma. *Lancet*, **i**, 948–949

Chan, S. H., Goh, E. H., Khor, T. H., Chew, T. S., de-Thé, G. & Shanmugaratnam, K. (1977) *General immunological status of NPC patients in Singapore.* In: *Proceedings of the International Symposium on Etiology and Control of Nasopharyngeal Carcinoma (NPC), Kyoto, Japan, April 1977*, Lyon, International Agency for Research on Cancer (in press)

Chan, S. H., Lee, S. K. & Simons, M. J. (1976) Levamisole augmentation of lymphocyte hyporesponsiveness to phytohaemagglutinin in patients with pulmonary tuberculosis. *Proc. Soc. exp. biol. Med.*, **151**, 716–719

Chan, S. H., Wallen, W. C., Levine, P. H., Periman, P. & Perlin, E. (1977) Lymphocyte responses to EBV-associated antigens in infectious mononucleosis, and Hodgkin's and non-Hodgkin's lymphoma patients using the leukocyte adherence inhibition assay. *Int. J. Cancer*, **19**, 356–363

Cooper, J. E. & Levin, A. G. (1976) A disease resembling murine hepatitis in nude mice in Kenya. *Trans. Roy. Soc. trop. Med. Hyg.*, **70**, 529–530

Crowther, J. S., Drasar, B. S., Hill, M. J., MacLennan, R., Magnin, D., Peach, S. & Teoh-Chan, C. H. (1976) Faecal steroids and bacteria and large bowel cancer mortality in Hong Kong by socio-economic indicators. *Brit. J. Cancer*, **34**, 191–198

Day, N. E. (1976) *A new measure of age-standardized incidence, the cumulated rate.* In: Waterhouse, J. A. H., Muir, C. S., Correa, P. & Powell, J., eds, *Cancer Incidence in Five Continents*, Vol. 3, Lyon, International Agency for Research on Cancer (*IARC Scientific Publications* **No. 15**), pp. 443–452

Day, N. E. & Simons, M. J. (1976) Disease susceptibility genes—their identification by multiple case family studies. *Tissue Antigens* (in press)

Desgranges, C. & de-Thé, G. (1977) *Presence of EBV-specific IgA in saliva of nasopharyngeal carcinoma (NPC) patients: their activity, origin and possible clinical value.* In: *Proceedings of the International Symposium on Etiology and Control of Nasopharyngeal Carcinoma (NPC), Kyoto, April 1977*, Lyon, International Agency for Research on Cancer (in press)

Desgranges, C., de-Thé, G., Ho, J. H. C. & Ellouz, R. (1977) Neutralizing EBV-specific IgA in throat washings of nasopharyngeal carcinoma (NPC) patients. *Int. J. Cancer*, **19**, 627–633

Desgranges, C., Lenoir, G., de-Thé, G., Seigneurin, J. M., Hilgers, J. & Dubouch, P. (1976) *In vitro* transforming activity of EBV. I. Establishment and properties of two EBV strains (M81 and M72) produced by immortalized *Callithrix jacchus* lymphocytes. *Biomedicine*, **25**, 349–352

Desgranges, C., Li, J. Y. & de-Thé, G. (1977) Secretory nature and tumorous origin of IgA antibodies to Epstein-Barr virus in saliva of nasopharyngeal carcinoma (NPC) patients. *Int. J. Cancer* (in press)

de-Thé, G. (1976) Epidémiologie et nature de l'association entre virus herpès (EBV et herpès simplex) et certaines tumeurs humaines. *Bull. Cancer*, **63**, 417–426

de-Thé, G. (1977) *Viruses as causes of some human tumours? Results and prospectives of the epidemiological approach.* In: Hiatt, H. H., Watson, J. D. & Winsten, J. A., eds. *The Origins of Human Cancer*, Cold Spring Harbor (*Cold Spring Harbor Cell Proliferation Series* **Vol. 4**) (in press)

de-Thé, G. (1977) Is Burkitt's lymphoma (BL) related to a perinatal infection by Epstein-Barr virus (EBV)? *Lancet*, **i**, 335–338

de-Thé, G. (1977) *Comparative EBV serological data and markers in nasopharyngeal carcinomas from high, intermediate and low risk areas.* In: *Proceedings of the International Symposium on Etiology and Control of Nasopharyngeal Carcinoma (NPC), Kyoto, April 1977*, Lyon, International Agency for Research on Cancer (in press)

de-Thé, G. & Lenoir, G. (1977) *Comparative diagnosis of Epstein-Barr virus related diseases: infectious mononucleosis, Burkitt's lymphoma and nasopharyngeal carcinoma.* In: Kurstak, E., ed., *Comparative Diagnosis of Viral Diseases*, New York, Academic Press (in press)

Drevon, C., Kuroki, T. & Montesano, R. (1977) *Microsome mediated mutagenesis of a Chinese hamster cell line by various chemicals.* In: *Proceedings of the International Mutagenesis Conference, July 1977* (in press)

Geser, A., Charnay, N., Day, N. E., de-Thé, G. & Ho, H. C. (1977) *Environmental factors in the etiology of nasopharyngeal carcinoma. Report of a case control study in Hong Kong.* In: *Proceedings of the International Symposium on Etiology and Control of Nasopharyngeal Carcinoma (NPC), Kyoto, April 1977*, Lyon, International Agency for Research on Cancer (in press)

Glaser, R., Lenoir, G., Desgranges, C. & de-Thé, G. (1977) Cell surface markers on epithelial/Burkitt hybrid cells superinfected with Epstein-Barr virus. *Cancer Res.* (in press)

Goh, E. H., Chan, S. H. & Simons, M. J. (1976) *Immunological aspects of nasopharyngeal carcinoma (NPC). IV. Mantoux responsiveness and patient survival.* In: Shanmugaratnam, K., Nambiar, R., Tan, K. K. & Chan, L. K. C., eds, *Liver Cancer: Cancer Problems in Asian Countries*, Singapore, pp. 290–292

Goh, E. H., Chan, S. H. & Simons, M. J. (1977) *Effect of levamisole on cell mediated immune responses in patients with nasopharyngeal carcinoma.* In: *Proceedings of the International Symposium on Etiology and Control of Nasopharyngeal Carcinoma (NPC), Kyoto, Japan, April 1977*, Lyon, International Agency for Research on Cancer (in press)

Griciute, L. (1976) *Les composés* N-*nitrosés: analyse et carcinogénicité éventuelle chez l'homme.* In: Rosenfeld, C. & Davis, W., eds, *Environmental Pollution and Carcinogenic Risks*, Lyon, International Agency for Research on Cancer (*IARC Scientific Publications* **No. 13**), pp. 375–386

Griciute, L. (1976) [*Comparative aspects of experimental lung tumours*] In: *Proceedings of the Conference on the 100th Anniversary of Experimental Cancer Research, Moscow, 1976* (in press)

Griciute, L. (1977) [Concerning the monitoring of chemical carcinogens in the human environment.] *Vop. Onkol.*, **23**, 49–53

Hawkins, B. R. & Simons, M. J. (1976) Blood group genetic studies in an urban Chinese population. *Human Heredity*, **26**, 441–453

Higginson, J. (1976) *Importance of environmental factors in cancer.* In: Rosenfeld, C. & Davis, W., eds, *Environmental Pollution and Carcinogenic Risks*, Lyon, International Agency for Research on Cancer (*IARC Scientific Publications* **No. 13**), pp. 15–24

Higginson, J. (1976) Future role of epidemiology in food safety evaluations. *Clin. Toxicology*, **9**, 691–708

Higginson, J. (1977) The role of the pathologist in environmental medicine and public health. *Amer. J. Pathol.*, **82**, 459–484

Higginson, J. (1977) The necessity for an international system of cancer surveillance and monitoring in selected environments. *Rev. Epidém. Santé publ.*, **25**, 179–184

Higginson, J. (1977) Primary cancer control in relation to community and individual responsibility. *Cancer Forum (Australia)* (in press)

Higginson, J. (1977) *The role of epidemiology in environmental cancer research.* In: *Proceedings of the AACR Symposium, Houston, 1975* (in press)

Higginson, J. (1977) Environmental carcinogens in modern society. *Vop. Onkol.* (in press)

Higginson, J. (1977) *Worldwide review of epidemiology of gastric cancer.* In: Hirayama, T., ed., *Epidemiology of Stomach Cancer: Key Questions and Answers*, WHO Collaborating Center Monograph, WHO Collaborating Center for Evaluation of Methods of Diagnosis and Treatment of Stomach Cancer, pp. 81–92

Higginson, J. & Jensen, O. M. (1977) *Epidemiological review of lung cancer in man.* In: Mohr, U., Schmähl, D. & Tomatis, L., eds, *Air Pollution and Cancer in Man*, Lyon, International Agency for Research on Cancer (*IARC Scientific Publications* **No. 16**), pp. 169–189

Higginson, J. & Muir, C. S. (1977) *Cancer epidemiology (updating).* In: Holland, J. F. & Frei, E., eds, *Cancer Medicine, Part IV*, Philadelphia, Lea & Fibiger (in press)

Higginson, J. & Muir, C. S. (1977) Détermination de l'importance des facteurs environnementaux dans le cancer humain: rôle de l'épidémiologie. *Bull. Cancer (Paris)* (in press)

IARC Intestinal Microecology Group (1977) Dietary fibre, transit-time, faecal bacteria, steroids and colon cancer in two Scandinavian populations. *Lancet* (in press)

Jensen, O. M. (1975) Lung cancer and smoking in Danish women. *Int. J. Cancer*, **15**, 954–961

Joint Iran/IARC Study Group (1977) Oesophageal cancer studies in the Caspian littoral of Iran: results of population studies—a prodrome. *J. nat. Cancer Inst.* (in press)

Kinnier-Wilson, L. M. & Muñoz, N. (1977) *Congenital cancer.* In: *Proceedings of the VIth Perugia Quadrennial International Conference on Cancer, Perugia, 1977* (in press)

Kirk, R. L., Blake, N. M., Serjeantson, S., Simons, M. J. & Chan, S. H. Genetic components in susceptibility to nasopharyngeal carcinoma (submitted for publication)

Kirk, R. L., Keats, B., Blake, N. J., McDermid, E. J., Ala, F., Karimi, M., Nickbin, B., Shabazi, H. & Kmet, J. (1977) Genes and people in the Caspian littoral of Iran. *Amer. J. phys. Anthrop.* (in press)

Kuroki, T., Drevon, C. & Montesano, R. (1977) Microsome-mediated mutagenesis in V79 Chinese hamster cells by various nitrosamines. *Cancer Res.*, **37**, 1044–1050

Kuroki, T., Drevon, C., Saint Vincent, L., Tomatis, L. & Montesano, R. (1977) *Studies on the use of liver parenchymal cells in* in vitro *carcinogenesis.* In: *Méchanismes d'Altération et de Réparation du DNA, Relations avec la Mutagenèse et la Cancérogenèse chimique*, Paris, Editions du CNRS, pp. 307–314

Lamelin, J. P., Revillard, J. P., Chalopin, J. M., Ho, J. H. C., Souissi, T., Schwaab, G. & de-Thé, G. (1977) Cold lymphototoxic antibodies in nasopharyngeal carcinoma. *Brit. J. Cancer*, **35**, 426–432

Lamelin, J. P., Revillard, J. P., Gabbiani, G. & de-Thé, G. (1977) *Autoantibodies (cold lymphocytotoxins and antiactin and antinuclear factors) in nasopharyngeal carcinoma patients.* In: *Proceedings of the International Symposium on Etiology and Control of Nasopharyngeal Carcinoma (NPC), Kyoto, April 1977*, Lyon, International Agency for Research on Cancer (in press)

Lamelin, J. P., Souissi, T., de-Thé, G. & Revillard, J. P. (1977) Lymphocyte subpopulations and mitogenic responses in nasopharyngeal carcinoma, prior to and after radiotherapy. *Int. J. Cancer* (in press)

Lamelin, J. P., Williams, E. H., Souissi, T., de-Thé, G. & Gabbiani, G. (1977) Smooth-muscle antibody in Burkitt's lymphoma and in nasopharyngeal carcinoma. *J. clin. exp. Immunol.*, **27**, 157–162

Lenoir, G. & de-Thé, G. (1977) *Epstein-Barr virus epithelial cell interaction and its implication in nasopharyngeal carcinoma etiology.* In: *Proceedings of the International Symposium on Etiology and Control of Nasopharyngeal Carcinoma (NPC), Kyoto, April 1977*, Lyon, International Agency for Research on Cancer (in press)

Likhachev, A., Margison, G. P. & Montesano, R. (1977) Alkylated purines in the DNA of various rat tissues after administration of 1,2-dimethylhydrazine. *Chem.-biol. Interact.*, **18**, 235–240

Linsell, C. A. & Higginson, J. (1976) *The geographical pathology of liver cancer.* In: Cameron, H. M., Linsell, C. A. & Warwick, G. P., eds, *Liver Cell Cancer*, Amsterdam, Excerpta Medica, pp. 1–16

Linsell, C. A. & Peers, F. G. (1977) Aflatoxin and liver cell cancer. *Trans. Roy. Soc. trop. Med. Hyg.* (in press)

Linsell, C. A. & Peers, F. G. (1977) *Field studies on liver cell cancer.* In: *The Origins of Human Cancer*, Cold Spring Harbor (*Cold Spring Harbor Cell Proliferation Series* **Vol. 4**) (in press)

Loprieno, N., Barale, R., Barnoncelli, S., Bartsch, H., Bronzetti, G., Cammellini, A., Corsi, C., Frezza, D., Nieri, R., Leporini, C., Rossellini, D. & Rossi, A. M. (1976) Induction of gene mutations and gene conversions by vinyl chloride metabolites in yeast. *Cancer Res.*, **36**, 253–257

MacLennan, R., Jensen, O. M., Mosbech, J. & Vuori, H. (1977) Diet, transit time, stool weight and colon cancer in two Scandinavian populations. *Amer. J. clin. Nutr.* (in press)

Malaveille, C. & Bartsch, H. (1977) *Short-term tests and mycotoxins.* In: *Proceedings of a Meeting on Mycotoxins in Foodstuffs, Paris, July 1976* (in press)

Malaveille, C., Kuroki, T., Sims, P., Grover, P. L. & Bartsch, H. (1977) Mutagenicity of isomeric diol-epoxides of benzo[*a*]pyrene and benz[*a*]anthracene in *S. typhimurium* TA 98 and TA 100 in V79 Chinese hamster cells. *Mutation Res.*, **44**, 313–326

Malaveille, C., Planche, G. & Bartsch, H. (1977) Factors for efficiency of the *Salmonella*/microsome mutagenicity assay. *Chem.-biol. Interact.*, **17**, 129–136

Malaveille, C., Tierney, B., Grover, P. L., Sims, P. & Bartsch, H. (1977) High microsome-mediated mutagenicity of the 3,4-dihydrodiol of 7-methylbenz[*a*]anthracene in *S. typhimurium* TA 98. *Biochem. biophys. Res. Commun.*, **75**, 427–433

Margison, G. P., Brésil, H., Margison, J. M. & Montesano, R. (1976) Effect of chronic administration of dimethylnitrosamine on the excision of 0^6-methylguanine from rat liver DNA. *Cancer Lett.*, **2**, 79–86

Margison, G. P., Margison, J. M. & Montesano, R. (1977) Accumulation of 0^6-methylguanine in non-target tissue deoxyribonucleic acid during chronic administration of dimethylnitrosamine *Biochem. J.*, **165**, 463–468

Menezes, J., Seigneurin, J. M., Patel, P., Bourkas, A. & Lenoir, G. (1977) Presence of Epstein-Barr virus (EBV) receptors, but absence of virus penetration in cells of an EBV genome-negative human lymphoblastoid T line (MOLT 4). *J. Virol.*, **22**, 816–821

Mohr, U., Schmähl, D. & Tomatis, L., eds (1977) *Air Pollution and Cancer in Man*, Lyon, International Agency for Research on Cancer (*IARC Scientific Publications* **No. 16**)

Montesano, R. (1976) Cancérogénicité des composés *N*-nitrosés. *Ann. Nutr. Alim.*, **30**, 867–871

Montesano, R., Bartsch, H. & Tomatis, L. (1977) *Carcinogenesis and mutagenesis.* In: *Principles and Guidelines for the Evaluation of the Toxicity of Chemicals*, Geneva, World Health Organization (in press)

Montesano, R., Drevon, C., Kuroki, T., Saint Vincent, L., Handleman, S., Sanford, K. K., DeFeo, D. & Weinstein, I. B. Tests for malignant transformation of liver cells in culture: cytology, growth in soft agar, and production of plasminogen activator (submitted for publication)

Montesano, R., Margison, G. P. & Likhachev, A. (1977) *The possible role of nucleic acids alkylation in the organ specificity of carcinogens.* In: Farber, E., *et al.*, eds, *Physiopathology of Carcinogenesis in the Digestive Organs*, Tokyo, University of Tokyo Press and Baltimore, University Park Press (in press)

Montesano, R. & Tomatis, L. (1977) Considerations on legislation concerning chemical carcinogens in several industrialized countries. *Cancer Res.*, **37**, 310–316

Montesano, R. & Tomatis, L. (1976) In vitro *neoplastic transformation of rat liver cells by dimethylnitrosamine and* N-*methyl*-N′-*nitro*-N-*nitrosoguanidine.* In: *Seminari Biologici della Facoltà di Medicina 1973–1974*, Milan, Vita e Pensiero, Università Cattolica, pp. 152–161

Montesano, R. & Tomatis, L. (1977) Les cancérogènes chimiques. *Lyon méd.*, **238**, 107–117

Muenz, L. R., Sizaret, P., Bernard, C., Chayvialle, J. A., Kithier, K., Kohn, J., Krebs, B. P., Lehmann, F. G., Martel, N., Masseyeff, R., McIntire, R., Nishi, S., Orr, A. H., Princler, G. L., Reynaud, S. & Waldmann, T. (1977) Results of the second international study on the WHO alpha-foetoprotein standard. *J. biol. Stand.* (in press)

Muir, C. S. (1977) Predictive value of cancer statistics. *J. environm. Path. Toxicol.* (in press)

Muir, C. S., MacLennan, R., Waterhouse, J. A. H. & Magnus, K. (1976) *Feasibility of monitoring populations to detect environmental carcinogens.* In: Rosenfeld, C. & Davis, W., eds, *Environmental Pollution and Carcinogenic Risks*, Lyon, International Agency for Research on Cancer (*IARC Scientific Publications* **No. 13**), pp. 279–294

Muir, C. S. & Nectoux, J. (1977) The role of the cancer registry. *J. nat. Cancer Inst. Monogr.* (in press)

Muir, C. S. & Wagner, G., eds (1977) *Directory of On-Going Research in Cancer Epidemiology*, Lyon, International Agency for Research on Cancer (*IARC Scientific Publications* **No. 17**) (in press)

Muir, C. S., Waterhouse, J. A. H., Correa, P. & Powell, J., eds (1976) *Cancer Incidence in Five Continents, Vol. III*, Lyon, International Agency for Research on Cancer (*IARC Scientific Publications* **No. 15**)

Muñoz, N. (1976) Geographical distribution of pediatric tumors. *Tumori*, **62**, 145–156

Muñoz, N. (1976) Prenatal exposure and carcinogenesis. *Tumori*, **62**, 157–162

Muñoz, N., Davidson, R. J. L., Witthoff, B., Ericsson, J. E. & de-Thé, G. Hodgkin's disease preceded by infectious mononucleosis (submitted for publication)

Muñoz, N., Matko, I. & Kmet, J. Intestinal metaplasia and gastric cancer. A cohort study (submitted for publication)

Napalkov, N. P., Tomatis, L., Likhachev, A. Y. & Kolodin, V. I. (1977) Tumours in BALB/c mice after transplacental exposure to *N*-nitrosoethylurea in progenesis. *Vop. Onkol.*, **23**, 3, 41–51

Napalkov, N. P., Tomatis, L., Likhachev, A. Y. & Kolodin, V. I. (1977) Blastomogenesis in mice: post-natal effect of *N*-nitrosoethylurea and X-ray irradiation. *Vop. Onkol.*, **23**, 4, 49–57

Napalkov, N. P., Tomatis, L., Likhachev, A. Y. & Kolodin, V. I. (1977) The effect of *N*-nitrosoethylurea dosage and post-natal roentgen irradiation on transplacental blastomogenesis in mice. *Vop. Onkol.*, **23**, 5, 66–70

Nyandieka, H. S. & Levin, A. G. (1976) Distribution of glyoxalase activity of lymphocytes in cancerous tissues. *E. Afr. J. med. Res.*, **3**, 65–68

Olweny, C. L. M., Atine, I., Kaddu-Mukasa, A., Owor, R., Anderson, M., Klein, G. & de-Thé, G. (1977) Epstein-Barr virus genome studies in Burkitt and non-Burkitt lymphomas in Uganda. *J. nat. Cancer Inst.*, **58**, 1191–1196

Peers, F. G. & Linsell, C. A. (1976) Dietary aflatoxins and human primary liver cancer. *Ann. Nutr. Alim.* (in press)

Pignatelli, B., Castegnaro, M. & Walker, E. A. (1976) *Effects of gallic acid and of ethanol on formation of nitrosodiethylamine.* In: Walker, E. A., Bogovski, P. & Griciute, L., eds., *Environmental N-Nitroso Compounds—Analysis and Formation*, Lyon, International Agency for Research on Cancer (*IARC Scientific Publications* **No. 14**), pp. 173–178

Ponomarkov, V. & Mackey, L. J. (1976) Tumours of the liver and biliary system. *Bull. Wld Hlth Org.*, **53**, 187–194

Ramioul, L. & Tuyns, A. J. (1977) La distribution géographique des cancers du tube digestif en Belgique. Inégale répartition de la mortalité par cancer de l'estomac et par cancer du rectum. *Acta gastro-ent. Belg.*, **40**, 129–147

Rosenfeld, C. & Davis, W., eds (1976) *Environmental Pollution and Carcinogenic Risks*, Lyon, International Agency for Research on Cancer (*IARC Scientific Publications* **No. 13**)

Rosenfeld, C., Goutner, A., Venuat, A. M., Choquet, C., Pico, J. L., Doré, J. F., Liabeuf, A., Durandy, A., Desgranges, C. & de-Thé, G. (1977) An effective human leukaemic cell line: Reh. *Europ. J. Cancer*, **13**, 377–379

Saracci, R. (1976) Survival after myocardial infarction. *Lancet*, **i**, 1020

Saracci, R. (1977) *Epidemiology of lung cancer in Italy*. In: Mohr, U., Schmähl, D. & Tomatis, L., eds, *Air Pollution and Cancer in Man*, Lyon, International Agency for Research on Cancer (*IARC Scientific Publications* **No. 16**), pp. 205–215

Saracci, R. (1977) Asbestos and lung cancer: analysis of the epidemiological evidence on the asbestos-smoking interaction. *Int. J. Cancer*, **20**, 323–331

Seigneurin, J. M., Desgranges, C., Lavoué, M. F. & de-Thé, G. (1976) Herpes simplex-Raji (A44), a new cell line for serologic testing by immunofluorescence. *J. Immunol.*, **117**, 950–952

Seigneurin, J. M., Vuillaume, M., Lenoir, G. & de-Thé, G. Replication of Epstein-Barr virus: ultrastructural and immunofluorescent studies of P_3HR1 superinfected Raji cells (submitted for publication)

Shabad, L. & Griciute, L. (1976) Le centenaire de la première transplantation de tumeur. *Bull. Cancer*, **63**, 305–308

Shabad, L. M. & Ponomarkov, V. I. (1976) Mstislav Novinski, pioneer of tumour transplantation. *Cancer Lett.*, **2**, 1–3

Siemiatycki, J. (1977) Mantel's space-time clustering statistic: computing higher moments and a comparison of various data transforms. *J. Statist. Comput. Simul.* (in press)

Simarak, S., de Jong, U. W., Breslow, N., Dahl, C. J., Ruckphaopunt, K., Scheelings, P. & MacLennan, R. (1977) Cancer of the oral cavity, pharynx/larynx and lung in North Thailand: case-control study and analysis of cigar smoke. *Brit. J. Cancer*, **36**, 130–140

Simons, M. J. (1976) *Immunodeficiency to hepatitis B virus infection and hepatocellular carcinoma*. In: Shanmugaratnam, K., Nambiar, R., Tan, K. K. & Chan, L. K. C., eds, *Liver Cancer: Cancer Problems in Asian Countries*, Singapore, pp. 61–66

Simons, M. J. & Amiel, J. L. (1977) *HLA and malignant diseases*. In: Dausset, J. & Svejgaard, A., eds, *HLA and Disease*, Copenhagen, Munksgaard (in press)

Simons, M. J. & Chan, S. H. (1977) *HLA and nasopharyngeal carcinoma*. In: *Proceedings of the International Symposium on Etiology and Control of Nasopharyngeal Carcinoma (NPC), Kyoto, Japan, April 1977*, Lyon, International Agency for Research on Cancer (in press)

Simons, M. J., Chan, S. H., Day, N. E., Wee, G. B. & Shanmugaratnam, K. (1977) *HLA and nasopharyngeal cancer*. In: Murphy, G. P., Cohen, E., Fitzpatrick, J. E. & Pressman, D., eds, *HLA and Malignancy*, New Ork, Alan R. Liss, Inc., pp. 145–148

Simons, M. J., Chan, S. H., Wee, G. B. & Mathews, J. (1977) *HLA and hepatocellular carcinoma in Singapore Chinese.* In: *Proceedings of the Hepatitis Symposium, Tokyo, Japan, 1976* (in press)

Simons, M. J. & Day, N. E. (1977) HLA patterns and NPC. *J. nat. Cancer Inst. Monogr.* (in press)

Simons, M. J., Wee, G. B., Chan, S. H., Shanmugaratnam, K., Day, N. E. & de-Thé, G. (1976) Immunogenetic aspects of nasopharyngeal carcinoma associated with the Chinese-related HLA profile (HL-A2, Singapore 2). *J. nat. Cancer Inst.*, **57**, 977–980

Simons, M. J., Wee, G. B., Shanmugaratnam, K., Goh, E. H., Ho, J. H. C., Chau, J. C. W., Darmalingam, S., Prasad, U., Betuel, H., Day, N. E. & de-Thé, G. (1977) *NPC and histocompatibility antigens.* In: *Proceedings of the International Symposium on Etiology and Control of Nasopharyngeal Carcinoma (NPC), Kyoto, Japan, April 1977*, Lyon, International Agency for Research on Cancer (in press)

Sizaret, P. & Anderson, S. G. (1976) The international reference preparation for alpha-foetoprotein. *J. biol. Stand.*, **4**, 149

Sizaret, P., Martel, N. & Reynaud, S. (1977) Mean alpha-foetoprotein values of 1,333 males over 15 years by age groups. *Digestion*, **15**, 97–103

Soeripto, Jensen, O. M. & Muir, C. S. (1977) Cancer in Yogyakarta, Indonesia. A relative frequency study. *Brit. J. Cancer*, **36**, 141–148

Sohier, R., Charnay, B. & de-Thé, G. A sero-epidemiological study on Epstein-Barr virus in 360 families (submitted for publication)

Steel, C. M., Ennis, M., Levin, A. G. & Wasunna, A. (1977) The mitogenic response of cryopreserved human lymphocytes in a microculture system. *Cytobios* (in press)

Tomatis, L. (1976) *Extrapolation of test results to man.* In: Rosenfeld, C. & Davis, W., eds, *Environmental Pollution and Carcinogenic Risks*, Lyon, International Agency for Research on Cancer (*IARC Scientific Publications* **No. 13**), pp. 261–262

Tomatis, L. (1977) *Validity and limitations of long-term experimentation in cancer research.* In: Mohr, U., Schmähl, D. & Tomatis, L., eds, *Air Pollution and Cancer in Man*, Lyon, International Agency for Research on Cancer (*IARC Scientific Publications* **No. 16**), pp. 299–307

Tomatis, L. (1977) *The value of long-term testing for the implementation of primary prevention.* In: *The Origins of Human Cancer*, Cold Spring Harbor (*Cold Spring Harbor Cell Proliferation Series* **Vol. 4**) (in press)

Tomatis, L. Chemical carcinogens: animals and man (submitted for publication)

Tomatis, L., Ponomarkov, V. & Turusov, V. (1977) Effects of ethylnitrosourea administration during pregnancy on three subsequent generations of BDVI rats. *Int. J. Cancer*, **19**, 240–248

Tuyns, A. J. (1977) *Relation alcool-cancer.* In: *Les Boissons Alcooliques, Situation Actuelle et Recherches Récentes*, Paris, La Documentation Française, pp. 66–72

Tuyns, A. J. & Audigier, J. C. (1976) Double wave cohort increase for œsophageal and laryngeal cancer in France in relation to reduced alcohol consumption during the Second World War. *Digestion*, **14**, 197–208

Tuyns, A. J., Jensen, O. M. & Péquignot, G. (1977) Le choix difficile d'un bon groupe de témoins dans une enquête rétrospective. *Rev. Epid. Santé publ.*, **25**, 67–84

Tuyns, A. J. & Péquignot, G. (1977) Alcool et cancer de l'œsophage. *Cah. Nutr. Diét.*, **XII**, 63–66

Tuyns, A. J., Péquignot, G. & Jensen, O. M. (1977) Le cancer de l'œsophage en Ille-et-Vilaine en fonction des niveaux de consommation d'alcool et de tabac. Des risques qui se multiplient. *Bull. Cancer*, **64**, 45–60

Wagoner, J. K., Infante, P. F. & Saracci, R. (1976) Vinyl chloride and mortality. *Lancet*, **ii**, 194–195

Walker, E. A., Bogovski, P. & Griciute, L., eds (1976) *Environmental* N-*Nitroso Compounds—Analysis and Formation*, Lyon, International Agency for Research on Cancer (*IARC Scientific Publications* **No. 14**)

Walker, E. A. & Castegnaro, M. (1976) *New data on collaborative studies on analysis of volatile nitrosamines.* In: Walker, E. A., Bogovski, P. & Griciute, L., eds, *Environmental* N-*Nitroso Compounds—Analysis and Formation*, Lyon, International Agency for Research on Cancer (*IARC Scientific Publications* **No. 14**), pp. 77–83

Walker, E. A. & Castegnaro, M. (1976) *Problems in trace analysis of nitrosamines in foodstuffs.* In: *Proceedings of the 7th International Materials Research Symposium, Gaithersburg, Md., October 1974*, National Bureau of Standards Special Publication **No. 422**, pp. 727–736

Waterhouse, J. A. H., Muir, C. S., Correa, P. & Powell, J., eds (1976) *Cancer Incidence in Five Continents, Vol. III*, Lyon, International Agency for Research on Cancer (*IARC Scientific Publications* **No. 15**)

Zardi, L., Saint Vincent, L., Barbin, A., Montesano, R. & Margison, G. (1977) Effect of split doses of *N*-methyl-*N*-nitrosourea on DNA repair synthesis in cultured mammalian cells. *Cancer Lett.*, **3**, 183–188

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