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Studies on Environmental Chemical Carcinogenesis in Japan

TAKASHI SUGIMURA

The historical background of studies in Japan on chemical carcinogenesis from environmental sources is described from personal experience.

IN 1915, YAMAGIWA FIRST SUCCEEDED IN PRODUCING CANcers in experimental animals at Tokyo Imperial University. This opened the door for studies of environmental chemical carcinogenesis throughout the world. He and his co-worker Ichikawa repeatedly applied coal tar that had been dissolved in benzene to the ears of rabbits. After persistent experimentation, squamous cell carcinoma–like tumors were produced (*1*). Yamagiwa was inspired (*1*) by Virchow's irritation theory of carcinogenesis and by Pott's observation of the high incidence of scrotum cancers in chimney sweeps. This was followed by the work of Tsutsui (*2*), of Chiba Medical College, who produced cancers in a shorter time by painting coal tar on the skin of mice.

In England, scientists under the direction of Kennaway fractionated coal tar into the active compounds that caused cancer with Tsutsui's method as a bioassay. The compound 1,2,5,6-dibenzanthracene was synthesized as the first pure chemical substance capable of producing mouse skin cancers in 1930 (*3*). Benzo[*a*]pyrene was also isolated by the same group from coal tar in 1932 (*4*). The scientific atmosphere in Japan at Yamagiwa's time did not encourage him to pursue collaborative work with scientists from different disciplines. Nevertheless, his contribution in demonstrating the first man-made cancer in

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animals was internationally accepted as a great milestone in the studies on environmental carcinogenesis.

The second major breakthrough in the study of chemical carcinogenesis was made in 1932, again in Japan. Sasaki, of the Sasaki Institute, spent years in Fischer's laboratory and concentrated his efforts on the organotropism of chemicals. Under the guidance of Sasaki, Yoshida (*5*) was successful in producing hepatomas in rats fed a mixture of rice with *o*-aminoazotoluene in oil, a diet supplemented occasionally with vegetables and dried fish. Soon after, Kinoshita, at the Osaka Imperial University (*6*), discovered that 4-dimethylaminoazobenzene (DAB) was a much more potent hepatocarcinogen than *o*-aminoazotoluene. DAB and its derivative 3'-methyl-DAB have been widely used in experimental hepatocarcinogenesis studies throughout the world (*7*).

Studies of Chemical Carcinogenesis in Japan During World War II

Nakahara, at the Cancer Institute of the Japanese Foundation of Cancer Research, observed that the addition of dried yeast powder to the DAB diet inhibited hepatocarcinogenesis in rats. This inhibition was caused by breakdown of the dye by azo reductase, a flavoprotein. This was one of the first demonstrations that nutrients are important modulators of chemical carcinogenesis. A paper by Nakahara *et al.* (*8*) appeared in English in the journal *Gann* (meaning "cancer" in Japanese) which was founded by Yamagiwa in 1907, but did not draw much attention internationally. *Gann*, compiled in Japan and printed in English, remained a poor medium for distributing information throughout the world. As a result, Japanese scientists tended to publish their best quality works in journals published in the United States and Europe to reach a wider readership. Recently *Gann* changed its title to the *Japanese Journal of Cancer Research* and invited guest editors from overseas with the aim of encouraging rapid international exchange of information.

Discovery of the Carcinogenicity of 4-Nitroquinoline 1-Oxide

In 1957, Nakahara and co-workers (*9*) discovered the carcinogenicity of 4-nitroquinoline 1-oxide (4NQO), which has mutagenicity toward microbes. Most typical carcinogens such as benzo[*a*]pyrene are not mutagenic per se in microbes. Most of them are activated metabolically by mammalian enzymes to form products that react readily with DNA, but cannot be activated by enzymes in microbes. We found that 4NQO is converted to 4-hydroxyaminoquinoline 1-oxide (4HAQO) by reduced nicotinamide adenine dinucleotide phosphate [NAD(P)H] oxidoreductase (*10*). 4HAQO and its ester forms produced by subsequent metabolism can bind with DNA. Interestingly, microbes are as well supplied with the enzymes for 4NQO metabolism as are mammalian cells. The availability of 4NQO, which was first synthesized by Ochiai of Tokyo University facilitated collaboration between Japanese cancer researchers and microbial geneticists. We reported the formation of adducts with 4NQO derivatives and DNA bases, strand scission of DNA by 4HAQO, and formation of the radical and hydrogen peroxide in the presence of oxygen from 4HAQO (*10*). Furthermore, it was noted that ultraviolet (UV)-sensitive bacterial strains were also 4NQO-sensitive (*11*). A parallel between UV and 4NQO sensitivities was also shown in cell lines derived from patients with xeroderma pigmentosum (*11*). Reports from other Japanese and foreign laboratories have been summarized in a review and a monograph (*12*).

Carcinogenicity of the Mutagen MNNG and the Food Additive AF-2

Schoental (*13*) and our group (*14*) reported in 1966 that *N*-methyl-*N'*-nitro-*N*-nitrosoguanidine (MNNG), a potent bacterial mutagen, was carcinogenic in rodents. Rats given MNNG in drinking water developed a high incidence of typical adenocarcinomas in the glandular stomach (*15*). This experimental model is still the most widely used system for the study of stomach cancer available today.

Soon afterward Japanese cancer researchers had readily adopted the Ames bacterial mutagen assay to detect carcinogens (*16*, *17*). This assay was very important in the analysis of AF-2 [2-(2-furyl)-3-(5-nitro-2-furyl)acrylamide], an efficient food preservative widely used in Japan since 1965. AF-2 had been reported to be noncarcinogenic; however, AF-2 was found to be mutagenic in *Escherichia coli* WP2 by the working groups of Kada (*18*), of the National Institute of Genetics, and Kondo (*19*), from Osaka University, and by us (*20*). It was later shown to be carcinogenic in rodents, and therefore its usage was legally prohibited in 1974 (*21*). One of the problems that we encountered was that AF-2 was not mutagenic to *Salmonella typhimurium* strains TA1538 and TA1535 (*20*), the original strains used in the Ames method. By including an R-factor plasmid in these two strains, Ames developed *Salmonella* strains TA98 and TA100; these strains have been more useful for the detection of environmental carcinogens in general, as well as AF-2 (*17*). Establishment of Environmental Panels under the United States–Japan Cooperative Medical Science Program between the National Institutes of Health and the Japanese Ministry of Health and Welfare provided an efficient vehicle for information exchange between the two nations.

Mutagens and Carcinogens in Cooked Food

Accounts of food additive carcinogenicity provided strong motivation for us to study the mutagenic activity of chemicals in our daily foods and to look for new environmental carcinogens (*22*, *23*). We have some reservations about the use of short-term assays for mutagenicity or carcinogenicity for regulatory purposes. The compound quercetin, a common flavonoid in vegetables and fruits, is mutagenic but noncarcinogenic in mice, rats, and hamsters (*24*). However, by means of microbial assays for mutagenicity we discovered environmental mutagens in pyrolysates or charred material of amino acids, proteins, and proteinous foods as well as in ordinarily cooked foods. All of these substances were later proven to be carcinogenic to mice or rats or both (*25*, *26*).

By means of a close collaboration among scientists from diverse disciplines [including Kosuge (*26*) of the Shizuoka College of Pharmacy], a series of heterocyclic amines were isolated quickly from pyrolysates and ordinarily cooked food. Their chemical names, mutagenicity, and carcinogenicity are listed in Table 1 and their structures are shown in Fig. 1 (*25*, *26*). Swedish and Japanese scientists demonstrated that creatine and creatinine are precursors for IQ, MeIQx, 4,8-DiMeIQx and 7,8-DiMeIQx (*27*). These heterocyclic amines are metabolized to their hydroxyamino derivatives by cytochrome P-450's (*28*). They may bind to DNA and cause strand scissions (*28*). After esterification, they become more active. Chemically synthesized *N-O*-acetyl-Glu-P-1 can form 2-(C^8-guanyl)-amino-6-methyldipyrido[1,2-*a*:3',2'-*d*]imidazole, a DNA base adduct as revealed by Shudo of Tokyo University and co-workers (*29*). Pentachlorophenol, an inhibitor of sulfate ester formation, suppressed their mutagenic potentials (*30*). Ingested heterocyclic amines are metabolized and excreted into bile, and probably converted further to active substances by microbial enzymes in the gut

Table 1. Heterocyclic amines isolated from cooked foods and pyrolysates of amino acids and proteins (*26, 27*). Mutagenicities are shown as revertants of *Salmonella typhimurium* per microgram of chemical obtained with metabolic activation. Carcinogenicities are expressed for target organs in mice and rats that were fed heterocyclic amines mixed in the diet.

Chemical name	Abbreviation	Year of discovery	Mutagenicity		Carcinogenicity	
			TA98	TA100	Mouse	Rat
2-Amino-3-methylimidaz[4,5-*f*]quinoline	IQ	1980	433,000	7,000	Liver, forestomach, lung; 1983	Liver, intestine Zymbal gland, clitoral gland, skin; 1984
2-Amino-3,4-dimethylimidazo[4,5-*f*]quinoline	MeIQ	1980	661,000	30,000	Liver, forestomach; 1985	Ongoing
2-Amino-3,8-dimethylimidazo[4,5-*f*]quinoxaline	MeIQx	1981	145,000	14,000	Liver; 1986	Ongoing
2-Amino-3,4,8-trimethylimidazo[4,5-*f*]quinoxaline	4,8-DiMeIQx	1985	183,000	8,000	Not tested	Not tested
2-Amino-3,7,8-trimethylimidazo[4,5-*f*]quinoxaline	7,8-DiMeIQx	1984	163,000	9,900	Not tested	Not tested
3-Amino-1,4-dimethyl-5*H*-pyrido[4,3-*b*]indole	Trp-P-1	1977	39,000	1,700	Liver; 1981	Liver; 1985
3-Amino-1-methyl-5*H*-pyrido[4,3-*b*]indole	Trp-P-2	1977	104,200	1,800	Liver; 1981	Liver; 1981
2-Amino-6-methyldipyrido[1,2-*a*:3′,2′-*d*]imidazole	Glu-P-1	1978	49,000	3,200	Liver, blood vessel; 1982	Liver, intestine, Zymbal gland, clitoral gland; 1984
2-Aminodipyrido[1,2-*a*:3′,2′-*d*]imidazole	Glu-P-2	1978	1,900	1,200	Liver, blood vessel; 1982	Liver, intestine, Zymbal gland, clitoral gland; 1984
2-Amino-9*H*-pyrido[2,3-*b*]indole	AαC	1978	300	20	Liver, blood vessel; 1982	Ongoing
2-Amino-3-methyl-9*H*-pyrido[2,3-*b*]indole	MeAαC	1978	200	120	Liver, blood vessel; 1982	Ongoing

(*26, 31*). After the ingestion of fried ground beef, mutagenic heterocyclic amines, possibly MeIQx and its metabolites, were recovered from human feces and urine (*31*). Another report indicated that people who frequently eat charred fish have a higher incidence of gastric cancer (*32*). Sensitive and simple methods for determining the concentration of these newly found heterocyclic amines became available (*33*). The use of specific antibodies against modified amino acid residues as well as modified DNA bases would provide valuable information on in vivo human exposure. The actual content of these heterocyclic amines in ordinary foods is usually low (*26*).

In addition to these heterocyclic amines, many new mutagens/carcinogens are present in our environment. Dinitropyrenes (DNP's) are extremely potent mutagens (*34*) and are carcinogenic in mice and rats (*35*). They were first found in the toner of photocopy machines by Rosenkranz *et al.* (*34*). DNP's have been removed from toner, but they are still present in the air as pollution from the exhaust of combustion engines. Recently Ohnishi *et al.* (*36*) of Tokushima University found a mononitropyrene and a tiny amount of dinitropyrene in the charred portion of chicken meat that had been broiled over an open flame.

When we began this study, I often recalled a talk with Yoshida about the fact that a century ago we already had a high frequency of stomach cancer in autopsy records at Tokyo Imperial University Hospital. We speculated that causative agents and conditions for such a high incidence of stomach cancer among us must somehow be related to our traditional life-style. We have realized that mutagens and carcinogens are not only chemicals produced by modern industries but also chemicals produced in daily foods as a result of cooking and broiling (*25, 26*). Intensive surveys by Doll and Peto (*37*), Higginson and Muir (*38*), and Wynder and Gori (*39*) suggested that life-style, including dietary habits, is one of the most important factors responsible for the different incidences of cancer in various organs of the body in different countries of the world.

Mutagen Precursors

During our research we were informed by Tannenbaum and colleagues (*40*) that the high incidence of stomach cancer in Colombia might somehow be associated with nitrate/nitrite in well water and nitrosable mutagen precursors derived from broad beans. They identified 4-chloro-6-methoxyindole as a mutagen precursor (*40*). We investigated Japanese foods and found that Chinese cabbage had indole derivatives that serve as new mutagen precursors. One of them was indole-3-acetonitrile, which is converted to a direct-acting mutagen, 1-nitrosoindole-3-acetonitrile, after nitrite treatment (*41*). This vegetable also contained other mutagen precursors, namely, 4-methoxyindole-3-acetonitrile and 4-methoxyindole-3-aldehyde (*41*).

Two isomers of 1-methyl-1,2,3,4-tetrahydro-β-carboline-3-carboxylic acid (MTCA) and tyramine were isolated from soy sauce as mutagen precursors (*41*). MTCA's and tyramine were converted to direct-acting mutagens after nitrite treatment (*41*). A direct mutagen from tyramine was proven to be 4-(2-aminoethyl)-6-diazo-2,4-cyclohexadienone; administration of this chemical to rats in their drinking water resulted in the production of squamous cell carcinomas in the oral cavity (*41*). Toth and co-workers (*42*) reported carcinogenicity of a structurally related chemical, obtained from a mushroom that induces adenocarcinomas in the stomach of mice.

The concentration of nitrite required to form direct-acting mutagens from mutagen precursors is often higher than physiological amounts. However, an endogenous nitrite production associated with oxidation in activated macrophages would suggest a possibility

of the presence of a higher local concentration of nitrite (*43*). These research areas remain exciting challenges for understanding the basis of the reality and diversity of human cancer risk.

New Tumor Promoters

The carcinogenic process seems more complicated than the classical two-step concept. Nevertheless, tumor promotion has been intensively studied by means of 12-*O*-tetradecanoylphorbol-13-acetate (TPA) (*44*). It was our desire to discover new tumor promoters and open our own avenue to explore tumor promotion. In our laboratory, a group headed by Fujiki (*45*) found many new tumor promoters—the teleocidin class of compounds from *Streptomyces* and a blue-green alga, *Lyngbya majuscula*, and the aplysiatoxin class from a variant of the same blue-green alga. These discoveries were initiated by a serendipitous event, a brief conversation with Umezawa of the Institute of Microbial Science, at a meeting of a governmental committee totally unrelated to chemical carcinogenesis. He spoke about the fact that in an industrial process that uses *Streptomyces* to produce an antibiotic, workers were annoyed by an occasional irritation of the skin.

The irritant was dihydroteleocidin B (*46*), which was previously isolated and given to us by Takashima and Sakai of the Fujisawa Pharmaceutical Industry. Teleocidin is composed of four isomers of teleocidin B (molecular weight 451) and two isomers of teleocidin A (molecular weight 437) (*45*). Lyngbyatoxin A, which was first isolated from *Lyngbya majuscula* by Moore and colleagues (*47*), was identical to one of two stereoisomers of teleocidin A (*45*). Teleocidin has a lactam ring structure of *N*-methyl-L-valyl-L-tryptophanol attached to a hydrophobic side chain (Fig. 2). Olivoretins A, B, and C were recently isolated from *Streptoverticillium* by a group headed by Sakai of Chiba University (*48*). Olivoretins have a methoxy group on the ring and are inactive for tumor promotion, while *des-O*-methylolivoretins are as active as teleocidin, indicating that the free hydroxy group at C-14 is important (*45*). Aplysiatoxin was originally isolated by Kato and Scheuer from the midgut gland of a sea hare (*49*). Aplysiatoxin has the structure of acetogenic phenolic bislactone with a bromine atom (Fig. 2). Debromoaplysiatoxin without a bromine and bromoaplysiatoxin with two atoms of bromine also retain tumor-promoting activity (*45*).

The tumor-promoting activities of these two classes of tumor promoters, teleocidin and aplysiatoxin, are similar to that of TPA (*45*). Despite the absence of any apparent structural similarity, [except for some common features depicted by computer-graphic analysis (*50*)] they share the same receptors with TPA on cell membrane surfaces (*45*). New compounds and TPA activate protein kinase C (*45*). Over 60 biological phenomena shown by TPA in in vivo and in vitro systems were also observed in exactly the same form with both teleocidin and aplysiatoxin, for example, prostaglandin E_2 production and induction of differentiation of HL-60 cells (*45*). We called these three classes of tumor promoters the TPA-type tumor promoters.

Ito of Kyoto University and others (*51*) reported the induction of Epstein-Barr virus antigen from Raji cells by TPA, and this observation was also confirmed with the new tumor promoters. It was suggested that the exposure to various TPA-type tumor promoters in traditional herb remedies may be partly related to the incidences of nasopharyngeal cancers in Southeast Asia and of Burkitt lymphomas in Africa.

There are many other types of tumor promoters that are not mediated through the receptor used by TPA, teleocidin, and aplysiatoxin. Examples found in our laboratories are palytoxin from a coelenterate and thapsigargin from a plant (*45*). They cannot

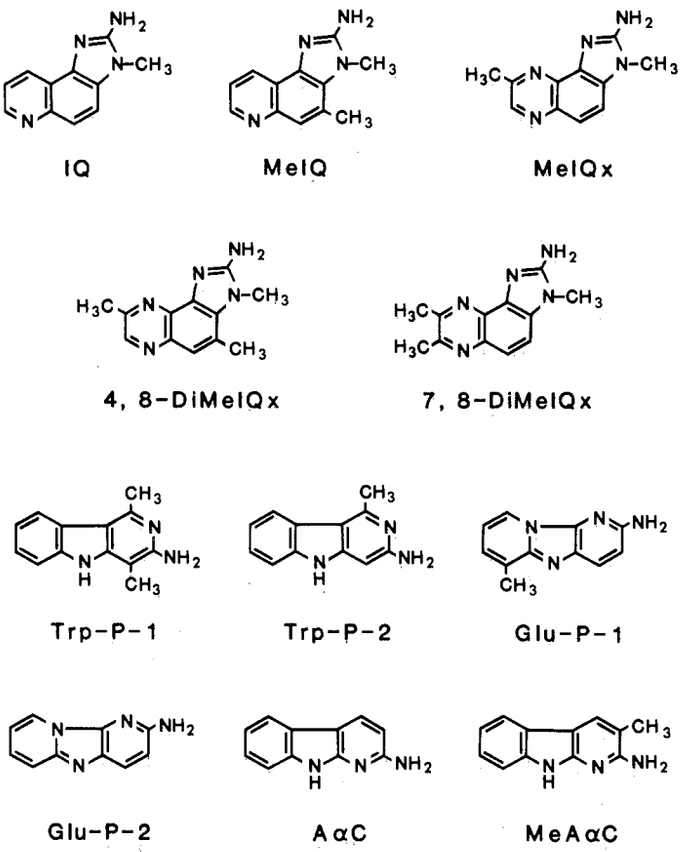

Fig. 1. Structures of heterocyclic amines isolated from cooked foods and pyrolysates of amino acids and proteins.

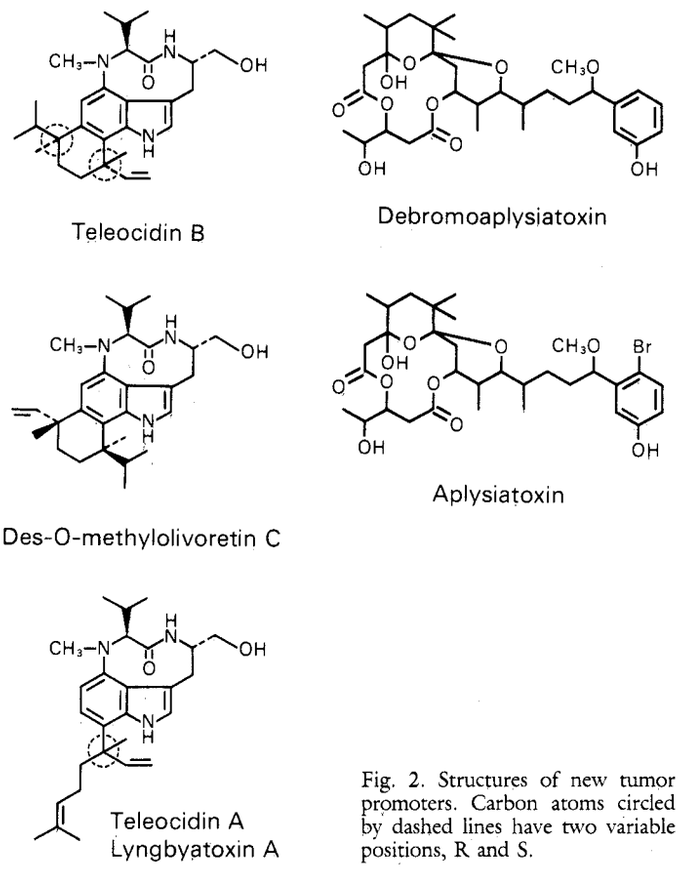

Fig. 2. Structures of new tumor promoters. Carbon atoms circled by dashed lines have two variable positions, R and S.

induce ornithine decarboxylase (ODC), produce adhesion of HL-60 cells, or activate protein kinase C, but they can cause skin irritation and promote tumor formation on the skins of mice to which a limited amount of *7,12*-dimethyl benzanthracene (DMBA) was applied previously. We termed this category of tumor promoters as non-TPA–type tumor promoters (*45*).

Takahashi of the National Institute of Hygienic Sciences and our group suggested that sodium chloride promoted stomach carcinogenesis of rats when administered with MNNG (*52*). A higher intake of sodium chloride has been shown to be related to higher incidence of stomach cancer and of intestinal metaplasia of the stomach among the inhabitants of Japan (*53*). Intestinal metaplasia is produced after prolonged inflammation of the stomach mucosa. Adenocarcinomas often arise from areas of intestinal metaplasia (*52*). Other hidden tumor promoters in seemingly natural guises may exist in our environments. L-Leucine and L-isoleucine were proved to be urinary bladder tumor promoters by means of a long-term animal test (*54*). We developed a short-term assay for urinary bladder carcinogens by assaying the agglutinability of free cells by concanavalin A. These cells are obtained from the urinary bladder of rats treated first with a limited amount of *N*-butyl-*N*-(4-hydroxybutyl)nitrosamine followed by administration of other test compounds. With this test, oral administration of L-leucine and L-isoleucine was as effective as saccharin in promoting bladder carcinogenesis (*54*).

As has been mentioned by many researchers, high fat intake gives rise to increased formation of bile acids and to changes in the hormonal condition, which could lead, possibly through a promotional pathway, to a higher frequency of colon and breast cancers in humans (*55*). In Japan, colon and breast cancers are becoming much more common while the incidence of stomach cancer declines (*56*). This can be accounted for by a gradual shifting of the Japanese lifestyle from traditional to Western, especially with regard to diet (*57*).

Fusion of Studies of Chemical and Viral Carcinogenesis in Japan

In 1910, Fujinami and Inamoto (*58*) of the Kyoto Imperial University reported the transmission of Fujinami chicken sarcoma by the filtrate of a tissue extract. The oncogene of Fujinami virus (*fps*) was proved to be different from the *src* gene of Rous sarcoma by Hanafusa *et al.* (*59*) 70 years after the original discovery. Domestic and international collaboration has led to the quick determination of the total base sequences of HTLV-I by Yoshida of the Cancer Institute and co-workers (*60*), and HTLV-II by Shimotohno of the National Cancer Center Research Institute and co-workers (*61*).

Studies on oncogenes in experimental cancers that have been induced by new mutagens or carcinogens are progressing in our laboratories. H-*ras* gene is activated in IQ-induced rat hepatoma (*62*). N-*ras* gene is activated in Glu-P-2–induced rat intestinal cancer (*62*). A new rearrangement of the *raf* oncogene has been found that occurs during transfection of DNA of IQ-induced rat hepatoma (*62*). This rearrangement is probably responsible for the acquisition of the transforming activity of the *raf* gene (*62*). A fibrosarcoma induced in rats by 1,8-DNP contains an activated K-*ras* oncogene with a mutation at the codon for the 12th amino acid, glycine to cysteine (GGT → TGT) (*62*). Activation of H-*ras* and N-*ras* was observed in 1,6-DNP–induced sarcomas (*62*). Studies on oncogenes in human cancers are also progressing. We have recently isolated a new transforming gene, *hst*, from patients with stomach cancer (which is still the most common cancer in Japan) (*62*).

Necessity for a New Science for Risk Estimation in Human Cancer Development

Kato of Keio University and colleagues (*63*) and Nishino of Kyoto Prefectural University and colleagues (*64*), after initially applying DMBA, painted quercetin together with either TPA or teleocidin and found a dramatic effect of suppression of tumor formation. We found many other substances that significantly modify the carcinogenic process (*45*). Ito of Nagoya City University and colleagues (*65*) found that butylated hydroxyanisole, an antioxidant, was carcinogenic at high doses, but anticarcinogenic at lower doses in some organs.

An astute approach to the risk estimation of environmental chemicals for the causation of human cancer is equally important for scientists in both academic and industrial laboratories, as well as in regulatory agencies and among the general public. Several mathematical models have been proposed, for which values such as the doses causing tumors in 50% of experimental animals (TD_{50}) and the virtually safe dose (VSD) are often determined (*66*). The TD_{50} is available for many carcinogens, including the heterocyclic amines newly discovered to be carcinogenic.

People are exposed to a vast number of mutagens for different durations and frequencies (for example, minute, repetitive doses of the heterocyclic amines) and also to various modulators of carcinogenic potency. Risk can be increased or decreased easily by 100-fold by modulating substances, and this makes proper risk estimation very difficult for these carcinogens. The different sensitivities of animals and humans makes extrapolation of animal data more complicated. Risk estimation is being given a high priority by committees of many organizations that provide grant funds for cancer research in Japan.

Importance of Primary Cancer Prevention

Our National Cancer Center, consisting of Hospital, Research Institute, and Administration Office, was established in 1962 on the land in downtown Tokyo where the Imperial Navy Hospital and Navy Medical School and the U.S. Army Hospital functioned before and after World War II. The National Cancer Center Hospital has nearly 600 beds, and each year approximately 700 cancer patients are cured as measured by the 5-year survival rate. The cure rate is 52%. The operating budget of the hospital is about $60 million. Annually 350,000 new cancer patients appear in Japan. How many National Cancer Center Hospitals are required to fulfill the needs of these people? Perhaps it is more reasonable to think there is an alternative way, namely, "Primary Cancer Prevention." Total elimination of environmental carcinogens and promoters is impractical, and contact with some environmental carcinogens remains virtually unavoidable. In addition, active oxygen radicals (*67*) and endogenous formation of nitrite and mutagen/carcinogen precursors cannot be eliminated with our present knowledge. Yet, a dramatic change of cancer incidence in individual organs (such as that encountered among immigrants) or a steep reduction of certain occupational cancers encourages our idea of primary cancer prevention as a practical and plausible challenge. The latent period of cancer development may sometimes be measured over decades. The beneficial effects of our efforts to prevent cancer cannot be achieved overnight. However, one can already see marvelous signs of victory in the decreases in lung cancer incidence and mortality in England and Norway and in the white male population of the United States, most probably as a result of the antismoking campaigns in these countries (*68*).

Japanese Cancer Control Policy

The Ministry of Education, Science and Culture and the Ministry of Health and Welfare in Japan provided grants for cancer research to many scientists in different universities, institutes, and hospitals, as a way of supporting "invisible cancer institutes." In the past, grants were given to a group of scientists who shared common interests but who possessed different knowledge and skills. This is called the "Kenkyu-han" system of funding in Japan. Grants are distributed to scientists ("Han-in") in the group by each group leader. This system functions well, especially when the size of the budget is limited and many promising scientists want to work closely on interrelated subjects. This is especially true in Japan, where most staff scientists are permanent employees, obtaining their stipends from the government or other institutions without depending on grants. There are many advantages to this system, as in the case of member scientists who do not make their promised progress; they are compensated by other members whose progress has been better. Now, a new granting system with a more strict project orientation that provides a relatively large sum of money to a limited number of scientists is becoming more popular in Japan.

The Japanese government headed by Prime Minister Yasuhiro Nakasone launched the Comprehensive 10-Year Strategy for Cancer Control in 1984 for basic research. From the outset of this project, six subjects were chosen: oncogenes, oncogenic viruses, suppression of carcinogenesis, diagnosis based on new principles, therapy based on new principles, and host defense mechanisms. The project represents a collaboration among the Ministry of Health and Welfare; the Ministry of Education, Science and Culture; and the Agency of Science and Technology. In addition, mainly through the Foundation for the Promotion of Cancer Research and the endorsement of governmental and nongovernmental organizations, a new program was started for international personal exchange and a campaign for cancer prevention was resumed. Twelve points for cancer prevention, originally made by us in 1978 (*26*), have been recently polished by a committee headed by Takayama of the National Cancer Center and distributed as brochures with 1986 calendars all over Japan, recommending: (i) Have a nutritionally balanced diet; (ii) have a variety of types of food; (iii) avoid excess calories, especially as fat; (iv) avoid the excessive drinking of alcohol; (v) smoke as little as possible; (vi) take vitamins in appropriate amounts; eat fiber and green and yellow vegetables rich in carotene; (vii) avoid drinking fluids that are too hot and eating foods that are too salty; (viii) avoid the charred parts of cooked food; (ix) avoid food with possible contamination by fungal toxins; (x) avoid overexposure to sunlight; (xi) have an exercise program matched to the individual's condition; and (xii) keep the body clean.

Although these recommendations are based on laboratory science and epidemiological studies, we avoided absolute prohibitions. The reason is that the science of cancer prevention is still far from an exact discipline. Freedom of individual taste determines individual choice and with it responsibility for personal health. Of course, regulatory actions against smoking in public places and intense smoking controls to limit smoking in minors are crucial to protect the general public. However, it is also true that nobody has the right to stop an adult from smoking at home in his own room, if at the same time he pays attention to undesirable passive smoking which can expose family members to a higher risk. General consensus without coercion is a traditional way for cooperative development of ideas in a population of over 100 million people on a highly unique, small island. So far these 12 points are well accepted by the general public, and slowly but steadily the program of action for primary prevention of cancer is now moving ahead.

We expect much closer and more efficient collaboration between cancer researchers and clinical doctors to make the outcomes of basic research more quickly available for immediate benefit of cancer patients. Hybridization between Oriental intuitionalism and Western rationalism may provide solutions for the worldwide reduction of human cancer.

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Organic Synthesis in Japan: From Natural Products to Synthetic Control

TERUAKI MUKAIYAMA

Organic chemistry in present day Japan, which has developed from early interest in natural products chemistry, now includes total synthesis, physical organic chemistry, synthetic methods, and organometallic chemistry. In this article, the current state of Japanese organic chemistry is briefly reviewed and a representative aspect of today's organic chemistry—exploration of new synthetic methodology—is discussed.

JAPAN IS A LEADING NATION IN THE FIELD OF ORGANIC chemistry, and Japanese chemists have made important contributions to the recent growth of this subject. Initially, the chemistry of natural products was their main area of interest. At present there are major schools of research in total synthesis, physical organic chemistry, synthetic methods, and organometallic chemistry, and, in this article, I take the opportunity to introduce the work of some of these prominent research chemists.

Nozoe has been one of the leading figures in the development of Japanese natural products chemistry. Among his achievements, the most significant is the elucidation of the chemistry of the troponoid system. This work originated from his studies on hinokitiol [2-hydroxy-4-isopropyl-2,4,6,-cycloheptatrien-1-one], which he isolated from a natural source in 1935. His experiments led him to recognize the aromatic properties of the novel seven-membered ring independently of Dewar's tropolone hypothesis. He later synthesized tropolones and a wide variety of other nonbenzenoid aromatics, such as S- and N-analogs of tropolones; heptafulvenes; cyclic, cross-conjugated quinarenes; azulenes; and heterocyclic compounds annulated to the tropylium system. He also studied the physical and chemical properties of these diverse systems and thus established the chemistry of nonbenzenoid aromatic compounds as a new field in organic chemistry (*1*).

The development of physical organic chemistry has contributed to the basic understanding of reaction pathways. For example, Fukui has developed a theoretical approach to predict the reactivity of the organic molecule, which has proved useful for designing reaction sequences. Also well known in the field of physical organic chemistry are: Oki, for the isolation of rotational isomers and studies of their reactivity; Sakurai, for his interest in theory (chemical bonding and bond interactions) and new synthetic methods of organosilicon compounds; Misumi, for the synthesis of layered aromatic com-

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