Mr. Chairman and fellow students: “Komban wa”.

We are gathered here this evening to pay tribute to Professor Yamagiwa and his outstanding achievements. I had been asked by the Committee to talk about the era from Yamagiwa to Kennaway. However, my task has been much simplified by the address of the eminent speaker who preceded me, and I do not propose to bore you with repeating what he has already said.

Carcinogenesis, as we now know, is a far more complicated subject than merely applying a chemical to a tissue. The endocrines are involved. Viruses are involved. Diet is involved. And, in more recent years, we’ve come to learn about the carcinogenic potency of nitroso compounds, of metabolites of fungi such as the aflatoxins, and even constituents of food in some quarters of the world like cycasin.

These topics lie outside the scope of the subject that I’ve been asked to discuss. I therefore will restrict my comments to tar cancer and to some closely related aspects of the work of Yamagiwa and of other pioneers here in Japan.

Now I do not propose to overwhelm you with minutiae. Most of you have had the beneficial experience of exposure to papers and lectures during the morning and the afternoon. So I think it would hardly be agreeable to be fed additional factual information in this after-dinner lecture. It would be more fitting if I were to reminisce and regale you with some anecdotes, to show that research is not accomplished by disembodied intelligences. Research is accomplished by people, really much like other people, who have their emotions, ambitions, disappointments, and excitements, their rivalries and their joys. To correct the public image of the scientist as a non-human thinking machine, we ought to get across that scientists are also human.

As Professor Henschen has stated there has been a long background to Yamagiwa’s work. Professor Haddow also alluded to the clinical observations made during the past two hundred years. They summarized the attempts, on the part of many investigators, to produce tumors in experimental animals with a variety of substances. These all led to failure, as you have heard, until Yamagiwa and Ichikawa succeeded.

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*1 A paper read at the Yamagiwa Memorial Lecture, held during the Ninth International Cancer Congress in Tokyo, October 24, 1966.
With the benefit of hindsight, the reasons for those failures appear to be threefold. One is the selection of appropriate animal species for specific types of experiments. For example, in the elicitation of skin tumors the skin of the rat is not particularly useful, for it is refractory; the skin of the rabbit is more responsive. If one wishes to get subcutaneous tumors, the subcutaneous tissues of the rat are far more sensitive than those of the rabbit or the mouse, perhaps even too sensitive. There is such a thing as too great refractoriness and also too great sensitivity. In the early attempts to produce malignant skin tumors in animals, one reason for the failures was the use of rats and dogs; Yamagiwa used rabbits.

A second reason for failure was that tars vary in carcinogenic potency. “Tar” is not a simple substance. Tars are enormously complex mixtures. Whether a tar is prepared in a vertical retort or a horizontal retort, the temperature at which it is distilled, and the amount of oxygen that is present all make a great difference in the carcinogenic potency of the tar.

Thirdly, knowing when to terminate a biological experiment may spell the difference between success and failure. Unfortunately, every person who emerges from a university with a doctor’s degree is not of equal quality. Some have great imagination but lack “Sitzfleisch.” Some can work hard and persistently, but not with enough imagination. If a research is going to yield negative results even if kept up for years, how does one know that the horse is dead? No matter how long you kick a dead horse he will not get up. On the other hand, in this kind of experiment several of the workers had the right sort of animal and the right kind of tar, but they didn’t keep the experiment going long enough — they thought they were kicking a dead horse.

So you can imagine his elation when Yamagiwa finally accomplished what he felt had to be. He was so happy when he achieved success that he wrote a poem, a Haiku, which Professor Henschen has already mentioned.

Dr. Iwao Hirono, who is currently working on carcinogenesis, as a Fellow of the UICC, in the laboratory of Dr. Gert Laqueur in Bethesda, has helped me in this. According to Dr. Hirono this poem reads: “Gan”, which is cancer, “dekitsu, iki kōzen to niho sampo.” Hirono tells me that, literally, this is a condensation of a summary of an abstract. In other words the beauty of this type of poetry lies in its terseness, with no words wasted. Literally, it means something like this, according to Hirono: “Cancer developed, the spirit proudly walks.” In order to convey the meaning to those more familiar with English poetry I have, with Hirono’s approval, paraphrased it. This is not a literal translation, but he tells me it conveys the feeling: “Success in producing cancer lifts our spirits high and causes us joy.”

In 1915, a horrible catastrophe was engulfing much of the world — the First World War. Because of the war it was not until the conclusion of hostilities that confirmation of the findings of Yamagiwa and Ichikawa was reported from other countries. In 1926 William H. Woglom, of the Institute of Cancer Research at Columbia University in New York, wrote a critical review of the literature on “Experimental Tar Cancer”; in that short period 290 publications had already appeared and were listed in the bibliography. Woglom stated that, in the course of their preliminary experiments, Yamagiwa and Ichikawa had tried various other mechanical and chemical irritants but
found tar to be the most effective. Woglom listed 17 published contributions by Yamagiwa and Ichikawa and other co-workers between 1914 and 1926.

There have been great minds and great achievements in cancer research, and Professor Rudolf Virchow is among the giants. Nevertheless, those who have worked for years and decades with carcinogenesis are not at all convinced that irritation is involved. One may apply for long periods chemicals that produce obvious irritation and even sloughing off of the skin, or necrosis of subcutaneous tissue, and never produce a tumor. On the other hand, after a low dose of a carcinogen; there is no obvious damage, there is no obvious cell death, and there is no obvious regeneration and replacement, yet a malignant tumor develops. So if "irritation" is used to explain the mechanism of carcinogenesis the meaning of the word irritation would have to be changed from what it ordinarily means.

As Dr. Haddow has mentioned, Bruno Bloch and W. Dreifuss in Switzerland were among those who made pioneer progress in the fractionation of tar. In the first place, they found that potency resided in the high boiling fraction, the so-called anthracene fraction. In the second place, they found that the acid fraction, that is, the part that's extractable with alkali, was inactive. They found that the basic fraction, the part that's extractable with acid, was also inactive. These findings were made as early as 1921. By 1924, Bloch and Dreifuss found that the neutral, high boiling fraction, which contained the carcinogenic potency, was free of nitrogen.

In the folklore of cancer research is an anecdote that when young men from other countries came to work in Bloch's laboratory he would keep an eye on them, noting their social proclivities. There were several good-looking young female technicians in Bloch's laboratory, and any young men who failed to try and date the girls Bloch got rid of, because he said if their endocrine system isn't functioning properly, their brains won't function properly!

By far the most valuable investigations on the relationship between the chemistry of tar and its carcinogenic power were those of Kennaway and his colleagues in London. In 1924, Kennaway discovered that isoprene, a compound which contains only carbon and hydrogen, when heated in hydrogen gas at about 800°, formed ring compounds with double bonds, and that the product produced in this way was more potent than coal tar. In 1925, Kennaway found that when acetylene — how much simpler can you get chemically? just carbon and hydrogen — when Kennaway heated acetylene to 700° a potent tar was obtained.

Professor Haddow mentioned something about the work of Leitch, Kennaway's predecessor. Leitch had been studying "mule spinners" cancer which occurred in workers in factories that spun cotton. These workers were spurted with lubricating oil from the machines ("mules") and developed scrotal carcinoma in an incidence higher than the general population. So lubricating oils were examined for their carcinogenic potency. In 1924, Leitch tested a mineral oil from California and found it to be noncarcinogenic. But when he heated it to 880° in an atmosphere of hydrogen it became carcinogenic.

About the same time, in 1924 and 1925, Kennaway made another remarkable discovery, viz., that biologic materials when heated in sealed containers became converted to tars that were carcinogenic. Among the substances which he treated in this way
and converted from non-carcinogenic to carcinogenic were skin, cholesterol, hair, and yeast.

Pyrolysis, i.e., heating in the absence of oxygen at a sufficiently high temperature, confers carcinogenic potency on some biologic materials. The question therefore arises, and it has not been answered — although I see Professor Shabad sitting over there — whether substances which occur in the body, other than the hormonal, are capable of evoking malignant tumors. In other words, are there endogenous carcinogenic compounds? Exposure of the skin to ultraviolet ray converts a normally-occurring sterol to vitamin D. Exposure of the skin to UV ray, in the mouse and in man, can eventuate in malignant tumors. Does the radiation transform an endogenous precursor to a carcinogen?

Different tars have different potencies. Deelman published at least nine reports on this subject between 1921 and 1924. In an extensive study of experimental tar cancer, he reported in 1923 that one of the tars he used, a product of the gas factory Ooster gas Fabrik in Amsterdam, which had been distilled in a horizontal retort, was much more carcinogenic than one produced in a vertical retort in a more modern gas factory, at the Zuider gas Fabrik, also in Amsterdam.

I mention Amsterdam at this point because I am reminded of Peyton Rous, who fortunately lived long enough to be awarded the Nobel Prize. Now, this is something you might bear in mind — "Don’t die too young!" The Nobel regulations stipulate that the award be given to a living person. If you would have gotten the prize had you lived to 92, and if you die, say at the age of 65, bear in mind that the Prize is not awarded posthumously!

Well, anyhow, Deelman was among those who fractionated tar and examined the fractions for potency. He found, curiously enough, that scarification of the skin prior to applying tar speeded up tumor production. Later on, Peyton Rous would find that punching a hole in the ear of the rabbit speeded up tumor production with either coal tar or virus. Now, is this non-specific irritation, or is it some specific factor which is brought into play when the skin is damaged?

In 1952, at the annual meeting of the American Association for Cancer Research, Rous presented a paper on carcinogenesis which evoked considerable discussion. In the course of the discussion, Rous said that all of the experiments which he had carried out over many, many years had been performed with aliquots of the same specimen of tar. He said that Karl Landsteiner — who had received the Nobel Prize many years ago for his work on blood groups, which made transfusions possible — that Landsteiner had brought over from Holland a barrel of tar which Rous then used in all his carcinogenesis work. Rous told us that he had almost reached the bottom of the barrel, and that when this supply was all used up he didn’t know what he was going to use in further experiments on carcinogenesis!

I have mentioned the chemical complexity of tar and Dr. Haddow has mentioned the work of Mayneord in London. In the early 1930’s it was as difficult to isolate the active agent from complex tars as finding proverbial needle in a haystack. But, with imagination and courage a magnet, the fluorescence spectrum, was applied by workers in London to track down the “needle”. Mayneord had observed that some bands in the fluorescence spectra seemed to be characteristic of various carcinogenic tars. This
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clue was followed by Hieger in an investigation of the spectra of polycyclic aromatic carbons, particularly those related to anthracene and 1,2-benzanthracene.

By a fortunate coincidence, in 1929 the previous year, Clar had reported the convenient synthesis of dibenzanthracene — just for fun, you know, the organic chemist in the laboratory playing with benzene rings — by sticking five rings together. This compound, 1,2:5,6-dibenzanthracene, was found to have a fluorescence spectrum quite similar to those of the carcinogenic tars. By painting it on the skin of mice, and by subcutaneous injection, it evoked malignant tumors. This was the first chemical compound to be shown to be a carcinogen.

Using the fluorescence spectrum as a guide, 3,4-benzopyrene was isolated from a large amount of coal-tar pitch. The details were reported by Izrael Hieger in his thesis for the Ph.D. degree at the University of London in 1936. 3,4-Benzopyrene was found to be more strongly carcinogenic than 1,2:5,6-dibenzanthracene. (I am using the old chemical designations in this historical resume.)

On the Continent a compound, methylcholanthrene, was prepared from a bile acid which was even more potent than benzopyrene. A few years later, in Fieser's laboratory in Cambridge, Massachusetts, methylcholanthrene was synthesized by Arnold Seligman.

The London group came to the conclusion that five benzene rings were necessary for conferring carcinogenic potency on a compound. When I started work with polycyclic hydrocarbons in 1932, the only one available to me was dibenzanthracene. By 1934, I wanted others, particularly of my own selection. A young organic chemist had just been brought back to Harvard. His name was Louis Fieser. He had been teaching chemistry at Bryn Mawr, a girls' college near Philadelphia. My chief, Dr. J. W. Scherschewsky, and I journeyed from Boston to Cambridge to see him and ask him if he would care to collaborate in cancer research. He said "I know nothing about cancer research." I said, "You have already been doing cancer research."

"Oh, no" he said, "It must have been someone else."

I said, "Remember you and Emma Dietz synthesized 1,2:5,6-dibenzanthracene and published the synthesis in the Berichte in 1929?"

"Yes, what's that got to do with cancer?"

"Well," I said, "the group in London has shown that this compound can produce cancer."

You can imagine how his interest grew. A cooperative arrangement was made. One of his associates was an attractive girl student at Bryn Mawr, who came to Harvard as a graduate student and became Mrs. Mary Fieser. She collaborated with him in production of several important books in chemistry.

Fieser recruited some brilliant young men — Mel Newman, E. B. Herschberg, Byron Riegel, and others, after Arnold Seligman, the first one — and provided me with almost 200 new polycyclic compounds over a period of some five years. Then I brought the joint arrangement to a close when it became obvious that there was no unique relationship between chemical structure and carcinogenic potency. Besides, I'd gotten interested in the work of another pioneer with a totally different kind of chemical carcinogen, namely Yoshida's compound, "o-Amidoazotoluol" (2-amino-5-azotoluene).
We sometimes hear complaints from uninformed people about “unnecessary” duplication of research. They watch carefully the allocation of funds and if the wording of a proposed project seems similar to that of another one, they say, “Out! No duplication!” As a matter of fact, we must have duplication.

Fibiger won the Nobel Prize but nobody since then has been able to produce carcinoma of the glandular stomach with his method. This is not to say that he didn’t do it. But unless other people in other places can reproduce the results, they are of dubious scientific value. It is important and, in fact, essential to have work repeated for still another reason. Frequently, when work of a similar nature is done in different places, it is not done in identical fashion. For example, the London group painted the skin; in my laboratory the polycyclic compounds were injected subcutaneously. In London, they used mice of heterogeneous genetic origin; we used pure strain mice. Out of these differences came information that would not have been available had all the work been done with a uniform technique.

An entirely different type of chemical carcinogen was also pioneered in Japan, first by Tomizo Yoshida and then by Riojun Kinosita and their colleagues. Time does not permit me to do justice to Yoshida’s systematic work over many years. I will merely refer to his success, with the oral administration of this azo dye mixed in the food, in the production of hepatic carcinomas in the rat. When rabbits were used, he did not get any tumors. However, the rat was also used in his investigations and this yielded highly successful results. I was happy to have extended his findings to mice in 1935; in a paper published in 1937, I reviewed the many reports by Yoshida which had appeared by that early date.

Among the substances used by Yamagiwa and Ichikawa in attempting to produce malignant growth was Scarlet Red. They used it alone and in combination with tar. Hyperplasia was produced by Scarlet Red but, for various reasons, its use was discontinued and coal tar was used alone.

This line of work with Scarlet Red had originated with Bernhardt Fischer. When it was published in 1906, it created a sensation. A large number of investigators had their interest aroused; they used, in their subsequent publications, the words “sensational finding”. A great deal of work was done with Scarlet Red, with 2-amino-5-azotoluene, and with similar substances between 1906 and 1915. When, in the latter year, Yamagiwa and Ichikawa reported that they had succeeded in producing true malignant growths in rabbits by application of coal tar, the attention of those interested in carcinogenesis shifted from the azo compounds to coal tar. We owe a great debt to Dr. Yoshida for bringing our attention back to this subject and to Dr. Kinosita for working with other Butter Yellows.

With Scarlet Red, Fischer obtained epithelial proliferation which many agreed was, histologically, indistinguishable from cancer. But the growths were not progressive, they always regressed. When the molecule was split, one fragment was the compound called o-aminoazotoluene or 2-amino-5-azotoluene. Hepatic carcinomas were produced on oral administration to the rat. On subcutaneous injection in the mouse I got a similar picture of multiple liver cell carcinoma. However, with Kinosita’s analogous compound — p-dimethylaminoazobenzene — which produces liver tumors when given to rats in the food, failed, in my hands, to yield tumors on subcutaneous injection into mice.
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I should like to make one more comment — on the question of diet — and then close. I was a young man at the time when I obtained liver tumors in mice, and I was flattered when Professor Wolbach, the Chairman of the Department of Pathology at Harvard Medical School, invited me to collaborate with him in a repetition of Yoshida's work. Professor Wolbach had maintained a fine strain of rats. We fed the best diet, and I used the same purified sample of azo dye that had been successful in inducing hepatomas in the mouse. At the end of a year, the rats were healthy and happy, with no sign of any tumors. I couldn't figure out what was wrong. The only thing I could think of to account for the failure to obtain hepatomas was the highly unlikely rôle of the diet. In the United States the rats get wheat and corn while Yoshida presumably used rice. I repeated the experiment the following year, substituting rice for the usual American cereal. Yoshida's findings were confirmed — this time we obtained multiple liver cell carcinomas. Since, in the experiment in the previous year, the rats had eaten the same amounts of the same yellow dye, the carcinogenic agent must obviously be the rice!

Mr. Chairman, it is quite fitting to establish the Yamagiwa Memorial Foundation on the occasion of the fiftieth anniversary of his great achievement.

Many young Japanese scientists have studied and worked in the biomedical sciences in other countries. They have had, Your Highness, high qualifications. I cite only one bit of evidence. In the open competition for Fellowships from the UICC, the percentage won by young Japanese scientists has been extraordinary high and out of proportion to the size of the population of Japan.

But this is not good enough as I have told my friend Tomizo Yoshida. It should be a two-way street. People from other countries should come to Japan to study and to work and to learn from Japan. So that we get the full benefit and not merely half of the pie. One of the admirable things, in my opinion, about this new Foundation is that it proposes to bring people from other countries to work in Japan in cancer research. You need have no false modesty about what foreigners can learn here in that regard!

In closing, I would like to end on a personal note. It is my pleasure to be in Japan for the fourth time. My only regret is that other obligations have prevented my coming more often and staying longer.

I wish to offer my congratulations to Japan and to our Japanese colleagues in cancer work for their past achievements and for setting up this new forward-looking Foundation. I have every expectation that the future will bring further high achievements in cancer from Japan.