MINUTES OF THE MEETING OF THE SUBCOMMITTEE ON CARCINOGENICITY
Held at the Blackstone Hotel, Chicago
Friday, September 27, 1957

A meeting of the Subcommittee on Carcinogenicity of the Medical Advisory Committee of the American Petroleum Institute was convened in Dr. R. E. Eckardt's suite at the Blackstone Hotel at 9:30 a.m. on September 27, 1957. The full Subcommittee was in attendance, plus Mr. Paul Halley of the Standard Oil Company (Indiana) as a guest.

The Subcommittee discussed preliminarily the work at the Kettering Laboratory and wishes to point out that work now being performed by Dr. Horton is far more fundamental in nature than was originally anticipated by the Subcommittee. For this reason the Subcommittee believes that its decision to discontinue sponsoring this research under its direction has been a correct one, and the Subcommittee plans to continue with the recommendation that this work be discontinued as of July 1, 1959. However, the Subcommittee does wish to point out that there are fundamental aspects in the petroleum chemistry now being conducted by Dr. Horton, which might warrant a continuation of this type of research, either at Kettering Laboratory or some other laboratory, under the sponsorship of the Committee for Fundamental Research of the API. The methodology now being pursued by Dr. Horton is primarily chemical in nature rather than biological and might therefore well be considered by the Committee for Fundamental Research. In addition, the Subcommittee discussed in a preliminary fashion, the possibility of suggesting to the MAC that Dr. Kehoe be put on a retainer fee at the conclusion of the work at the Kettering Laboratory on July 1, 1959, in order that Dr. Kehoe might attend all meetings of the MAC and offer us his advice and guidance in some of the problems which the petroleum industry is facing now and might face in the future. It is the Subcommittee's belief that Dr. Drinker's attendance at and participation in the affairs of the MAC have been highly worthwhile and if Dr. Kehoe were willing to accept such a retainer agreement, his participation in the affairs of the MAC would also be worthwhile.

At 11 a.m. the group was joined by Dr. R. A. Kehoe, Dr. A. W. Horton and Dr. J. J. Phair of the Kettering Laboratory. Dr. Horton began by presenting a progress report of the work at the Kettering Laboratory during the past five months. He indicated that all the experimental work for the past five months has been in the field of analytical methods. He divided this work into two sections: that work done on carcinogens and that work done on accelerators.

Carcinogens

To date the work of the Kettering Laboratory suggests that the most significant carcinogens in petroleum oils are polycyclic aromatic compounds, most hydrocarbons.

He emphasized the difference in concept between the basic carcinogenic potency of an oil, which is that due to the presence of carcinogens vs. the actual carcinogenic potential of the oil, which is the carcinogenic activity resulting from both the presence of carcinogens and of accelerators and inhibitors. He indicated that certain data developed at the Kettering Laboratory have suggested that some sulfur compounds may act as accelerators but that they only do so in the presence of other accelerators. Thus, if there are sulfur-containing accelerators they would not be active as accelerators in the absence of hydrocarbon accelerators. Present data at the Kettering Laboratory indicated that the accelerators are probably long-chained hydrocarbons.

Suggestive evidence at the Kettering Laboratory indicates that there may be inhibitors present in petroleum oils. These inhibitors do not inhibit the carcinogens, but rather inhibit only the accelerators. This evidence has arisen from some work done by the Kettering Laboratory with two different white mineral oils which were used as diluents of accelerators. It is important to emphasize that these inhibitors are inhibitors of the accelerating action, rather than inhibitors of the basic carcinogenic activity due to the carcinogens.

In the past five months almost all of the work done by the Kettering Laboratory on carcinogens has been done on what they have previously identified in the April 1957 report, as 3 1/2-ring aromatics. These consist of pyrenes, fluoranthrenes, cyclopentanophenanthrenes, and benzfluorenes. Dr. Horton believes that these 3 1/2-ring aromatics are probably the most important class of carcinogens in most petroleum oils.

The work has been done principally on API-8 and fractions of API-8, as diagramatically indicated in the attached diagram. Descriptivly, API-8 was subjected to a theoretical 80 plate distillation. Examination of the 660°F fraction suggested that only 0.001% of carcinogens was present in this fraction but that it contained a very high concentration of accelerators. This evidence is supported in part by the fact that tumors appear rapidly but only at a very low total incidence of tumors.

Following the work on this fraction, Dr. Horton next concentrated his attention on three fractions which boiled in the range of 730°F to 760°F. These fractions contain very little accelerators. These three fractions were subjected to maleic anhydride treatment in order to remove the anthracenes and their derivatives. Testing suggested that .03 out of .21 of the carcinogens present in this fraction was due to carcinogens of this type. Roughly, therefore, 1/7 of the basic carcinogenic activity of these three fractions is due to anthracenes and their derivatives. The raffinate from the maleic anhydride treatment of these three fractions was next combined and chromatographed, and two fractions were obtained, one of which contained the benzfluorenes and experimental work has suggested

that these are unimportant in the total carcinogenicity of these three fractions. The other fraction contains the remaining three classes of carcinogens, namely, pyrenes (and their methyl and dimethyl derivatives) cyclopentanophenanthrenes, and fluoranthrenes(principally the dimethyl derivatives). At the present time, Dr. Horton is working on the separation of these three types of aromatic hydrocarbons in the raffinate from the maleic anhydride treatment of the 730°F to 760°F cuts of API-8. This is being done by liquid-liquid partition between 99% sulfuric acid and iso-octane at -30°F. Dr. Horton says that this treatment will separate the pyrenes from the other two classes of compounds. The raffinate of this extraction then is chromatographed and treated with picric acid which may lead to the separation of the cyclopentanophenanthrenes from the fluoranthrenes.

Dr. Horton did indicate that of seven oils carried through the maleic anhydride extraction step, only one was found to have much benzanthracene which contributed to carcinogenic activity. This one oil was from a coker gas oil (API-33) in which 7/11 of the basic carcinogenicity was due to benzanthracene-type compounds. This contrasted with other types of oils in which only 1/7 of the basic carcinogenicity is due to this type of compound.

In work which Dr. Horton did with a thermally cracked, cat cracked, oil he found that the benzpyrene and 4-ring carcinogens almost doubled in concentration but the activity of the oil decreased. This result might be due to the destruction of accelerators or to the destruction of the 3 1/2-ring carcinogens. It is Dr. Horton's belief that possibly this reduction in activity is most likely due to the destruction of the accelerators rather than to the destruction of the 3 1/2-ring carcinogens.

Accelerators

At the present time, Dr. Horton is following clues which have been provided for the presence of inhibition of accelerators in petroleum oils. He believes that these inhibitors probably fall into a class of compounds referred to as polycycloparaffins. These have essentially the same structure as the aromatic carcinogens except that they are saturated compounds. In partial support of this, Dr. Horton pointed out that Zechmeister and Kotin reported about a year ago in SCIENCE, that hydrogenated aromatics inhibit sarcoma production when injected subcutaneously. However, they used ethyl laurate as a solvent for their carcinogens and ethyl laurate would be expected to be a highly accelerating solvent. Therefore it was not clear from their work whether they were inhibiting carcinogens or inhibiting accelerators. It is Dr. Horton's belief that the inhibition they observed is most likely the inhibition of the ethyl laurate as an accelerator rather than true inhibition of the carcinogens.

In discussing the so-called preconditioning of an animal by accelerators, it is Dr. Horton's belief that this occurs through an accelerator "setting up" the animal for other accelerators. In other words, preconditioning does not precondition the animal for carcinogenic activity but only for accelerating activity. Support for this belief is contained in the fact that if the animal is painted with an oil essentially free of accelerators following preconditioning it is not possible to demonstrate the preconditioning effects, but if an oil high in accelerators is used after the preconditioning treatement, the preconditioning effect can be demonstrated.

The white oils being studied by Dr. Horton for so-called inhibiting effects (principally API-80) are free of n-paraffins. They contain some iso-paraffins and condensed cycloalkanes which represent the greatest amount of material present. The suggestion at the moment, therefore, is that these condensed cycloalkanes are inhibitors of accelerators.

In an attempt to separate these inhibitors from other compounds present in the white oils, Dr. Horton is going to use distillation, thiourea separation, and liquid thermal diffusion. In addition, he may possibly consider the use of molecular sieves.

Dr. Horton submitted his Interim Report on his work dated September 26, 1957 which was distributed to the Subcommittee at this meeting.

Epidemiology

Next, Dr. J. J. Phair gave a progress report on the epidemiological work. Dr. Phair indicated that he had completed the coding and punching of the cases. In this process he felt it necessary to discard some cases since the information contained on the report was inadequate. Hence, the total number of cases reported in his final report will not be the same as in previous reports from Dr. Phair. At the moment, the status of this final report is that the introduction, the objectives, and so forth have already been written up and the data tables are now being prepared and the comparisons and correlations will be made. It is hoped by both Dr. Phair and Dr. Kehoe that this final report can be made available to the Subcommittee by the first of the year.

In Dr. Phair's work he pointed out that in the cases occurring after 1950, 28% of the tumors have been found to be skin tumors, whereas, only 15.3% of the cases prior to 1950 were skin tumors. In general population figures available, 16.6% tumors are skin tumors. Whether this increase in the relative number of skin tumors in the post-1950 group of cases is significant, is not now known. Dr. Phair is at present making age, geographic location, primary site and occupational-group breakdowns of the cases which he analyzed.

Following Dr. Phair's presentation, the Subcommittee discussed in private with Dr. Kehoe certain phases of the relationships between the Subcommittee and the Kettering Laboratory and it was concluded that these relationships remain on a friendly, amiable, and professional level.

The Subcommittee meeting broke up at approximately 4 p.m. on September 27, 1957, without having set a date for its next meeting.

R. E. ECKARDT, M. D./cjh
Chairman, Subcommittee on Carcinogenicity
October 1, 1957

Attachment

API-8

80 Plate Distillation

660 fraction –
.001% carcinogen.
Much accelerators
Tumors appear rapidly
but at low incidence

730-760 (3 fractions) little accelerators

Maleic
Anhydride of the 3 cuts ——→ anthracenes and their derivatives – .03 out of .21, i.e., 1/7

Raffinate

Chromatographed ——→ benzfluorenes (unimportant)

Pyrenes - methyl & dimethyl
Cyclopentanophenanthrenes
Fluoranthrenes - dimethyl derivatives

Now working on separation of these 3 hydrocarbons

Liquid-liquid
Partition between 99% sulfuric acid and iso-octane at -30° separates pyrenes from other 2

Raffinate

Chromatography & picric acid

MINUTES OF THE MEETING OF THE SUBCOMMITTEE ON CARCINOGENICITY
Held in the Conrad Hilton Hotel in Chicago
Wednesday, November 13, 1957

The Subcommittee on Carcinogenicity held a meeting at the Conrad Hilton Hotel in Chicago at 9 a.m. on Wednesday, November 13, 1957. The following members were in attendance:

E. W. Adams
C. H. Hine, M. D.
E. K. Linder, M. D.
M. N. Newquist, M. D.
A. C. Pabst
R. E. Eckardt, M. D. - Chairman

In addition, the following individuals attended the meeting as guests:

R. D. Bent
Lamson Blaney, M. D.
C. F. Boys, M. D.
C. T. Brown
R. C. Cole
L. E. Curtis, M. D.
T. M. Frank, M. D.
H. W. Gerarde, M. D.
N. J. Gothard
J. H. Hege, M. D.
R. M. Landon
B. B. Reeve, M. D.
F. J. Sanders
L. J. Wade, M. D.

The purpose of the meeting was to discuss the report of the Subcommittee to the MAC that afternoon. In addition to the three points mentioned in the Chairman's letter to the Subcommittee, it was decided to add a fourth point; namely, that the Subcommittee solicit comments and criticisms from all members and advisors of the MAC about the outline of the final report of the work at Kettering Laboratory which was attached to Dr. Horton's Interim Report dated September 26, 1957.

The Chairman of the Subcommittee then summarized for those in attendance the present status of the wax situation. Briefly, the situation is that a Mr. Kellogg of the Milk Industry Foundation in Washington had collected 40 wax samples from dairies which he had forwarded to Dr. Hueper. Twenty-four of these were forwarded to Dr. Shubik and UV absorption data on these 24 samples obtained by Dr. Shubik's chemist indicated that the UV absorption at 290 millimicrons varied from 0.04 to infinity. From one sample, in which the total absorption at 290 had been greater than 3.0, he chromatographed and isolated 4 micrograms of 1,2,5,6-dibenzanthracene

per gram of wax. It was the considered opinion of Dr. Shubik that he believed this quantity of the known carcinogen, 1,2,5,6-dibenzanthracene, in milk carton wax is undesirable and that the petroleum industry should establish an immediate standard which does not permit this much carcinogen in the finished product. Long range work then would attempt to establish a standard based upon biological testing.

In discussions which the Chairman had with Dr. Hueper at the recent Lung Cancer Workshop at the American Cancer Society, Dr. Hueper indicated that he was willing to sit still as long as the industry was willing to approach this problem with a reasonable, rational attitude, and as long as he was kept informed of the results of any work which was done. The Chairman indicated to the Subcommittee that the Three-Man Wax Committee appointed by the Medical and Health Committee of the Board, had met in Chicago on October 24 and unanimously recommended to the Medical and Health Committee, that work be done on waxes to the extent of approximately $100,000 a year for the next three to five years in Dr. Shubik's laboratory. Copies of the report of the Three-Man Wax Committee will be made available to the members of the MAC through Mr. E. O. Mattocks' office of the API.

The meeting was adjourned at approximately 10:30 a.m.

R. E. ECKARDT, M. D./cjh
Chairman, Subcommittee on Carcinogenicity
November 14, 1957