



PHS

Research

SRE/AIV/3.5

19th September, 1973

FOR FILING

Dr. E. Karbe,
Battelle-Institut, EV.,
6 Frankfurt am Main 90,
Postfach 900160,
West Germany.

Dear Eberhard,

Following my visit last week, I would like to confirm the main points of the discussion as follows : -

Battelle Research Conference

A conference on "Experimental Carcinogenesis and Bioassays" is being held at Battelle Seattle in June, 1974. Yourself and Jim Park (Battelle North West?) are joint organisers of the conference. Approximately 45 speakers will be invited and about 60 "auditors" also by invitation.

Skin Painting

a) Long-term Experiments

Only a few animals are still alive in the B10 experiment now at week 125. These animals were found to have a respiratory infection on arrival in Battelle in 1971 and the death rate is higher than that found in other experiments. Nevertheless, no recurrence of the infection has been observed. In contrast, the mice for the B11 experiment are of high quality and the death rate is low.

The design of the next long-term experiment was discussed in detail. Because of the delays in sample manufacture and in the availability of Grade 4 mice, I suggested that it might be possible to start all the long-term samples at the same time. This would be preferable from a statistical point of view since it would permit direct comparison of all ten samples. I accepted that this plan would delay the start of the promotion experiment. You agreed to advise me on the practicability of this plan within two weeks; a provisional order has been placed with Carworth for the animals.

The final sections of the report on long-term experiment B2 were discussed; the report will be printed in September. I asked that a start on the B3 report should be made as soon as possible; the revised computer reports, incorporating the new definition of a tumour bearing animal, will be sent to Battelle.

100995403

GK is currently undertaking the histology of specimens from experiment B8. Approximately 50% of the B6 data is coded and the remainder will be finished in the near future.

b) Promotion Experiments

The current experiments are proceeding satisfactorily. It was agreed that the experiment, now at week 26, should continue to week 32 and that a decision on the exact date of termination would be taken on the basis of the results to week 28.

The design of the promotion experiment was discussed. As with the long-term test, it was suggested that the number of mice used for the eight variants should be lower than that used for the control tobacco and reconstituted sheet samples. The numbers suggested were 84 and 126 respectively.

Inhalation Studies

a) Promotion Study (MNU pre-treatment)

The report on this study has been delayed so that you had the opportunity to refine the scheme used for classifying the various lesion classes. The method has been discussed with some other pathologists, eg. at Battelle North West and also with Dentenwill. It was also appropriate to re-discuss the classification scheme, since DW was present at the meeting.

In the classification system, the total number of lesion classes has been reduced to 6 including 0, ie. no change or hyperplasia. Grades 0 and 1 are exactly analogous to those used by Dentenwill. Dentenwill's grading of 2, 3 and 4 has been condensed into classes 2 and 3, while the Battelle grades 4 and 5 are equivalent to Dentenwill's 5 and 6. The descriptions you used are given in histological terms only, but it should be noted that grade 4 would include carcinoma in situ, while grade 5 (epidermoid carcinoma) would describe an invasive squamous cell carcinoma.

When you visited Dentenwill, you were encouraged to develop your own scheme. I was interested in your comment that Dentenwill had been over concerned with terminology used in human pathology and that the term leucoplakia, which is specified in four of Dentenwill's lesion classes, is not well defined.

DW had the opportunity of examining some of the slides and in general agreed with the classifications including those of grades 4 and 5 which he examined. The only minor difference appeared to be one of terminology.

In discussing the results, it was apparent that the modifications to the classification scheme have resulted in only minor changes to the overall results, ie. there is still a 4:1 ratio between smoke exposed and control (MNU treated) hamsters when considering the most important dose-response groups.

100995404

If the other groups (MNU in saline, etc.) are included, the ratio is even greater. The report on this experiment will be finalised when a "blind test" has been undertaken on the slides.

A considerable number of lesions were found in the hypopharynx, but in this case, the ratio of class 3 lesions is in favour of the hamsters not exposed to smoke. It should be noted, however, that the lesions had a papillematous appearance which is not found in laryngeal lesions. DW asked whether there was any relation between the incidence of lesions in the larynx and the hypopharynx, but EK did not think one existed. You commented that Dentenwill had also found quite a number of pharyngeal lesions.

A few detailed aspects of the follow-up inhalation promotion study were discussed. This experiment is now delayed pending the manufacture of reconstituted tobacco sheet and cigarettes. You advised me that the starting date of the inhalation exposure had to be defined 3 months in advance, since the minimum time necessary to produce the 400 hamsters was 3 months. Shipment, quarantine and pre-treatment with NNU would take one further month.

b) Vitamin A Study

Satisfactory progress with this experiment is being maintained. The levels of vitamin A determined by analysis of liver samples show that this ranges progressively from 156,000 IU/100g to zero at the two lowest levels of vitamin diet. In contrast to hamsters reared on the special diet, a small group of animals placed on the special diet at 4 weeks had an appreciable level of vitamin (2,100 IU/100g) in the liver after 5 months. The effect of the very low levels of dietary vitamin A have been found in that high proportions of the hamsters have died, the effect being most marked in the smoke exposed groups. Although only a limited number of sections have been examined, the finding of metaplasia with hyperkeratosis and purulent laryngitis are consistent with marked vitamin A deficiency. The groups maintained on the three intermediate levels of vitamin A are healthy and the death rate in the smoke exposed groups is only slightly higher than that in the control groups.

Miscellaneous

a) Analysis of Inhalation Experiments

A brief discussion was held with Dr. Langbein following a further exchange of detailed comments between Southampton and Battelle on the statistical methods to be used for the analysis. It was agreed that the stage had been reached when it would be appropriate for Dr. Langbein to visit Southampton to discuss the various outstanding points. This was arranged for 27/28th Sept.

b) Coding Forms for Skin-Painting Data

In the early long-term experiment, Königemann recorded on each mouse record card all the histological data from the slides.

100995405

-4-

Subsequently, this data was translated into the computer codes and recorded on standard coding forms. To simplify this process new mouse record cards were produced so that the data could be coded simultaneously and entered directly onto the mouse record card. Various techniques for transmitting the coded data were considered and I agreed to discuss the alternatives in Southampton.

As usual I will expect you to write to me with comments on any points which are not adequately covered.

On behalf of David Walker and myself, I would like to thank you for your kind hospitality.

With kind regards,

Yours sincerely,



S. R. EVELYN

cc: Dr. F. A. Sacherer
Dr. R. M. E. Sullivan,
Battelle, London.

noo. Dr. S. J. Green ✓

100995406