

INH/LT /46D

27th February, 1963.

PRELIMINARY SUMMARY OF THE RESULTS OF THE EXPERIMENTS
ON THE CARCINOGENICITY OF TOBACCO SMOKE CONDENSATE
DURING THE YEAR 1961 - 1962

BY

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SUMMARY

The following paper is not a draft for publication. It is preliminary to that and above all it is only intended for internal use of those interested. It is also "one-sided" in that it must be remembered that the co-workers of the scientific staff of the B-A.T. in addition have a say.

The experiments were begun at the instigation of the Board of B-A.T. The foundation of the scientific research constant in VdC is concerned with this. It is also well to remember that it is accomplished only in co-operation with the scientific staff of the B-A.T.

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EXPERIMENTAL PROCEDURE

The series of experiments can be divided into four groups:

A. Controls.

30 mice - not shorn or treated in any way with chemicals or tobacco smoke ingredients, designated as unshorn controls. 40 mice were shorn but received no application of anything - designated as shorn controls.

Paper A - medium porosity 31 ℓ /hr., high burn rate

Paper B - medium porosity 48 ℓ /hr., high burn rate

Paper C - low porosity 15 ℓ /hr., low burn rate

B. Standard Experiment.

The mice were painted three times weekly or three times a week with tobacco smoke condensate. The standard cigarette has medium porosity paper.

C.

Same tobacco as standard cigarette with low porosity paper and high porosity paper. Reconstituted tobacco in medium porosity paper.

D. "Catalytic" Experiments.

The tobacco and paper same as in the standard cigarette. The tobacco was mixed with additives, namely ammonium sulfamate, ammonium persulphate, and it is known that these additives reduce the carcinoma producing materials in tobacco smoke tar. These carcinoma inhibiting properties will occur when the tobacco is impregnated with 5% solution in small doses, namely 1% cerium oxide and n-propyl gallate also in 1% doses.

It was the carcinogenicity of the various cigarettes to be examined therefore the tar content of the tobacco smokes were determined; the smoke condensate from the cigarette was painted on to the mice in a 50% acetone solution. In the case of the other condensates, the concentration of the solution was determined by the yield of tar. Each mouse was painted with 0.07 g. of solution. In total therefore each mouse

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received in 191 paintings, 13.37 g. of tobacco tar solution. This quantity is, when one examines the literature, relatively small, but as the results show is sufficient.

When a tumour was formed and its diameter reached 5 mm., the tumour was removed. In the case of some of these animals operated on, a new growth later appeared.

RESULTS

Surprisingly, tumours were found in the case of the control, this peculiarity was not to be expected. In the case of the shorn controls it is noteworthy that the carcinoma was found on the 'clipped face'. In the case of the unshorn controls and of the acetone treated controls lip tumours and the sebaceous gland tumours were found.

The finding of these tumours showed that the strain of mouse used can develop spontaneous tumours and this must be borne in mind throughout these experiments. The mouse strain is unsuitable therefore for further experiments. For new experiments the use of other mice strains is foreseen.

In the case of the mice treated with condensate from the smoke from standard cigarettes there was found one papilloma which regressed, leading to malignant tumour and carcinoma. Among the others was found a sarcoma and a sebaceous gland tumour. Malignant tumours were found in 23% including papillomas 25.7% (round figures 26%). In absolute terms 39 mice gave 8 animals with tumours, 2 animals have 2 tumours. After correcting for the controls an induced incidence of 20% was found. In all other experimental groups less tumours were found. The next highest was found with cigarettes having high porosity paper: 15% malignant tumours, 22.2% mild and malignant tumours. This result was close to the standard cigarette.

In this group the lowest tumour incidence was found in the case of cigarettes with the low porosity paper and in the case of the cigarettes containing reconstituted tobacco. In both cases further experiments are required and in the case of the

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reconstituted tobacco new experiments are planned and should start this month (??). It is not going to be discontinued and further work will be done to clarify the effects of paper porosity.

In the catalytic experiments the tumour incidence in the case of ammonium sulfamate and ammonium persulphate and ceric oxide was lower than in the case of the standard cigarette: on the basis of malignant tumours the incidence is half, after mild papillomas are included two thirds.

For these calculated effects the concentration of added materials was 1% and the results are not to be neglected. For a safe cigarette with these chemicals higher concentrations would be required.

Next text difficult to translate but appears to suggest he is commenting on the possible limits of concentration in relation to the detection of smoke.

In the case of n-propyl gallate, the tumour incidence is less than that of the control. It gives the same order as the low porous paper. The question is then raised of what happens with a combination of both, would they for example cancel each other out.

Summarising then, using either low porosity paper or n-propyl gallate a reduction in carcinogenicity is effected. The results are then quoted.

Then follows a section which I think suggests that it would be a good idea to measure the benzpyrene, phenols, coal temperature etc., to find out what is happening.

The significance of spontaneous tumours on mice by brushing with tobacco.

In addition to the spontaneous skin tumours, which have already been mentioned, other spontaneous tumours are also observed on the mouse variety under investigation: leukaemia (tumours of the blood cells, in the present case exclusively by a type of lymphocytes), lung tumours, mammary tumours, and tumours of the germ glands (gonads). It is interesting to note that among the 315 experimental animals there is more leukaemia and more lung tumours and tumours of the gonads than among the 109 control animals used

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for comparison, but that they have fewer mammary tumours. It seems quite possible that the lower incidence rate of cancers of the breast is caused by the fact that the females, which were originally expected to perish from these tumours, had in fact already died from other tumours (leukaemia, lung tumours, and gonad tumours). This is because tumours of the breast glands are mainly found on aged animals, i.e. they are a typical disease of old age.

In the case of statistical investigations among non-smokers and smokers, a slight general increase in the incidence rate of tumours not localised in the lung has again been established among smokers, this increase being comparable with our own results, but nevertheless this characteristic parallel has not hitherto been recognised. Indeed, it would be wrong to equate this observation merely with the increased incidence rate of lung cancer among smokers. Like the skin cancers of mice, the lung cancer of smokers is in fact a genuine induced cancer, whilst the increased incidence rate of the other tumours is not likely to indicate genuine induction. Instead, other causes must be responsible for this fact, possibly an influence of nicotine on the vascular system or other changes brought about by tobacco smoke and its tar.

Yet another note regarding the mastocytomas: These are tumours with a special type of cell, the mastocytes. It has only recently been recognised that clusters of these tumours appear on mice after brushing with tobacco smoke. Our pictures of mouse skin sections from the regions which had been subjected to brush application provide a good material in evidence that the mastocytes first accumulate in the cutis, that they increase there, forming more or less loose clusters, and that later these coagulate to form genuine mastocytomas. It is not known which constituents of tar are responsible for their development. It is rare to find mastocytes among men. This may be the reason for the fact that so far no attempt has been made to investigate whether their incidence rate is greater among smokers than among non-smokers.

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Another problem relates to tumours of the bladder, which are found more frequently among smokers than among non-smokers. It has also been described that after the induction of cancer by the brush application of polycyclic compounds to the skin of rats and mice, changes occur in the urinary bladders of the animals which in part even degenerate to form tumours. We have again observed these changes which were quite extensive on the epithelia of the wall of the bladder of our animals during the time when the brushing of tar was first applied, but they faded away later on and disappeared when the brush treatment was discontinued. Thus, at the end of the experiments, no difference could be established between the bladders of the experimental animals and those of the control animals. Consequently, brush application of tar first led to abnormal changes in the epithelia of bladders which resembled an inflammation of the bladder (cystitis), but this decreased in intensity as the experiment proceeded (probably owing to adaptation or familiarisation), and which ceased entirely when brush application was stopped.

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