The Carcinogenicity of Wood Soot from the Chimney of a Smoked Sausage Factory

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In the course of a study of the estrogenic activity of plant materials in this laboratory (21) our attention was drawn to the fact that wood soot extracts have a slight estrogenic activity. It seemed of interest to ascertain whether this estrogenic activity is combined with a carcinogenic action.

In 1922 Passey (4) reported that extracts of soot collected from chimneys in England can evoke skin warts in animals. The well-known occurrence of scrotal carcinoma in chimney-sweeps, first described by Percivall Pott (1775), has been associated with the formation of carcinogenic substances in burning coal. Brownlie (1) has suggested that organic products of the decomposition of carbonaceous materials under high temperature conditions (especially bituminous coal, petroleum oil, coal-tar pitch, briquettes, illuminating gas, smoked foods, shale oils, etc.) can act as carcinogens. In numerous papers Roffo (5-17) has asserted that sometimes carcinogenic substances are formed by the burning at high temperatures of tobacco and coffee. Widmark (20) produced mammary adenocarcinomas in 40 per cent of female mice by applying extracts of roasted (275° C.) horse muscle, coffee, and brown butter. The spontaneous frequency of malignant tumors among his females was, however, rather high (10 per cent). Liu and Hu (3) produced papillomas in 4 out of 230 mice with tar obtained by destructive distillation of various vegetable foodstuffs. Dickens and Weil-Malherbe (2) produced 1 experimental skin papilloma and 8 pulmonary adenomas by means of wood smoke in 17 mice. Rats fed with smoked fish and meat did not, however, develop tumors during 20 months of observation. The experiments of Steiner, Steele, and Koch (18) suggest the possible carcinogenicity of overcooked meat, heated cholesterol, acrolein, and heated sesame oil. Whereas heating up to 300° C. did not produce carcinogenic substances, sesame oil that had been heated to 350° C. was carcinogenic in 3 out of 9 mice.

In view of these findings we undertook to ascertain:

(a) whether soot from wood is carcinogenic in our rat and mouse strains, which normally have a natural tumor incidence of about 0.1 per cent;

(b) whether the daily consumption of unlimited quantities of smoked sausage is carcinogenic for animals.

TECHNIC

The soot was obtained from the smoking chamber of a sausage factory in which eucalyptus timber was used for the smoking, where it had formed thick, black, coal-like masses. It was used either as fragments for implantation, or extracted from pulverized material with ether and afterwards with alcohol in a Soxhlet, and then injected. The extracts were pooled, the ether was evaporated at 40° C., and the alcoholic remnant was diluted with 96 per cent alcohol to a concentration that was one-tenth of the lethal dose for mice when 1 drop of the solution was licked. This precaution was taken in order to avoid primary mortality in mice painted with the extract, and secondary mortality in those sharing the same cage, for it is well known that mice sometimes lick extracts applied to the skins of their cage mates. Since separation of the mice used for this experiment was not advisable, care was taken to rub the soot extract well into the skin; thus optimal resorption ensued and little residue remained on the skin to be licked.

First Series

Thirty-six adult rats in all were implanted with pieces of soot weighing 5 to 20 mgm. in 18 (females, weighing 120 gm.) the particles were implanted subcutaneously near the right axilla; in the other 18 (males, weighing 150 gm.) the implantation was made in the scrotal sac between the testicles. In the male group tumor formation did not occur in the course of $2\frac{1}{2}$ years, a result that may have been due to low responsiveness of the site of implantation. Of the 18 females that had been implanted with the soot subcutaneously 3 reacted with sarcoma formation at the site of implantation after 12, 17, and 24 months respectively. These sarcomas did not metastasize in the original animals. When transplanted into 30 rats a take of about 50 per cent was recorded. In 36 male and female controls observed during the same interval no tumor was found.

Second Series

Ten adult female mice were treated with an alcoholic extract of soot on the dorsal skin of the neck once daily for 2 years. All in this group developed a cataract within a quarter of a year, a finding that sug-

gests the presence of derivatives of the naphthalene group. All the animals, furthermore, showed permanent estrus, which would indicate that the extract contained an estrogenic phenol derivative. The latter finding is hardly astonishing. Neoplasms occurred in 3 of the animals (30 per cent) and they died of their tumors after 5, 12, and 21 months respectively. Two of the tumors were sarcomas, situated in the vicinity of the urinary bladder, and one was a carcinoma of this viscus itself. No metastases were found. Implantations were made from each of the 3 tumors intraperitoneally near the kidney in 20 mice; only 1 take was recorded, this from a carcinoma implant. No neoplasms developed in 20 control mice of the same strain during an observation period of 2 years.

Third Series

Twenty adult rats were fed for 2 years on a diet that contained smoked sausage *ad libitum*. None of this group reacted with tumor formation during the period of the experiment.

DISCUSSION

The consumption of smoked sausage over a period of more than 2 years did not cause tumors in rats. However, the wood smoke used for its preparation was shown to contain substances that were carcinogenic for rats when introduced subcutaneously, and for mice when rubbed into the skin. The sarcomas obtained in 16.6 per cent of the rats following the implantation of soot particles may not appear significant, in view of the finding by Turner (19) that sterile bakelite disks, when similarly implanted in rats, elicited sarcomas in 31 per cent of the animals. Our findings become significant, however, by reason of the carcinogenicity of the soot extracts for mice (33.3 per cent) when rubbed into the skin. A further study of the possible carcinogenic role of wood soot extracts therefore seems desirable.

SUMMARY

1. Thirty-six female rats implanted subcutaneously with fragments of soot from the chimney of a sausage factory developed sarcoma in 16.6 per cent of the cases. No tumor developed in 36 male rats implanted intrascrotally with bits of the same soot.

2. Ten female mice treated for 2 years with an ether and alcohol extract of wood soot showed tumor formation in 3 cases (2 sarcomas and 1 carcinoma).

3. Twenty rats fed for 2 years on a diet containing an unlimited amount of smoked sausage failed to develop tumors.

4. The conflicting finding, carcinogenic effect in parenteral treatment versus absence of carcinogenic effect after oral administration, indicates the need for further study of the carcinogenic activity of smoked food, in view of the practical importance of the problem for human nutrition.

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REFERENCES

1. Brownlie, D. The Cause of Cancer. London: Chapman and Hall, Ltd. 1938, pp. 8, 9, 32, 56, 67, 69, 182, 191.
2. Dickens, F., and Weil-Malherbe, H. Investigation into the Possible Carcinogenic Activity of Wood Smoke. Cancer Research, **2**:680-684. 1942.
3. Liu, Y., and Hu, C. H. Experimental Production of Tumors by Tars from Various Foods. Proc. Soc. Exper. Biol. & Med., **48**:226-227. 1942.
4. Passey, R. D. Experimental Soot Cancer. Brit. M. J., **9**: 1112-1113. 1922.
5. Roffo, A. H. El tabaco como cancerigeno. Bol. Inst. Med. Exp. & Cancer, Buenos Aires, **13**:287-336. 1936.
6. Roffo, A. H. El tabaco rubio como cancerigeno. Bol. Inst. Med. Exp. & Cancer, Buenos Aires, **15**:1-22. 1938.
7. Roffo, A. H. Unidad cancerigena de los alquitránes de diversos tipos de tabaco. Bol. Inst. Med. Exper. & Cancer, Buenos Aires, **15**:349-406. 1938.
8. Roffo, A. H. El alquitrán de café cancerigeno. Bol. Inst. Med. Exp. & Cancer, Buenos Aires, **15**:741-762. 1938.
9. Roffo, A. H. 1,2-Benzopirene, cancerigeno extraido del alquitrán del tabaco. Bol. Inst. Med. Exp. & Cancer, Buenos Aires, **16**:1-38. 1939.
10. Roffo, A. H. Tumours malignes développées dans l'appareil digestif par l'ingestion de graisses oxydées par chauffage. Bull. Assoc. franc. p. l'étude du cancer, **28**: 556-588. 1939.
11. Roffo, A. H. Sobre los filtros en el tabaquismo. El narguilé, y el algodon como filtro del alquitrán de tabaco. Bol. Inst. Med. Exp. & Cancer, Buenos Aires, **16**:255-268. 1939.
12. Roffo, A. H. Alquitrán de café cancerigeno. Bol. Inst. Med. Exp. & Cancer, Buenos Aires, **16**:445-446. 1939.
13. Roffo, A. H. Cancerizacion gástrica experimental por ingestion de grasses oxidadas. Bol. Inst. Med. Exp. & Cancer, Buenos Aires, **19**:503-530. 1942.
14. Roffo, A. H. Carcinoma tabáquico de gran malignidad desarrollado en un conejo. Bol. Inst. Med. Exp. & Cancer, Buenos Aires, **20**:1-50. 1943.
15. Roffo, A. H. Carcinoma del pulmón experimental par alquitrán tabáquico. Bol. Inst. Med. Exp. & Cancer, Buenos Aires, **20**:103-122. 1943.
16. Roffo, A. H. Sobre la malignidad de los tumores producidos par alquitrán tabáquico. Bol. Inst. Med. Exp. & Cancer, Buenos Aires, **20**:189-202. 1943.
17. Roffo, A. H. Cancerización gástrica por el alquitrán de café. Bol. Inst. Med. Exp. & Cancer, Buenos Aires, **20**: 425-470. 1943.
18. Steiner, P. E., Steele, R., and Koch, F. C. The Possible Carcinogenicity of Overcooked Meats, Heated Cholesterol, Acrolein, and Heated Sesame Oil. Cancer Research, **3**: 100-107. 1943.
19. Turner, F. C. Sarcomas at Sites of Subcutaneously Implanted Bakelite Disks in Rats. J. Nat. Cancer Inst., **2**: 81-83. 1941.
20. Windmark, E. M. P. Presence of Cancer-Producing Substances in Roasted Food. Nature, London, **143**:984. 1939.
21. Zondek, B., and Bergman, E. Phenol Methyl Ethers as Oestrogenic Agents. Biochem. J., **32**:641-645. 1938.