SOME PHYSIOLOGICAL EFFECTS OF NON-GASEOUS EXHAUST MATERIAL FROM AN INTERNAL COMBUSTION ENGINE^{1, 2}

ALLEN W. SCHOLL

Department of Chemistry, Ashland College, Ashland, Ohio

ABSTRACT

Exhaust materials were collected from a four-cylinder automobile engine that consumed 1 quart of lubricating oil for each 5 gal of gasoline.

The oily condensate was separated into three fractions. These fractions contained paraffins, olefins, and cycloparaffins as fraction I, aromatics as fraction II, and non-hydrocarbons and oxygenated compounds as fraction III.

The solvents produced no noticable effects on test mice after 279 days of testing. Fractions I and II produced ulcerated sores which formed thick scabs. Fraction II also produced a small amount of cancerous cells at the site of application. Fraction III produced thin scabs and was associated with hyperirritability and other evidence of central nervous system changes. This fraction was lethal in doses of 100 mg per week.

A search of the literature discloses that little work had been reported prior to 1952 on the analysis of non-gaseous materials exhausted from internal combustion engines. Maillard and Friedrich (1937) found that when gasoline was burned under certain conditions, acetic and formic acids, formaldehyde, acrolein, and phenol could be detected in the products of combustion. Maillard (1937) examined the oily and water layers condensed from automobile-engine exhaust fumes and identified acids, aldehydes, and neutral cracked hydrocarbons, but he did not examine these further.

Passano (1942) found that some combustion products of fuel oil and highboiling petroleum fractions were capable of producing carcinogenic tumors in rats. Cook, Hewett, and Hieger (1933), in studying the high incidence of cancer cases among workers of the coal tar industry, discovered that polycyclic aromatic compounds, especially those containing from two to six fused benzene rings, were the principle carcinogenic agents.

MATERIALS AND METHODS

Analytical grade, petroleum ether, benzene, and methanol from the J. T. Baker Co. were used as the eluting solvents. Methylcholanthrene from Eastman Organic Chemicals, a known carcinogen, was used as a control compound.

Chromatographic columns were constructed from 4-ft sections of 45 mm ID Pyrex tubing. Each column was filled with 3 lb of silica gel, 28 to 200 mesh, which had been activated for 5 hr at 150°C. Chromatographic grade silica gel was purchased from the Fisher Scientific Company.

A four-cylinder automobile engine was operated on regular gasoline to which S.A.E. 20 paraffin base lubricating oil free of additives was added. The gasolineoil ratio was twenty to one. The exhaust materials were condensed by passing the fumes through coils of $\frac{3}{4}$ -inch copper tubing. The condensate was then collected in two 5-liter flasks connected in series and cooled in an ice bath.

The condensate was fractionated into three fractions by the chromatographic procedure outlined by Scholl, Bubernak, and Galford (1951). Fraction I, eluted by petroleum ether, contained paraffins, olefins, and cycloparaffins. Fraction II, eluted by benzene, contained aromatic compounds, and fraction III, eluted by methanol, contained non-hydrocarbon and oxygenated compounds. Each eluted

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No. 3

fraction, the eluting solvents, and the original condensate were tested by the skin painting technique.

All test animals were 8-week-old male mice. They were housed in standard mouse cages in groups of ten. The average weight per animal for each test group was 23 g. The animals were of the C_3H (Jax) strain reared in this laboratory from stock obtained from the Jackson Memorial Laboratory, Bar Harbor, Maine. The animals were fed ad libitum, Rockland Rat Diet, purchased from the Rockland Farms, New City, New York. Water was supplied from glass bottles attached to the cages. The floor of the cages was covered with wood (Pine strobus) shavings. The cages were cleaned and sterilized twice weekly.

A group of ten animals were used as controls. Animals were tested by applying the test material to the bare scapular area daily Monday through Friday. Number three artist's brushes were adjusted by trimming so that 10 mg of material would be deposited by a single brush stroke.

Each animal was weighed and examined once weekly.

EXPERIMENTAL DATA

The conditions of testing are summarized in table 1. The number of test animals used, the weekly dosage in milligrams, duration of the test in days, and the survival are given in columns 2, 3, 4 and 5 respectively.

Test material	No. of animals	Weekly dosage* (Mg)	Duration of test (Days)	Survival
None (Control)	10	None	279	10
Petroleum ether	10	100	279	10
Benzene	10	100	279	10
Methanol	10	100	279	10
Orig. Condensate	20	100	279	18
Fraction I	20	100	279	19
Fraction II	20	100	279	19
Fraction III	20	100	-33	0 after 33 days
Fraction III	10	50	103	5 after 68th day
Fraction III	10	25	244	9 after 202nd day
Methvlcholanthrene				5
0.05% sol. Benzene	30	180	152	26
Methylcholanthrene				
0.10% sol. benzene	30	280	152	30

TABLE 1							
Skin test data for exhau	st fume condensa <mark>te an</mark> d	solvents on	$C_{s}H$ mice				

*Application twice daily Monday through Friday, of one-tenth dose listed.

DISCUSSION

The original condensate did not show effects of dermatitus after 279 days of testing. All animals showed a normal growth rate similar to that of the control animals. The solvents used in the separation of fractions I, II, and III did not produce ill effects, and the growth rate of these animals was normal when compared to the controls.

Fraction I produced ulcerated sores which formed thick hard scabs. These sores increased in size as testing continued. The underlying tissue cells were not found to be malignant.

Fraction II formed thick hard scabs, or encrustations, which would form, drop off, and then reform. The underlying skin was not ulcerated as was the case with fraction I. Neither fraction I nor II produced detectable papillomas. The skin tissue of mice painted with fraction II showed some cancerous cells when examined pathologically. (Examination by F. C. Hodges, M.D., Pathologist, First Huntington National Bank Building, Huntington, West Virginia.)

Fraction III produced effects different from fractions I and II. Animals receiving 100 mg per week began to show signs of hyperirritability on the tenth day of testing. These animals were easily excited by noise or movement. The animals would leap into the sides of the cages. When they were allowed on the table top they would leap as much as 2 to 3 ft. They would try to hide whenever possible. The control animals were very tractable and undisturbed by the same After 24 days of testing, eleven of the animals were dead, and all 20 stimuli. were dead after 33 days of testing.

Fraction III did not appear to form sores or scabs. A darkening of the underlying tissue, at the area of application, was found in these animals. All organs appeared normal.

Doses of 50 and 25 mg per week were investigated, since the 100 mg dose was These produced the same hyperirritability as described above in proporlethal. tion to the size of the dose. Fraction III was much more toxic than either fraction I or II.

Control tests were carried out with the known carcinogen methylcholanthrene. These tests were continued for 152 days. The first papillomas appeared in 56 and 26 days, for weekly doses of 180 and 280 mg respectively.

The average latent period (i.e., the time it takes for 50 per cent of the surviving animals to develop tumors) for the 0.1 per cent methylcholanthrene was 103 days, as compared to 99 days as given by Greenstein (1947).

CONCLUSIONS

The oily condensate from the exhaust of internal combustion engines contains compounds that produce ulcerated sores, some cancerous tissue cells, hyperirritability, and evidence of central nervous system changes.

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