

## SKIN TUMORS FOLLOWING A SINGLE APPLICATION OF METHYLCHOLANTHRENE IN C57 BROWN MICE \*

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Connective tissue tumors have been produced with extremely small amounts of carcinogenic hydrocarbons.<sup>1</sup> A few tumors have followed the application of a single drop of hot tar to mouse skin.<sup>2</sup> It is generally believed that protracted painting with carcinogenic agents is necessary for the development of skin neoplasms. Fieser states that "in order to produce skin tumors in mice with even a highly potent carcinogen in benzene solution some 30 deliberate applications must be made."<sup>3</sup> This statement may be true for many strains of mice. Kreyberg<sup>4</sup> and Bonser<sup>5</sup> have established strains by selective breeding whose skin is especially sensitive to the action of carcinogenic substances.

In investigating the influence of methylcholanthrene on the development of internal tumors in mice the chemical was applied successively at 9 different sites.<sup>6</sup> Papillomas were found at the site of the first painting within 4 to 5 weeks among mice of the C57 brown strain. Investigation of this phenomenon seemed warranted.

C57 brown mice were obtained from the Roscoe B. Jackson Memorial Laboratory where the strain originated in 1925. They have a relatively high incidence of spontaneous internal tumors and less than 5 per cent of mammary carcinoma. The mice were painted when 4 to 5 weeks old with methylcholanthrene 0.5 per cent in benzene. It was applied to the back, from the occiput to the lumbar region, with 2 strokes of a No. 8 camel's hair brush. The animals were not allowed to breed. Sex distribution was equal. They were kept in glass cages and were fed Purina dog chow, water being available at all times.

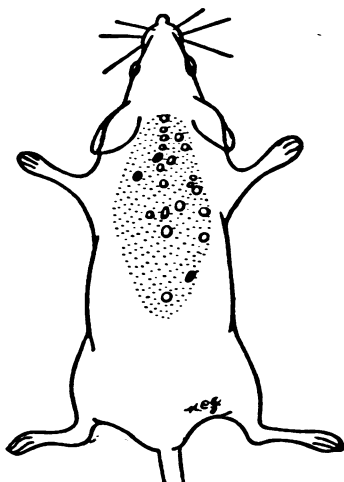
Forty-four mice were painted with methylcholanthrene. Epilation occurred within 10 days. Ulceration was rarely found. Six-

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teen of the animals developed a total of 22 papillomas in the painted area (Text-Fig. 1). The first tumor appeared in each mouse between the 31st and 48th days after painting. These lesions had all the gross characteristics of benign skin tumors produced by multiple paintings with carcinogenic hydrocarbons (Fig. 1). They varied from 2 to 8 mm. in diameter, grew rapidly at first and then passed into a stationary period during which their appearance was not altered. Marked variation in the duration of



TEXT-FIG. 1. Distribution of skin tumors in the area painted once with methylcholanthrene 0.5 per cent in benzene. Stippling indicates the painted area; circles represent papillomas; dots indicate carcinomas.

the tumors was noted (Table I). Fifteen of the papillomas regressed completely. The sites of the tumors showed no gross evidence of scarring after the lesions had disappeared. Hair eventually grew in the epilated areas. It was frequently white instead of brown.

Of the 7 papillomas that remained, 3 became malignant and 4 persisted beyond the 225th day after painting. The latter did not change in appearance for more than 100 days, and showed no evidence of progressive growth. The 3 carcinomas occurred in 1 male and 2 female mice on the 228th, 114th, and 124th days respectively (Fig. 2). Diagnosis was confirmed histologically, invasion of muscle being used as the criterion of malignancy (Fig. 3). No metastases were noted but the mice were sacrificed

as soon as clinical evidence of carcinoma was present. In contrast with mice painted twice weekly with methylcholanthrene 0.5 per cent in benzene, these animals remained in good health throughout the experiment.

This reaction of C57 brown mice is probably another instance of genetically determined increase in tissue susceptibility to an ad-

TABLE I  
*Skin Tumors Following a Single Application of Methylcholanthrene in C57 Brown Mice*

Number of mouse	Sex	Papillomas		Duration
		Appeared	Disappeared	
		<i>days</i>	<i>days</i>	<i>days</i>
1	M	34	102	68
2	M	46	81	35
3	M	38	94	56
4	M	48	76	28
5	M	48	76	28
6	M	41	107	66
		60	105	45
7	F	46	56	10
8	F	39	145	106
9	F	34	76	42
10	F	39	165	126
		42	137	95
11	F	32	105	73
		54	108	54
12	M	26	Carcinoma at 228 days	
		50	72	22
13	F	35	Carcinoma at 114 days	
14	F	31	Carcinoma at 121 days	
15	M	42	Persists at 225 days	
16	M	35	Persists at 225 days	
		54	Persists at 225 days	
		60	Persists at 225 days	

verse environmental stimulating factor. It differs from those produced by Kreyberg and Bonser only in the degree of susceptibility. This is in accord with the concept that carcinoma is not handed down in heredity as such. Cellular susceptibility or resistance to environmental factors is probably the mechanism concerned. A second explanation of the phenomenon would entail a change in the growth and differentiation of skin epithelium by long continued contact with methylcholanthrene. This seems less probable. The fluorescence associated with the chemical cannot be detected in the skin by ultraviolet light 1 week after its application. This is a

crude test and better analytical methods may yield a different result. Furthermore, it is entirely conceivable that a metabolite of methylcholanthrene may initiate neoplasia. Additional experimentation is necessary for the solution of this problem.

The production of skin tumors by a single painting with methylcholanthrene in C57 brown mice appears to afford further opportunity for the investigation of the fundamental physiological changes involved in carcinogenesis. Attempts to affect the biology of the benign tumors may be worth while.

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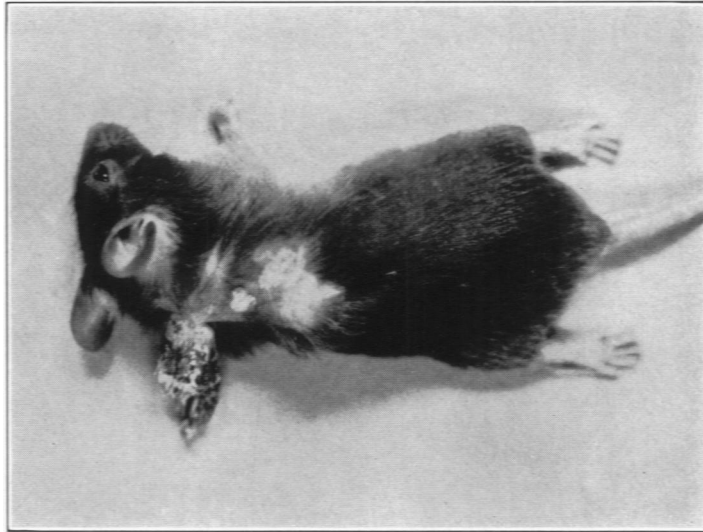
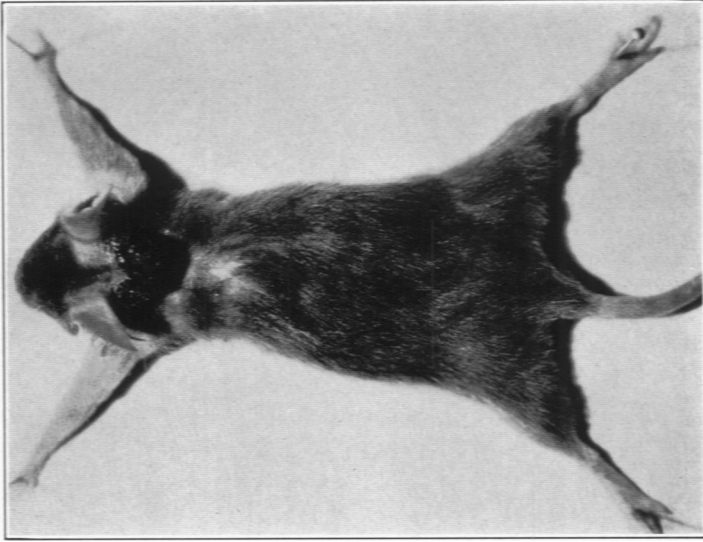
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#### DESCRIPTION OF PLATES

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##### PLATE 53

- FIG. 1. Papillomas in a C57 brown mouse painted once with methylcholanthrene 0.5 per cent in benzene. These tumors have been present for more than 225 days.
- FIG. 2. Carcinoma in a C57 brown mouse painted once with methylcholanthrene 0.5 per cent in benzene. A malignant tumor appeared 228 days after painting.



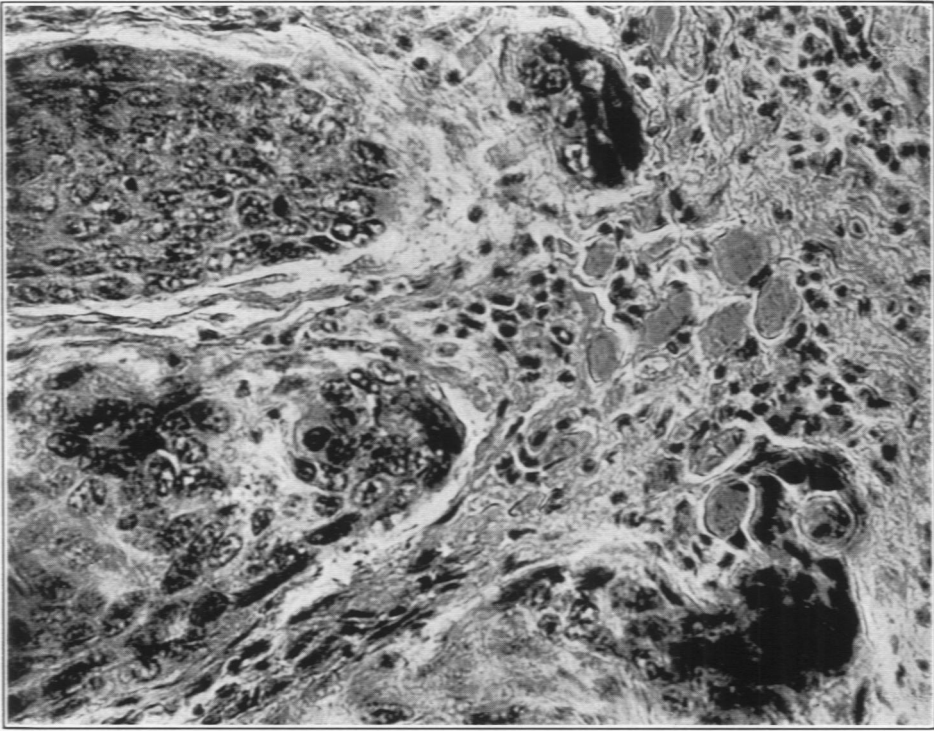
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Skin Tumors in C<sub>57</sub> Brown Mice

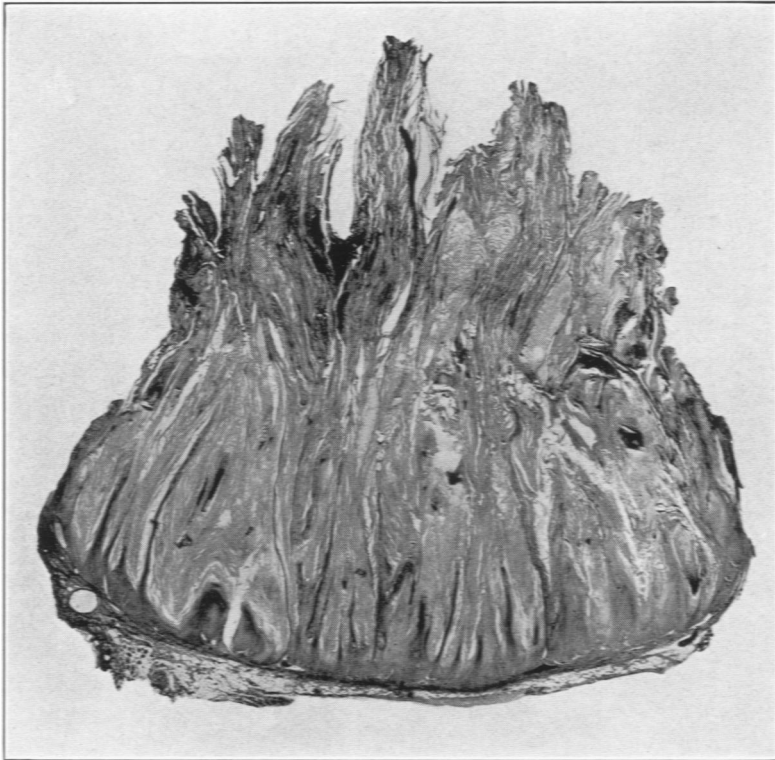
PLATE 54

FIG. 3. Microphotograph of carcinomatous growth shown in gross in Fig. 2. The tumor cells have invaded the muscle.  $\times 600$ .

FIG. 4. Microphotograph of a papilloma of the skin arising in a C<sub>57</sub> brown mouse after a single application of methylcholanthrene 0.5 per cent in benzene.  $\times 10$ .



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