

(9). Repeated efforts were made in our laboratory to produce chlorophyll deficiency in citrus by inoculation with *Aspergillus flavus*, *Aspergillus niger*, and other *Aspergillus* species isolated from the coats of albino-producing seeds (10). Also included were trials with *Aspergillus flavus* sent to us by Durbin. The results were completely negative.

The evidence indicates that the albinism occurring sporadically in citrus seedbeds is probably due to the action of *Alternaria tenuis* growing on the seed coats and producing a substance which interferes with chlorophyll formation in the developing seedling. The chemical nature of the active substance is being investigated.

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#### References and Notes

1. J. Perlberger and I. Reichert, *Palestine Agr. Research Sta. (Rehovot) Bull.* 24 (1939).
  2. J. M. Tager and S. H. Cameron, *Phyiol. Plantarum* 10, 302 (1957).
  3. H. B. Frost, in *The Citrus Industry* (Univ. of California Press, Berkeley, 1948), vol. 1, pp. 817-913.
  4. G. F. Ryan and Enrique Stein, *Calif. Citrograph* 43, 293 (1958); also in *Calif. Agr.* 12, 7, 12 (1958).
  5. F. A. Minessy, *Nature* 183, 553 (1959).
  6. Courtesy of E. C. Calavan, Department of Plant Pathology, University of California, Riverside.
  7. Courtesy of Lily H. Davis, Department of Plant Pathology, University of California, Los Angeles.
  8. R. D. Durbin, *Plant Disease Reprtr.* 43, 922 (1959).
  9. B. Koehler and C. M. Woodworth, *Phytopathology* 28, 811 (1938).
  10. Two cultures of *Aspergillus flavus* and one of *A. niger* obtained from Karl Olsen, Department of Plant Pathology, University of California, Los Angeles, were also tested.
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### Transmissible Agent Associated with Some Mouse Neoplasms

**Abstract.** The association between some forms of murine cancer and an agent that increases the lactic dehydrogenase activity in the plasma of normal mice is confirmed. This agent, however, is not associated with all mouse neoplasms.

All 26 murine tumors studied by Riley *et al.* had a transmissible agent associated with them. This was demonstrated by injecting normal mice intraperitoneally with 0.1 ml of plasma from mice bearing any of these 26 tumors,

Table 1. Lactic dehydrogenase activity in plasma of tumor-bearing mice and of mice injected with 0.1 ml of such plasma.

Donor mice		Enzyme activity in plasma	
No. and strain	Tumor	Donor mouse	Recipient mice
<i>Spontaneous tumors</i>			
1 (BR6)	Mammary carcinoma	74	61 ± 5(8)
2 (C3H)	Mammary carcinoma	38	44 ± 9(5)
3 (AKR)	Leukemia	540	43 ± 2(3)
4 (AKR)	Leukemia	750	43 ± 7(2)
<i>Transplanted tumors*</i>			
5 (C57/BL)	Sarcoma B/BP	47	56 ± 21(2)
6 (A)	Mammary carcinoma (i/36)	78	80 ± 24(5)
7 (C57/BL)	Sarcoma MC48	220	220 ± 12(6)
8 (A)	Fibrosarcoma (SA/γ)	790	235 ± 15(2)
9 (C57/BL)	Sarcoma 37	350	305 ± 35(2)

\* Originally spontaneous (i/36, Sa/γ, and 37) or chemically induced (B/BP and MC48).

whether spontaneously or as transplants. The lactic dehydrogenase activity in the plasma of the recipients increased several fold, and apparently the capacity to increase this enzyme activity can be passaged serially through normal mice indefinitely (1).

A similar agent is associated with some of the mouse tumors available to us, but not with all, so that the association is more probably coincidental than significant.

Our methods are exactly the same as those used by Riley *et al.*, but we express our results in units 10 times larger than theirs. (We define our unit as the activity reducing the optical density by 0.01 per minute per milliliter of plasma, and all results are expressed as average activity plus or minus the standard error of the mean, followed by the number of animals in parentheses.)

The plasma of normal mice of the four strains we have used (C57/BL, BR6, C3H, and A) has a lactic dehydrogenase activity of  $50 \pm 3(30)$ ; the activity is not affected by the strain or sex of the mouse. The lactic dehydrogenase activity in the plasma of mice bearing any sort of tumor tends to be high. (This is not unexpected since this enzyme activity is increased in a variety of diseased conditions.) In the case of a mammary adenocarcinoma that is transplantable into only a proportion of recipients, those in which the transplants are growing have an elevated enzyme activity in their plasma [ $138 \pm 5(4)$ ], and those in which it fails to grow have a normal activity [ $35 \pm 6(4)$ ]. Similarly, a mouse with a successfully transplanted lymphoma had an enormous lactic dehydrogenase activity in its plasma (1600) while mice in which transplantation failed had normal values [ $51 \pm 11(3)$ ].

Plasma (0.1 ml) from nine mice bearing various tumors (2) was injected intraperitoneally into normal mice. The lactic dehydrogenase activity in the plasma of the recipients was measured 2 days later. The results (Table 1) showed that the agent was clearly present in three mice (Nos. 7-9) bearing three sorts of transplanted sarcoma. The agent from one of these mice (No. 7) has been transmitted serially through seven passages; 2 days after injection of 0.1 ml of plasma, the lactic dehydrogenase activity in the plasma of each recipient was increased to 223-470. The activity in the plasma of the first recipient remained elevated for at least 1 month.

No agent was demonstrable in the plasma of the other six mice, and this was still so when higher volumes of plasma or cell-free extracts of the tumors were tested. These negative tumors were spontaneous mammary carcinomas (mice 1 and 2) or leukemia (mice 3 and 4) and transplanted sarcoma or carcinoma (mice 5 and 6).

If the difference between the positive and negative results is merely a quantitative one, then it must be enormous because active plasma is still fully effective after a 1000-fold reduction of

Table 2. Response to injection of diluted plasma from mice carrying agent derived from mouse 7 (Table 1).

Dose of plasma (ml)	Lactic dehydrogenase activity in plasma of recipient mice	
	Expt. 1	Expt. 2
0	34 ± 4(2)	
0.2	228 ± 24(5)	
0.1	222 ± 5(5)	293 ± 17(5)
0.05	288 ± 8(4)	
0.025	229 ± 10(5)	
0.0125	209 ± 23(4)	
0.01		280 ± 6(5)
0.001		298 ± 8(5)
0.0001		317 ± 20(5)

the normal dose (Table 2). It seems more likely to us that the difference between the two sets of tumors is an all-or-none difference: tumors either have the agent or they have not.

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#### References and Notes

1. V. Riley, F. Lilly, E. Huerto, D. Bardell, *Science* 132, 545 (1960).
2. We are very grateful to Dr. J. Craigie for supplying some of these mice.

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### Sign of Taxis as a Property of the Genotype

**Abstract.** Previous studies have assumed *Drosophila* to be negatively geotactic and found this to be the case. New methods of observation now yield a spectrum of geotactic behavior both positive and negative. Two-way selection produces races of animals performing in diametrically opposite fashion to the same stimulus conditions.

In the study of behavior it is usually assumed that for a given species there is a characteristic way of responding which defines a behavioral process under analysis. Though individuals may differ somewhat in the way they respond, the assumption is made that individual differences represent minor variations around the normal (1). In particular, taxes have been considered an invariant property of a species under a given set of conditions—for example, moths are positively phototactic, cockroaches negatively phototactic, and flies negatively geotactic (2).

In previous behavior genetic studies of *Drosophila melanogaster*, it was assumed that certain tactic orientations were normal, and individual differences were measured as deviations from the assumed normal orientation. Furthermore, these assumptions were often incorporated into the methods of observation and analysis, effectively guaranteeing that the results would accord with the preconception of the normal. In the study of geotaxis (3), individuals were scored on the strength of their negative geotaxis. The alternatives on every trial were those of responding or not responding, that is, ascending the walls of a test tube or failing to do so. In studies of phototaxis, individual (4) and population (5) differences were measured in terms of the strength of positive

phototaxis. The alternatives in these studies were effectively those of approaching a light source or not.

Recently, Lewontin (6) has shown how the sign of phototaxis can depend on the conditions of observation. *Drosophila pseudoobscura* that were agitated while responding were predominantly photopositive while those that were not agitated were predominantly photonegative.

The present study places no restrictions on the sign of the taxis. Apparatus has been developed which affords objective and automatic measurement of both positive and negative geotactic behavior in populations under a single set of stimulus conditions as well as reliable mass screening measurements of individual differences in the expression of each. The observations have been made in a 15-trial modification of the multiple unit classification maze (Fig. 1), which has been described elsewhere (7). The alternatives at each choice point in the maze require diametrically opposite responses, namely, going against the pull of gravity by climbing up (negative geotaxis) or going toward the pull of gravity by climbing down (positive geotaxis).

With the maze, geotaxis has been studied in an unselected wild-type population of *Drosophila melanogaster* developed by mixing in a population cage the Formosa stock from Berkeley, Calif., with freshly trapped Capetown and Syosset stock from Cold Spring Harbor, N.Y. The middle ogive in Fig. 2 shows that observation of the performance in the maze of large numbers of individuals from the unselected population reveals a response distribution which contains a spectrum of geotactic scores. The expression of geotaxis in this population ranges from a score of -7.5, which represents the extreme of negative geotaxis, through 0, a score indicating a neutral response to gravity, to +7.5, the extreme of positive geotaxis. The distribution is slightly skewed; 47 percent receive positive scores and 53 percent negative scores.

With a system of assortative mating, two-way selection pressure is applied. The maximum separation so far obtained was reached at the 48th generation of selection and is shown by the outer ogives in Fig. 2. The curve near the ordinate shows that 96 percent of the individuals in the strain selected for negative geotaxis receive a negative score in the maze, with 50 percent receiving the extreme negative score of

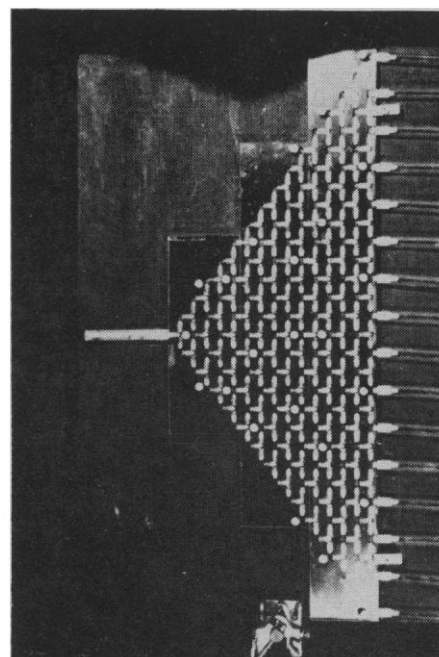


Fig. 1 Photograph of a 15-unit vertically placed maze. Large groups of flies are introduced into the vial at left and collected from vials at right. The flies are attracted by food in the vials at right and by a fluorescent light in a vertical position at the right. Small trap-like funnels discourage backward movement in the maze.

-7.5. The curve farthest from the ordinate shows that 95 percent of the individuals in the other strain receive positive scores in the maze, with 16 percent receiving the extreme positive score of +7.5.

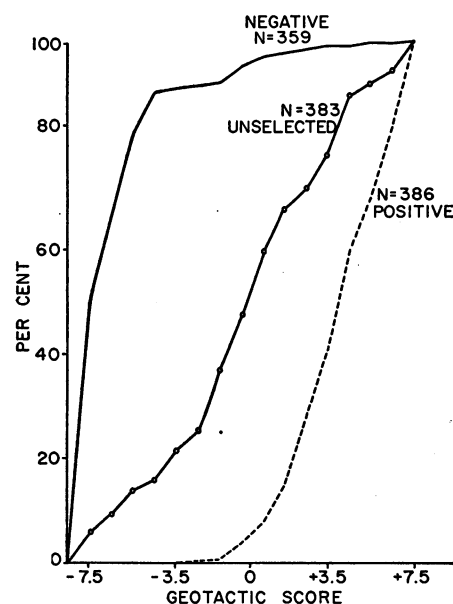


Fig. 2. Cumulated percentage of animals (males and females) receiving various geotactic scores in the maze in the unselected foundation population (middle ogive) and in the two selected strains (outer ogives).