triplicate set of cultures was incubated in a dark cabinet at 22–24° C, and the other set under fluorescent light at 450 ft-c at the same temperature.

Sample loopsful were taken from the tubes at intervals of 1 hr, 30 hr, 65 hr, and 168 hr, streaked on 2% agar plates of Maerten's solution plus 0.5% glucose, and incubated in the dark cabinet.

A straight-line relationship between streptomycin concentration and killing, as well as between duration of treatment at a given concentration and killing, was observed. *Scenedesmus, Stichococcus,* and *Chlorella* sp. were similarly affected by the drug, showing few surviving cells after 30 hr. *Chl. vulgaris* was least sensitive to streptomycin, a few cells remaining viable after 65 hr in the lowest concentration in the dark.

The fact that the longest survival time (65 hr) was found with cells incubated in the dark is consistent with the larger number of colonies appearing from the dark-incubated tubes in all cases.

Appearing among the colonies on the 65-hr plate just discussed was a colony that was half yellow and half green. When cells from the yellow half were examined, they also proved to be *Chlorella,* so they were streaked on Maerten's-glucose plates, incubated in the dark, and studied further.

Cells of the "mutant"[1] grown in the dark are slightly smaller and more ovoid (4.0μ × 4.6μ) than the wild type (4.6μ in diam). They appear colorless under the microscope, and their chloroplasts are slightly distorted. The dark-grown colonies are yellow to light-green and have never been observed to vary in color or shape during numerous plating experiments. The wild type retained its original color through three dark transfers extending over two months.

The behavior of the "mutant" is entirely different in light and dark. Although it is yellow-green in the dark, in contrast to the dark-green wild type, it is dark-green in the light and capable of growth on a medium devoid of carbon source.

All transfers of the dark-green photosynthesizing cells of the "mutant" give rise to yellow-green colonies in the dark, thus indicating that this is a stable mutant.

Preliminary determinations of the absorption spectra of methyl alcohol extracts of the pigments in the Beckman spectrophotometer indicate that, although the light-grown cells have an absorption spectrum similar to that of the wild type in light and dark, chlorophyll *a* is lacking in the dark-grown mutant. There is also evidence that a compound similar to protochlorophyll is present.

Although wild types of *Chlorella* possess the same pigments in light or dark (*4*), mutants have been obtained which do not synthesize chlorophyll (*5*). One of these mutant strains (*6*), which accumulates Mg vinyl phaeoporphyrin, greens in light as does the "mutant" described here. It has not yet been determined whether this "mutant" accumulates the same compound, or whether there is a blocking at the next stage as postulated by Granick (*5*); i.e., that of formation of chlorophyll *a* from protochlorophyll.

[1] There is a possibility that this form is not a mutant in the usual sense of the word: that is, that its nuclear genes have not been altered. If streptomycin brought about some defect in the chloroplast, and if the chloroplast were a self-duplicating unit, this would be a cytoplasmic mutation. It is equally likely, however, that a true genetic mutation has occurred and that it was picked up by chance during the scanning of a large population.

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Effect of Prior Injection of Non-Mouse Tissues on Growth of Tumor Homoiografts in Mice[1]

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With the exception of the so-called nonspecific transplantable tumors, grafts of mouse tumors do not survive when the donor and host animals are from genetically unrelated inbred lines. With certain experimental procedures, however, the normal resistance of an animal to a tumor homoiograft (i.e., grafts of a tumor indigenous to one inbred line into mice of a genetically unrelated inbred line) can be broken down. This can be effected either by the prior injection into the host of lyophilized normal or cancerous mouse tissues (*1–4*), fresh tissue homogenates (*5*), tissue antiserums (*6*), frozen tumor tissue (*7, 8*), or by repeated inoculations of the foreign tumor (*9*). As a rule, the best results follow the administration of normal tissues from animals of the inbred line to which the donor graft is indigenous, or the injection of homologous tumor tissue. "Cross reactions," however, are occasionally obtained when normal or cancerous tissues from an inbred strain to which the donor tumor is not indigenous are used (*3, 10*).

The present paper is directed to the question of species specificity of the effect. Three different sets of experiments are reported, dealing with the results of injection of lyophilized tissues from rats, hamsters, and guinea pigs on mouse tumor homoiografts.

The test tumor used was the tumor 15091a, an anaplastic mammary gland carcinoma that is indigenous to the inbred A strain of mice. It grows rapidly, on subcutaneous inoculation, in 100% of strain A animals of both sexes, killing most of the hosts within 3–5

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weeks after implantation. The test hosts were mice of the inbred strains C57BL/6Ks (a C57 black subline) and C57BR/cdJax (a C57 brown subline). These strains are not related to the A strain. The recipient mice were about equally divided by sex, and ranged in age from 2 to 4 months at the start of the experiment. All appeared healthy and vigorous.

Liver, kidney, and spleen were taken from rats, hamsters, and guinea pigs. The tissues were secured under sterile conditions, frozen over dry ice within 1 hr after being excised, and immediately thereafter subjected to freeze-drying. The lyophilized tissues were powdered and stored in ampoules sealed under vacuum. For purposes of injection, the tissues were ground in glass homogenizers in a vehicle of sterile double-distilled water. Injections of the resultant suspensions were given intraperitoneally twice weekly in the amount of 0.5 ml/injection, containing 10 mg dry weight of tissue. Control groups of mice received injections of lyophilized homologous tissue or saline.

One week after the last injection, single inoculations of bits of minced live tumor were made by trocar, under sterile conditions. The grafts were placed subcutaneously in the suprascapular region. The subsequent course of growth of the implants was followed by palpation. Animals were classified as "takes" if they died with progressively growing tumors, and as "negatives" if there was no sign of a graft for at least two consecutive months.

The data are presented in Table 1. It is apparent that only the prior injections of the lyophilized homologous mouse tumor tissue abrogated the resistance of the hosts to the homoiografts. The numbers of "takes" in the groups injected with lyophilized guinea pig kidney or spleen are not statistically significant, as compared with the saline-injected controls.

TABLE 1

EFFECT OF PRIOR INJECTIONS OF LYOPHILIZED TISSUES ON THE GROWTH OF TUMOR HOMOIOGRAFTS IN MICE

Substance injected	Total mg/mouse (dry wt)*	Mice dying with tumors†	
		C57BL/6Ks	C57BR/cdJax
Rat tissue			
Liver	100	—	0/17
Kidney	100	—	0/19
Spleen	100	—	0/10
Saline (control)	(5 ml)	—	0/20
Hamster tissue			
Liver	100	0/19	—
Kidney	100	0/20	—
Spleen	100	0/5	—
Saline (control)	(5 ml)	0/10	—
Guinea pig tissue			
Liver	50	0/23	0/23
	100	0/22	0/15
Kidney	50	1/22	0/18
	100	0/19	2/9
Spleen	100	1/6	—
Saline (control)	(2.5 or 5 ml)	0/24	1/25
Homologous tumor	50	9/10	7/10

* Injections given intraperitoneally twice weekly in 10 mg/injection.

† Numbers in numerators are the numbers dying in each experimental group; numbers in denominators are the total numbers of mice in each group.

The failure of the out-of-species tissues to abolish the resistance of the host to the homoiografts indicates that we are dealing with a specific reaction evoked by substances, present in both the lyophilized mouse tissues and the fresh tumor inoculum, which must have moieties in common. This supposition is further supported by the predominant absence of cross reactions (*3, 7, 8, 10*) when tissues from inbred mouse strains not homologous to the tumor inoculum are used.

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The Role of Darkness in Sexual Activity of the Quail[1]

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Studies with plants have demonstrated that the duration of the night or dark period is an essential factor in photoperiodism (*1*). The requirements of some plants for short day lengths for flowering were shown to be constituted of two parts: (*a*) a requirement for a specific minimum amount of light followed by (*b*) a specific minimum of darkness (*2*). The possible role that periods of darkness may play in the photoperiodic responses of higher animals has not been conclusively shown.

Evidence that the dark period is critical to the photoperiodic responses of lower animals has been obtained in two instances. Shull (*3*) has shown that wing production by aphids is dependent upon a requirement for a light period followed by a long dark period, entirely comparable to the plant requirements mentioned above. Jenner (*4*) has found that reproduction in the pulmonate snail (*Lymnaea palustris*) requires long photoperiods, but short photoperiods with brief light interruptions of the night will bring on reproduction.

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