TABLE 1

Patient	Age	Visual acuity		Dates (1952)	No. injections
		O.D.	O.S.		
Mrs. S. W.	34	Light and dark	20/20–3	Feb. 27	
		20/40	20/20–3	July 9	40
Mr. S. S.	64	20/100	20/200	Feb. 28	
		20/30	20/50	Apr. 29	29
Mrs. E. D.	54	Light and dark	20/100	Feb. 24	
		20/100	20/30	May 21	40
Mr. M. F.	67	20/60	Amblyopic	Jan. 9	
		20/15		Mar. 6	22
Mrs. G. R.	49	20/150–1	20/30–2	Feb. 18	
		20/40–3	20/15–3	Apr. 29	30
Mrs. S. J.	54	20/200	20/150	May 12	
		20/200	20/40+	July 16	28
Mrs. M. M.	60	Aphakial	20/150	Jan. 22	
			20/60	Mar. 19	30
Mrs. C. P.	70	20/70	20/80	Apr. 23	
		20/30	20/30	July 9	36
Mr. P. de P.	39	20/50–2	> 20/200	Apr. 3	
		20/20+	20/100	June 25	41
Mrs. I. L.	57	20/70	20/30	Apr. 30	
		20/40	20/30	May 28	12
Mrs. J. A.	67	Fingers at 5 ft	20/80	May 21	
		20/100	20/40	July 16	26
Mrs. P. B.	70	20/80	20/60	Jan. 2	
		20/20–2	20/20–1	Apr. 17	34

subject of an extensive and more detailed clinical report, are shown in Table 1. No attempt is made to amplify the mitigating factors that affect visual acuity, such as retinopathy, which is not discovered until a clearing of the lens permits a detailed examination of the fundus; nor are the sketches and written descriptions of the ophthalmoscopic appearances of the opacities included. These data, as well as all other pertinent observations regarding coexisting pathology, such as diabetes, hypertension, etc., and subjective changes reported by the patients, have been a normal part of the critical evaluation procedures employed. For the purpose of this brief and preliminary paper, a record of the initial and final visual acuities, together with the ages of the patients and the number of injections received, should answer that first and most important question that will be asked in regard to this nonsurgical therapy of cataract: "Is the patient's vision improved?"

Another clinical series involving approximately 50 patients is being observed and studied under conditions of the most critical scrutiny, and these results will be submitted for publication as soon as available. In view, however, of the promise that this therapy has shown so far, this report is submitted in order that those interested in the subject may be advised of the work that is in progress.

References

1. DAVIS, A. E. *Trans. Am. Ophthalmol. Soc.* (1924).
2. ELLIS, Z. *Arch. Ophthalmol.* (Chicago), **57**, 46 (1928); SELLINGER. Quoted in D. T. Atkin (Ed.), *External Diseases of the Eye.* Philadelphia: Lea & Febiger, 569 (1937).
3. HEKTOEN, L., and SCHULKOF, K. *J. Infectious Diseases*, **34**, 433 (1924).
4. SHROPSHIRE, R. F. *Arch. Ophthalmol.* (Chicago), **17**, 508 (1937).
5. DISCHE, Z., and ZIL, H. *Am. J. Ophthalmol.*, **34**, (5), Pt. II, 104 (1951).

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Observations on a Chlorophyll-Deficient Strain of *Chlorella vulgaris* Obtained after Treatment with Streptomycin

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It has been demonstrated that streptomycin brings about a loss of chloroplasts in the flagellate *Euglena* (*1*) and in the seedlings of certain Gramineae, and that it causes the loss of color in crown gall tumors of carrot (*2*). Therefore, research was recently initiated to determine whether streptomycin would induce the loss of chloroplasts in algae other than *Euglena. Stichococcus bacillaris, Scenedesmus dimorphus,* and two species of *Chlorella* (sp. and *vulgaris*) were investigated. All these species are capable of heterotrophic growth in the dark, with glucose as a carbon source.

Vegetative cells of all these algae were treated in tubes containing Maerten's solution ($Ca(NO_3)_2 \cdot 4 H_2O$, 1.0g; $MgSO_4 \cdot 7 H_2O$, 0.2g; K_2HPO_4, 0.2g; 1 ml of trace element solution [*3*]; and distilled H_2O, 1000 ml), 0.5% glucose, and four concentrations of streptomycin sulfate (500, 2500, 5000 and 7500 μg/ml). One

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.

References

1. PROVASOLI, L., HUTNER, S. H., and SCHATZ, A. *Proc. Soc. Exptl. Biol. Med.,* **69**, 279 (1948).
2. HUTNER, S. H., and PROVASOLI, L. In A. Lwoff (Ed.), *Biochemistry and Physiology of Protozoa.* New York: Academic Press, 79 (1951).
3. BURKHOLDER, P. R., and NICKELL, L. G. *Botan. Gaz.,* **110**, 426 (1949).
4. MEYERS, J. *Plant Physiol.,* **15**, 575 (1948).
5. GRANICK, S. *Ann. Rev. Plant Physiol.,* **2**, 15 (1951).
6. ———. *J. Biol. Chem.,* **183**, 713 (1950).

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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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