Table 1. Average weight response to vitamin  $B_{12}$  and pseudo-B12.

	$\begin{array}{c} Basal \\ + B_{12} \end{array}$	Basal - B <sub>12</sub>	$Basal + pseudo-B_{12}$
No. of pigs	5	5	5
Avg. individual wt. (kg)	1.93	1.89	1.62
Avg. 4-wk wt. (kg)	10.19	5.13	5.06
Avg. gain (kg)	8.26	3.24	3.44

tion: alpha-protein 29.4; DL-methionine 1.3; cerelose 30.5; lard 30.5; minerals 8.3. A complete vitamin premix was added at the time of feeding with the exception of vitamin  $B_{12}$  or pseudo-vitamin  $B_{12}$ , which were given by weekly injection at the rate of  $0.8 \,\mu/\text{kg}$  of body weight per day.

Three groups of five pigs each were used. Weight gains for the  $B_{12}$ -deficient and pseudo-vitamin  $B_{12}$ groups were essentially equal for the 4-wk period, as is shown in Table 1. Erythrocyte, leucocyte, reticulocyte, and hemoglobin determinations were made every other day and were relatively equal for both the deficient and pseudo-vitamin  $B_{12}$  groups, showing a gradual decrease in red cells and reticulocytes. Injection of  $B_{12}$  produced an immediate appetite response in both groups, followed by a reticulocyte response in 8 to 10 days. Untreated pigs in the deficient and pseudo-vitamin  $B_{12}$  groups began to die of the  $B_{12}$ deficiency during the fifth week.

Pseudo-vitamin  $B_{12}$  is apparently inactive for the baby pig.

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# Reversible Changes in the Absorption Spectrum of Chlorella upon Irradiation

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When a suspension of purple bacteria is irradiated with visible or near infrared radiation, a change in the absorption spectrum occurs within one or a few seconds (1, 2). In the dark, the original absorption spectrum is restored within the same time. When the bacteria are photosynthesizing and the intensity of irradiation does not much exceed the intensity at which saturation of photosynthesis occurs, the difference between the absorption spectra in light and darkness is strikingly similar to the difference between the absorption spectra of oxidized and reduced cytochrome c(2).

This finding, combined with what was known about the photosynthesis of purple bacteria, strongly suggests that a cytochrome pigment is an intermediate in the photosynthetic oxidation of the reductant in purple bacteria. The question now poses itself whether in green plants the photosynthetic oxidation of the reductant-water-also is mediated by a cytochrome pigment.

We found that Chlorella, too, showed a change in the absorption spectrum upon irradiation, which was reversed upon darkening. This change was measured by a method similar to that used with purple bacteria (1). Red light ( $\lambda > 670 \text{ m}\mu$ ) was used to illuminate the suspension, contained in a square cell, and the change in absorption brought about by this illumination was measured by a second, weak beam of variable wavelength at a right angle to the illuminating one. The optical density of the suspension, corrected for scattering, was about 0.5 at 680 mµ. The difference spectrum determined in this way is plotted in Fig. 1.



Fig. 1. Change in absorption spectrum of a Chlorella suspension upon irradiation with red light;  $\Delta \varepsilon$  is change in optical density.

It shows a maximum, corresponding to a pronounced increase in absorption during irradiation, at about 515 mµ, and minima, indicating decreased absorption, at about 478 and 420 m $\mu$ .

Similar effects around 515 and 480 mµ were found also in a leaf, in the thallus of a sea alga, and in blades of Vallisneria. The region around 420 mµ was measured only for Chlorella. Thus it seems that the difference spectrum found for Chlorella is characteristic of green plants and algae in general.

As a whole, this spectrum of Chlorella cannot be identified with the difference between the absorption spectra of oxidized and reduced cytochrome c or f; it may, however, be the sum of two difference spectra: one belonging to a cytochrome, with the peak at 420 mµ, and one belonging to another pigment, with peaks at 515 and 478 mµ. If this is the correct interpretation, then the absorption drop at 420 mµ can be considered as revealing the oxidation of a cytochrome upon irradiation of Chlorella and its reduction in the dark. This would be in accordance with the suggestion of Hill and coworkers that cytochrome f, which they found in the chloroplasts of green plants (3) and in

Chlorella (4), functions in photosynthesis as an oxidation-reduction catalyst.

Further experiments are in progress to identify the pigment or pigments responsible for the irradiation effect on the absorption spectrum of Chlorella.

#### **References** and Notes

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# Negative Effects of Antibiotics on Thyroid Gland\*

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Recently Calesnick, Harris, and Jones (1) reported that penicillin and aureomycin produced a goitrogenic and antithyroid effect in young rats as indicated by increased thyroid weight and decreased uptake of I<sup>131</sup>. Both of these antibiotics were fed in concentrations of 1 mg/kg of food to only two groups of four rats each. Previous work in our laboratories (2, 3)failed to show that antibiotics exert any effect on the thyroid gland itself in either thiouracil-treated chicks or in Protamone-treated rats, although body weight losses produced by these drugs were partially counteracted by the use of antibiotics. Inasmuch as antibiotics are widely used in human and animal practice, it was considered important to determine further whether they could alter the activity of the thyroid gland.

Thirty immature male white rats of the Carworth strain were divided into three groups of 10 animals each and were fed a basal diet with or without antibiotics for 21 days as follows: (i) basal (3); (ii) basal and 50 mg/kg potassium penicillin G; (iii) basal and 50 mg/kg aureomycin. On the 21st day, and 16 hr prior to sacrifice, each rat was injected intraperitoneally with 1.0  $\mu c$  of I<sup>131</sup>. The thyroids were removed, weighed on a Roller-Smith balance, and counted for radioactivity by the standard procedure. In another phase of this study, 20-day-old White Leghorn cockerels were divided into two groups of 10 each and fed the following diets: (i) basal (2); (ii) basal and 2 g/ton pencillin. At the end of 5 wk, the chicks were killed and the thyroids were removed and weighed. Radioactive iodine was not given to the chicks.

It can be seen in Table 1 that neither penicillin nor aureomycin altered the size of the thyroids or the uptake of I<sup>131</sup> by the young rats, although both antibiotics increased body weight. Penicillin also failed to alter thyroid weight in the chicks, but it did not affect body weight. An additional group of 10 chicks was fed 90 g of arsanilic acid per ton of feed, and this substance also failed to influence thyroid weight.

Thus, we have not been able to confirm the claim that penicillin and aureomycin are goitrogenic or decrease the uptake of I<sup>131</sup> by the thyroids of young rats, even though 50 times more of these antibiotics was fed to our rats than in the experiment reported by Calesnick et al. (4). Neither was it possible to demonstrate any antithyroid action by penicillin in chicks. In general, these findings are in agreement with previous indications of a lack of direct action by antibiotics on the thyroid gland (2, 3).

#### **References** and Notes

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- In our opinion, the measure of  $I^{131}$  in the thyroids of young rats 16 hr after injection of the isotope is a more accurate index of thyroid function than one taken at 48 hr, as reported by Calesnick *et al.* (1), since the latter measure may be modified to a varying degree by the amount of thyroid output that has taken place.

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Table 1. E	fects of	antibiotics	on	thyroid	gland	of	five	groups	of	10	animals	each.
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Treatment	· Avg. bod	Avg. body weight		yroid weight		
	Start (g)	Final (g)	Actual (mg)	per 100 g B.W. (mg)	I <sup>131</sup> uptake (c/sec mg thyr.)	
Rats		-				
Controls	82	157	13.1	$8.35 \pm 0.97 *$	$185.6 \pm 64.4*$	
Potassium penicillin G	83	168	14.1	$8.42 \pm 1.85$	$174.1 \pm 74.1$	
Aureomycin	83	199	14.8	$7.47 \pm 0.99$	$233.8 \pm 52.9$	
Chicks						
Controls	36	434	32.8	$7.56 \pm 1.27$		
Penicillin	36	402	26.0	$6.46 \pm 0.61$		

\* Standard error of mean.