what aspects of the previous breeding experience are required in order that a bird should act like an "experienced" bird in this situation, what contribution is made by the purely hormonal aspects of the earlier events, what the exact details of the differences in behavior between experienced and inexperienced birds are, and so forth (*9*).

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Thioctic Acid Contained in Visual Cell

Abstract. The D-α-thioctic acid content of the eye of the frog (*Rana nigromaculata*) was determined quantitatively. Outer segments of rods, whole retinas, and choroids contained, respectively 2.0, 8.6, and 13.0 mμg of D-α-thioctic acid per milligram (dry weight). DL-α-Thioctic acid had no accelerating effect on the regeneration of rhodopsin in vivo.

Several years ago, Strauss (*1*) proposed a hypothetical role of 6-8-dithioctic acid in vision. He assumed that rhodopsin should contain one molecule of 6-8-dithioctic acid per molecule of retinene and that the -S-S- bond of 6-8-dithioctic acid might be split by illumination. Since that time, however, there has been no report about this subject. In the experiment reported here the D-α-thioctic acid content of the dark-adapted frog eye (retina, choroid, and outer segments of rods) was measured, and the effects of DL-α-thioctic acid on the regeneration of rhodopsin in vivo were observed by means of the intraocular injection method (*2*).

To measure the D-α-thioctic acid content of the frog eye (*Rana nigromaculata*), 75 dark-adapted frogs were used. They were kept in a dark room overnight, and all operations were carried out under dim red light. The outer segments of rods were isolated, by Kimura's sugar flotation method (*3*) at 0°C, from 60 frogs. Retinas and choroids were separated from the rear half of eyeballs of 15 frogs at room temperature (about 15°C). To determine D-α-thioctic acid, tissues were homogenized in a Potter-Elevehjem homogenizer and were hydrolyzed with 6*N* H_2SO_4 for 1 hour at 120°C. For the microbiological assay of D-α-thioctic acid, *Streptococcus faecalis* (10 CL) was used.

Outer segments of rods, whole retinas, and choroids contained, respectively, 2.0, 8.6, and 13.0 mμg of D-α-thioctic acid per milligram (dry weight). According to Hubbard (*4*), the rhodopsin content of outer segments of rods in the frog is about 35 percent of the dry weight, and frog rhodopsin has a molecular weight of about 40,000 and contains only one retinene per molecule. If Strauss's hypothesis were correct, the thioctic acid content of the outer segments of rods might be about 1600 mμg per milligram (dry weight). However, only 2.0 mμg of D-α-thioctic acid per milligram (dry weight) was contained in the outer segments of rods.

Table 1 gives data for the effect of DL-α-thioctic acid on the regeneration of rhodopsin in vivo. In each experiment, ten frogs (*Rana nigromaculata*) were light-adapted for 2 hours under white light (2000 lux). A DL-α-thioctic acid solution (0.02 ml neutralized with NaOH, containing 400 μg of DL-α-thioctic acid) was injected into the vitreous humor of the left eye (*2*). The right eye was used as the control. After the injection, the frogs were placed in the dark at about 20°C for 1 hour. Then the outer segments of rods were isolated under dim red light at 0°C. For the quantitative determination of rhodopsin contained in separated outer segments of rods, Kimura's method was used (*5*). The DL-α-thioctic acid had no accelerating effect on the regeneration of rhodopsin in vivo, as shown in Table 1.

From these results it cannot be concluded that D-α-thioctic acid is directly related to the construction of rhodopsin. However, the observation that D-α-thioctic acid is found abundantly in the choroid as well as in the liver is particularly interesting, suggesting a possible relationship to the function of visual cells. Further studies are being carried out in our laboratory on the role of D-α-thioctic acid in vision.

Table 1. The effect of DL-α-thioctic acid on the regeneration of rhodopsin in vivo. All data are expressed as follows: $R = [\Delta E/\text{Dry wt. (mg) of separated outer segments}] \times 100$. Here ΔE is the difference in optical density at 500 mμ before and after illumination.

Experiment No.	With thioctic acid	Control
1	5.6	6.2
2	6.0	7.0
3	4.5	5.1
4	7.0	7.5
5	7.0	7.8

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Brightness Scaling of White and Colored Stimuli

Abstract. Brightness scales for chromatic and achromatic test stimuli have been obtained by the methods of ratio production and magnitude estimation for the dark-adapted observer and conditions of brief foveal stimulation. All scales adhere to the form of a power law for brightness, and scales obtained under the differing psychophysical procedures agree.

There is now considerable evidence from several laboratories both here and abroad relating to the form of the brightness scale for the dark-adapted observer (*1*). The universal finding is that brightness is related to luminance by a power law with an exponent of about 0.3. That the results of studies obtained under widely different conditions should happen to agree might be attributed in part to biases associated with one or another scaling procedure, or to the interaction of the scaling method with a fortuitous choice of stimulus conditions for measurement. Earlier studies employed differing stimulus areas (Stevens, 3.3°; Hanes, 4.5°; Hopkinson, 2°), differing durations of test stimulus (2 to 3 seconds, or prolonged viewing), and differing means of controlling the effects of simultaneous contrast. These studies all employed stimuli large enough to stimulate extrafoveal areas and long enough to permit some local adaptation to the test flash. Hanes employed the method of ratio production, Stevens and Hopkinson the method of magnitude estimation.

The purpose of the investigation reported here was to compare brightness scales obtained by differing scaling

methods (ratio production and magnitude estimation) when these techniques are applied under identical stimulus conditions (*2*). Stimuli were presented in a 1° field, fixated centrally to assure foveal stimulation. To preclude changes in adaptation due to the presence of the test flash itself, we used brief (275 msec) test stimuli. The generality of our comparison between psychophysical methods has been extended by an investigation of the observers' responses to both chromatic and achromatic stimuli.

For the ratio-production procedure we used a variation of the binocular matching technique (*3*), presenting test and comparison stimuli simultaneously to adjacent noncorresponding points of the two eyes. This haploscopic presentation avoids the effects of monocular simultaneous contrast. After dark-adapting, each observer made four judgments of "half as bright" and of "twice as bright" by adjusting the comparison stimulus to correspond to those ratios for each of a series of standard test stimulus luminances. White, red (W29), green (W65), and blue (W48) test stimuli were investigated for two observers with normal color vision. Stimulus fields were presented once every 6 seconds, after the extinction of a dim fixation target. An observer could view as many flashes as he required to reach his judgment.

These same stimulus series were also studied by the method of magnitude estimation. Observers made numerical estimations of the brightness of each stimulus relative to an assigned standard luminance (*4*). Observations were monocular, by the preferred eye. Twelve observers with normal color vision participated in this part of the experiment. The duration of the test flash, the interstimulus interval, and the size of the test field were identical with those employed in the ratio-production procedure.

Standard techniques for the derivation of psychological scales were applied to the data for each scaling procedure.

Figure 1 summarizes the brightness scales derived for the dark-adapted observer by the psychophysical methods of ratio production and magnitude estimation. All functions have been equated arbitrarily at the point indicated by the arrow. The brightness scales obtained for two observers by the method of ratio production fall well within the range of estimations of brightness made by the group of 12 observers. Although the range of magnitude estimations is somewhat greater for chromatic than for achromatic stimuli, the general form of the brightness scale as derived from median estimates remains invariant: to a good first approximation, this function indicates that brightness is related to luminance by a power law.

Despite the impressive agreement shown here between scales derived by differing methods, certain qualitative differences appear. In the ratio-production result, judgments of "half as bright" consistently produce scales which have greater curvature than those derived from judgments of "twice as bright." It has been suggested (*5*) that this curvature results from adoption by the observer of an equal-interval criterion when making "half" judgments of test luminances near threshold.

Results from the method of magnitude estimation reflect considerable variability in the absolute magnitudes reported, although the scales based on medians provide fairly stable functions. From observers' comments about their estimates of brightness magnitude, we find that those showing the greatest deviance from the median tend to be individuals who are relatively unfamiliar with the use of numbers to express ratios; one observer, in fact, tended merely to *order* the stimuli numerically.

Within the limits of errors of estimate, brightness scales obtained by the method of ratio production and those obtained by magnitude estimations agree, when these methods are applied

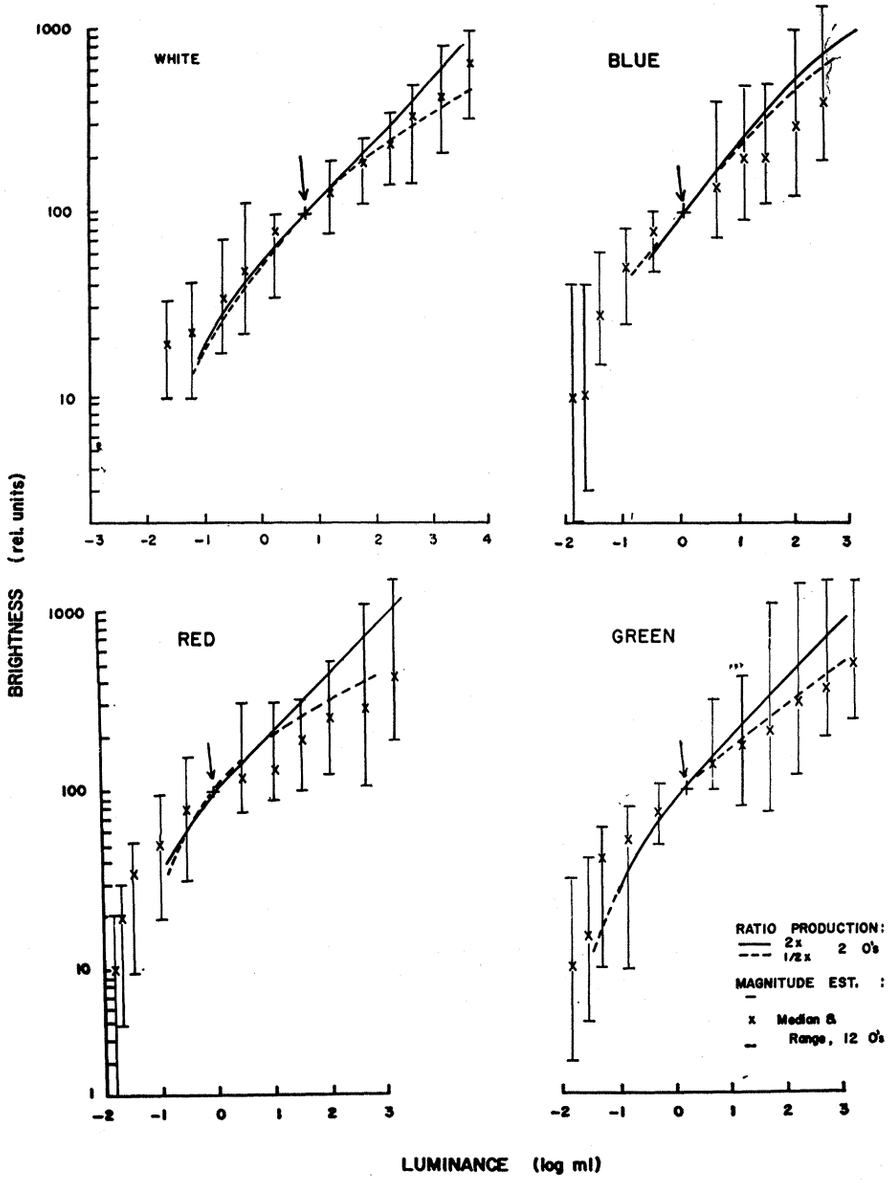

Fig. 1. Brightness scales for chromatic and achromatic test stimuli, methods of ratio production, and magnitude estimation. All scales have been equated arbitrarily at the point shown. Linearity on these coordinates indicates a power function, with exponent equal to the slope of the linear function.

under identical stimulus conditions, with brief foveal stimulation. The general form of the brightness scale for the dark-adapted observer and for stimulus conditions which preclude adaptation to the test stimulus approximates a power function, as had been predicted. The form of the function as well as its exponent remain essentially invariant with changes in the color of wide-band test stimuli.

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Sexual Recombination in a Homothallic, Antibiotic Producing Fungus

Abstract. The presence of a perfect or sexual stage in the fungus *Emericellopsis salmosynnemata* has made possible an investigation of the effect of meiotic recombination on yields of antibiotic. While most of the fruiting bodies produced by this organism are the result of self-fertilization, conclusive evidence of cross-fertilization and recombination between two mutants was obtained. Cross-fertilization occurred rarely.

As pointed out by Raper (*1*), the application of the breeding techniques used by geneticists in producing better varieties in higher plants have frequently been looked upon with great interest by microbiologists engaged in improving strains of microbes of economic and industrial significance. This type of approach has been barred to them, however, by the absence of a sexual stage in the penicillia, aspergilli, and actinomycetes commonly employed in industrial fermentations.

Emericellopsis salmosynnemata, a fungus belonging to the order Eurotiales, has both a sexual or cleistothecial stage and an imperfect *(Cephalosporium)* stage (*2*). Since it is homothallic, the sexual fruiting bodies formed on corn-meal agar are normally the result of self-fertilization. However, since cross-karyogamy and recombination in homothallic fungi have previously been described (*3*), an investigation of the potential for sexual recombination in this organism was undertaken.

Table 1. Data on progeny from three hybrid cleistothecia resulting from the following cross: *al-4 lys⁻-1 ser⁺ f⁺* (75 unit/ml)* × *al⁺-4 lys⁺-1 ser⁻ f* (0 unit/ml)*

Genotypes recovered	No. recovered	Radius of inhibition (mm)	Yield of synnematin (unit/ml)
al-4 lys⁻-1 ser⁺ f⁺	15	3.5	75
al-4 lys⁻-1 ser⁺ f	3	0	0
al-4 lys⁺-1 ser⁻ f	1	0	0
al-4 lys⁻-1 ser⁻ f⁺	7	0	0
al-4 lys⁺-1 ser⁺ f	4	0	0
al-4 lys⁺-1 ser⁺ f⁺	1	6	400
al⁺-4 lys⁺-1 ser⁻ f	23	0	0
al⁺-4 lys⁺-1 ser⁻ f⁺	1	0	0
al⁺-4 lys⁻-1 ser⁺ f	3	0	0
al⁺-4 lys⁻-1 ser⁺ f⁺	3	0	0
al⁺-4 lys⁻-1 ser⁻ f	2	0	0
al⁺-4 lys⁻-1 ser⁻ f⁺	3	0	0
al⁺-4 lys⁺-1 ser⁺ f	4	0	0
al⁺-4 lys⁺-1 ser⁺ f⁺	3	7	600

* Yield of antibiotic of the parental auxotroph.

A factor of added interest in these studies is the production by *E. salmosynnemata* of the antibiotic substance synnematin B. The presence of this factor offered the opportunity for investigation of the effect of meiotic recombination on yields of the antibiotic.

After induction of a number of morphological and physiological mutants by ultraviolet irradiation of conidia, a series of sexual crosses were made for the purpose of demonstrating recombination of the selected markers and the effect of recombinant genes on yields of the antibiotic.

Crossing was accomplished by placing mycelial inocula about 1 in. apart on corn-meal agar. After a 2- to 3-week period of incubation at 28°C, single fruiting bodies were removed from the line of contact and single spores were isolated with a De Fonbrune micromanipulator. Colonies obtained from these single spores were analyzed for recombination by growth on suitable media.

Of a total of 98 cleistothecia of *E. salmosynnemata* in which single-spore analysis was performed, only nine showed conclusive recombinant types; in most instances all products of meiosis were not recovered. All recombinants from these crosses were then investigated for their antibiotic-producing ability, and yields were compared with those of the parental strains used in the cross, and with those of the original wild-type strain. Fermentations were run with the medium of Nara and Johnson (*4*), and filtrates were assayed by the agar-well technique against a sensitive strain of *Bacillus subtilis.*

Table 1 shows the results of one such cross between an albino, lysine-requiring mutant with fluffy mycelium (*al-4 lys⁻-1 ser⁺ f⁺*), and a serine-requiring mutant of wild-type color with flat mycelium (*al⁺-4 lys⁺-1 ser⁻ f*).

This cross proved interesting because almost all of the expected recombinant types were recovered. From the point of view of antibiotic production, a more critical evaluation of the effect of recombination on yields was not possible because of the low antibiotic activity of both parent strains. However, an examination of Table 1 suggests that genetic reconstitution to a prototrophic state by meiotic recombination is not sufficient to re-establish the original antibiotic-yielding capacity of the wild-type strain (average radius of inhibition, 7 mm). The results obtained from this particular cross indicate that possibly several factors active in the synthesis of the antibiotic may be linked to the chromosome carrying the gene for fluffy mycelium (*f⁺*); when other, unselected factors are brought together in a prototroph with *f⁺*, the proper gene combinations may be achieved, and the result may be production of the antibiotic. Mycelial color appears to have no effect on yields of the antibiotic.

These results indicate that a potential for hybridization and meiotic recombination exists in this homothallic fungus, and that an application of breeding techniques in studies of antibiotics may give promising leads in this field of research (*5*).

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