do not appear, as a means of eliminating the misleading effects to which they refer. This program has in fact already been carried out by Herget and Musen (*4*) and forms the basis for the Vanguard orbital calculations. Second, the distribution of the Minitrack stations is, as pointed out by the Stanford Research Institute group, concentrated along the 70th meridian. However, the location along the meridian does not imply a bias favoring a particular portion of the orbit, as they suggest. The rotation of the earth actually spreads the successive observations out along the orbit at very reasonable intervals of about 35 degrees.

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24 November 1959

Colors of All Hues from Binocular Mixing of Two Colors

Abstract. Land has recently studied the perception of colors resulting from appropriate mixtures of two colors or of one color and light from an incandescent lamp. In an "image situation," colors of all hues may result from such mixtures. The findings presented demonstrate that the mixing which Land accomplished by superimposing two projected images on a screen can be achieved when the two color separation images are presented simultaneously but separately to the two eyes.

The problem of binocular fusion of colors has interested investigators since Hecht's demonstration in 1928 that presenting red to one eye and green to the other led to a subjective sensation of yellow (*1*). Hurvich and Jameson (*2*) confirmed these results; it is today generally accepted that such fusion is readily obtainable in most subjects.

Land (*3, 4*) has recently considerably extended the early work of Fox and Hickey (*5*) and of Bernardi (*6*) on colors resulting from mixtures of two colors or of one color and light from an incandescent lamp. In the "image situation" (that is, a complex array of natural objects), colors of all hues may result from such mixtures. We decided to study the question of whether or not such mixing occurs when the two color separation images were presented simultaneously but separately to the two eyes.

Our experimental procedure followed Land's closely, deviating primarily in the technique of viewing. A complex scene was photographed on Kodak 35-mm direct positive film through various Kodak Wratten filters. Pairs of positive transparencies were selected in which the scale of grays was complete. The positives were then viewed in a Kodak stereoscopic viewer (however, the pictures used were not stereo pairs) with appropriate filters place in each half. Thus, in a typical experiment a scene was photographed through a Kodak Wratten filter No. 29 (red) and through a Kodak Wratten filter No. 58 (green). The black and white positive photographed through the red filter was placed on the left with a No. 29 filter and the positive photographed through the green filter was placed on the right with either a No. 58 filter or with no filter at all. A pair of crossed polarizing screens (Kodak Polascreen) placed on the brighter side was adjusted for optimum color. The brightness control of the viewer was then adjusted for maximum color saturation.

Under these conditions of binocular mixing, as full a range of colors was seen as had been obtained by projecting the two images on a screen, as in Land's experiments. If the filters were interchanged, "color reversal" occurred (that is, greens appeared as reds and vice versa).

The balancing of brightness in the two images is of critical importance since, otherwise, many subjects have great difficulty in fusing the colors, seeing predominantly with one eye. This effect varies from subject to subject, presumably because of varying degrees of retinal rivalry. Retinal rivalry probably also explains the fact that the perception of the colors may vary in time, in contrast to their constancy when projected and superimposed.

Under the conditions of this experiment, the perception of colors of all hues from two-color mixtures cannot be a purely retinal effect but must involve the interaction of higher centers.

Students of simple binocular color mixing have tended to explain their experiments in terms of color mixing at some level higher than the retina (*1*) or as the central "cancellation" of the hues not common to the two eyes, leaving the hue common to the two eyes to be perceived centrally (*2*). Another equally acceptable explanation is that the images in each eye initiate reflexes at higher levels whose efferent limbs modify the response of the opposite retina. Because of the possibility that this explanation is correct, it cannot be excluded that under some conditions the perception of colors of all hues from two-color mixture is a retinal effect, or that parallel mechanisms are present in the retina and higher centers (*7*).

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3 December 1959

Interspecific Transformation of Neisseria by Culture Slime Containing Deoxyribonucleate

Abstract. Genetic change of *Neisseria meningitidis* is elicited by deoxyribonucleate preparations obtained from *N. sicca*. Such interspecific transformation is effected not only by deoxyribonucleate obtained from cells by conventional methods but also by crude deoxyribonucleate-containing slime which accumulates without experimental intervention in some cultures incubated for a period as short as 44 hours.

Deoxyribonucleates (DNA) of high molecular weight are recognized as determinants of bacterial heredity (*1*). Deoxyribonucleate which has been extracted from donor bacteria after lysis by added deoxycholate or other lytic agents, and extensively purified, elicits heritable change (transformation) when applied in minute quantities to suitable recipient bacteria. The genetic changes which have been studied affect numerous properties of the bacteria, including virulence, specificity of antigens, cellular and colonial morphology, resistance to various antibacterial agents, and capacity to utilize certain compounds for nutritional purposes (*1, 2*). Accumulated evidence led Hotchkiss (*1*) to the conclusion that the transforming DNA contains biologically specific entities operationally equivalent to bacterial genes.

Recently, highly polymerized DNA has been found extracellularly in ordinary cultures of a variety of bacteria; it is observable when a slimy sediment or pellicle accumulates (*3*). In cultures of *Micrococcus halodenitrificans* (*4*), *Pseudomonas fluorescens,* and *Neisseria meningitidis* (*5*), slime DNA accumulated under conditions associated with loss of viability of some cells of the population (obscured in young cultures by multiplication of viable cells), and was accounted for as intracellular DNA liberated by lysis. Crude culture slime of *N. meningitidis* strain 15, as well as purified preparations of DNA from intracellular and from extracellular sites, exhibited transforming activity for recipient cells of the same species (*5*).

The possible occurrence in nature of transformation has been suggested (*6*) as a means by which surviving cells of a mixed population might acquire, and thus preserve, certain heritable properties of cells which have been killed and lysed by a selectively deleterious agent (for example, penicillin). The presence of genetically active DNA released into the culture medium without experimental intervention offers further support for this possibility (*5*). Slimes containing DNA may provide actual "gene pools" from which by transformation microorganisms may randomly withdraw genetic units. The range of possible transformation thus becomes a problem of considerable interest. If genes from members of other species or more distantly related groups can be incorporated into the genetic structure of a cell, the range of consequences of transformation will be extended. The idea that transformation might play an important role clinically by changing the inheritance of the large reservoir of nonclassifiable *Neisseria* found in the human nasopharynx was suggested (*7*), though no evidence was available. More recent investigations have demonstrated transformation reactions between different species of *Haemophilus* (*8*), and between pneumococci and streptococci (*9–11*).

Possible interspecific transformation of *Neisseria* has been studied, as part of an investigation (*12*) concerning the range of transfer of genetic information. Apart from the two common pathogenic members of *Neisseria, meningitidis* and *gonorrhoeae,* the definition of species within this genus is far from clear (*13*); however, for determinative purposes, various species have been characterized (*14*). *Neisseria sicca* strain 12, used in this study, was isolated several years ago from the nasopharynx of a healthy student, and was retained (at −60°C) in the culture collection because it was typical (*14*).

A single-step mutant resistant to 1000 μg of streptomycin (*str*) per milliliter (strain 12 *str-r*) was obtained from the sensitive *N. sicca.* Both strains characteristically developed slimy sediments in cultures of brain heart infusion (Difco; 200 ml in liter flasks, mechanically shaken for the initial 20 hours) incubated at either 37° or at 25°C. Slimes aseptically removed from cultures of various ages were tested for transforming activity. That an essential component of these slimes was extracellular DNA was shown, as previously described (*3*), by their rapid loss of slimy consistency upon addition of crystalline pancreatic deoxyribonuclease. Preparations of intracellular DNA, obtained from detergent-lysed cells of *N. sicca* 12 *str-r* and from two *str-r* strains of *N. meningitidis* strain 15, were purified by methods (*5*) which included deproteinization with detergent.

Transforming activity was investigated by exposing competent meningococci (suspended in heart infusion broth, Difco, at 36°C) to various dilutions of each DNA-containing material. Crystalline deoxyribonuclease and magnesium ions were added after 15 to 30 minutes to destroy the transforming activity of unbound DNA. The total number of cells (or colony-forming units) exposed to DNA (*E*) was determined quantitatively by plating a suitably diluted sample in *str*-free medium (*15*). *Str-r* transformants (*T*) were assayed as follows: A sample of the reaction mixture (0.1 to 2.0 ml, taken within 1 hour) was mixed with 40 ml of liquefied *str*-free HIY-1 soft agar, and 4-ml aliquots were pipetted on top of supporting layers (20 ± 0.5 ml) of 10 HIY-1 hard agar plates. Cells were thus confined in agar before the *str-r* trait was developed or the transformants multiplied; plates were placed at 37°C without delay. Phenotypic expression was complete 5 hours after initial exposure to transforming DNA (*5*), and the cells were challenged with *str* by overlaying the inoculated agar with 4 ml of HIY-1 soft agar containing a quantity of *str* sufficient to make 500 μg/ml after diffusion throughout the underlying agar. Colonies developing on plates incubated at 37°C for 3 to 4 days were counted, and the transformation ratio was calculated (*T*/*E*, multiplied in this paper by 10^5 to avoid undesirably small numbers). Controls for each test consisted of dilutions of culture slime or of DNA which had been inactivated by crystalline deoxyribonuclease 5 minutes before recipient

Table 1. Transforming activity for *Neisseria meningitidis* of deoxyribonucleate-containing materials from *N. meningitidis* (strain 15) and from *N. sicca* (strain 12).

Recipient cells (strain; No. exposed/ml) (*E*)	Material tested	Exposure (min)	*Str-r* transformants (No./ml) (*T*)	(*T*/*E*) × 10^5
	Experiment A			
15 *str-s;* 2.5 × 10^6	15 *ery-r str-r* DNA, 0.8 μg/ml	15	4546	182
	15 *ery-r str-r* DNA, 0.1 μg/ml	15	2175	87
	15 *ery-r str-r* DNA, 0.001 μg/ml	15	65	2.6
	12 *str-r* DNA, 10.0 μg/ml	15	290	11.6
	12 *str-r* DNA, 2.0 μg/ml	15	223	8.9
	12 *str-r* DNA, 0.8 μg/ml	15	191	7.6
	12 *str-r* DNA, 0.1 μg/ml	15	82	3.3
	12 *str-r* DNA, 0.01 μg/ml	15	21	0.8
	12 *str-r* DNA, 0.001 μg/ml	15	3	0.1
	Experiment B			
15 *str-s;* 8.6 × 10^5	15 *ery-r str-r* DNA, 5.0 μg/ml	15	1066	124
	12 *str-r* DNA, 5.0 μg/ml	15	63	7.3
	Experiment C			
15 *car-r str-s;* 4.3 × 10^6	15 *ery-r str-r* DNA, 5.0 μg/ml	25	9340	217
	12 *str-r* DNA, 5.0 μg/ml	25	364	8.5
	12 *str-r* 44-hr slime, dild. 1:10	25	2	0.05
	12 *str-r* 44-hr slime, dild. 1:100	25	12	0.28
	Experiment D			
15 *car-r str-s;* 3.2 × 10^6	15 *str-r* DNA, 5.0 μg/ml	30	4120	129
	15 *str-r* DNA, 1.0 μg/ml	30	3715	116
	15 *str-r* DNA, 0.1 μg/ml	30	2308	72
	15 *str-r* DNA, 0.01 μg/ml	30	589	18
	12 *str-r* DNA, 5.0 μg/ml	30	379	12
	12 *str-r* DNA, 1.0 μg/ml	30	346	11
	12 *str-r* DNA, 0.1 μg/ml	30	200	6.3
	12 *str-r* DNA, 0.01 μg/ml	30	58	1.8
	12 *str-r* 62-hr slime, dild. 1:25	30	35	1.1
	12 *str-r* 62-hr slime, dild. 1:100	30	121	3.8
	12 *str-r* 62-hr slime, dild. 1:250	30	108	3.4
	12 *str-r* 16-day slime, dild. 1:25	30	288	9.0
	12 *str-r* 16-day slime, dild. 1:100	30	262	8.2
	12 *str-r* 16-day slime, dild. 1:250	30	267	8.3

cells were added; in all other respects similar to the transformation tests, these controls were uniformly negative in all tests reported.

Sterile solutions of purified DNA preparations were examined for capacity to transform cells of *N. meningitidis* 15 *str-s*, a strain inhibited by less than 1 μg of *str* per milliliter which had been isolated from spinal fluid of a fatal non-epidemic case of meningitis. Representative results are compiled in Table 1 (experiments A and B). Meningococci were transformed to *str*-resistance by as little as 10^{-3} μg of DNA per milliliter of the preparation extracted from *N. sicca* 12 *str-r*, and the number of transformants was a function of the quantity of DNA employed. Transformation ratios for this meningococcus strain ranged from 17 to 26 times greater with corresponding concentrations of a DNA preparation from meningococcus (a strain resistant to both erythromycin and to *str*) than with one from *N. sicca* (compare with *8*, *9*).

To investigate the transforming activity of *N. sicca* culture slime, carbomycin (*car*, *16*) was used to eliminate the growth of viable slime-donor cells, without affecting the slime DNA or growth of the recipient cells (in this case, a mutant of *N. meningitidis* 15 resistant to 50 μg of *car* per milliliter). Recipient cells were exposed to the DNA-containing material in the usual antibiotic-free medium; thereafter, they were plated in agar containing 12.5 to 25 μg of *car* per milliliter, which did not interfere with the selective action of *str* added later. With this strain, also, preparations of purified DNA, which were examined for comparative purposes, elicited both intraspecific and interspecific transformations (Table 1, experiments C and D).

Transforming activity was exhibited by crude slimes removed from cultures ranging in age from 2 to 16 days. Viable cells numbering over 10^8/ml were present in all slimes from *N. sicca* 12 *str-r* cultures in brain heart infusion broth sampled during the first 5 days of incubation. Indeed, the slime harvested at 44 hours (and tested for transforming activity, Table 1, experiment C) contained 1.3×10^9 colony-forming units per milliliter; the culture incubated for 16 days (Table 1, experiment D), on the other hand, was essentially sterile. An inverse correlation was observed between transforming activity and number of viable cells present in the culture slime. This would be anticipated if increased cellular lysis associated with aging of cultures released more DNA. In addition, intact *N. sicca* cells, which could undergo some metabolic activities but could not form colonies in the presence of *car*, appeared to exert an adverse effect on *str-r* transformation or on colony formation by meningococcus transformants. Thus, slimes obtained from cultures incubated for periods of 44 to 116 hours were more effective as transforming agents when diluted 1:100 immediately before use, than when used in final dilutions of 1:5 to 1:25. On the other hand, no increase of transformation ratio with dilution was observed with the sterile (16-day) culture slime. A dilution of 1:500 examined for one culture slime (not shown) resulted in fewer transformants than were obtained with the 1:250 dilution of the same slime, as would be expected from results with decreasing concentrations of purified DNA preparations.

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15. The plating medium, HIY-1 agar, was heart infusion broth with 0.3 percent Bacto yeast extract and agar (0.7 percent for "soft agar," or 1.4 percent for "hard agar," all Difco), further supplemented aseptically with 250 μg of sodium ribonucleate per milliliter, 0.00005*M* sodium glutamate, and 0.0005*M* $CaCl_2$.
16. Magnamycin, a gift of Chas. Pfizer & Co., Inc.

28 September 1959

Effect of Acclimation on the Preferred Body Temperature of the Lizard, Sceloporus occidentalis

Abstract. The preferred body temperature was determined for several groups of *Sceloporus occidentalis* previously acclimated to several constant temperature levels. Acclimation to a high temperature (35°C) resulted in the selection of a lowered mean preferred body temperature, whereas acclimation to lower temperatures (12°C and 25°C) produced no change in the preferred body temperature.

In recent years considerable interest has been directed towards the controlling mechanism of thermoregulation in poikilotherms. Current interest in reptilian thermoregulation was initiated by Cowles (*1*), and the entire subject has been reviewed recently by Saint-Girons and Saint-Girons (*2*). Concerning the control of thermoregulation, Rodbard (*3*) described a temperature-sensitive area in the brain of the turtle, and some other aspects of control, especially behavioral, have been reviewed by Bogert (*4*). More recently, Stebbins and Eakin (*5*) have reported on the parietal eye of lizards as influencing over-all exposure to heat.

Of equal importance is the role of acclimation in modifying temperature tolerances and preferred body temperature of certain poikilotherms. Lowe and Vance (*6*) demonstrated a direct relationship between acclimation and the critical thermal maximum of lizards, and Dawson and Bartholomew (*7*) have shown a relationship between acclimation and oxygen consumption. Garside and Tait (*8*) have reported results of acclimation on preferred temperatures in fish. This paper is a discussion of experiments designed to ascertain the effects of acclimation on thermoregulatory behavior and the preferred body temperatures of lizards.

Fifty-three fence lizards, *Sceloporus occidentalis*, collected in Berkeley, California, were used in the experiments, which were conducted in two stages, the first during September and October of 1958, and the second during March and April of 1959. Fall and spring groups were treated equally, except that during the fall no controls were utilized. The spring group was divided between control and experimental animals of approximately equal size, weight, and