gives isomaltose and glucose, but no maltose. These findings mean, first, that the trisaccharide must consist of three glucose residues united by one  $\alpha$ -1. 4 bond and by one  $\alpha$ -1,6 bond (the high positive rotation,  $[\alpha]D + 154^{\circ}$ , excludes the possibility of  $\beta$ -linkages); second, of the three possible structures for such a trisaccharide, the only one that could give rise to isomaltose on partial hydrolysis of the aldonic acid is that in which a glucosyl unit is joined to the sixth position in the nonreducing glucose residue of maltose; i.e.,  $4 - [6 - (\alpha - D - glucop v ranos v]) \alpha - D - glucop v rano$ syl] D-glucose.

The more exact identification of the products of partial acid hydrolysis, together with confirmatory tests for other aspects of the structure, will be reported elsewhere.

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## Physiological Mechanisms of **Color Blindness**

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Retinal processes of color-blind subjects were investigated by Motokawa's method (1). The electrical excitability of the eye (the reciprocal of threshold) is determined with an electrical phosphene as an index after preliminary dark adaptation of about 20 min. Stimulating currents, single constant current pulses of 100 msec in duration are applied to the eye through a pair of silver electrodes of  $2 \times 1.5$  cm<sup>2</sup> in size, placed one on the forehead slightly above the evebrow and the other on the homolateral temple of the subject. The electrical excitability of the eve is measured at varying intervals after 2 sec exposure of the eye to a preilluminating patch of 2° in visual angle. The excitability recovers in general very rapidly, becomes supernormal, and then gradually decreases to the initial level. It is, however, sometimes observed that a few humps appear in the recovery curve, especially in those obtained from color-anomalous subjects. The time course depends upon the wavelength of the light used for preillumination and upon the kind of color blindness, but little on the intensity of the preilluminating light; the latter changes only the height of the excitability curve without altering its form. Examples of excitability curves obtained from the fovea of typical dichromats as tested by Ishihara's test, as well as by Nagel's anomaloscope, are shown in Fig. 1, in which percentage increases in electrical excitability above the resting level are plotted as ordinates against time

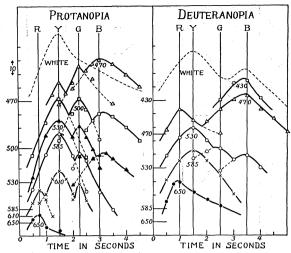


FIG. 1. Excitability curves of dichromats. Ordinates: percentage increases in electrical excitability above the resting level. Abscissae: time after the end of preillumination. Figure by each curve indicates wavelength of light used for preillumination. Zero level for each curve is indicated on the axis of ordinate.

after removal of the preilluminating light as abscissae. As can be seen in the figure, the curves generally show distinct humps, which may be classified into four groups according to their temporal location. It is very probable that these humps represent different retinal processes, but not different phases of one and the same process, because for one and the same abscissa 2 different values of threshold can be obtained, one of which belongs to the one hump, and the other to the neighboring hump, as indicated by broken lines in Fig. 1. In such cases, the subject, at a certain definite voltage, finds it very difficult, or almost impossible, to distinguish the stimulus in question from one far below the threshold, but on further decreasing the voltage, discrimination becomes easier, and thus the second threshold is reached.

The four kinds of processes may be denoted by R, Y, G, and B in the order of rapidity of processes. In color-blind subjects, any one of these processes is especially weak, e.g., the R process is weak in the protanope, the G in the deuteranope. It is another characteristic common to the two kinds of color blindness that a dominant Y process can be seen in the fovea. Motokawa (2) has provided evidence that the Y process, an independent process underlying a sensation of yellow, does exist in the periphery of the retina of normal and color-blind subjects, but he has not yet succeeded in proving the same process in the fovea itself of normal human subjects. The relative magnitudes of the processes are naturally different according to the wavelength of the preilluminating light. This relation is most clearly shown in the curves for spectral distribution of the four processes shown in Fig. 2. in which the ordinates represent the magnitudes of each process or the heights of each kind of hump plotted against the wavelengths of the spectral lights used for preillumination as abscissae. Fig. 2

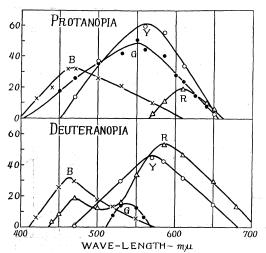


FIG. 2. Curves for spectral distribution of 4 retinal processes of dichromats.

shows that the distribution curves for the deteriorated processes, viz., the R in the protanope and the G in the deuteranope, are much lower and narrower than the other curves.

The well-known theory of Young assumes that one of three components is lacking in dichromats, but this claim obviously does not conform to our findings, (1) because there are, in reality, four components instead of three; and (2) because no component is entirely lacking, although the R or G component, in the respective color-blind types, is weaker than any others. It is to be expected from Young's theory that the sensitivity to yellow of dichromats would be deteriorated on account of the absence of the R or G, but in reality no such serious deterioration as would be expected from the theory can be demonstrated. Moreover, in rare cases such as those reported by v. Kries (3) and by Judd (4), in which one eye was color-

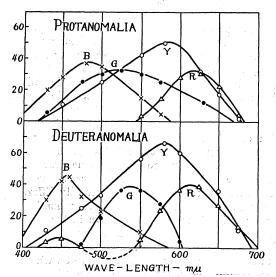


FIG. 3. Curves for spectral distribution of 4 retinal processes of anomalous trichromats.

anomalous and the other was normal, the part of the spectrum which looked yellow to the normal eye also looked yellow or yellowish to the anomalous eye. These facts which the trichromatic theory fails to interpret can easily be understood on the basis of our finding that the Y component is well developed in the fovea of dichromats. It is generally believed that there is a qualitative difference between anomalous trichromatism and dichromatism. In the light of our findings the difference is, however, merely quantitative (Fig. 3).

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3. 4.

Chromatographic Separation of Amino

# Sugars and Amino Acids, Using the N-(2:4-dinitrophenyl) Derivatives

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In the study of bacterial polysaccharides and mucocomplexes, the need has been found for more sensitive methods of detection and estimation of microguantities of amino sugars in the presence of amino acids, neutral sugars, and uronic acids.

The ordinary procedures of paper chromatography (1), investigated independently (2), were found to be suitable for the separation of mixtures of neutral partly methylated sugars. The amino sugars, studied by this method, separated much less readily, because of a slow rate of migration and the diffuse character of the spots-since thought to be due to the ion-exchange behavior of the papers. Some reducing amino sugars could only be observed by their reaction with alkaline silver nitrate or with ninhydrin, whereas others, mainly nonreducing amino sugar derivativese.g., glucosides-could only be detected by spraying the chromatogram with dilute alkaline potassium permanganate (3). The introduction of aniline hydrogen phthalate (4) as an improved general reaction for reducing sugars gave little or no coloration with free amino sugars or N-acetylated sugars. It has now been found that N-(2:4-dinitrophenyl) reducing amino sugars (5) did react when warmed with aniline hydrogen phthalate.

Investigation showed that amino sugars would react readily under mild conditions (viz., room temperature in the presence of sodium bicarbonate) with 2:4-dinitrofluorobenzene forming stable N-(2:4-dinitrophenyl) derivatives and that the colored nature of these compounds rendered them preeminently suitable for chromatographic analysis. A mixture of neutral sugars, a uronic acid, and DNP<sup>2</sup> glucosamine was

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