(5) *Disturbance.* When the trains are appearing almost continuously they nevertheless are interrupted every few seconds by some sort of disturbance. This disturbance may interrupt the trains coming from one part of the head without stopping those from another part.

(6) *Mental activity* as such does not seem to affect the trains. A person may be read to, converse or do *simple* problems in mental arithmetic, without affecting the trains.

(7) *Emotional states* seem to have a profound effect upon the waves. If a subject is embarrassed or apprehensive practically no waves appear. The trains can usually be stopped if a subject is asked to solve a difficult problem in the shortest possible time (that is, if he becomes "rattled"). One subject could stop his trains at will by imagining himself in a terrifying situation (a phantasy of fear).

(8) *Effect of light.* The appearance of light stops the waves in 0.1 to 0.2 second. Seven tenths seconds or more may elapse before they again start after the light disappears. This influence of light is certainly not a direct effect but rather a secondary one, for the following reasons. If the eyes are kept open continuously and the subject is lying quietly in a lighted room, a few trains will usually appear after a time. On the other hand, if the subject is lying in a completely dark room and a faint light has been flashed on and off several times at regular intervals, then if he is told that the light will be flashed on, the trains will stop, even though the light is not flashed on. This effect is best seen (with the subject in a dark room) by simultaneously turning on a light and sounding a tone at regular intervals. After a time the sounding of the tone alone will stop the trains, although before this "conditioning" the tone alone has no such effect. In the case of a hypnotized subject the converse was also true, *i.e.*, the trains would start up at once when he was told he could not see the light, even though it continued to shine directly into his eyes.

(9) *Flicker.* We find, as does Adrian, that flickering lights give rise to potentials in some persons that exactly follow the flicker frequency over a limited range. Occasionally the brain response will be double the flicker frequency. In one subject who showed unusually regular trains no flicker frequency could be induced.

(10) *Sleep.* As a subject is going to sleep the trains appear regularly, but they cease after a time during deep sleep. And yet they will immediately appear if the subject is disturbed. Noises, etc., that the subject is accustomed to hear during sleep, such as the blowing of an automobile horn, do not necessarily have any effect, but anything that indicates that another person is near almost always produces a marked effect with the immediate appearance of the trains. Thus, a faint cough, a whisper, faint footstep, rustling a piece of paper, have all produced trains when loud noises and bright lights have failed. We are inclined to believe that this starting of trains by sound is not a direct result of the sound stimulus but is connected with a change in the general level of brain activity.

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LEAF XANTHOPHYLLS

As a result of the improvement of methods for the isolation of carotenoids, it has been found that leaves contain not less than twelve, and probably more, xanthophylls. Many of these are very soluble in aqueous alcohol and are not recovered from the dilute alcoholic solutions employed in the partition and crystallization methods of Stokes and of Willstaetter. Several of the xanthophylls are adsorbed with the chlorophylls upon Tswett columns and, under the conditions previously employed, are not observable in the presence of the green pigments. Others occur in such small amounts that they can be detected only by the use of Tswett columns composed of very active adsorbents.

In order to demonstrate the presence of such a great number of xanthophylls in leaves, it has been necessary to develop methods for the isolation of all the xanthophylls free from other leaf constituents. This mixture of xanthophylls is then separated by adsorption upon Tswett columns. The successful application of the latter method has been due to the selection of highly active adsorbents which do not decompose the plant pigments and to the availability of a number of solvents which may be used to vary the quantity of pigment held by the adsorbent.

When leaf xanthophyll, prepared by the improved method, is adsorbed upon Tswett columns, three or four pigments pass rapidly through the column. These pigments resemble Cryptoxanthin with respect to solubility and to partition between immiscible solvents. They are followed on the column by lutein, which comprises somewhat more than half the recoverable xanthophyll. The lutein is followed by a pigment not previously described, for which the name "isolutein" is proposed. The isolutein is optically inactive, its absorption maxima are twenty Ångström units toward the violet from those of lutein and, in contrast to lutein and zeaxanthin, its solutions in ether exhibit a deep blue color when treated with concentrated hydrochloric acid. Zeaxanthin, which is present in small amounts, is found above the isolutein on the adsorption column. The former has been found in all the leaves examined (those of barley, carrots, filaree, miner's lettuce, sunflower, squash and spinach) and is identical with zeaxanthin isolated from the calyx of *Physalis alkekengi* with respect to melting point, optical activity, quantitative absorption spectrum of its solutions, absorption maxima in different solvents, adsorption upon Tswett columns and elementary composition. The zeaxanthin is followed upon the column by a pigment, the absorption curves of which are similar to those of flavoxanthin (absorption maxima 4510 and 4220 Ångström units in ethanol). The pigment is optically active ($[\alpha]^{18}_{6678}=+75°$ in chloroform). Another similar pigment, which exhibits a like absorption curve (absorption maxima 4510 and 4220 Ångström units in ethanol) but opposite optical rotation ($[\alpha]^{18}_{6678}=-56°$ in chloroform), is adsorbed next on the column. It is followed by considerable quantities of a xanthophyll, the absorption curves of which are intermediate in shape between those of lutein and those of the flavoxanthin-like pigments (absorption maxima 4667 and 4374 Ångström units in ethanol). Upon partition between 60 per cent. methanol and petroleum ether, this pigment was distributed almost entirely in the methanol. It is followed on the column by several xanthophylls, which occur in small quantities and which remain adsorbed near the top of the column, even after the latter has been washed repeatedly with fresh solvent.

It now seems established that several of the adsorbed pigments observed by Tswett are mixtures of two or more xanthophylls. Many of these are not identical with previously described substances of this nature and, even though many of them occur in small quantities, together they constitute a considerable proportion of leaf xanthophyll.

The physical and chemical properties of the leaf xanthophylls are being redetermined. When this is accomplished, a complete description of the compounds isolated and of the methods used in their preparation will be published elsewhere.

The observations recorded in this note are of particular significance in relation to methods for the quantitative estimation of leaf xanthophyll. The variations in the composition of leaf xanthophyll, which result from different methods of preparation, and the great differences in the absorption spectra of the different constituents preclude the use of most xanthophyll preparations as standards or reference substances for the determination of xanthophyll concentration by comparative spectral absorption methods. The heterogeneity of leaf xanthophyll and the variations in properties of the constituents account, no doubt, for the divergent values which have been reported for the xanthophyll content of leaves.

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THE AVERAGE PORE SIZE OF DIAPHRAGMS[1]

IN connection with some investigations of the relationship between surface charge density of adsorbed ions and ionic antagonism,[2] it became necessary to evaluate the mean pore radius of cellulose and powdered quartz diaphragms.

The expression for electrical conductivity,

$$C=\frac{l}{\pi r^2 n},$$

(where C is the cell constant and n the number of capillaries of mean length, l, and mean radius, r) was substituted into Poiseuille's law,

$$V=\frac{\pi P r^4 n}{8\eta l}.$$

Here V represents the rate of flow in cc per sec. under a pressure, P, in dynes per cm.[2] The resultant equation,

$$r=\sqrt{\frac{8\eta C V}{P}},$$

can be used to estimate r, assuming a capillary of circular cross-section (as, in general, with most of such methods) and a membrane of non-conducting material. A relationship of this degree of simplicity does not appear to have been previously described.

Diaphragms were made by packing the desired material into a streaming potential cell of the type used by Martin and Gortner.[3] The origin of the quartz and cellulose used has been described before.[4] The cell constant was measured with the diaphragm filled with N/10 KCl to eliminate the effect of surface conductance. Since the pierced gold electrodes form the ends of the cell, electrical conductance and liquid flow data can be directly compared. Any narrowing of the effective pore radius by imbibition of water should be accounted for by this method.

The rate of flow was found to be linear with pressure and independent of time; no blocking effects could be noted. The radius of the quartz diaphragms investigated ranged from 1.4 to 30 μ, depending upon the degree of grinding. On the other hand, cellulose membranes were more uniform in behavior, with a

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[1] Paper No. 1355 Journal Series, Minnesota Agricultural Experiment Station.

[2] L. S. Moyer and H. B. Bull, *Jour. Gen. Physiol.*, 19: 239, 1935.

[3] W. M. Martin and R. A. Gortner, *Jour. of Phys. Chem.*, 34: 1504, 1930.

[4] H. B. Bull and R. A. Gortner, *Jour. of Phys. Chem.*, 35: 309, 1931; 36: 111, 1932.