

Lamm,<sup>9</sup> in calculating the diffusion constant, and neglecting the viscosity, values of  $2.0$  and  $2.1 \times 10^{-8}$  were obtained from the initial and final photographs. It seems fair to state, however, that one is justified in calculating the molecular weight of the virus protein from these data, providing he can account for the peculiar diffusion behavior in a quantitative manner.

In Fig. 2, values for the ratio of the short to the

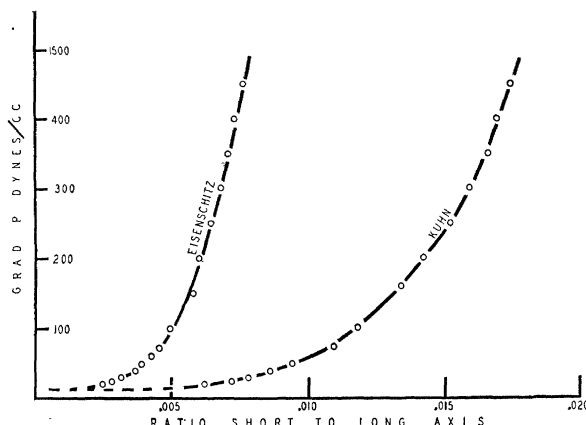


FIG. 2.

long axis of the virus protein at various shearing stresses, as calculated from Frampton's<sup>10</sup> data using the Kuhn and EisenSchitz equations, are plotted against the pressure gradient along the capillary of the capillary viscometer. These two equations may legitimately be used for solutions of elongated particles that obey Poiseuille's law in the case of a random orientation of the particles—ostensibly for a solution at rest. The extrapolated value of the short to long axis is zero; that is, the extrapolated value for the ratio of the long to the short axis for the quiescent fluid is infinite. In the Perrin equation

$$\frac{1}{R} = \frac{\left(\frac{a}{b}\right)^{\frac{2}{3}}}{\sqrt{1 - \left(\frac{a}{b}\right)^2}} \log \frac{1 + \sqrt{1 - \left(\frac{a}{b}\right)^2}}{\frac{a}{b}},$$

where  $a/b$  is the ratio of the short to the long axis and  $R$  is the asymmetry constant, as  $a/b$  becomes smaller and smaller, the value of

$$\frac{\left(\frac{a}{b}\right)^{\frac{2}{3}}}{\sqrt{1 - \left(\frac{a}{b}\right)^2}}$$

approaches zero more rapidly than

$$\log \frac{1 + \sqrt{1 - \left(\frac{a}{b}\right)^2}}{\frac{a}{b}}$$

<sup>9</sup> O. Lamb, *Zeit. Physik. Chem.*, Abt. 143: 177, 1929.

<sup>10</sup> V. L. Frampton, *Jour. Biol. Chem.*, 129: 233, 1939.

approaches infinity. That is, in the limit,  $1/R$  is zero, and  $R$  is infinite. Substituting the value of  $R$  in the well-known equation

$$R = \frac{M(1 - \zeta v)}{6 \pi \eta_0 N S_{20} \left( \frac{3 M v}{4 \pi N} \right)^{1/3}}$$

where  $M$  is the molecular weight, we see that in the legitimate use of these methods the corrected value for the molecular weight of the virus protein, as determined by ultracentrifuge means, is infinite. From a similar consideration, the corrected value for the molecular weight as determined by means of diffusion is zero.

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### UPWARD TRANSPORT OF MINERALS THROUGH THE PHLOEM OF STEMS

In 1937 Gustafson and Darken<sup>1,2</sup> showed that radioactive phosphorus was conducted upward in the stem of a plant through the bark. In the second paper it was suggested that to get a quantitative comparison between the conduction in the xylem and the phloem the part of the plant above the cut of xylem or phloem should be ashed and the total activity determined. This has now been done with *Bryophyllum calycinum* and *Salix* sp.

The experiments were conducted as before, except that the activity was determined with a Geiger counter. Usually three plants as alike as it was possible to select were used in each experiment; one was used as control, the second had a section of the xylem removed and the third plant a girdle of bark removed. In some plants the xylem was separated from the bark for a distance of several centimeters, but a piece was not removed, only a cut was made at the lower end of the xylem-phloem separation. The lower end of this bare xylem was kept in a test-tube of water during the experiment to supply the top with an abundance of water, as it was found in some experiments, where a portion of xylem had been removed, that the leaves wilted soon after the experiment was set up.

At the end of the experiment the plants were cut off at the level where the girdle had been made and the two parts ashed separately. The lower part has been designated roots, even though several inches of the base of the stem was included. The ashing was done in a muffle furnace at  $650^\circ \text{C}$ .

The ash was dissolved in 10 to 20 ml of 10 per cent. HCl, depending upon the amount of ash. An aliquot (.2 to 1.0 ml, depending upon the activity of the phosphorus) was allowed to be absorbed by a piece of blot-

<sup>1</sup> F. G. Gustafson and Marjorie Darken, *SCIENCE*, 85: 482-493, 1937.

<sup>2</sup> F. G. Gustafson and Marjorie Darken, *Amer. Jour. Bot.*, 24: 615-621, 1937.

ting paper somewhat smaller than the window over the target of the counter. This piece of blotting paper was then placed on the window over the target. The number of counts were noted for a certain length of time and the counts per minute calculated, and from this was subtracted the background count to give the number of counts due to the plant material. This count per minute was multiplied by the proper factor to obtain the count for all the ash, per minute. The total active phosphorus in the tops or roots was determined only in experiment 37, but as determined the activity in the different parts is strictly comparable and the figures in the table denote the number of counts that would have been observed per minute if all the ash of the particular part of the plant had been placed above the target of the counter.

Table 1 gives the number of beta particles originating in the irradiated phosphorus of the ash that would have hit the target of the Geiger counter per minute if all the ash had been placed above the target.

Table 1 shows clearly that if the top was supplied with water through the cut end of the xylem rather than through the phloem, when the xylem was removed, there was much less phosphorus in the top than when all the water had to pass through the phloem and there was a transpiration stream connecting the top with the root.

The actual amount of active phosphorus in the tops of the plants in experiment 37 was 0.0024 g for the control, 0.0012 g for the plant carrying on conduction

TABLE 1

Plant and experiment number	Hours in solution	Control		Conduction through xylem		Conduction through phloem	
		top	roots	top	roots	top	roots
Bryophyllum							
16 ....	41	310	1020	350	1030	15 (2)	1100
20 ....	48	360	1920	550	990	27 (1, 3)	350
21 ....	48	365	860	447	770	30 (1, 3)	593
22 ....	46	87	548	103	254	20 (1, 3)	684
36 ....	69	6100	6300	4200	4450	375 (3)	5900
37 ....	42	10200	10850	5880	6950	60 (1, 3)	6950
						820 (3)	15400
48 ....	47	.....	.....	1100	2460	500 (1)	2040
Willow							
52 ....	22	195	720	300	810	40 (2, 4)	300
						20 (1, 3)	585
53 ....	48	3060	765	135	345	45 (2)	825
						0 (1, 2)	540
54 ....	7	40	580	112	920	0 (2)	720

Note: (1) Lower end of xylem in a tube of water; (2) leaves wilted badly long before the end of the experiment; (3) partly wilted at end of experiment; (4) plant was in solution only 5 hours.

only through the xylem, 0.0002 g for the plant with phloem as the conduction element and only 0.000014 g when the conduction of the mineral took place through the phloem, but most of the water was supplied to the leaves through the cut end of the xylem.

From these experiments it would seem that not as much minerals are conducted through the phloem as Gustafson and Darken had supposed. Nevertheless, there is undoubtedly some upward conduction of minerals in the phloem under normal conditions.

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## SCIENTIFIC APPARATUS AND LABORATORY METHODS

### AN ACETO-CARMINE METHOD FOR BIRD AND MAMMALIAN CHROMOSOMES

EXCELLENT aceto-carmine preparations, either temporary or permanent, can be made of bird and mammalian chromosomes (and no doubt of other animals) by the simple procedure described below. The method is applicable to any tissue which can be readily subdivided by teasing with needles prior to fixation.

The tissue—usually young embryos, embryonic ovary or adult testis—is removed with the greatest possible speed and with the usual precautions to insure that the cells are living and normal, and quickly dissociated by teasing with needles for four or five seconds and then plunged in one of Carnoy's fixatives, where it can be further teased apart. Both the 3:1 and 6:3:1 solutions give excellent results with bird embryo somatic cells and ovaries, and with adult rat testes. (3 parts absolute alcohol and 1 part of glacial acetic acid, or 6 parts of absolute alcohol, 3 parts of chloroform and 1 part of glacial acetic acid.)

The tissue is fixed for 20 minutes to several hours and then placed directly into aceto-carmine, where it

should stain for half an hour, more or less, depending on the tissue. *Small* bits of tissue are next removed with a pipette, along with aceto-carmine, and placed on a clean slide and covered with a coverglass. The preparation is now treated just as you would a salivary gland. The excess aceto-carmine is blotted off with filter paper and the soft tissue mashed out with considerable pressure from the finger tips. If necessary, complete spreading of the cells may be accomplished by rubbing a blunt needle over the coverglass. During this whole process the coverglass must not be allowed to slip about. For temporary mounts, the coverglass is sealed with vaseline or some other like agent and studied at once. For permanent slides the coverglass is left unsealed and the slide is placed at once into a jar saturated for alcohol vapor. (This is Bridges's alcohol-euparal method.) After standing in alcohol fumes for a few hours, the slides are placed in 95 per cent. alcohol, where they should remain for several hours or longer. If the coverglass doesn't loosen and come off it should be pried off with a spade-shaped needle in a Petri dish filled with 95 per cent. alcohol. When this is done part of the cells adhere to