

covers by chipping. Improvement in methods of manufacture are expected to produce a material with a greatly reduced yellow color, but it is not likely to ever be water-white. The experienced microscopist will readily detect the ease with which transmission of the green ray for which most objectives are corrected is effected.

Upon heating, the material becomes so fluid it readily passes through filter papers and this property makes it very superior in diatom-work because of the freedom of the slides of bubbles. Its high index of refraction is its most valuable property, of course, and it should make chitinous structures, such as insects, crustaceans, etc., readily visible without staining.

Experiments are still in progress by the chemists, L. A. Penn and Paul Ruedrich, of Berkeley, California, in order to determine the best method of preparation of the resin and the chemical reactions involved.

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A NEW USE FOR THE NEPHELOMETER AND REFRACTOMETER

DURING the course of certain experiments with a large number of samples of sweet corn, it became essential to determine with a minimum of time and effort the relation between vigor and the condition of the distilled water in which it had been soaked. Some years ago, when the question of the toxicity of distilled water was under discussion, evidence was presented that the injurious effect produced on seedlings grown in distilled water was at least partially due to the extraction of electrolytes from the roots. These determinations were made in several instances by means of conductivity measurements. Apparently none of the investigators attempted to measure the effect of distilled water on dormant seeds. A number of experiments were undertaken along this line, but the Abbé refractometer was used instead of the hydrogen-ion concentration as the means of determining the relative quantities of solids leached from the seeds. The method is extremely simple, consisting merely of soaking 5 or 10 gm of seed in 50 cc of distilled water in well-stoppered bottles for forty-eight to seventy-two hours at a temperature of 30° C. Two or three drops of the liquid are all that is required for the reading. In order to check the test thousands of readings were taken of distilled water in which sweet corn seed had been immersed. In nearly all cases duplicate samples from the same ear were planted on the greenhouse bench and the growth of the seedlings compared with the instrument read-

ings. A representative portion of one of the experiments is given in the accompanying table. This will give an idea of the responsiveness of the test. The refractive index tends to increase in inverse proportion to the vigor of the seedlings. When the refractive indices (x) are correlated with the seedling characters (y) of the entire series of 116 ears in this particular experiment, $r_{xy} = .592 \pm .041$ when y = per cent. germination; $r_{xy} = .360 \pm .055$, when y = height of seedling; and $r_{xy} = .343 \pm .055$ when y = green weight. Higher correlations have been obtained in other series.

No.	Refractive index (21° C.)	Colloidal index (mm) (Std. 30 mm)	Greenhouse test		
			Pct. germination	height (mm)	Increases over check greenweight (gm)
28	1.33595	1.05	0	- 93.0	- 0.26
29	1.33470	3.05	0	- 93.0	- 0.26
30	1.33440	6.90	15	- 82.5	- 0.23
31	1.33455	3.55	0	- 93.0	- 0.26
32	1.33445	7.05	45	- 23.5	- 0.06
33	1.33420	7.25	65	- 18.0	- 0.07
34	1.33540	4.00	0	- 134.4	- 0.30
35	1.33510	1.95	0	- 134.4	- 0.30
36	1.33440	3.85	30	- 44.5	- 0.10
82	1.33410	20.25	95	5.6	- 0.08
83	1.33400	25.20	100	116.6	0.38
84	1.33410	30.95	100	112.0	0.42
85	1.33405	26.15	100	73.2	0.21
86	1.33415	17.45	90	50.4	0.16

Early in the experiments it became apparent that high refractive indices were accompanied by the presence of dense suspensions of colloids. Upon reading these with a Leitz nephelometer it was at once apparent that in many respects the colloidal index of the leachings is superior to the refractive index as a measure of the potential vigor in sweet corn. The colloidal indices in the above table are typical. The standard used in these experiments consisted of 0.5 per cent. c.p. soluble starch dissolved in 0.5 per cent. sodium toluene para sulphochloramid.

It should be noted that the coefficients of correlation for the entire series are even higher than for the refractive indices. The values are $r_{xy} = .634 \pm .037$ when y = per cent. germination; $r_{xy} = .680 \pm .034$ when y = height of seedlings; and $r_{xy} = .693 \pm .033$ when y = green weight. The correlation between the refractive and colloidal indices is $r_{xy} = .713 \pm .015$.

A considerable tendency exists for the coefficients of correlation to increase inversely as the percentage of germination in the case of the colloidal index. This is illustrated by the following:

Mean pct. germination	Correlation between colloidal index and		
	(1) pct. germination	(2) height	(3) green weight
55.8	.634 $\pm$.037	.680 $\pm$.034	.693 $\pm$ 0.33
76.5	.498 $\pm$.051	.534 $\pm$.049	.547 $\pm$ 0.48
86.5	.374 $\pm$.026	.341 $\pm$.027	.313 $\pm$.027

According to this the colloidal index test is increasingly accurate as the sweet corn becomes more inferior. This is a very decided advantage.

A number of students in the laboratory of plant physiology at the University of Illinois have used the nephelometer with decided success in measuring the reduction of vitality as affected by disease or following treatment with various chemical and physical agents. It is possible that these tests may prove useful in determining viability in grains and in other seeds having a fairly large endosperm.

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SPECIAL ARTICLES

EXCYSTATION IN VITRO OF HUMAN INTESTINAL PROTOZOA¹

FOR many years it was believed that the cysts of intestinal protozoa would not excyst until subjected to the digestive juices peculiar to the normal host of the species. Recent experimental work, however, indicates that moisture and a temperature of about 37° C. for several hours are the only factors necessary to stimulate excystation in the intestinal protozoa of man. Darling² (1913) noted the disappearance of cysts and the appearance of trophozoites in feces containing cysts of *Endamoeba histolytica* that were kept in a moist chamber. It is not at all certain, however, that the trophozoites observed came from the cysts, since amoebae of other species often appear in fecal material kept under similar conditions. Yorke and Adams³ (1926) observed the process of excystation in this species; Allen⁴ (1926) describes

¹ From the Laboratory of Protozoology, Johns Hopkins School of Hygiene and Public Health. The writer is greatly indebted to Mr. Conrad Bauer for his valuable assistance.

² Darling, S. T., 1913. "Observations on the cyst of *Entamoeba tetragena*." *Archiv. Int. Med.*, 11: 1-14.

³ Yorke, W. and Adams, A., 1926. "Observations on *Entamoeba histolytica*. I. Development of cysts, excystation and development of excysted amoebae, in vitro." *Ann. Trop. Med. and Parasit.*, 20: 279-302.

⁴ Allen, E. A., 1926. "Excystment of *Councilmania lafleuri* Kofoid and Swezy in culture in vitro." *Univ. Cal. Pub. Zool.*, 29: 175-178.

what she believes to have been excystation in the form named by Kofoid and Swezy⁵ (1921) *Councilmania lafleuri*; and Smith⁶ (1927) has observed, and shown to the writer, excystation in *Iodamoeba williamsi*. The writer⁷ is now able to add to this list *Endamoeba coli*, *Endolimax nana* and the flagellate *Chilomastix mesnili*; he has also observed early stages of what appears to be excystation in vitro in *Giardia lamblia*. The other intestinal protozoa of man are *Trichomonas hominis* and *Endamoeba gingivalis*, which have no cyst stage, and *Embadomonas intestinalis*, *Tricercomonas intestinalis* and *Dientamoeba fragilis*, which are rare species not easily obtained for study.

Endamoeba histolytica. Excystation in vitro in this species has been described by Yorke and Adams (1926). Material containing cysts was sealed under a cover glass and examined in a warm microscope chamber. Pseudopodia were formed inside of the cyst; then a break appeared in the cyst wall through which the amoeba escaped. Moisture and a temperature of about 37° C. seemed to be the essential factors in excystation. Cysts that had been in Locke-egg-serum medium at 37° C. for two hours proved more satisfactory than unincubated specimens.

Councilmania lafleuri. Allen (1926) saw what she believed to be the last of eight amoebulae to escape from the cyst wall of this species. According to her observations the entire amoeba does not leave the cyst, but the eight amoebulae into which it divides pass out one by one through a pore in the cyst wall.

Iodamoeba williamsi. Excystation in this species has been observed by Septima C. Smith (1927). She found that cysts fifteen hours old, when placed in an incubator at 37° C. for two hours and then in a warm chamber at about 40° C. for three hours, would excyst in a saline medium. Minute pseudopodia were noted within the cyst; then followed a break in the wall and the escape of the amoeba. In some cases the amoeba emerged part way and then returned, escaping only after several passages back and forth. The newly excysted organisms were very active. She concluded that the stimuli necessary for excystation are moisture and a temperature of about 37° C. for several hours.

⁵ Kofoid, C. A. and Swezy, O., 1921. "On the free, encysted and budding stages of *Councilmania lafleuri*, a parasitic amoeba of the human intestine." *Univ. Cal. Pub. Zool.*, 20: 169-198.

⁶ Smith, Septima C., 1927. "Excystation in *Iodamoeba williamsi* in vivo and in vitro." *SCIENCE*, 65: 69-70.

⁷ Hegner, Robert, 1927. "Excystation and infection in the rat with *Giardia lamblia* from man." *Amer. Journ. Hyg.* (in press).