taken the very considerable fossil record into account beyond being influenced by the entirely speculative essay of Arber and Parkin. Floral morphology, perhaps the most variable feature in plants, is greatly overweighted in this as in all classificatory schemes, and I fancy that I can see the ghosts of Bentham and Hooker influencing some of the author's decisions.

It seems to me that more consideration should be given to the evidence of the probable late origin of such herbaceous groups as the Cruciferae and Caryophyllaceae. For example, in seeking to justify his separation of woody and herbaceous plants Hutchison says, "Many of the primitive families are either entirely woody or entirely herbaceous" (page x), and then names under the second category the Ranunculaceae, Papaveraceae, Crassulaceae, Saxifragaceae, Caryophyllaceae and Cruciferae. That any of these are primitive is almost entirely a matter of opinion. Surely the Crassulaceae are rather specialized, all are practically unrepresented in the fossil record, and there are many facts that suggest their relative modernity. It should be recognized that extreme habitat specializations, *e.g.*, to shores and salt pans (Frankeniaceae, Plumbaginaceae) or to aridity (Cactaceae, some Euphorbiaceae) do not give issue to new types, but represent blind endings which are likely to be comparatively modern, as is further indicated by their restricted distribution.

From a fair knowledge of both distribution and geology I venture to think that the so-called Wegener hypothesis of continental displacement raises many more problems than it solves.

I am heartily in sympathy with Hutchison's sharpening and multiplying the orders. The old aggregations that passed as Ranales and Geraniales have been sadly in need of segregation for a long time, and I am glad to see the Laurales set apart and the shifting of the Piperales from a primitive to a derivative position. I much doubt the suggested alliance between the ancient (despite the aforementioned ghosts) Proteaceae and the Thymeleaceae, the separation of the Hydrangeaceae and Saxifragaceae, or of the Myrsinaceae and Primulaceae, although I concede that the last may have a foundation.

Hutchison considers the Juglandales as derived rather late from the Sapindales, although the former appear early in the geological record, as do also the Moraceae; and the Fagales are among the earliest clearly recognized angiosperms.

The Platanaceae, although antedating both in the rocks, is considered to have been derived from the Rosales through the Hamamelidales, and to lead to the Amentiferae. The question of direction of evolution is a puzzling one in all groups of organisms, but I believe that the direction has been reversed in this instance.

There is a vast amount of inertia among systematists, whether it be in the writing of regional floras, or the arrangement of herbaria, and it is doubtful and probably not desirable that Hutchison's scheme should be adopted. A pious wish for international uniformity in taxonomy is probably as elusive a will-o-wisp as international comity, or the nomenclatural stability that we have heard so much about during the last thirty years, but that the first general phylogenetic presentation of dicotyledons after nearly forty years of systematic activity should not receive the attention and discussion which it deserves is unthinkable.

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SCIENTIFIC APPARATUS AND LABORATORY METHODS

THE CULTIVATION OF OPALINA

IN a recent paper appearing in SCIENCE,[1] a report was made on the reaction of Opalinas to various laboratory media. The maximum period over which these animals could be made to live was seventy-three hours. In working upon a problem of the specificity of Opalinas in frogs, we found that this period was too short for the purposes of our experiments.

A search of the literature on this subject revealed a method used by Konsuloff[2] in 1922 which succeeded in maintaining Opalinas in culture for two months. There was one fault to be found with this method, however; he had to change the medium every day. For our purposes this was unsatisfactory. Therefore we refined Konsuloff's technic until we have succeeded in cultivating Opalinas for twenty-five days without having to change the medium.

The procedure is as follows. Pütter's fluid, which has the following composition, is used.

NaCl	.8 per cent. sol.	100 parts
Rochelle salts	30.0 per cent. sol.	5 parts
Distilled water		400 parts

This is made with boiling distilled water to ensure as complete sterility as possible. After this has cooled it is subjected to the action of a suction which is maintained until bubbling ceases. This removes any excess air, a factor which has been demonstrated by Konsuloff to be lethal if permitted to remain. To this fluid is added, in the proportion of .25 grams to 200

[1] Larson, Van Epps, and Brooks, SCIENCE, Vol. LXII, No. 1604, page 289, (1925).

[2] Konsuloff, S., "Untersuchungen über Opalina," *Archiv. f. Protistenkunde,* Band 44, Heft 3, (1922).

cc fluid, powdered egg white. This is added just slowly enough so that no flocculation occurs. The fluid is then put in 15 cc centrifuge tubes to the amount of 9 cc per tube. The Opalinas are introduced by a capillary pipette after having been removed from the cloaca of the infected frog and having been washed once in either Ringer's or Pütter's fluid, without the egg white. This last procedure tends to reduce the amount of extraneous matter. About .1 cc of fluid containing a heavy concentration of Opalinas is introduced into each tube. These tubes are then corked with one-hole rubber stoppers. As a final seal, the holes of the stoppers are closed with small glass plugs. This arrangement permits the escape of the air that would otherwise be confined under pressure in the tubes. The presence of this pressure was found by Konsuloff to kill the cultures. All the glassware and corks were sterile before using.

In order to observe the animals without removing them from the tubes and thereby introducing air and bacteria which would lessen the length of the period of cultivation, a binocular microscope was set up horizontally and the tubes held singly at the correct focal distance.

We hope that this technic in the cultivation of Opalinas will be of some help to others working in this field.

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A SIMPLE METHOD OF OBTAINING SINGLE-SPORE CULTURES

WHILE working with species of Fusarium the writer developed a method of obtaining single-spore cultures that seems simpler and more rapid in operation than methods previously described. A piece of soft glass tubing is heated over a Bunsen flame and drawn out to an inside diameter slightly greater than that of the spores to be isolated. Care in the amount of heat to be applied and a little practice in manipulation is necessary to obtain suitable diameters. The prepared capillaries are broken up into lengths of about three cm and filled by means of capillary attraction from a spore suspension made in warm nutrient agar. After a few trials a spore dilution is easily made that will give from one to four spores to the tube.

The filled tubes are then examined under the microscope and broken up according to the number and position of the spores observed. The resultant pieces are again examined to make sure that each contains one spore only, then picked up with forceps, immersed in alcohol to sterilize the outside and placed in the desired medium. By using a solid rather than a liquid medium the filled tubes may be broken without disturbing the contents and sterilized without injury to the spores.

Should it be desirable to obtain cultures from a single hypha, then the piece of glass tube with the spore is pushed to the bottom of the medium in a Petri dish where germination may be followed under the microscope; as soon as the germination hypha has grown beyond the end of the tube it is cut off and the glass removed. A single hypha is very easily obtained in this manner because of the macroscopic size of the glass tube.

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SPECIAL ARTICLES

DIET AND BODY FAT

IN a study[1,2] of the influence of diet upon the quality of fat produced in the animal body we found when rats were fed diets containing dried skimmed milk and either peanut oil or soybean oil or corn oil, these dietary oils furnishing about 60 per cent. of the total food calories in each case, the fat or rather oil yielded by the rat in each case was quite similar in iodine number value to that of the food oil. On the other hand, when a diet containing dried skimmed milk and starch (the latter being substituted equicalorically for the oil ingredient of the above diets) was fed, a so-called "hard" fat was obtained. Under all experimental conditions, cod liver oil and yeast were added to the ration as sources of vitamins.

Furthermore, we found it possible to convert the "soft" body fat into a "hard" body fat by changing the oily diet to the carbohydrate-rich diet, provided the change of food took place when the rats were of adolescent age (140–150 gm.) and the feeding of the "hardening" diet was continued over a comparatively long period. For example, the "soft" body fat of 140 gm. rats produced on a soybean oil diet was completely "hardened" on the carbohydrate-rich diet when the latter was fed until rats attained the weight of about 250 gm.

The question naturally arose: What would be the effect of fat depletion through selective starvation on the subsequent rate of "hardening" of the body fat?

In seeking an answer we subjected rats, grown to various weight levels on the oil-containing diets, to a starvation process before feeding the "hardening" diet. We then compared the fat obtained from other

[1] Anderson, W. E., and Mendel, L. B., "A Technique for the Study of Fat Production in Animals," Proc. Soc. Exp. Biol. and Med., 1923–24 (21), 436.

[2] Anderson, W. E., "The Influence of Diet on Fat Production in the Animal Body," Proc. Am. Soc. Biol. Chem., *J. Biol. Chem.*, 1925 (63), XLVI.