pointed base. In this way it was noticed that now and then a leaf contained a tree frog. These frogs having exceptionally long legs and slender trunks seem especially well shaped to fit into any narrow cavity and as they sat holding to the side of the trumpet some distance down from its mouth where they yet had ample room, they presented very quaint and attractive pictures. The color of these frogs being a yellow-green with some golden spots, they harmonized excellently with the background illuminated by light in part transmitted through the walls of the leaf.

The frogs sat with their heads up toward the mouth of the pitcher or trumpet, and were advantageously placed to seize any insect that might come down into the cavity of the leaf. That this was a chance worth considering was perhaps indicated by the fact that in some cases small spiders had spun their webs across the opening of the leaves, thus being placed well to intercept the insects that might else have gone down to the frog.

In the latter part of June and the first of July as many as twelve frogs were found, on two days, a week or more apart, while looking into about six hundred leaves. That this occurrence of frogs in the leaves was not limited to the above cases was proved by a third examination made July 9, when just two hundred leaves were looked into as they came; one hundred in the locality previously visited and the other hundred in a new locality. In both cases the leaves examined were on plants growing along the edges of the woods where the frogs were probably more numerous than out in the open where the pitcher plants were more numerous. The first hundred leaves contained three frogs; the twenty-ninth leaf harbored two frogs and the thirty-sixth one. The second hundred contained five frogs; one in the fourth, in the fifty-sixth, fifty-eighth, sixty-sixth and eighty-seventh. Thus in these leaves skirting the woods some two to five per cent. contained frogs.

The season being then very dry the occurrence of the green-tree frog in the leaves of the pitcher plant may have been an unusual occurrence due to the frogs seeking protection from drying; on the other hand, this frog may have developed a special appreciation of the advantages of these retreats, both as affording food and as giving the comparative immunity from attacks of enemies that the protective resemblance of the frog to the leaf suggests.

E. A. ANDREWS

THE JOHNS HOPKINS UNIVERSITY

THE OCCURRENCE OF CONJUGATION IN PARAMECIUM CALKINSI

Paramecium calkinsi was described as a new species in 1921 by Woodruff[1] after he had carried pedigreed cultures of it for nearly a year and had made various tests to determine that the new species belonged to the genus Paramecium. Woodruff states that every effort was made to secure conjugation, but without success. He further states that during the life of his pedigree culture an intensive study of the nuclear conditions was made to detect endomixis if it occurred. There was no indication of endomixis, although there were rhythms in the rate of fission. So far as we are aware, neither Woodruff nor any other investigator has succeeded in inducing conjugation in *P. calkinsi* and we have seen no record of endomixis having been observed. One of us (Wenrich) has made repeated but unsuccessful efforts to induce conjugation in a strain of this species obtained from Professor Woodruff.

As a result of all these failures to observe conjugation or endomixis, the impression has been developing that these phenomena do not occur in this species. In view of the important theoretical implications of such an exceptional behavior on the part of a distinct species of Paramecium, it seems worth while to record here the fact that conjugation has been observed in cultures of *Paramecium calkinsi* secured originally at Woods Hole, Mass.

The original material was a sample of water taken from the pond on the east side of the Eel Pond at Woods Hole. The water in this pond is brackish, since at high tide sea water from the Eel Pond flows into it; at other times during the day, it drains into the Eel Pond. The material collected from the source pond on August 18, 1927, contained large numbers of *Paramecium calkinsi,* and some of it was distributed into syracuse watch glasses for more convenient culture and study. At the beginning of September, each of these cultures was placed into a vial and transported to Philadelphia. On September 5 each of these cultures was reestablished in a syracuse watch glass without change of media. On September 7 it was noticed that in culture number 5 many individuals had died, so a new culture, 5A, was made, by transferring about 200 individuals to another watch glass containing an infusion made by boiling together some timothy hay and wheat grains. The old culture was made over by taking out most of the fluid and replacing it with the infusion just mentioned. Within the next few days other cultures were replenished with the hay-wheat infusion. Examination on September 16 showed heavy populations in both cultures 5 and 5A and some dozens of pairs of conjugants. Conjugation has since been observed in five others of the

[1] "The Structure, Life History, and Intrageneric Relationships of Paramecium calkinsi," Sp. Nov. *Biol Bull*., Vol. 41, pp. 171–180.

cultures brought from Woods Hole; and in the original culture, No. 5, a smaller or larger number of conjugating pairs were observed almost daily up to October 15, 1927.

The question presents itself as to the causes of the failure of Woodruff's strain to conjugate, in comparison with the apparent readiness to conjugate on the part of the Woods Hole strains. The differences may be attributed either (1) to the difference in the original habitat, (2) to different culture methods in the laboratory or (3) to inherent racial differences, or some combination of these. In regard to the first possibility, it will be remembered that Woodruff's strain came from a fresh-water source while the Woods Hole strains have come from brakish water. Experimental tests demonstrated that these brakish-water strains would live in an apparently normal condition in fresh water and in various strengths of sea water up to pure sea water, provided the changes to the higher strengths were made gradually. The brakish-water habitat may therefore be considered a normal one.

In regard to the second possibility, it may be pointed out here that we have subjected Woodruff's strain to the same cultural conditions that we used for the Woods Hole strains, but have not as yet been able to induce conjugation in this strain. The evidence at present available rather favors the third possibility—that of inherent racial differences.

Segregated strains are being established from ex-conjugants and it is hoped to make an intensive study of the conditions which will induce conjugation as well as to investigate thoroughly the cytological details of the process.

D. H. WENRICH,
C. C. WANG

ZOOLOGICAL LABORATORY,
UNIVERSITY OF PENNSYLVANIA

CENTRIFUGING FILTERABLE VIRUSES

I READ with interest the note by M. S. Marshall, entitled "Centrifuging Filterable Viruses," which appeared on page 219 in SCIENCE of September 2, 1927. There seems to be little doubt as to the accuracy of Marshall's computations, and it seems likely that his conclusions are correct for a pure virus in water. However, it should be pointed out that his conclusions do not hold for virus which is in the plant extract. The writer's studies show that the virus of tobacco mosaic can be concentrated by means of the supercentrifuge. These investigations were published in *The Journal of Agricultural Research,* vol. 35, pp. 13–38, July 1, 1927. It should be pointed out that the supercentrifuge has been used in this and in other laboratories for concentrating bacteria and other micro-organisms. See the article by C. Juday, in the *Transactions* of the Wisconsin Academy of Science, Arts and Letters, vol. 22, pp. 299–314. 1926.

The writer's studies indicate that physical and chemical treatments which cause coagulation and precipitation to take place in plant extracts, also cause or assist the virus to settle out of the extract. However, the relative advantages of the various treatments, and the exact relations between the virus particles and other particles which are precipitated out of the extracts, are not fully known. Some treatments are less desirable than others because they are toxic to the virus in varying degrees. Some treatments produce only very finely divided coagula which do not settle out on long standing. Frequently these are heavily charged with virus, and they can be removed almost completely by means of the supercentrifuge.

It should be emphasized that centrifuge methods are of unquestionable value in studies on the virus of tobacco mosaic, and thus far the writer has found the supercentrifuge to be one of the most useful pieces of apparatus in the laboratory.

H. H. MCKINNEY

CEREAL VIRUS DISEASE INVESTIGATIONS,
BUREAU OF PLANT INDUSTRY

COLORIMETRIC METHODS IN BIOLOGY

PAST discussions of colorimetry, in the pages of SCIENCE, and particularly the recent appeal by Irwin G. Priest for bibliographic references and reprints bearing on this subject, have emboldened me to call attention in your columns to a paper of my own, published during the past year. I refer to an article entitled "Linear and Colorimetric Measurements of Small Mammals," which appeared in *The Journal of Mammalogy,* vol. 8, no. 3, August, 1927, pp. 177–206.

I hope that this unseemly bit of self-advertising on my part will be condoned for the following reasons. The scope of the journal in which the paper was published would doubtless tend to conceal it from the view of many biologists who are not especially interested in the Mammalia. On the other hand, the methods therein described are doubtless applicable to a wide range of biological and even of inorganic objects.

The writer is far from wishing to pose as an expert on colorimetry, either practical or theoretical, but he has been dealing for many years with color differences in certain species of rodents, and has been obliged to treat these differences quantitatively. Since no recognized technique was available for the purpose, it was necessary to work this out through protracted experimentation. A type of instrument (the Ives Tint Photometer) was finally adopted, which was already in use for industrial purposes. Some further equipment was necessary, however, and