NON-TRANSFER OF MATERIALS BETWEEN PLASMODIA OF DIFFERENT SPECIES OF MYXOMYCETES[1])

by

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(with 8 figs.)

INTRODUCTION

Until the relatively recent work of DANIELS & RUSCH (1), opinion differed (2) as to whether myxomycete plasmodia were able to absorb materials in solution from their substrata or whether they were obligately holozoic. With the successful culture of *Physarum polycephalum* in a non-particulate medium this controversy was settled once for all, at least for that particular species. It has long been known that plasmodia of differents species when placed in intimate contact one with another do not merge, but that the protoplasm of one may flow over that of the other just as it migrates over the surface of any suitable substratum, With the knowledge that the plasmodium of *P. polycephalum* is able to absorb materials in solution, the question posed itself as to the possible transfer of soluble substances from one plasmodium into another when two protoplasts are in intimate contact. This paper presents the first results of a study which attempts to answer that question.

MATERIALS AND METHODS

The plasmodia employed were those of *Physarum polycephalum*, *Physarum gyrosum*, and *Fuligo cinerea*. The strains were freed from bacteria before they were used in these experiments. The plasmodia were grown in separate Petri dishes at 25° C. for five days

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on Difco plain agar in which sterile rolled oats had been incorporated as follows: Difco plain agar 1½ % was prepared, distributed in culture tubes, 20 ml to a tube, and autoclaved for 15 minutes at 15 lbs. pressure. Rolled oats were similarly sterilized in tubes. The contents of one tube of agar was then poured into a sterile Petri dish and allowed to cool. Before solidification, sterile oats were poured slowly at one edge of the Petri dish and allowed to sink into the cool agar without scattering. When the agar solidified, a por-

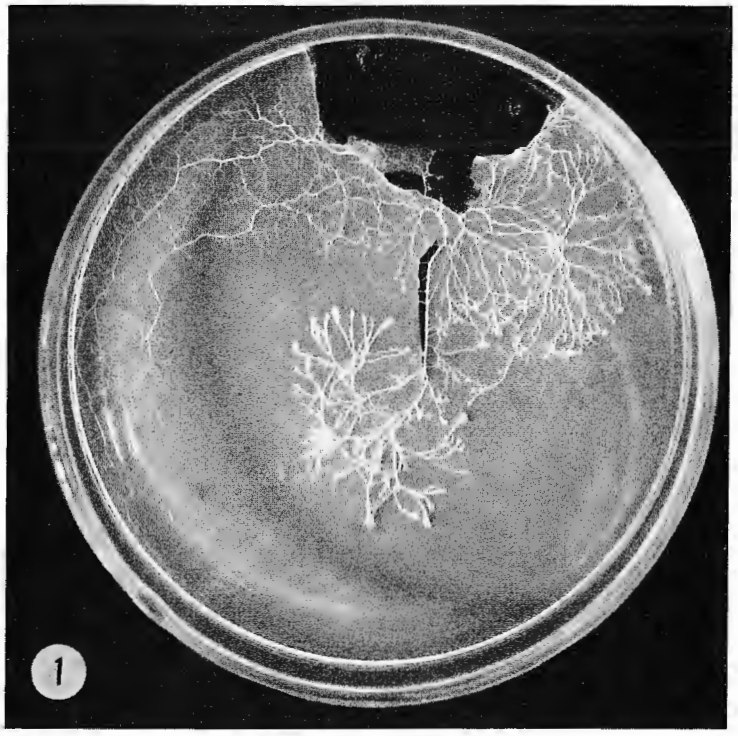

Fig. 1. Radioactive ("hot") plasmodium of *Physarum polycephalum* growing on non-radioactive ("cold") agar. × 0.66

tion of a bacterium-free plasmodium was transferred onto the agar surface over the oats. Within five days the vigorously growing plasmodium had spread over the entire agar surface and agar blocks bearing well-formed plasmodial fans could be cut out for experimental purposes.

Radioactive agar was prepared by adding 1 ml of an aqueous solution of $NaHPO_3$, containing 15 μc P—32, to 20 ml of melted Difco plain agar. The tube was rolled between the palms of the hands to mix the agar well and the contents were then poured into a sterile Petri dish and allowed to solidify.

Agar blocks bearing vigorous plasmodial fans of *P. polycephalum* were then transferred to radio-active ("hot") agar whereas fans of *P. gyrosum* were transferred to non-radioactive ("cold") agar

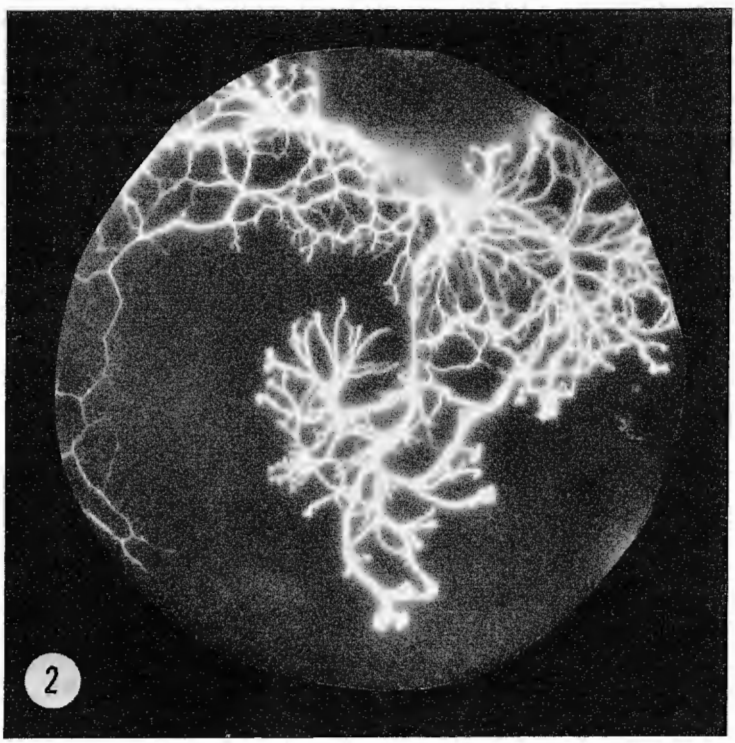

Fig. 2. Radioautograph of culture shown in Fig. 1. × 1.0

and permitted to spread. Within a few hours the plasmodia on "hot" agar had absorbed enough P-32 to render them intensively radioactive. Blocks of "hot" agar bearing radioactive plasmodial fans were now transferred to Petri dishes containing "cold" agar

and permitted to migrate off the block and spread onto the "cold" agar (Figs. 1 and 2.) The "hot" agar block was removed as soon as the plasmodial fan had migrated from it. A block of "cold" agar bearing a "hot" fan of *P. polycephalum* was now transferred into a Petri dish in which a "cold" plasmodium of *P. gyrosum* had spread and the two plasmodia were then permitted to intermingle and spread one over the other. The results are recorded in a series of photographs and corresponding radioautographs.

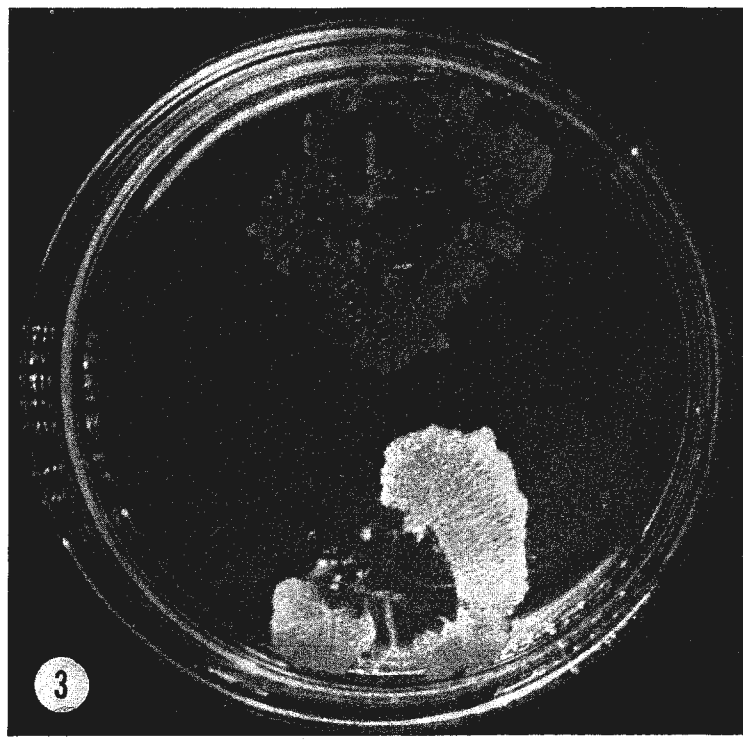

Fig. 3. "Hot" plasmodium of *Physarum polycephalum* (top) and "cold" plasmodium of *P. gyrosum* approaching each other on "cold" plain agar. × 1.0

Results

Figs. 1 and 2 — photograph and radioautograph respectively — illustrate a hot plasmodium of *Physarum polycephalum*. Fig. 3

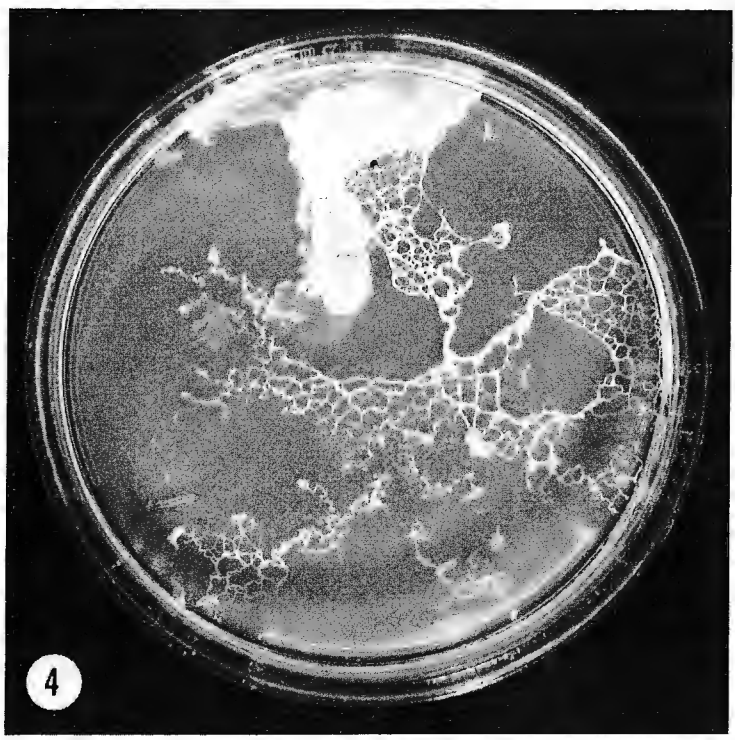

Fig. 4. Extensive contact established between the plasmodia shown in Fig. 3. × 0.84

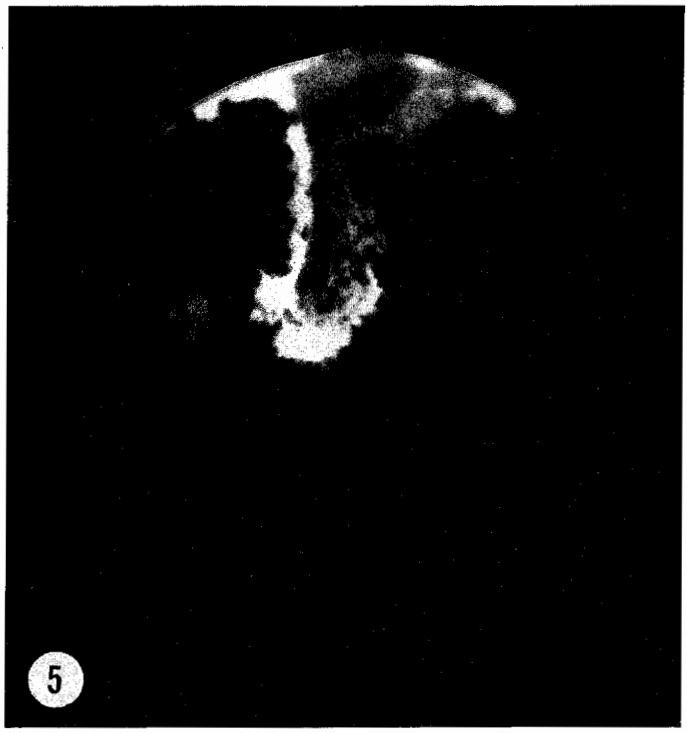

Fig. 5. Radioautograph of culture shown in Fig. 4 indicating that only *P. polycephalum* is radioactive and that no transfer of P-32 has taken place from one plasmodium to the other. × 1.0

is a photograph of a "hot" plasmodium of *P. polycephalum* and a "cold" plasmodium of *P. gyrosum* spreading toward each other. In Fig. 4 (photograph) the two plasmodia are in intimate contact over a large area, the "cold" plasmodium having spread over the "hot" plasmodium. From the radioautograph of Fig. 5, it is evident that only one plasmodium is "hot" and that radiophosphorus has not been transferred to the other plasmodium. The radioautographs in Figures 6 and 7 were made 7 and 24 hours respectively after that

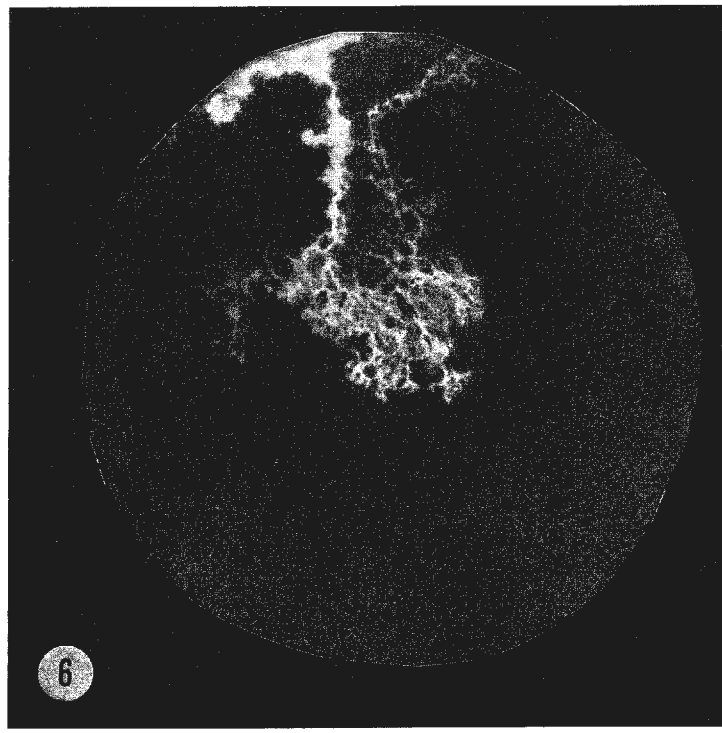

Fig. 6. Radioautograph of the same culture 7 hours after stage shown in Fig. 5. No radioactivity evident in *P. gyrosum*. × 1.0

of Fig. 5. The photograph of Fig. 8 shows the position of the two plasmodia at the time the radioautograph of Fig. 7 was made. It is clear from this series of illustrations that no transfer of P-32 had taken place after 24 hours of contact between the two living plasmodia.

To determine whether such transfer could take place by simple diffusion between non-living plasmodia, the culture shown in Figure 8 was exposed to formalin fumes for one hour. A radioautograph taken 48 hours later was similar to that in Fig. 7 indicating no transfer of P-32 had occurred.

These experiments were repeated using "hot" *P. polycephalum* and "cold" *Fuligo cinerea* plasmodia. Extensive contact was established between the two species, but again no transfer of radiophorus was evident.

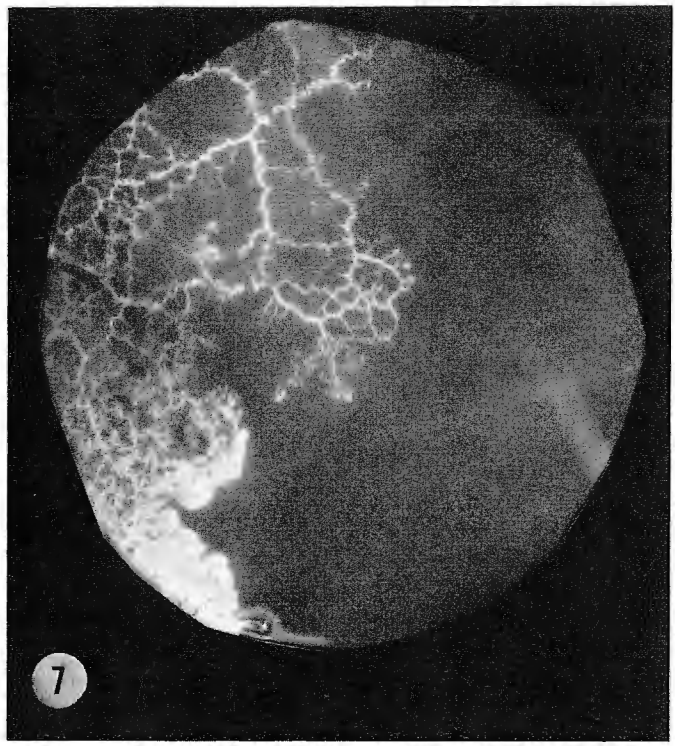

Fig. 7. Radioautograph of the same culture 24 hours after stage shown in Fig. 5. No radioactivity evident in *P. gyrosum*. × 1.0

Summary

Under the experimental conditions of this study no transfer of radiophosphorus occurred from one living plasmodium to another

when a radioactive plasmodium of *Physarum polycephalum* and a non-radioactive plasmodium of *Physarum gyrosum* or *Fuligo septica* were in intimate contact for 24 hours.

References

1. DANIEL, J. W. & H. P. RUSCH (1956). Growth of a plasmodial slime mold in pure culture on a soluble medium (Abs.) Fed. Amer. Soc. exp. Biol. Fed. Proc. **15** (1, pt. 1): 513.:
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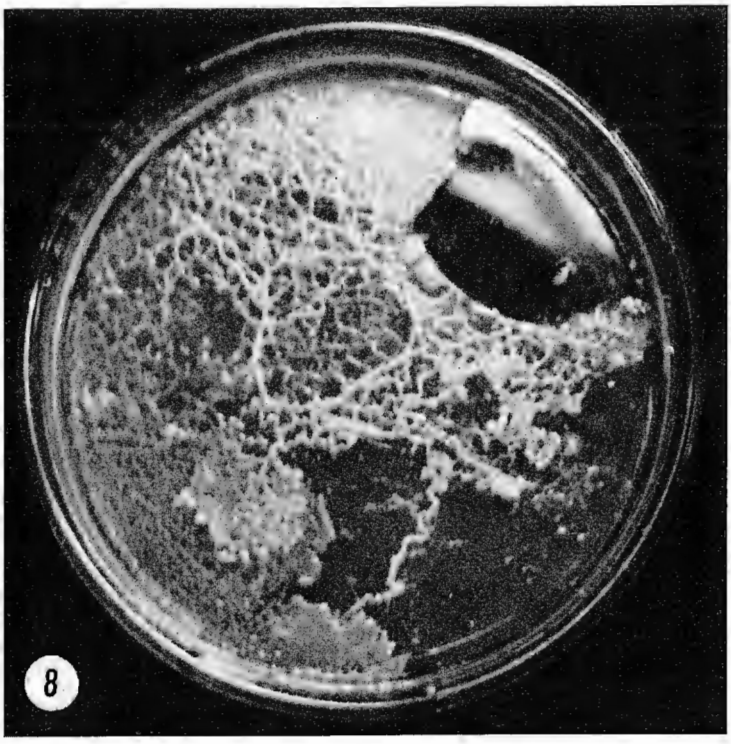

Fig. 8. Photograph of same culture at the stage corresponding to Fig. 7. × 1.0