

terms of arrays of dislocations, and the disorientation was found to be comparable to that produced in a crystal of a very heavily cold-worked polycrystalline aggregate in which it is well known that the number of dislocations is about 10^5 times that in an annealed crystal. This implies that the increase in the number of dislocation lines cutting the surface after bombardment at 77 eV. is of the order of 10^5 times the number after bombardment at 14 eV. This increase is similar to that in the number of sites found from the catalytic experiments. We therefore consider that the electron diffraction evidence gives strong support to the conclusion that dislocations cutting the surface provide sites for the reaction.

This work will be fully reported elsewhere.

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Tritium and Paper Chromatography

SINCE hydrogen is an element of fundamental interest in life processes, it is important to have a method for studying its pathway through living systems. If, in paper chromatography, one is using a radioactive tracer, a sheet of X-ray film can be placed on top of the chromatogram and the labelled compounds then produce darkened spots on the film². Unfortunately, in the case of hydrogen, only tritium is radioactive, and this decays by the emission of a very soft β -particle (18 keV.), the range of which is so low that it is absorbed before it can leave the paper and enter the film. This communication describes a technique of 'scintillation autoradiography' whereby this difficulty is overcome. The technique involves soaking the chromatogram in a liquid scintillator so that the energy of the disintegrating tritium atoms is converted into light which is detected by a film. Thus a weak β -particle is converted into light quanta which can travel into the film to produce an image. By the use of fast film, small amounts of compounds labelled with tritium can be separated and identified in only a few days.

This technique is used in our laboratories for researches on the path of hydrogen in photosynthesis. A typical experiment is described to illustrate the technique.

25 mgm. of washed algae (*Chlorella*) are centrifuged to the bottom of a conical centrifuge tube. As much water as possible is removed, since it will lower the specific activity of the tritiated water. 50 mc. tritiated water (4 c./ml.) are added under the particular conditions of the experiment and the whole rapidly agitated with a stream of air with 4 per cent carbon dioxide. At the end of the experiment absolute methanol is added to kill the algae. After extraction with alcohol and water, the extracts are evaporated under vacuum and placed on one corner of a piece of Whatman No. 4 filter paper (15 cm. \times 15 cm.) washed in oxalic acid. After development the chromatogram is attached to some suitable backing material with wire staples (used X-ray film is suitable). A spot of extract from algae or yeast labelled with tritium is placed on a piece of filter paper and when dry cut into segments. These seg-

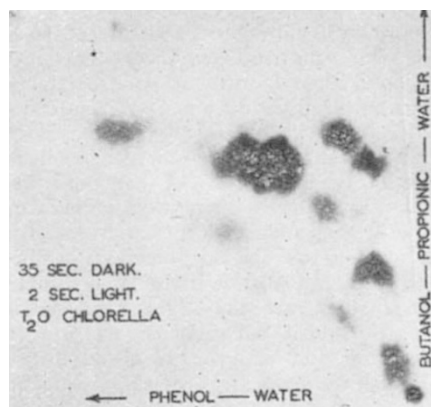


Fig. 1. Example of scintillation autoradiogram

ments are then attached to the corners of the backing sheet with staples. These later provide reference points on the backing sheet and also on the film with which to line up the film and the original chromatogram, thus identifying a spot with a definite area on the paper chromatogram. The backing sheet thus prepared is laid in the bottom of a shallow tank and covered with a scintillating liquid (for example, 3 gm./l. *p*-diphenyl benzene in sulphur-free toluene). A sheet of film is next placed on top of the chromatogram, taking care to eliminate any bubbles. The tank is then covered with an air- and light-tight top. After a few days, the film is removed from the tank, drained of scintillating liquid and allowed to dry. It is then developed and fixed in the normal manner. Such a 'scintillation autoradiogram' is shown in Fig. 1. Thus, as in carbon-14 radioautography, compounds can be tentatively identified by their position and this identification confirmed by removal from the paper and co-chromatography with known compounds, or by chemical treatment and re-chromatography.

As can be seen, the quality and sharpness compares favourably with radio-autograms produced by beta-particles from carbon-14. Although this technique has been used to study the 'photo' part of photosynthesis in our laboratories, it could also be applied to many other biological and chemical problems. In view of the cheapness and availability of tritium (10^{-4} that of carbon-14), and also the ease of its introduction into complicated organic molecules (by exposure to tritium gas³), it should be possible to use it to discover the biological fate of some of the more complex organic materials which are difficult to synthesize with carbon-14. This new technique also provides a method of preparing chromatographically pure tritium-labelled compounds. This overcomes the principal difficulty in the Wilzbach synthesis³, which yields a product contaminated with small amounts of tritium-labelled material of high specific activity, the removal of which by other means is both tedious and difficult.

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