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# Detail and Survey Radioautographs[1]

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QUASI-MICROSCOPIC TECHNIQUES in radioautography have left much to be desired in the study of the cellular distribution of radioactive elements and labeled compounds (*19*). The introduction of nuclear track emulsions in radioautographic techniques (*8*) has brought with it the possibility of studying localization of radioactive substances at oil immersion magnification. Alpha tracks have been studied at high magnification (*1, 8*) in conjunction with soft tissue sections mounted on the emulsion (*8, 9*). The localization of beta emitters has been studied at high magnification in tissue sections and blood smears (*5, 6*). The principles of radioautographic technique have been reviewed recently (*1, 4, 10, 11*) and techniques for special purposes have been described (*3, 5, 13, 14, 17*).

The much used *survey* or *contact* type of radiograph (*2, 12*) illustrated in Fig. 1 is indispensable for a study of the gross distribution of radioactive materials. This figure shows the distribution of plutonium in the tibia of a young rat. The section was cut unsoftened (*15*) at 10 μ. The survey examination, however, gives only the gross picture and does not reveal directly the detailed localization of this heavy metal.

A *detail* radioautograph is equally indispensable. Its application to bone is seen in Fig. 2, which was prepared by mounting a second thin section of the tibia on nuclear track emulsion. The detail radioautograph makes it possible to determine the localization of plutonium with respect to cells.

The interpretation of a detail radioautograph depends upon the determination of the point of origin of the emission. Although this is far more easily accomplished for alpha particle emitters, as in Fig. 2, much information may be obtained by examining at high magnification a beta particle sensitive nuclear track emulsion (*5, 7*). A description of the types of nuclear tracks is given by Powell and Occhialini (*18*).

Nuclear particles leave their points of origin in the tissue and travel in random directions. About half of the particles enter the emulsion and strike silver grains. The series of grains in a single path, when developed, reveal a nuclear track (A, Fig. 2). The mean free path is determined by the type and energy of the emission. Thus, the track can be retraced towards the locus in the tissue from which it origi-

[1] This document is based on work performed under Contract No. 7405-eng-36 for the Atomic Energy Commission.

[2] Grateful acknowledgment is made to Mrs. Julie Wellnitz and Mrs. Norma Lanter for technical assistance.

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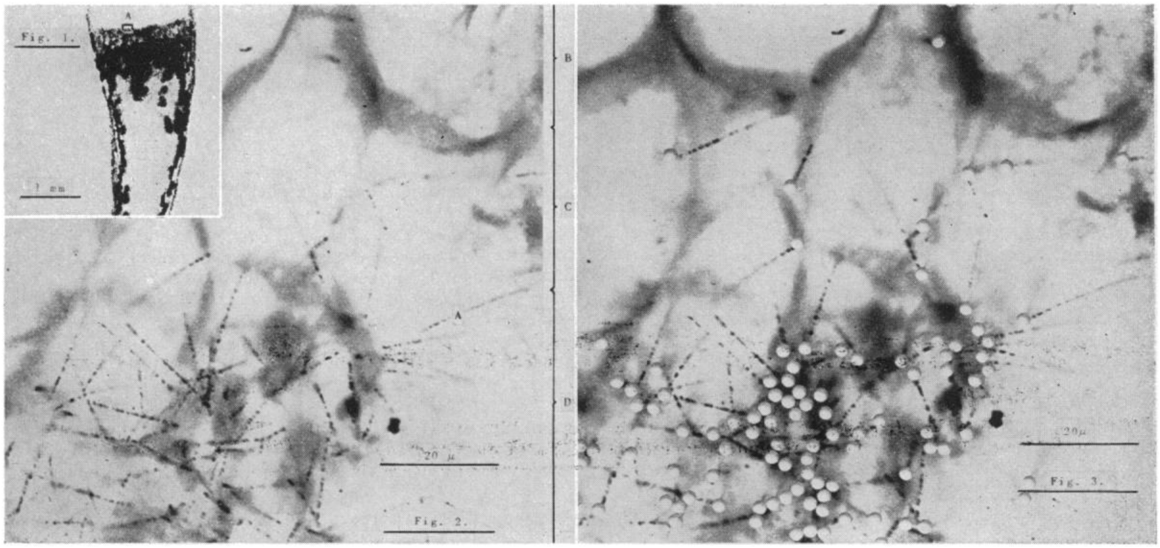


Section of tibia of 9-day-old Wistar rat, 16 g, injected intraperitoneally at 7 days of age (approx. 10 g) with 5.9 μg plutonium as citrate. Ten-μ sections of unsoftened bone (Rat 13; spec. 133; embedded in nitrocellulose, R.S. ½ sec). Fig. 1 shows a *survey* radioautograph, by contact with Eastman ultra-speed dental X-ray film, exposed 31 days. *A*—indicates area of the *detail* radioautograph. (Pl. 001494.) Fig. 2 shows a *detail* radioautograph, tissue section mounted on Eastman NTA plate, exposed 7 days. *A*—Alpha track (tissue end in focus); *B*—zone of vesicular cartilage cells (tissue plane out of focus); *C*—zone of eroded cartilage cells; *D*—primary spongiosa. (Plate 063; stained with hematoxylin eosin; Bausch and Lomb Ampliplan 6, Zeiss oil immersion apochromatic 60; extension of camera 50 cm.) Fig. 3 shows localization of plutonium, in the *detail* radioautograph of Fig. 2, marked by placing map tacks at the tissue end of each nuclear track. Note the orderly distribution of plutonium as against the apparent haphazard arrangement of tracks seen in Fig. 2. (Pl. 001496.)

nated by focusing up and down with the fine adjustment of the microscope at high magnification. For purposes of orientation the points of origin may be marked on an enlarged photomicrograph with map tacks. As marker after marker is placed at the beginning of the tracks the pattern of localization of the radioactive substance can be visualized. The orderly localization of plutonium in the region of the zone of eroded cartilage cells of the tibial epiphysis is shown in Fig. 3.

*Outlines of survey and detail* radioautographic techniques are given in Table 1. The necessity for the use of pressure to insure intimate contact between tissue and emulsion for *survey* radioautographs has been demonstrated by Sherwood's studies (*20*) of microradiographic technique. Nitrocellulose sections are mounted on nuclear track plates by carrying a flat section wet with approximately 75 percent alcohol on a section lifter to the emulsion. The water and alcohol render the gelatin surface sticky enough to hold the section securely throughout processing. The section is quickly covered with filter paper (Whatman's No. 1) and the excess alcohol is taken up by moving a tissue roller back and forth over the plate. The nitrocellulose is immediately removed by repeated flooding with ether-alcohol.

*Selection of the emulsion* is greatly simplified by an understanding of the functions of the survey and detail radioautographs. For survey radioautographs large grain size is not objectionable. Even the shortest tracks, such as those from an alpha particle, may

TABLE 1

OUTLINE OF RADIOAUTOGRAPHIC TECHNIQUE

| | *Survey* radioautograph | *Detail* radioautograph |
|---|---|---|
| | *Contact* | |
| *Mount:* | *Mount* tissue on microscope slide (stain after radioautographic exposure). *Place* film against tissue. *Wrap* appropriately to prevent scatter to next film. *Clamp* between glass slides with two 7-lb paper clamps. *Expose* in box, painted black inside and out (*5*). | *Mount tissue on emulsion.* |
| *Emulsion:* | *Fast X-ray film.* | Alphagraph: Alpha or beta sensitive *nuclear track emulsion.* Betagraph: Beta sensitive *nuclear track emulsion.* |
| *Exposure:* | *Estimate* exposure time from experience with comparable material. | Approximately ⅛ exposure time of survey radioautograph. |

radiate a distance of 20 μ. Thus, the total width of the radioautographic image of a point source of alpha particles may be as great as 40 μ (*4*). For beta emitters the images will be much wider (*4, 16*). Therefore, since the fastest emulsions have a grain size sufficiently small to show all the detail possible by the contact radioautographic technique, the selection of an emulsion is determined by the emulsion speed desired. Fast X-ray films offer the advantage of the shortest possible exposure time for survey radioautographs. *Processing* of the emulsions must be carried out according to the manufacturers' recommendations.

In summary, radioautographic examination of thin tissue sections is accomplished by two complementary procedures. Survey radioautographs may be obtained by the well-known method of pressing the tissue, mounted on a glass microscope slide, in contact with a fast X-ray emulsion. Detail radioautographs for examination at high magnification under high dry and oil immersion objectives may be obtained by recently announced methods in which the section is *mounted directly on a nuclear track emulsion.*

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