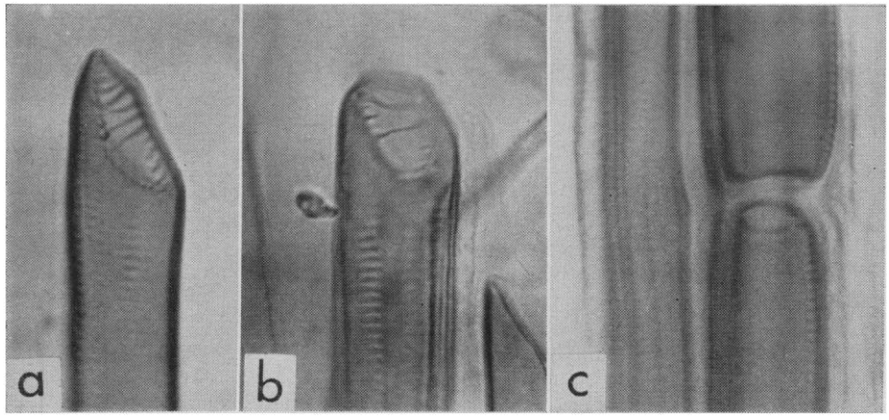

Fig. 1. Vessel members of *Marsilea quadrifolia*. Vessel members wtih scalariform-perforated end walls (*a, b*). The articulation of two vessel members with transverse, simple-perforated end walls (*c*).

are, indeed, sieve-tube members, their occurrence in *Marsilea* will be, as far as is known, unique within the Filicales and the only known occurrence outside of the angiosperms.

On the basis of frond form, *Marsilea* is generally regarded as the most primitive genus of the Marsileaceae. On the basis of sporangial type, however, *Marsilea* is evidently the most specialized (*5*). The evidence from the vascular tissue tends to support the latter.

The vessels of the bracken fern, *Pteridium*, are very different from those of *Marsilea*. They are located in the rhizome, petiole, and root and are not confined solely to the roots, as in *Marsilea*. Furthermore, the vessels of *Pteridium* possess members with only the scalariform type of perforation plate. Pits on the lateral walls are typically scalariform, whereas those of *Marsilea*, although scalariform, are relatively smaller and more widely spaced. In addition, vessel members in *Pteridium* are much shorter than those of *Marsilea*.

Both *Marsilea* and *Pteridium* are members of families considered to be taxonomically advanced. The heterosporous family Marsileaceae is generally considered more highly advanced than the Pteridaceae. The vessel members of *Marsilea*, on the basis of characters used to determine degree of specialization in angiosperms, are, with the exception of their length, more highly specialized than those in *Pteridium*.

The discovery of vessel members in *Marsilea* demonstrates once again the homoplastic development of these structures in taxonomically widely separated plants. In view of their occurrence in these distantly related genera, and the differences in morphology and location in the plant, it is probable that the vessels have originated independently at least twice in the ferns—in the terrestrial *Pteridium* and in the aquatic *Marsilea*. This parallels the situation that seems to have occurred in the angiosperms (*6*).

Histological and developmental studies will supply detailed information concerning the nature and ontogeny of the vessel and supposed sieve-tube elements in *Marsilea*. A statistical study of the tracheary elements of the ferns in progress will provide a broader basis for a more critical interpretation of the significance of these cell types in the Filicineae (*7*).

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X-rays Affect the Incorporation of 5-Iododeoxyuridine into Deoxyribonucleic Acid

Abstract. When labeled with iodine-131, 5-iododeoxyuridine, an analogue of thymidine, is useful in estimating the effect of x-radiation on deoxyribonucleic acid metabolism. Although this compound is readily incorporated into deoxyribonucleic acid in the absence of ionizing radiation, we find that whole-body exposure to as little as 10 r will significantly inhibit its incorporation.

The debilitating effects of x-rays to higher animals are usually the delayed consequences of the inhibition of multiplication or death of cells in certain normally proliferative tissues. Tracer studies in turn suggest that the damage to proliferating cells can be correlated with altered metabolism of their genetic material, deoxyribonucleic acid (DNA). Tritiated-thymidine, while an excellent measure of DNA metabolism (*1*), is difficult to assay because of the weak energy of tritium's β-radiation; whereas an iodinated analogue would permit easy assay of whole tissues or animals by the gamma rays of radioiodine. Therefore, the present technique was developed, utilizing the thymidine analogue, iododeoxyuridine (*2*), which can be incorporated in place of thymidine into DNA (*3*). For this purpose the iododeoxyuridine was prepared labeled with I^{131} by iodination of deoxyuridine as described by Prusoff (*2*) [but with the addition of radioactive iodine (I^{131}) to the reaction mixture] and purified by four recrystallizations from water.

Groups of Swiss albino mice, 6 to 10 weeks old, were exposed to 250 kv (peak) x-rays filtered through 0.5 mm of copper and 1 mm of aluminum. The rate of irradiation was 50 to 52 r/min, different doses being attained by varying the duration of exposure. At a variable time interval before the onset or after the cessation of irradiation, each mouse received a single intraperitoneal injection of 0.1 μmole of the labeled compound having a specific activity of approximately 10 μc/μmole; the controls were unirradiated litter mates, which had been similarly injected. All mice were given 0.1 percent NaI in their drinking water for at least 24 hours before injection with labeled iododeoxyuridine to prevent retention in the thyroid of radio-iodide, the major catabolic product.

The radioactivity of each mouse was counted periodically in a large well-type crystal scintillator. Figure 1 illustrates the average radioactivity retained (plotted logarithmically) by groups of three to five mice as a function of time after injection. In both irradiated and unirradiated animals the initial rapid disappearance of radioactivity was followed by a slow phase (from about the 10th to the 40th hour) in which the retained amount of activity remained essentially constant and can be attributed to label in metabolically stable DNA within new cells. The more rapid loss of radioactivity after 40 hours undoubtedly reflects the death of labeled cells. Chemical data in support of this interpretation are as follows.

At intervals, two or three mice from a group were killed, and their organs were either assayed for radioactivity directly or for activity insoluble in 10 percent trichloroacetic acid after homogenization, precipitation with this reagent, and washing until the supernatant contained less than 2 percent of the radioactivity of the precipitated

Table 1. Incorporation of I^{131}-iododeoxyuridine into DNA after irradiation. The figures represent percentages relative to incorporation into unirradiated controls at 20 hours after injection of the iododeoxyuridine (three to five mice per tabulated number).

Exposure (r)	Incorporation at intervals between x-radiation and injection (%)			
	2 hr	6 hr	24 hr	48 hr
10	110	68		
25	52	39		
52	37	29	102	
104	24	23	55	
208	20	23	43	
312	18		39	
416	12		26	
520	13		22	
624	12		20	62

tissue. This precipitate contains the DNA together with most of the tissue proteins. Analysis of various organs revealed that the amount of acid-insoluble radioactivity reached a maximum within an hour after injection of labeled iododeoxyuridine. The acid-soluble radioactivity after this period was primarily due to the presence of iodide, resulting from degradation of the labeled compound. By 20 hours, almost all the body radioactivity was insoluble in trichloracetic acid. Of the radioactivity lost from the body, over 95 percent was excreted in the urine, largely as iodide.

The unirradiated animal, 21 hours after the injection of labeled iododeoxyuridine, retained an average of 8.3 percent of the injected dose. The retained activity was proportioned 41 percent to the gastrointestinal tract (none of this was in its contents), 21 percent to the skin (about half of this represented contaminating iodide), and 15 percent to the bones (including bone marrow). Only 2.7, 2.3, 1.7, and 1.1 percent were present in the skeletal muscles, liver, thymus, and spleen, respectively. While the rates at which radioactivity

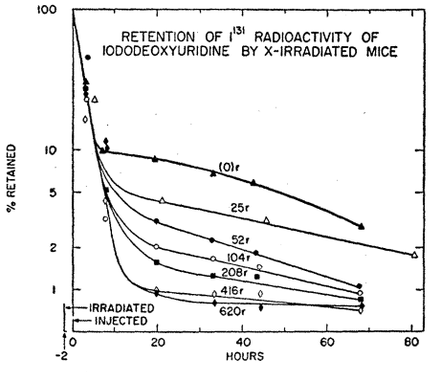

Fig. 1. Disappearance of total radioactivity in mice after injection of I^{131} iododeoxyuridine. Tracer was given 2 hours after x-radiation. "O" dose curve shows unirradiated controls. Three to five mice per point.

disappeared from the various organs were quite different, the preponderance of radioactivity in the gut and the steepness of the decline from this compartment after 40 hours largely account for the shape of the curve for whole-body retention.

Since the extent of incorporation of labeled iododeoxyuridine in the intact mouse could not be determined until after the degradation products had been eliminated, the amount of radioactivity present at 20 hours was chosen as a measure of incorporation. Table 1 shows that the incorporation was a function of the irradiation dose and of the time between irradiation and injection. Maximum inhibition of iododeoxyuridine retention for any given x-ray dose occurred approximately 6 hours after irradiation and was already close to maximum 2 hours after irradiation. Recovery, in that iododeoxyuridine retention returned to normal, began within 24 hours after administration of 52 r, but was not yet complete even 48 hours after administration of 624 r.

The inhibition produced by the larger doses is even greater than the graph indicates, for an iodide retention curve would approximate that for labeled iododeoxyuridine after administration of 624 r, and in fact, over 80 percent of the radioactivity "incorporated" when the labeled compound was given 2 to 24 hours after 624 r of radiation was found in the skin, largely as iodide (*4*). The greatest inhibition of incorporation was suffered by the gastrointestinal tract, and the greatest recovery, 48 hours after irradiation with 624 r, also occurred in this organ, little or no incorporation then being observed in the spleen, lymph nodes, or thymus. These results parallel those recently reported by Nygaard and Potter (*5*) for the effect of x-rays on the incorporation of thymidine-2-C^{14} into the DNA of rat intestine, thymus, and spleen.

Irradiation given 2 hours *after* the injection of I^{131}-iododeoxyuridine produced a small depression in the curve for retained radioactivity, and irradiation given 24 hours after injection produced barely detectable changes in the shape of the retention curve, indicating that early death of labeled cells can account for only a fraction of the inhibition observed. The maximum inhibition observed 6 hours postirradiation (Table 1) may result from inhibition of mitosis and the resultant depletion of the proliferating pool, since this interval is close to the generation time for the rapidly proliferating cells of the gut. However, inhibitions observed at earlier times (Fig. 1) obviously cannot be so explained and must therefore represent either a slowing of DNA synthesis within cells or a less efficient utilization of label because of altered cell permeability or increased pool size of DNA precursors.

In any event, the sensitivity of iododeoxyuridine incorporation in mice to as little as 10 r of precedent x-radiation suggests the possibility of developing this technique as an index of radiation injury in man after either accidental or therapeutic exposure. Of course, the possible mutagenic effects of labeled iododeoxyuridine, either from the compound itself or from its radiation, must be considered in any such study (*6*).

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Fallout Radioactivity in Cattle and Its Effects

Abstract. The levels of strontium-90 and cesium-137 in cattle grazed on the Nevada Test Site and elsewhere in Nevada are similar to those in cattle from other parts of the country. Gastrointestinal absorption of the relatively large amounts of radioactive cerium-praseodymium, ruthenium-rhodium, and zirconium-niobium present in the rumina is very small. Zinc-65 made its first appearance in samples of muscle and liver in November 1958 and has persisted in later samplings. There has been no evidence of biological damage to date, either histologically or grossly.

In 1957, the U.S. Atomic Energy Commission authorized a project (*1*) to determine what effects, if any, the radioactivity produced in atomic bomb tests at the Nevada Test Site was having on cattle grazing in areas heavily contaminated by fallout as contrasted