Administration of 5-Iododeoxyuridine-I\(^{131}\) in the Mouse and Rat*  

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SUMMARY

5-Iododeoxyuridine-I\(^{131}\) was administered to normal as well as tumor-bearing rats and mice, and the effect of several variables on the disposition of the administered compound and on the I\(^{131}\) content of selected tissues was studied. Incorporation of the iodouracil moiety into the DNA of tumor and intestine was demonstrated.

Simultaneous administration of a large excess of 5-iodouridine, together with the 5-iododeoxyuridine-I\(^{131}\), did not significantly affect the I\(^{131}\) content of tumor and intestine. Administration of the labeled compound intraperitoneally as an oil emulsion rather than as an aqueous solution appreciably increased the incorporation of the iodouracil moiety into the DNA of tumor tissue. Administration of 5-iododeoxyuridine-5'-phosphate did not appreciably alter the incorporation of the iodouracil moiety as compared with an equimolar dose of 5-iododeoxyuridine.

Recent studies by Prusoff, Welch, and their colleagues have shown that 5-iododeoxyuridine (5-iodo-2'-deoxyuridine) possesses antitumor activity (15, 23, 40), acts as a competitive antagonist of thymidine utilization (15, 22, 25, 26), and inhibits the utilization of pyrimidine precursors for the synthesis of DNA thymine (26, 27). The incorporation of the iodouracil moiety into the DNA of mammalian cells in culture as well as in the whole organism has been described (3, 4, 7, 8, 22, 23). The pharmacology of iododeoxyuridine in animals and man has been studied by Jaffe et al., Prusoff et al., and Welch et al. (15, 29, 39).

This paper presents the results of several experiments in which 5-iododeoxyuridine labeled with I\(^{131}\) was administered to normal as well as to tumor-bearing rats and mice. The effect of several variables on the disposition of the administered compound and on the I\(^{131}\) content of the tumor tissue was studied.

MATERIALS AND METHODS

Preparation of compounds.—5-Iododeoxyuridine labeled with I\(^{131}\) was prepared by a procedure previously reported (3). 5-Iodouridine was prepared following the procedure of Prusoff et al. (28). 5-Iododeoxyuridine-5'-phosphate was synthesized by a procedure to be published elsewhere. Deoxyribonuclease was purchased from Worthington Biochemical Company; Russell Viper Venom, from Wellcome Research Labs.; purified phosphodiesterase from snake venom was kindly supplied by J. Salsel of this Institute.

Radioactivity measurements.—I\(^{131}\) activity was measured with a well-type scintillation crystal counter. Counting of paper strips was carried out in a specially designed crystal scintillation counter connected with a recording rate meter. In the well-type scintillation crystal counter 1 \(\mu\)c. \((2.2 \times 10^6\) disintegrations/min\) of I\(^{131}\) furnished 1.0 \(\times 10^6\) counts/min.

Administration.—In most experiments the 5-iododeoxyuridine was injected intraperitoneally in physiological saline. Unless otherwise specified in the experiments described below this route of administration was used, and the stated dose was administered in one injection. In some experiments the medium was 16 per cent gelatin in saline, and in others an emulsion was used which contained saline, mineral oil, and Tween 80 in the proportions 10:10:1. This emulsion was prepared by dissolving the 5-iododeoxyuridine in saline and
mixing this solution with the other components in a blender. The emulsion was kept at refrigerator temperature and prior to use the liquid was taken up and blown out of a syringe repeatedly.

The animals included Swiss mice and Wistar rats. Tumor-bearing animals included Wistar rats bearing Murphy-Sturm lymphosarcoma and tumors of human origin, H.Ep. #1 and H.Ep. #3 (37, 38), as well as a human lung tumor referred to in this paper as A-4 (carried in routine transplantation in the laboratory of H. W. Toolan at this Institute, and obtained from J. V. Skiff, Jr., Albany Medical College [34]). Swiss mice bearing intramuscular H.Ep. #3 tumors were used in several experiments.

Chemical procedures.—The DNA fraction was isolated following a modification of the Schmidt-Thannhauser procedure (32, 33). The ribonucleic acid was hydrolyzed with NaOH at room temperature for 24 hours. The I\textsuperscript{131} content of the DNA was not appreciably reduced by this treatment, since the supernatant following DNA precipitation contained only 1–2 per cent of the total counts. Under the conditions of acid precipitation of DNA at 0°C, at least 95 per cent of the 5-iododeoxyuridine-I\textsuperscript{131} is unaltered. Colorimetric determination of DNA was performed by the method of Keck (17). Paper chromatography in butanol-acetic acid-water (5:2:3) by a descending technic, with Whatman #1 paper, gave the following R\textsubscript{f} values: 5-iododeoxyuridylic acid, 0.41; 5-iododeoxyuridine, 0.75; 5-iodouracil, 0.75; potassium iodide, 0.41; deoxyuridylic acid, 0.30.

To determine whether the I\textsuperscript{131} activity of the DNA fraction was due exclusively to the iodouracil moiety incorporated therein, the DNA preparation was hydrolyzed with deoxyribonuclease (DNase), and then separate aliquots of this digest were treated with phosphodiesterase and with crude snake venom, respectively, following the procedures of Dunn and Smith (5). After the initial treatment with DNase all the I\textsuperscript{131} activity of this solution moved slightly from the origin, corresponding to the region where oligonucleotides would be located. The product of this reaction was further hydrolyzed with phosphodiesterase from snake venom or simply with crude snake venom.

In the first case all the I\textsuperscript{131} activity was present in the region corresponding to the marker spot for 5-iododeoxyuridine-5'-phosphate. In the second case all the I\textsuperscript{131} activity was at the spot corresponding to 5-iododeoxyuridine. This I\textsuperscript{131} activity was rechromatographed in the ethyl acetate-phosphate buffer system (24) that separated 5-iododeoxyuridine from 5-iodouracil. The I\textsuperscript{131} activity moved with the marker spot for iododeoxyuridine.

RESULTS

Radioiodine Activities Following Administration of 5-Iododeoxyuridine-I\textsuperscript{131}

Rats bearing H.Ep. #3 tumors were given injections intraperitoneally of 5 mg. 5-iododeoxyuridine-I\textsuperscript{131} (50 X 10\textsuperscript{6} counts/min). After the injection, the animals were sacrificed after each of the periods indicated in Chart 1. An aliquot of finely minced and pooled tissue was counted with the well-type scintillation crystal counter. The I\textsuperscript{131} content of the wet tissue is expressed in terms of per cent of administered I\textsuperscript{131} present per gram of wet tissue.

![Chart 1: Radioiodine content of wet tissue following administration of 5-iododeoxyuridine-I\textsuperscript{131}. Rats bearing H.Ep. #3 tumors were given injections of 5.0 mg. 5-iododeoxyuridine-I\textsuperscript{131} (corresponding to 50X10\textsuperscript{6} counts/min) 9 days after tumor implantation. Groups of three animals were sacrificed at the indicated periods after the injection. The ordinate refers to the percentage of the administered I\textsuperscript{131} present per gram of wet tissue.](image-url)

In the case of the tumor tissue per gram was higher than that for intestine, liver, and blood after 4 days and also underwent the smallest percentage decrease over this period.

To follow the disposition of 5-iododeoxyuridine-I\textsuperscript{131} immediately following administration, this compound was administered to a 300-gm. rat through the jugular vein, and blood was collected via the carotid artery. Urine was collected directly...
from the bladder with a catheter at different intervals of time and analyzed by paper chromatography. The blood was extracted with 10 volumes of butanol, the butanol evaporated, and the residue subjected to paper chromatography. The I\textsuperscript{131} activity in the areas corresponding to iodide ion and 5-iododeoxyuridine plus 5-iodouracil was determined. The 5-iododeoxyuridine and 5-iodouracil have similar R\textsubscript{f} values in the solvent system used, and the I\textsuperscript{131} activity for each compound was not measured. Although the paper strip had other radioactive areas, these were not further investigated. The distribution coefficient for iodide ion (concentration in butanol layer/concentration in aqueous layer) of 0.34 was used together with the relative volumes of the two phases in applying a correction factor to the I\textsuperscript{131} activity as iodide.

**ADDITION OF EXCESS IODIDE**

The effect of administering a large excess of potassium iodide together with the 5-iododeoxyuridine-I\textsuperscript{131} on the I\textsuperscript{131} content of thyroid tissue is shown by the results in Table 2. The rapid dehalogenation to iodide ion-I\textsuperscript{131} was reflected by the high specific activity of thyroid tissue in the

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**TABLE 1**

<table>
<thead>
<tr>
<th>TIME AFTER INJECTION (MIN.)</th>
<th>PER CENT OF ADMINISTERED I\textsuperscript{131}</th>
<th>COUNTS/MIN/ML BLOOD</th>
<th>I\textsuperscript{131} AS IUDR+IU</th>
<th>COUNTS/ML BLOOD</th>
<th>I\textsuperscript{131} AS IUDR+IU</th>
<th>COUNTS/ML URINE</th>
<th>I\textsuperscript{131} AS IUDR+IU</th>
</tr>
</thead>
<tbody>
<tr>
<td>5</td>
<td>1.50</td>
<td>79</td>
<td>1150</td>
<td>44,000</td>
<td>14</td>
<td>0.17</td>
<td>0.09</td>
</tr>
<tr>
<td>15</td>
<td>1.20</td>
<td>1880</td>
<td>325</td>
<td>71,000</td>
<td>0.110</td>
<td>0.001</td>
<td></td>
</tr>
<tr>
<td>30</td>
<td>1.10</td>
<td>970</td>
<td>32</td>
<td>79,000</td>
<td>0.033</td>
<td></td>
<td></td>
</tr>
<tr>
<td>60</td>
<td>0.90</td>
<td>1180</td>
<td>124</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>120</td>
<td></td>
<td>970</td>
<td>32</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Abbreviations: IUDR—5-iododeoxyuridine; IU—5-iodouracil.

A 300-gm. rat was given an injection through the jugular vein of 0.046 mg. 5-iododeoxyuridine-I\textsuperscript{131} corresponding to $5.2 \times 10^6$ counts/min, and blood was collected via the carotid artery. Urine was collected directly from the bladder with a catheter at different intervals of time.

The corresponding distribution coefficients for 5-iododeoxyuridine and 5-iodouracil were 1.18 and 2.12, respectively.

The results are listed in Table 1. It is evident from these data that, although the I\textsuperscript{131} content per ml. of blood varies only from 1.5 to 1.0 per cent of the administered dose in the 5- to 120-minute interval, degradation to the form of iodide ion-I\textsuperscript{131} was rapid and almost complete. The urine sampling also demonstrated the rapid dehalogenation. In a sample obtained 30 minutes after administration, I\textsuperscript{131} present as iodide was 10 times greater than the I\textsuperscript{131} present as 5-iododeoxyuridine plus iodouracil. One hour afterwards almost all the radioactivity of the sample was present as iodide ion.

The thyroid gland secretes a mixture of di-, tri-, and tetra-iodinated amino acids, including thyroxine and 3,3'-diodothyronine. After the administration of KI-I\textsuperscript{131}, those compounds are labeled in the body fluids (30). It was necessary to investigate their R\textsubscript{f} values in the chromatographic system used to separate iododeoxyuridine and iodouracil from iodide ion, since in these experiments the thyroid hormones could be labeled by the iodide ion split off from the iododeoxyuridine-I\textsuperscript{131}. Thyroxine (R\textsubscript{f} 0.98) and 3,3'-diodothyronine (R\textsubscript{f} 0.84) were well separated from the compounds under study.

**INCORPORATION OF THE IODOURACIL MOIETY INTO DNA**

Groups of three rats bearing A-43 tumors were given injections of 5-iododeoxyuridine-I\textsuperscript{131}. The I\textsuperscript{131} content of the dry defatted tissue and the DNA fraction was determined. The DNA was hydrolyzed with deoxyribonuclease and phosphodiesterase as described above in the "Methods" section in order to demonstrate directly that the I\textsuperscript{131} was present in the iodouracil moiety. The results are given in Table 3. Approximately 60 per cent of the I\textsuperscript{131} content of the dry tumor and intestinal tissue was present in the DNA as the
iodouracil moiety. Of the total I$_{131}$ administered to the animal, 1.4 per cent was present in DNA of the tumor.

To determine the I$_{131}$ content of tumor tissue when the I$_{131}$ is administered in the form of potassium iodide rather than iododeoxyuridine, an experiment was performed with two rats bearing 7-day H.Ep. #3 tumor transplants. Carrier-free potassium iodide-I$_{131}$, corresponding to 10$^7$ counts/min was injected, and the animals were sacrificed 2 days later. The percentage of the administered I$_{131}$ per gram of wet tumor tissue was only 0.004 for each animal, whereas the percentage in the thyroid gland and immediately adjacent tissue was 0.78 and 0.52.

**Effect of Varying the Dose of 5-Iododeoxyuridine-I$_{131}$**

To determine whether the injected iododeoxyuridine-I$_{131}$ corresponded to a "tracer dose," an experiment was carried out in which the quantity of injected compound was increased by a factor of 10. Four rats bearing H.Ep. #3 tumor transplants (7 days) were given five injections on the 7th day at 2-hour intervals, five additional injections on the 8th day, and the animals were sacrificed on the 9th day. Two rats received a total dose of 0.5 mg. 5-iododeoxyuridine-I$_{131}$ (5 $\times$ 10$^6$ counts/min), while two received 5 mg. (50 $\times$ 10$^6$ counts/min). The percentage of the total I$_{131}$ administered that was found per gram of dry tumor tissue was 1.0 in each case. These results demonstrate that the amount of compound administered (over a tenfold range) did not affect the percentage of the administered I$_{131}$ present in the tumor tissue at the time of sacrifice.

**Attempts to Alter the Disposition of Administered 5-Iododeoxyuridine-I$_{131}$**

*Simultaneous administration of 5-iodouridine.*—To explore the effect of simultaneous administration of a compound closely related in structure to iododeoxyuridine, the unlabeled ribose-containing analog, 5-iodouridine, was synthesized and used in the experiment described in Table 4. Administration of a 60-fold molar excess of the ribose-containing analog did not appreciably affect the I$_{131}$-content of tumor and intestine. The I$_{131}$-content of liver tissue increased by a fivefold factor when 25 mg. 5-iodouridine was administered.

**Table 2**

<table>
<thead>
<tr>
<th>Compounds Administered</th>
<th>Per cent of administered I$_{131}$ per gm. wet tissue</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Thyroid plus Trachea</td>
</tr>
<tr>
<td>5-iododeoxyuridine-I$_{131}$</td>
<td>0.061</td>
</tr>
<tr>
<td>5-iododeoxyuridine-I$_{131}$ plus potassium iodide</td>
<td>0.024</td>
</tr>
</tbody>
</table>

Two rats bearing H.Ep. #3 tumors were given intravenous injections, 7 days after tumor transplantation, of 0.30 mg. (0.85 $\mu$moles) 5-iododeoxyuridine-I$_{131}$, corresponding to 7.5 $\times$ 10$^6$ counts/min. In addition, one rat received 4.7 mg. (28 $\mu$moles) of potassium iodide in three injections 2 hours before administration of 5-iododeoxyuridine-I$_{131}$, at the time of administration, and 24 hours later. The animals were sacrificed 48 hours after administration of 5-iododeoxyuridine-I$_{131}$.

**Table 3**

<table>
<thead>
<tr>
<th>Tissue</th>
<th>Per cent I$_{131}$ per gm. dry tissue</th>
<th>Per cent of administered I$_{131}$ incorporated into DNA per gm. wet tissue</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tumor</td>
<td>1.7</td>
<td>1.0</td>
</tr>
<tr>
<td>Intestine</td>
<td>0.85</td>
<td>0.88</td>
</tr>
<tr>
<td>Liver</td>
<td>0.03</td>
<td></td>
</tr>
</tbody>
</table>

Three rats bearing A-42 tumors were given one injection, 8 days after tumor transplantation, of 7.1 mg. 5-iododeoxyuridine, corresponding to 2.9 $\times$ 10$^7$ counts/min, and sacrificed 2 days later; 10 mg. potassium iodide was injected 5 minutes before the 5-iododeoxyuridine-I$_{131}$ was injected.
proximately 3 the I\(^{131}\) content of the wet tumor tissue, as well as the incorporation of the iodouracil moiety into the DNA of this tissue.

**Amethopterin.**—The effect of amethopterin, a drug known to interfere with nucleic acid metabolism (1, 21), was studied in the following experiment: Two groups of four mice bearing intramuscular H.Ep. \#3 tumors were used. The control group received three injections over a period of 1 week, totaling 0.8 mg. (2.6 \(\times 10^6\) counts/min)

**TABLE 4**

**EFFECT OF 5-IODOURIDINE ON I\(^{131}\) CONTENT OF WET TISSUE AFTER ADMINISTRATION OF 5-IODODEOXYURIDINE-I\(^{131}\) TO SWISS MICE BEARING H.Ep. \#3 TUMORS**

<table>
<thead>
<tr>
<th>5-IODOURIDINE (mg.)</th>
<th>Per cent of administered I(^{131}) per gm. wet tissue</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tumor</td>
<td>Intestine</td>
</tr>
<tr>
<td>0</td>
<td>0.00%</td>
</tr>
<tr>
<td>0.04</td>
<td>0.00%</td>
</tr>
<tr>
<td>0.04</td>
<td>0.01%</td>
</tr>
</tbody>
</table>

Ten Swiss mice weighing approximately 25 g.m. (in groups of 4, 8, and 8) were given injections of 0.40 mg. 5-iododeoxyuridine-I\(^{131}\), corresponding to 6 \(\times 10^6\) counts/min, 10 mg. potassium iodide, and 5-iodouridine as stated in column one. Injections were started 1 day after tumor transplantation. Two injections were given on the 7th day, two more on the 8th day, and the animals were sacrificed on the 9th day.

**TABLE 5**

**EFFECT OF MODE OF ADMINISTRATION ON THE I\(^{131}\) ACTIVITY FOLLOWING INJECTION OF 5-IODODEOXYURIDINE-I\(^{131}\)**

<table>
<thead>
<tr>
<th>Vehicle</th>
<th>Counts/min/gm wet tumor tissue</th>
<th>Counts/min/mg tumor DNA</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.9% NaCl</td>
<td>3.0 (\times 10^4)</td>
<td>4.9 (\times 10^4)</td>
</tr>
<tr>
<td>18% Gelatin</td>
<td>4.5 (\times 10^4)</td>
<td>1.4 (\times 10^4)</td>
</tr>
</tbody>
</table>

Groups of three mice each bearing intramuscular H.Ep. \#3 tumors were given injections intraperitoneally of 5-iododeoxyuridine-I\(^{131}\) on the 7th day after tumor implantation and an equal dose on the 8th day. Animals were sacrificed 24 hours later. The total dose administered to each animal was 0.47 mg., corresponding to 9.1 \(\times 10^6\) counts/min.

5-iododeoxyuridine-I\(^{131}\) and 0.75 mg. potassium iodide in the oil emulsion. In addition to this, the other group received a daily injection of 0.03 mg. amethopterin in saline. The injections were started 1 day after tumor implantation and were terminated 2 days before sacrificing the animals. The I\(^{131}\) content of the pooled tumor tissue was approximately 3 times greater for the group receiving the amethopterin—3.1 per cent of the injected dose per gm. of wet tissue for the control group and 8.8 per cent for the amethopterin-treated group.

**Comparison of 5-Iododeoxyuridine and Iododeoxyuridine-5'-Phosphate**

The effect of using the phosphorylated derivative of 5-iododeoxyuridine was studied in the experiment described in Table 6. An equinomol dose of each compound was used. The DNA fraction was hydrolyzed enzymatically as described above. It is seen from Table 5 that the magnitude of incorporation of the iodouracil moiety into DNA was not significantly different for the tumor tissue and was slightly smaller for the phosphorylated derivative in the case of intestine under these conditions.

**Estimation of Extent of Incorporation of Iodouracil Moiety into DNA**

The results in Tables 5 and 6 are expressed in terms of the directly measured counts/min/mg DNA.

**TABLE 6**

**COMPARISON OF INCORPORATION OF IODOURACIL MOIETY INTO DNA FOLLOWING ADMINISTRATION OF 5-IODODEOXYURIDINE-I\(^{131}\) AND 5-IODODEOXYURIDINE-5'-PHOSPHATE-I\(^{131}\)**

<table>
<thead>
<tr>
<th>Tissue</th>
<th>Counts/min/mg DNA</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tumor</td>
<td>6.8 (\times 10^3)</td>
</tr>
<tr>
<td>Intestine</td>
<td>5.95 (\times 10^3)</td>
</tr>
</tbody>
</table>

Ten mg. potassium iodide per animal was administered intraperitoneally to two groups of three rats each, bearing 8-day transplants of A-42 tumors. Five minutes later 0.85 mg. 5-iododeoxyuridine-I\(^{131}\) corresponding to 2.0 \(\times 10^7\) counts/min was administered to one group of rats. The other group of rats received an equimolar quantity of 5-iododeoxyuridine-5'-phosphate-I\(^{131}\) (IUDRP) corresponding to 2.0 \(\times 10^7\) counts/min. The animals were sacrificed 2 days after the injection.

DNA. The order of magnitude of the molar ratio of iodouracil to iodouracil plus thymine, IU/ (IU + T), may be estimated by converting the counts/min to moles of iododeoxyuridine and mg. DNA to moles of iodouracil plus thymine. In making this calculation it was assumed that the DNA had a phosphorus content of 10 per cent and that the mole fraction of the thymine moiety in the DNA purine and pyrimidine bases was 0.30 (2). By these assumptions IU/ (IU + T) is calculated to have been 0.7 and 2 \(\times 10^{-3}\) in the experiments described in Table 5 with physiological saline and oil emulsion as vehicles, respectively. In the experiments described in Table 6, IU/ (IU + T) is estimated to have been 0.9 and 0.8 \(\times\)
10⁻³ for tumor and intestine, respectively (column 2).

Observations Related to Toxicity

A systematic effort to ascertain toxicity levels for 5-iododeoxyuridine was not made in the course of the above experiments. However, the following observations were noted: The average weight of a group of five mice (30 gm., 2 months old) which received daily doses of 5-iododeoxyuridine at a 30 mg/kg level for 8 days was not appreciably different from that of a control group. To test the effect of administering a larger dose of I¹³¹₁₀ mg, 5-iododeoxyuridine-I¹³¹ corresponding to 1 × 10⁸ counts/min was administered in one injection to each mouse in a group of ten (average weight, 25 gm.). No significant weight difference relative to controls was observed in a 4-week period. A histological study of different organs after 1 month did not reveal any lesions produced by radiation.

Discussion

The results of the DNA analyses in Tables 3, 5, and 6 demonstrate the incorporation of the iodouracil moiety into DNA in mammalian cells when 5-iododeoxyuridine is administered and are in accord with the results of other investigations (3, 4, 7, 8, 22, 25). The extent of this incorporation in vivo will depend, among other factors, on the rate of DNA synthesis and degradation. The small percentage decrease in I¹³¹ content of tumor tissue in Chart 1 may reflect a larger incorporation of the I¹³¹ into a component turning over relatively slowly, such as DNA (9). The relative activities after 1 day for tumor, intestine, and liver approximately parallel the results of Prusoff et al. in mice after 21 hours (29). Krueger et al. have reported that the I¹³¹ content of several organs 24 hours following injection of 5-iododeoxyuridine-I¹³¹ in the mouse corresponded to their proliferative activity (18).

The antitumor activity of 5-iododeoxyuridine has been described (15, 23, 40). The mechanism of growth inhibition by this compound has been considered from the point of view of interference with normal DNA synthesis and formation of DNA containing the unnatural base iodouracil (3, 27, 29). The rapid dehalogenation to iodide ion shown by the results of Table 1 demonstrates that the iodouracil moiety is available for nucleotide formation and DNA synthesis for a relatively short time after administration of the 5-iododeoxyuridine. The level at which the de-iodination takes place was not studied in the experiments described above. This rapid dehalogenation is in agreement with the results of Prusoff et al. in the mouse (29). These authors have also shown that cleavage of iododeoxyuridine to iodouracil and subsequent dehalogenation is an important degradative pathway. Methods designed to extend the availability of the iodouracil moiety were therefore explored and preliminary results described above. The results in Table 4 demonstrate that the I¹³¹ content of tumor and intestinal tissue was not affected when a large excess of the ribose-containing analog, 5-iodouridine, was administered together with the 5-iododeoxyuridine-I¹³¹. The enhanced I¹³¹ content of the liver under these conditions is of interest. The distribution of the I¹³¹ among various chemical constituents was not further explored.

The use of the oil emulsion as a vehicle resulted in a threefold increase in the incorporation of the administered compound into the iodouracil moiety of the tumor DNA. This may reflect an increased time period over which the 5-iododeoxyuridine would be available for anabolic metabolism and is of interest for more extensive study.

Several compounds have been found to interfere with the de novo synthesis of the thymine moiety. Among these are 5-fluorodeoxyuridine (6, 14) and amethopterin (13, 26). Such interference has been shown under cell culture conditions to result in increased utilization of the preformed thymine moiety and of structural analogs of thymidine (11, 12). The results of the experiment with amethopterin described above show an increased I¹³¹ content of the tumor tissue. Additional studies will be required to determine the chemical nature of this enhanced I¹³¹ activity.

The studies by Lichtenstein et al. (19) indicated different patterns of utilization for deoxycytidine and its corresponding phosphorylated derivative. The results in Table 6 demonstrate relatively little difference in the utilization of the iodouracil moiety for tumor DNA synthesis when the nucleoside or the corresponding nucleotide is administered. These results are consistent with the results of Liebman and Heidelberger (20) and Roll et al. (31) pointing to the loss of the phosphate moiety following administration of several naturally occurring nucleotides and with experiments in this laboratory showing that dephosphorylation of 5-iododeoxyuridine-5'-phosphate in serum at 37°C is complete in 1 hour.

The possible use of 5-iododeoxyuridine-I¹³¹ as a radiotherapeutic tool remains to be explored (7). The recent studies by Śzybalski and Djordjevic have indicated an enhanced radiosensitivity (x-radiation and ultraviolet) of cells in culture which have incorporated the bromouracil and iodouracil.
moiety into DNA (4, 35, 36). Kaplan and Tomlin and Greer have reported similar results in a bacterial system (10, 16). This approach may be benefited by technics aimed to decrease the rapid dehalogenation and permit enhanced availability of the iodouracil moiety for DNA synthesis in the more rapidly growing tissues.

ACKNOWLEDGMENTS

The authors are indebted to P. C. Merker, K. Sugitara, M. N. Teller, and H. W. Toolan for making available tumor-bearing rats and mice. Several preparations of 5-iododeoxyuridine-1^3^ and the synthesis of 5-iodouridine were carried out by L. Cheong. The synthesis of iododeoxyuridine-5'-phosphate was aided by suggestions from A. Hampton. Histological studies were carried out in the laboratory of S. Sternberg.

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4. more rapidly growing tissues.
5. biological studies were carried out in the laboratory of S. Stern-berg.
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