Tritiated Digoxin

XIV. Enterohepatic Circulation, Absorption, and Excretion Studies in Human Volunteers


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SUMMARY

Metabolic turnover studies performed on six patients with surgically induced biliary fistula given tritiated digoxin reveal virtually all stool excretion of digoxin to be derived from bile. This establishes the relative lack of recycling of digoxin as an important factor in digoxin metabolism. This is probably related to its polar chemical structure and is a determinant of its clinical duration of action in human subjects. The amount of digoxin in the enterohepatic circulation (recycling) was calculated to be 6.5% of the administered dose and absolute absorption to be 85% in human subjects.

Additional Indexing Words: Biliary fistula Digoxin, metabolism of

The availability of $^3$H digoxin for experimental clinical use has made possible a number of meaningful studies during the last decade.\textsuperscript{1-17} These studies have outlined the serum turnover rates, serum half-time (the time required for one half of the radioactivity identified with the drug to disappear from the serum) and serum digoxin levels. The urinary and fecal excretion of digoxin was characterized, and the larger portion of the drug was found to be excreted unchanged in the urine. Comparative studies in patients given $^3$H digoxin by oral, intramuscular, and intravenous routes suggested that digoxin was more completely absorbed than previously supposed, as there were only small differences in stool excretion after different routes of administration.

Patients with surgical T-tube drainage provide a method for determining the contribution of biliary excretion to total stool excretion and provide an index of recycling of the drug in the intestine and its contribution to the metabolism of digoxin.

The purpose of this paper is to report tritiated digoxin turnover and excretion studies on six patients with surgically induced biliary fistula and T-tube drainage and to apply these data to the calculation of the enterohepatic circulation and absorption of tritiated digoxin. Previous studies with tritiated digoxin are utilized in these computations.\textsuperscript{1-13}

Methods

Six male patients with biliary tract disease who were candidates for T-tube drainage postoperatively were selected for study. Clinical diagnoses
Table 1

Clinical Information

<table>
<thead>
<tr>
<th>Case</th>
<th>Age (yr)</th>
<th>Weight</th>
<th>Race</th>
<th>BUN (mg/100 ml)</th>
<th>Dose of digoxin (mg)</th>
<th>Diagnosis</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>40</td>
<td>118</td>
<td>N</td>
<td>15</td>
<td>1.0</td>
<td>Cholelithiasis (preoperative), chronic pancreatitis (postoperative)</td>
</tr>
<tr>
<td>2</td>
<td>42</td>
<td>160</td>
<td>W</td>
<td>14</td>
<td>1.0</td>
<td>Chronic cholecystitis, cholelithiasis</td>
</tr>
<tr>
<td>3</td>
<td>47</td>
<td>159</td>
<td>W</td>
<td>14</td>
<td>1.0</td>
<td>Cholangitis, chronic pancreatitis, hepatitis</td>
</tr>
<tr>
<td>4</td>
<td>49</td>
<td>170</td>
<td>W</td>
<td>12</td>
<td>0.75</td>
<td>Cholelithiasis, muscular dystrophy</td>
</tr>
<tr>
<td>5</td>
<td>66</td>
<td>173</td>
<td>W</td>
<td>14</td>
<td>0.5</td>
<td>Chronic cholecystitis, cholelithiasis</td>
</tr>
<tr>
<td>6</td>
<td>71</td>
<td>146</td>
<td>W</td>
<td>18</td>
<td>1.0</td>
<td>Choledocholithiasis</td>
</tr>
</tbody>
</table>

are shown in table 1. None were in congestive heart failure, nor did they exhibit any cardiac arrhythmia. They were not jaundiced at the time of the surgical procedure. Cholecystectomy, exploration of the common bile duct, or both were performed under general anesthesia, with barbiturate induction and halothane anesthesia supplemented with succinylcholine chloride, agents not known to affect digoxin metabolism. Cyclopropane was used to anesthetize one patient (case 2). A T-tube was placed in the common bile duct and drained externally. None of the bile was returned to the patients. The patients were taken from the operating suite to the recovery area, and about 1 hour following closure of the abdominal incision, when consciousness had returned, each was given 0.5 to 1.0 mg of 3H digoxin intravenously. The 3H digoxin was 99.5% chemically pure by the manufacturer's statement,* and radiochromatographically pure. The specific activity was 125 μc/mg. Serum samples were obtained at 5, 10, 15, 30, 45, 60, and 90 min and 2, 2%, 3, 4, 5, 6, 8, 12, and 24 hours, and daily thereafter for a total of 7 days. All urine, stools, bile, and nasogastric aspirate were saved in aliquots for 7 days. Stool markers were not used because of the nasogastric suction and limited oral intake dictated by the surgical procedure. The radioactivity present in the samples representing digoxin and its major metabolites (digoxigenin mono- and bis-digitoxside and digoxigenin) was extracted by a method of column chromatography previously described. Total recovery of radioactivity was verified by a modification of the Schoniger combution technic.18 Metabolites were searched for by thinlayer chromatography with silica gel plates immersed in a solvent system which consisted of 65.3% cyclohexane, 32.7% acetone, and 2.0% glacial acetic acid.19

These data were analyzed and compared with those from patients receiving 3H digoxin intravenously2, 6 and orally4, 16 to compute equations for enterohepatic circulation, absorption, and excretion of tritiated digoxin. The basic techniques of study did not vary greatly between studies, and the data obtained are felt to be reliable and comparable to those of the current investigation. A total of 54 studies of tritiated digoxin turnover in human beings were analyzed in the computations included in this communication and are annotated in table 2. All the patients except those in the current study were in congestive heart failure and required digoxin for therapy.

Results

The clinical information on each of the patients with biliary fistula is recorded in table 1. None of the patients were in congestive heart failure; however, previous studies10, 16 have shown that there is no alteration in serum levels, turnover, and excretion rates of 3H digoxin between patients with and without

Table 2

<table>
<thead>
<tr>
<th>Tritiated Digoxin Half-Times and Excretion</th>
</tr>
</thead>
<tbody>
<tr>
<td>Oral (20*)</td>
</tr>
<tr>
<td>------------</td>
</tr>
<tr>
<td>Serum T 1/2</td>
</tr>
<tr>
<td>Urine T 1/2</td>
</tr>
<tr>
<td>Stool T 1/2</td>
</tr>
<tr>
<td>Bile T 1/2</td>
</tr>
</tbody>
</table>

Total excretion, 7 days, % of total dose administered

| Bile | – | – | 8.1 |
| Urine | 33.99 | 73.5 | 58.5 |
| Stool | 15.16 | 11.3 | 1.3 |

*Number of patients.
†Data insufficient for calculation.

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### Table 3

**Serum Digoxin Half-Time and Excretion**

<table>
<thead>
<tr>
<th>Case</th>
<th>Serum T1/2 (B) (hr)</th>
<th>Cumulative urine excretion*</th>
<th>Urine T1/2 (days)</th>
<th>Cumulative bile excretion*</th>
<th>Bile T1/2 (days)</th>
<th>Total stool excretion*</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>27.0</td>
<td>30.96</td>
<td>53.28</td>
<td>57.80</td>
<td>59.49</td>
<td>1.0</td>
</tr>
<tr>
<td>2</td>
<td>31.7</td>
<td>39.19</td>
<td>60.63</td>
<td>69.35</td>
<td>72.59</td>
<td>1.2</td>
</tr>
<tr>
<td>3</td>
<td>26.7</td>
<td>30.91</td>
<td>63.78</td>
<td>72.81</td>
<td>79.51</td>
<td>1.4</td>
</tr>
<tr>
<td>4</td>
<td>35.0</td>
<td>24.57</td>
<td>39.64</td>
<td>45.06</td>
<td>48.11</td>
<td>1.7</td>
</tr>
<tr>
<td>5</td>
<td>36.7</td>
<td>33.39</td>
<td>51.38</td>
<td>58.55</td>
<td>61.59</td>
<td>1.4</td>
</tr>
<tr>
<td>6</td>
<td>36.7</td>
<td>10.67</td>
<td>21.66</td>
<td>26.54</td>
<td>29.73</td>
<td>1.4</td>
</tr>
<tr>
<td>Mean</td>
<td>32.3</td>
<td>28.28</td>
<td>48.40</td>
<td>50.02</td>
<td>58.50</td>
<td>1.35</td>
</tr>
</tbody>
</table>

*Expressed as percent of the total dose administered.

†Data inadequate for calculation.

Abbreviations: sd = standard deviation; SEM = standard error of the mean.

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A surgically placed T-tube drain was only rarely completely occludes the common bile duct, and this probably accounts for the variation in cumulative biliary excretion rates. The stool excretion was noted to be 52.7% of the total dose administered in a 7-day study period. The correction was 1.5% similar but slightly reduced, the mean 7-day urinary excretion being 58.2% of the total dose administered. The stool excretion ranged from T/2 to 78% of the total dose administered. The excretion rates for the serum T/2 of digoxin are again noted (range 26.7 to 36.7 hours). The excretion rates for the serum T/2 of digoxin are again noted (range 26.7 to 36.7 hours).
utilized so that line B (the best straight line after the serum plateau is reached) can be extrapolated to zero and a dominant half-time for disappearance of the drug determined. The dominant $T\frac{1}{2}$ in this graph is 32.3 hours. This portion of the serum turnover curve is determined to be a result of metabolism and excretion of the drug and can be eliminated from the early portion of the curve A by subtracting the figure obtained for line B from simultaneous points of curve A above. This maneuver yields another straight line function for which a $T\frac{1}{2}$ can be expressed. This line, designated as line C, represents distribution and binding of digoxin to tissue (metabolism and excretion having been eliminated as factors). Line C has a half-time in these patients of 45 min. These figures resemble

Table 4
Recovery of Digoxin and Metabolites from Bile: Patient 5*

<table>
<thead>
<tr>
<th></th>
<th>Day 2</th>
<th>Day 3</th>
<th>Day 4†</th>
<th>Day 5</th>
<th>Mean</th>
</tr>
</thead>
<tbody>
<tr>
<td>Origin</td>
<td>1.6</td>
<td>BKG†</td>
<td>BKG</td>
<td>BKG</td>
<td>1.6</td>
</tr>
<tr>
<td>Trail</td>
<td>6.2</td>
<td>1.7</td>
<td>1.8</td>
<td>8.0</td>
<td>4.4</td>
</tr>
<tr>
<td>Digoxin</td>
<td>80.3</td>
<td>91.1</td>
<td>66.8</td>
<td>71.9</td>
<td>77.5</td>
</tr>
<tr>
<td>Digoxigenin bis-digitoxiside</td>
<td>7.2</td>
<td>6.2</td>
<td>8.3</td>
<td>6.3</td>
<td>7.0</td>
</tr>
<tr>
<td>Digoxigenin mono-digitoxiside</td>
<td>2.4</td>
<td>0.2</td>
<td>4.3</td>
<td>2.7</td>
<td>2.4</td>
</tr>
<tr>
<td>Digoxigenin</td>
<td>1.6</td>
<td>0.8</td>
<td>3.1</td>
<td>1.9</td>
<td>1.9</td>
</tr>
<tr>
<td>Solvent front</td>
<td>0.6</td>
<td>BKG</td>
<td>15.5</td>
<td>9.2</td>
<td>5.9</td>
</tr>
</tbody>
</table>

*Expressed as percent of total recovery from thin-layer chromatographic plate.
†Figure 2.
‡Background.
TRITIATED DIGOXIN

Figure 2

Thin-layer chromatogram of bile obtained after administration of tritiated digoxin. Markers of the specific glycosides (10 μg) were added to the sample for purposes of identification. This sample represents an extract of 10 ml of bile. The origin, glycosides, and solvent front are easily identified in this photograph in ultraviolet light. The per cent of radioactivity representing the actual amounts of digoxin and its metabolites is shown for this patient in Table 4 (day 4). The major product of biliary secretion is pure digoxin.

those obtained through similar analysis of data obtained from patients with congestive failure but without renal failure: Line B = 33 hours, and line C = 30 min. The data described are determined by the “peel-off” method, commonly employed in this type of study.

Table 2 compares the findings obtained in these studies to those previously reported after intravenous administration in subjects with congestive heart failure, and hyperthyroidism, hypothyroidism and normal human subjects. These data are utilized to compute absolute absorption and the percent of enterohepatic circulation in the next section.

Figure 2 is a thin-layer chromatogram of bile obtained from patient 5 on day 4. Note the identity peaks of digoxin, digoxigenin mono-digitoxiside, bis-digitoxiside, and digoxigenin in the figure.

Table 4 documents the distribution of digoxin and its metabolites by thin-layer chromatography on this patient. The figures shown in the table quantify the amounts of digoxin and its major metabolites in the bile on days 2, 3, 4, and 5 of the study on patient 5 and are representative of studies of bile from other patients. Digoxin is the major excretory product with small amounts of metabolites being identified. Chromatograms of urine reveal that 95 to 98% of the radioactivity present in the urine is unchanged digoxin.

These findings again are nearly identical to those obtained from patients with congestive heart failure.

Discussion

These studies demonstrate that the major portion of digoxin excreted in the stool is a product of biliary secretion. The total amount of digoxin recovered from stool and bile in this study suggests that the bile may be the only significant source of excretion of digoxin from the bowel, as the amount in the bile and the stool closely resembles total stool recovery by previously reported studies. Application of the formula proposed by Okita appears to show that recycling of digoxin in the bowel is of minor importance in its pharmacokinetic behavior.

The formula proposed by Okita for estimation of the enterohepatic circulation is of interest and deserves comment. He calculates the biliary excretion as:

\[
\text{Biliary excretion} = \text{Amount excreted via biliary fistula after intravenous dose of drug.}
\]
% absolute absorption = \[\frac{\text{oral dose} - \text{amount in GI tract} \times 100}{\text{oral dose}} \times 7\text{-day stool excretion in this instance}\]

\[\text{Enterohepatic circulation} = \frac{\text{biliary excretion} \times \% \text{ absolute absorption}}{\text{in} \times \text{travenous dose}}\times 100\]

It should be appreciated that refined pharmacokinetic studies are rarely obtainable for human subjects. We have chosen to base our calculations upon the data presented in this communication, as well as that obtained from six normal individuals who received digoxin by the oral route prior to nephrectomy as donors for renal transplant,10 12 patients with congestive heart failure,1 and 16 patients with thyroid disease6 whose excretion (but not their serum levels) approximates that of the other patients studied. These calculations reveal that the amount of absolute absorption of digoxin is 85% and the amount of enterohepatic circulation (recycling) is 6.5%. This figure can be compared to the estimate of Okita20 that 26% of an administered dose of digitoxin will participate in the enterohepatic circulation.

These data are in accord with the impression that polarity of the digitalis glycoside is of critical importance in determining excretion of the drug, its enterohepatic circulation, and thus the duration of therapeutic action in human subjects. Polar digoxin could be expected to be, and is, a short-acting glycoside (T ½, 34 hours), and nonpolar digitoxin, a long-acting glycoside (T ½, 105 hours).

A major reason for the short duration of action of digoxin, then, is the lack of recycling (or reabsorption) in the intestine. The polar characteristics of this glycoside also may account for its lack of degradation in the human body except by abnormal metabolic pathways.21 The nonpolar glycoside, digitoxin, on the other hand, appears to be extensively recycled during the process of its metabolism.20 The nonpolar characteristics of digitoxin thus contribute to its long duration of action and its properties of binding to serum protein.22, 23

A recent report by Caldwell and Greenberger24 reveals that the enterohepatic recycling of digitoxin may be partially interrupted by orally administered cholestyramine, thus shortening its metabolic half-life and duration of action. This experimental drug, studied primarily as a hypocholesterolemic agent, would appear to bind digitoxin similarly and interrupt its relatively large enterohepatic circulation. The physiologic T ½ as well as the serum turnover T ½ of digitoxin was shortened in patients given cholestyramine. A lesser effect was seen with digoxin as enterohepatic recycling plays only a minor role in its metabolism.

Digoxin is polar, virtually nonprotein bound,20 and excreted largely as unchanged glycoside. These properties all contribute to its short duration of action in human subjects in a clinical setting.

This study is in agreement with the mathematical evaluations of digoxin kinetics by Jelliffe,25-27 which reveal similar half-times and excretion.

Acknowledgment

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and Co. (USA), Inc., Tuckahoe, New York, provided and purified the tritiated digoxin.

References

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