Clinical Application of Serum Digitoxin Levels

A Simplified Plasma Determination

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SUMMARY
A simple, rapid, and reliable determination of plasma digitoxin levels is now available for clinical use. It can be determined in any clinical laboratory capable of determining a serum phosphorus if ATPase enzyme is available. Maintenance plasma levels range between 10 and 40 m\(\mu\)g/ml with a mean value of 25 m\(\mu\)g/ml. There is overlapping of maintenance and toxic serum levels in the higher ranges. Values over 45 m\(\mu\)g/ml warrant careful evaluation for digitalis intoxication.

Maintenance plasma levels can be achieved with or without the usual digitalizing dosage if time is not a factor. The daily regression rate is variable in cases of digitalis intoxication following drug withdrawal.

ECG changes (the degree of ST-segment depression, PR-interval prolongation, and multifocal premature contractions) were not good criteria for digitalis intoxication, although PR intervals above 0.24 sec consistently correlated with high plasma digitoxin levels. Variable second and third degree A-V block associated with mental confusion was a frequent presenting feature of digitalis toxicity.

Additional Indexing Words:
ECG changes Digitalis intoxication

An enzymatic assay of plasma digitoxin has recently been reported.\(^1\) It involves (1) the extraction of digitoxin from plasma by organic solvent, (2) the evaporation of an aliquot of extract to dryness in a test tube, and (3) the assay of transport adenosine triphosphate (ATPase) in the test tube where enzyme activity is determined by phosphate release. Enzyme inhibition by dried digitoxin in the tube is dose dependent, and this is the basis for the assay. Results are in agreement with more sophisticated and time-consuming methods previously reported.\(^2,3\)

ATPase enzyme is prepared from homogenized hog brain through a series of differential centrifugations and iodide extractions. A high-speed refrigerated centrifuge is required for the preparation.

The present study concerns (1) a further definition of the plasma maintenance and toxic digitoxin levels, (2) serial digitoxin levels during digitalization and digitalis withdrawal, and (3) a correlation of digitoxin levels with electrocardiographic changes suggestive of the pharmacologic effect of digitalis or of its toxicity, or both.

Methods
Levels of digitalis were determined in triplicate on 409 samples of blood from 233 adult patients at the VA Hospital and the University of Wisconsin Medical Center, Madison, Wisconsin. One hundred eighty-five patients were known to be taking either digitoxin or digitalis leaf. Sixteen patients were known to be taking digoxin, previously shown not to affect the assay system.\(^1\)

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and 32 patients not on any digitalis preparation served as the control sample. Although all patients were known to one of us (JDB), the clinical data were not available to either biochemist (RLC and GHB).

Thirty milliliters of venous blood was drawn in a heparinized syringe, preferably 3 hours after the last dose of digitalis was given. This could be refrigerated overnight if necessary. All determinations were available within 4 hours of blood drawing and were reported as millimicrograms (m\(\mu\)g) of digitoxin/ml* of plasma. Electrocardiograms were recorded on all patients at the approximate time of blood sampling and serial electrocardiograms were done if digitalis toxicity was clinically suspected. The electrocardiograms were interpreted by a reader who did not know the levels of plasma digitoxin. A blood urea nitrogen (BUN) was determined on all patients, and serum electrolytes and creatinine clearances were determined on 39 patients who either had BUN above 45 mg/100 ml or were clinically suspect of having digitalis toxicity.

Included in these data were 195 males and 37 females, with an age range of 20 to 90 years and a mean age of 64 years. The etiologic diagnosis included arteriosclerotic heart disease (47%), rheumatic heart disease (15%), chronic lung disease and cor pulmonale (13%), hypertensive cardiovascular disease (4%), cardiomyopathies (3%), and congenital heart disease (2%). Sixteen per cent of the patients were known to have no heart disease or the type was unclassified.

These data will be presented in the following four groups: Group I, an analysis of plasma digitoxin levels in a population known to be on a constant daily dose of digitoxin or Digitalis folia, who exhibited no clinical evidence of digitalis toxicity, in an attempt to determine the range of maintenance digitoxin levels; group II, determination of toxic digitoxin plasma levels; group III, determination of serial digitoxin levels during digitalization and following digitalis withdrawal, utilizing variable digitalization schedules; group IV, correlation of digitoxin levels with time-honored electrocardiographic changes of digitalis

*A This is equivalent of nanograms of digitoxin per milliliter of plasma.

Range of "minimal" digitalis effect

Range of "moderate-marked" digitalis effect

Figure 1

Examples of minimal and moderate-marked digitalis effect, based mainly on the degree of ST-segment sagging.
Table 1

Plasma Digitoxin Levels in 164 Patients Without Clinical Evidence of Toxicity

<table>
<thead>
<tr>
<th>Digitalis preparation and dosage</th>
<th>Patients</th>
<th>Plasma levels (μg/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Range</td>
</tr>
<tr>
<td>Digitoxin</td>
<td></td>
<td></td>
</tr>
<tr>
<td>0.05 mg/day</td>
<td>10</td>
<td>2-22</td>
</tr>
<tr>
<td>0.10 mg/day</td>
<td>115</td>
<td>2-50</td>
</tr>
<tr>
<td>0.15 mg/day</td>
<td>17</td>
<td>10-45</td>
</tr>
<tr>
<td>0.20 mg/day</td>
<td>4</td>
<td>43-50</td>
</tr>
<tr>
<td>Digitalis folia, 100 mg/day</td>
<td>18</td>
<td>5-35</td>
</tr>
</tbody>
</table>

Group II: Determination of Toxic Plasma Levels

Forty-one patients were studied for possible digitalis toxicity. Ten of these were suspected of having digitalis intoxication on clinical grounds (nausea, vomiting, bradycardia, and lightheadedness) (table 2). Others had ECG changes suggestive of toxicity (multifocal premature ventricular contractions [PVCs] or paroxysmal atrial tachycardia [PAT] with block and second and third degree A-V block).

When digitoxin (or Digitalis folia) was withheld, 21 of these 41 patients showed clinical or ECG improvement, with concomitant fall in their plasma digitoxin levels.

All patients felt to be in a toxic state were found to have initial plasma digitoxin levels at or above 41 μg/ml (table 3). Examples of electrocardiographic changes of toxicity and appropriate plasma digitoxin levels are shown in figures 3 to 6.

Ten other patients who had plasma levels of digitoxin greater than 45 μg/ml but who showed no evidence of toxicity were studied. Six of these patients had advanced pulmonary emphysema. Cessation of treatment with

Table 2

Composite Data on Digitalis-Toxic Patients

<table>
<thead>
<tr>
<th>Suspected digitoxin toxicity from</th>
<th>Patients</th>
<th>Toxicity proved</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Clinical diagnosis</td>
<td>10</td>
<td>7</td>
</tr>
<tr>
<td>ECG changes:</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Multifocal PVCs</td>
<td>17</td>
<td>6</td>
</tr>
<tr>
<td>1° A-V block</td>
<td>5</td>
<td>2</td>
</tr>
<tr>
<td>2° or 3° A-V block</td>
<td>9</td>
<td>6</td>
</tr>
<tr>
<td>Total</td>
<td>41</td>
<td>21</td>
</tr>
</tbody>
</table>

effect and toxicity. The electrocardiographic scoring of digitalis effect ranged from 0 to 3, where 0 shows no discernible digitalis effect; 1 + reveals minimal ST-segment sagging consistent with digitalis effect; 2 + exhibits moderate to marked ST-segment sagging associated with variable degrees of PR prolongation; and 3 + denotes variable degrees of second or third degree A-V block associated with marked ST-segment depression (fig. 1).

The PR intervals (ventricular rate in cases of atrial fibrillation) were plotted against the ventricular rates and the plasma digitoxin level.

Results

Group I: Determination of Maintenance Level of Digitalis Blood

A scattergram of serum digitoxin levels on the first 167 of 185 patients receiving a daily digitalis or Digitalis folia is shown in figure 2. From these composite data, 21 patients were excluded who were considered to exhibit unequivocal signs and symptoms of digitalis toxicity. The range and mean maintenance values for the various digitalis preparations are shown in table 1, with a fairly linear dosage relationship between the plasma digi-

toxin levels and increasing dosages of digitalis.

Thirty of the 32 patients who were not on any digitalis preparation yielded 0 digitoxin serum levels. The two remaining patients had levels of 2 μg/ml and 8 μg/ml, respectively. Retrospective questioning suggested that the latter patient had received a digitalis preparation 6 days prior to transfer from a nursing home. Conversely, two patients initially thought to be on maintenance digitoxin were found to have 0 plasma levels and were in fact not taking their digitoxin preparation.
Figure 2

Scattergram: digitalis dosages are plotted against the corresponding plasma digitoxin level on the first 167 patients on a maintenance digitalis preparation other than digoxin. All 10 patients taking 0.2 mg of digitoxin as a maintenance dosage had plasma levels above 42 μg/ml.

Figure 3

Atrial pauses. M.P., a 67-year-old man, entered the hospital with a 4-week history of dizziness and faintness associated with swallowing, resulting in loss of 20 pounds. He denied ingestion of digitalis, although this was subsequently refuted by his family physician. Repeated attempts at swallowing resulted in sinus pauses, temporary A-V block, and resultant ventricular slowing, reproducing his symptoms of dizziness.

The high serum digitalis level of 60 μg/ml fell to low maintenance levels in 17 days following cessation of digitalis; all central nervous symptoms cleared. Sinus arrest could not be reproduced by carotid body massage at the time of hospital admission.
Atrial flutter with AV block and ventricular bigeminy
10-3-68 Digitoxin level 75 μg/ml.

Atrial fibrillation
10-4-68 Digitoxin level 60 μg/ml.

S. Z. 82 yrs. Resolution of Bigeminal Rhythm

Figure 4
S.Z., an 82-year-old man, entered the hospital with mental confusion of 1 week’s duration, coinciding with a doubling of his maintenance dosage of digitalis. The atrial flutter (identified in V1), with multifocal ventricular bigeminy, was associated with an extremely high digitoxin blood level of 75 μg/ml. The bigeminy cleared, with a subsequent change in rhythm to atrial fibrillation 1 day following cessation of treatment with digitalis.

J.W. 76 yrs.

Figure 5
Complete heart block associated with digitalis toxicity. J.W., a 76-year-old man, entered the hospital for evaluation of mental confusion and nausea. His admission electrocardiogram confirmed the presence of complete heart block associated with an elevated digitoxin blood level of 62 μg/ml. The rhythm subsequently changed to a 1° A-V block, with clearing of all symptoms and a fall in digitoxin level following digitalis withdrawal.
digitalis again resulted in a fall in plasma digitoxin levels without change in clinical or electrocardiographic state.

Although there is no clear-cut plasma level that determines digitalis toxicity, levels above 45 mg/ml require consideration of toxicity. Regardless of the initial plasma level, cessation of digitalis resulted in a predictable fall in the plasma level.

**Group III: Determination of Digitalization Curves**

Fifteen patients were digitalized utilizing variable dosage schedules. All plasma digitoxin levels rose, with the rise dependent upon the speed and dosage of drug administration. Five representative plasma digitoxin curves are shown in Figure 7. Patients E. L. and E. H. achieved an average maintenance level of 20 to 30 mg/ml in approximately 30 days merely by the institution and continuation of a daily maintenance dose of digitalis. The drop at 3 to 5 days in serum levels noted in the three remaining patients corresponded to the institution of a maintenance digitalis schedule. All patients had normal renal function, as determined by BUN of less than 20 mg/100 ml and creatinine clearance greater than 70 ml/min.

**Figure 6**

Resolution of paroxysmal atrial tachycardia with block. L. D., a 72-year-old man, entered the hospital for evaluation of syncopal episodes. He had been receiving digitoxin, 0.1 mg daily. The admission ECG of 4-10-68 confirmed an atrial tachycardia with variable A-V block, associated with a high digitoxin blood level of 62 mg/ml. The latter fell to maintenance level 8 days following cessation of digitalis therapy, with a subsequent return to sinus rhythm and clearing of the syncopal episodes.

**Figure 7**

Five representative digitalization curves are shown, utilizing variable digitalization schedules. Maintenance digitoxin blood levels were achieved within 3 days by conventional methods of digitalization, namely, patients G. O., J. G., and R. H. Patient E. L., placed on a daily maintenance dose of 0.1 mg digitoxin, achieved a blood level of 20 mg/ml in approximately 30 days.
SERUM DIGITOxin LEVELS

Table 4
Electrocardiographic Signs of Digitalis Intoxication

<table>
<thead>
<tr>
<th>Serum digitoxin (mg/ml)</th>
<th>ECG grading</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>0</td>
</tr>
<tr>
<td>&gt;50</td>
<td>1</td>
</tr>
<tr>
<td>40-50</td>
<td>1</td>
</tr>
<tr>
<td>30-39</td>
<td>2</td>
</tr>
<tr>
<td>20-29</td>
<td>8</td>
</tr>
<tr>
<td>10-19</td>
<td>13</td>
</tr>
<tr>
<td>0-9</td>
<td>31</td>
</tr>
</tbody>
</table>

The decline in plasma digitoxin levels in seven patients considered to be toxic is shown in figure 8. The regression rates were variable (1 to 5.8 mg/ml/day) and were not always exponential with time. This is in contrast to the work of Lukas and Peterson who studied two patients with much lower plasma digitoxin levels for shorter periods.

Group IV: Electrocardiographic Scoring

Correlation between plasma digitoxin levels and the degree of ST-segment sagging was only fair in that the scoring of minimal digitalis effect correlated with low maintenance levels, and marked digitalis effect in general denoted high maintenance or toxic levels (table 4).

The presentation of abnormal ST segments and T waves with instances of left ventricular hypertrophy, left bundle-branch block, acute myocardial ischemic episodes, and chronic cor pulmonale were the chief causes of poor correlation, as these changes tended to obscure additive digitalis effect. Strict attention to J-junction depression on serial electrocardiograms was found to be of critical importance in assessing the presence of additive digitalis effect.

There was a wide scatter of PR intervals and plasma digitoxin levels, with only PR intervals above 0.24 sec consistently correlating with high plasma levels (fig. 9). In summary, one cannot predict blood digitoxin levels with any certainty on the basis of ECG change.

Discussion

Although technics exist to measure plasma
digitalis levels, they are either time consuming or technically difficult, and this limits their general clinical usefulness.

The method of Burnett and Conklin in determining plasma digitoxin levels, utilized in the current study, seems to have met the requirements of a simple, practical, and accurate assay of cardiac glycoside levels in man. The test is currently not applicable to digoxin preparation because of apparent differences in enzymatic binding.

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The current data suggest a maintenance range of plasma digitoxin to be 10 to 40 m\(\mu\)g/ml, with a mean of 25 m\(\mu\)g/ml. These levels are in agreement with the 5 to 40 m\(\mu\)g/ml maintenance range of Oliver and associates,\(^8\) the 10 to 56 m\(\mu\)g/ml levels of Lukas and Peterson,\(^2\) and the 10 to 50 m\(\mu\)g/ml range of Lowenstein and Corrill.\(^5\)

Serum digitoxin levels above 45 m\(\mu\)g/ml require consideration of digitalis toxicity and may merit a temporary cessation of maintenance therapy. No clear-cut plasma level differentiates maintenance from toxic levels.

Our observation that initiating digitalis therapy with only maintenance dosages in undigitalized patients will achieve a maintenance serum level within 30 days is in agreement with the previous data of Marcus et al.\(^11\) These workers, utilizing tritiated digoxin, noted that cumulation occurred even though an initial loading dosage was not given. By the sixth day of their study, there was no difference in plasma radioactivity between the group given only the daily maintenance dose and the group receiving a single digitalizing dose. The current data suggest that, although it will take a longer period to achieve a maintenance level utilizing digitoxin, the implication is the same. A loading dosage of digitalis is not necessary to achieve digitalization, except where maximum digitalis effect is required within hours or days.\(^12\)

Although the electrocardiographic changes attributed to digitalis are well described, we are unable to quantitate the plasma digitoxin level by scoring the degree of ST-segment depression or the prolongation of the PR interval. Minimal digitalis effect in general yielded low maintenance plasma levels, and marked ST-segment sagging, associated with variable degrees of PR interval prolongation, depicted high maintenance, or toxic levels.

Digitalis toxicity was first suggested by the appearance of a concomitant arrhythmia with gross ST-segment depression. Whereas the single instance of atrial tachycardia with variable A-V block and six episodes of transient second or third degree A-V block were all associated with toxic levels of digitoxin, only six of 17 patients with multifocal premature ventricular contractions had digitoxin levels above 45 m\(\mu\)g/ml that were thought to represent toxicity.\(^13\), \(^14\)

The consistent drop in plasma digitoxin levels following digitalis withdrawal is of considerable interest. The average regression rate for plasma digitoxin was found to be 3.2 m\(\mu\)g/ml/day, with a range of 1 to 5.8 m\(\mu\)g/ml/day; the higher regression values generally occurred where the initial plasma digitoxin levels were the highest.\(^15\) In a typical toxicity case with blood levels over 45 m\(\mu\)g/ml, it would take 6 to 10 days to achieve a maintenance level of 20 m\(\mu\)g/ml. These data are in agreement with the digitalis half-life determinations of Okita et al,\(^16\) and Braunwald and Klock.\(^6\)

Although we found two patients with plasma digitoxin levels above 60 m\(\mu\)g/ml with neither clinical nor electrocardiographic suggestions of toxicity, it was nonetheless felt advisable to withhold digitalis until the plasma levels fell below 35 m\(\mu\)g/ml. This is particularly true for patients in the older age group, because the only suggestion of digitalis toxicity may be the terminal arrhythmia or mental confusion.\(^17\), \(^18\)

The consistent association of plasma digitoxin levels above 43 m\(\mu\)g/ml in patients on a daily maintenance dosage of 0.2 mg/L of digitoxin serves to point out the potential hazard of this dosage schedule. This is particularly true for patients placed in a nursing home without close medical supervision.

Acknowledgment

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