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Digoxin Concentrations in Postmortem Specimens After Overdose and Therapeutic Use

The detection and evaluation of digitalis glycosides in autopsy specimens has been a serious problem for both medical examiners and toxicologists alike. Methods utilizing thin-layer chromatography or colorimetric techniques depended on a significant quantity of this drug remaining unabsorbed in the stomach or excreted into the urine. Digoxin, the most potent drug of this group and the form prescribed most frequently, is effective therapeutically in a dose range from 0.25 to 0.50 milligrams per day. This dose gives rise to therapeutic blood concentrations of 1.0 to 1.4 nanograms per millilitre [*1*]. Toxic effects of digoxin often begin to appear at 2.0 ng/ml. In view of these extremely low concentrations, even after an overdose with digitalis, detection of this drug in blood by conventional techniques could not be accomplished. The introduction of radioimmunoassay tests for these substances has provided an answer to this problem. Blood values of digoxin as low as 0.2 ng/ml can now be detected and measured reliably.

In a one-year period the Dallas County Medical Examiner's Office has detected four deaths due to an overdose of digoxin (see Table 1). In each of these cases a drug overdose, though not necessarily from digitalis, had been suspected from historical information. Drug screening for the presence of drugs usually associated with overdose was carried out and blood, vitreous humor, and urine were assayed for digoxin by radioimmunoassay.

In an attempt to accumulate information on digoxin in routine medical examiner cases, all cases seen in the medical examiner's office during a six-month period in which the deceased had a history of taking digoxin were studied. Specimens of blood, vitreous humor, and urine, when available, were assayed for the presence of digoxin.

Methods

Heart blood samples were centrifuged as soon as possible after removal from the body, the plasma separated, and then frozen or assayed within 24 hours. Vitreous humor and urine samples were treated likewise with the exception of not being centrifuged.

The procedure for digoxin employed the reagents supplied by Schwartz/Mann and is based on the technique of Smith et al [*1*]. The assay involves the competitive binding of

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TABLE 1—*Cases of digoxin overdose.*

Age, Race, Sex	Digoxin, ng/ml						Other Drugs
	Blood	Hemolysis	Vitreous	Vitreous:Blood Ratio	Urine	Other	
Case 1 87 w/f	38.6	0	2.8	0.07	1280.7	...	blood diphenylhydantoin 0.81 mg/100 ml; phenobarbital 0.80 mg/100 100 ml
Case 2 88 w/f	12.1	0	8.5	0.70	...	...	...
Case 3 20 w/m	10.0	++	1.0	0.10	...	stomach washings 796 ng/ml; intestinal contents 2909 ng/ml	blood propoxyphene 1.20 mg/100 ml
Case 4 25 w/m	...	++	...	...	2.40 (4.60 digitoxin)	stomach contents 0.60 ng/ml; 5.40 ng total	blood diazepam 0.05 mg/100 ml; blood ethanol 0.14% (w/v)

tritium-labeled digoxin and unlabeled digoxin contained in serum or plasma with digoxin-specific antibody. The digoxin-antibody complex formed a level of radioactivity which is inversely related to the concentrations of unlabeled digoxin in the sample.

The separation of digoxin bound to antibody from unbound digoxin is accomplished by absorption of the unbound (or free) digoxin, both labeled and unlabeled, on dextrose-coated charcoal. After centrifugation, the *bound* radioactivity is determined by decantation of the supernatant and counting by liquid scintillation counting. The percent tritium bound is plotted against nanograms per millilitre of sample and results are read from a standard curve prepared with each group of samples. Quench correction was accomplished by automatic external standardization.

Cases

Case 1

An 87-year-old white female was pronounced dead shortly after arrival at a hospital emergency room. The deceased had attempted suicide by overdose of medications six times in the past year. On the day of her death she had had an argument with her family, got out of bed, and went into the kitchen. A few minutes after getting back into bed she began to vomit. She subsequently told her family that she had taken almost a full bottle of 0.125-mg digoxin tablets. Autopsy findings were essentially unremarkable except for coronary artery disease and some myocardial fibrosis. Toxicological examination of the blood, vitreous humor, and urine revealed a blood diphenylhydantoin concentration of 0.81 mg/100 ml and a phenobarbital concentration of 0.80 mg/100 ml. Digoxin values were 38.6 ng/ml in the blood, 2.8 ng/ml in the vitreous, and 1280 ng/ml in the urine.

Case 2

An 88-year-old woman was admitted to the hospital with a clinical diagnosis of digitalis intoxication. The patient experienced cardiac arrest and died 30 minutes after admission. She had been under treatment by a family physician for arteriosclerotic and hypertensive cardiovascular disease. Her prescribed dose of digoxin had been increased from one to two 0.25-mg tablets per day over the last month. An electrocardiogram prior to death was consistent with the diagnosis of digitalis intoxication. Postmortem toxicology revealed digoxin concentrations of 12.1 ng/ml in the blood and 8.5 ng/ml in the vitreous.

Case 3

A 20-year-old white male was found dead lying on the bed in his bedroom. He had been seen alive 4½ hours before. The deceased had numerous personal problems and had talked of suicide to a number of his friends. At the time of autopsy it was not known to what drugs he had access. Postmortem toxicology revealed propoxyphene concentrations of 1.20 mg/100 ml in the blood, 3.27 ng/100 g in the kidney, 8.4 mg/100 g in the liver, 0.20 mg/100 ml in the vitreous, 112.0 mg/100 ml in the small bowel contents, and 17.60 mg/100 ml in the stomach contents. Digoxin concentrations were 10.0 ng/ml in the blood, 1.0 ng/ml in the vitreous, 796.0 ng/ml in the stomach washings, and 2909 ng/ml in the intestinal contents. It was not known in what order these medications were taken, but the high concentration of propoxyphene in the blood

and the low quantities found in the stomach contents indicate that sufficient time had elapsed for this drug to have been mostly absorbed. In contrast, a large amount of digoxin still remained in the stomach and in the small intestine. Both drugs had been taken in lethal quantities.

Case 4

A 25-year-old white male had been depressed over the breakup of his marriage. He had been drinking heavily that day when he announced to his father that he had taken an "overdose of pills." Members of the family tried to induce vomiting and gave him hot liquids to drink. He was pronounced dead approximately four hours later on arrival at a hospital emergency room. Death was initially thought to be due to salicylate intoxication as the deceased had pointed to a bottle containing aspirin in the medicine cabinet when he was questioned by members of his family as to medication he had taken. Findings at autopsy were generally unremarkable. The stomach appeared empty. A complete toxicological screen for drugs usually involved in overdoses, including salicylates, was negative except for a relatively low concentration of diazepam (0.05 mg/100 ml) and ethanol (0.14% weight/volume) in the blood. As a last resort, portions of the urine and stomach contents were analyzed for digitalis glycosides. Small quantities of digoxin were detected in the urine and stomach content and digitoxin was detected in the urine. Subsequent investigation revealed that the man's father had been taking digoxin and that the bottle was kept in the medicine cabinet next to the aspirin bottle. The deceased had been in good health and was not taking any cardiac medications prior to death. Assay for digoxin in blood could not be performed as no specimen was left at the time digitalis was considered as a possible intoxicant.

Control Group

Over a six-month period 35 cases were selected from all cases seen in the medical examiner's office. The sole criterion for selection of these cases was that the decedents were known to be taking digoxin prior to death. Analysis of fluids from these bodies revealed that the cases could be divided into two groups according to the digoxin concentrations in the blood. In 13 of the cases (37%), the digoxin concentrations were in the toxic range, that is, greater than 2.0 ng/ml. These comprised the first group and data from these cases are shown in Table 2. The remaining 22 cases (63%) were patients who died with plasma digoxin levels of 2.0 ng/ml or less (within the therapeutic range) and these comprised the second group. Digoxin concentrations in the first group ranged from 2.8 to 10.5 ng/ml. In one case, the deceased was dehydrated and showed evidence of renal damage, while in another, renal failure secondary to extensive burns of the body had occurred. Renal failure in both these cases probably accounted for the impaired excretion of the drug and consequent high blood levels. In other cases, however, no explanation could be found and as far as could be determined, the normal prescribed dose of 0.25 ng was not exceeded.

The concentration of digoxin in the vitreous humor in this first group of cases was always lower than that in the blood. The vitreous to blood ratio averaged 0.37 in these cases.

At this time, it should be pointed out that the hemolysis of blood which may occur post mortem has an effect on digoxin concentrations by the techniques presently used. Hemolysis was found to cause a falsely low value which can be reduced to one-half that of the actual level if up to 2% hemolysis is present. There was no attempt to compensate

TABLE 2—*Cases with toxic digoxin concentrations in blood.*

Age, Race, Sex	Digoxin, ng/ml					Circumstances
	Blood	Hemolysis	Vitreous	Vitreous:Blood Ratio	Urine	
59 w/f	4.7	+++	2.1	0.45	64.6	digoxin 0.25 mg[a]; found dead in bed; died during night; blood quinidine 0.48 mg/100 ml
1½ w/m	8.4	+++	<0.2	0.02	...	congestive heart disease; on pacemaker; at home; "screamed and died"
50 n/m	5.0	+++	1.2	0.24	7.8	found dead, 9:15 a.m.
68 n/m	4.0	+++	...	...	24.8	"took a pill 8:15 a.m."; died between 2 and 3 p.m.
78 n/f	9.5	+++	4.1	0.43	...	died in hospital, burns
72 w/f	2.8	+++	0.4	0.14	15.3	digoxin 0.25 mg; found dead 4:30 p.m.
77 n/m	10.5	+++	4.1	0.39	...	found dead at 11 p.m.; dehydrated; blood urea nitrogen 260
65 w/m	3.8	+++	2.1	0.55	34.9	digoxin 0.25 mg; collapsed and died, 4:30 p.m., watching TV
33 n/f	3.1	0	1.2	0.39	15.2	obese; found dead 6:10 p.m.
72 n/f	4.0	+++	1.6	0.40	...	digoxin 0.25 mg; died suddenly in bed, 3:15 a.m.
73 w/f	3.9	++	...	...	...	died at wheel of car, 5:10 p.m.
52 w/m	4.7	0	3.9	0.84	...	found dead, 8:20 p.m.
47 n/m	6.1	+++	1.2	0.20	...	digoxin 0.25 mg; found dead, 6:55 a.m.

[a] Daily dose given when known.

for this in measuring the samples reported here because of the difficulties in determining the degree of hemolysis. Consequently, cases where hemolysis occurred are indicated as such in the tables and the listed blood digoxin values are probably lower than the actual ones.

Data from the second group of cases are shown in Table 3. Most of these individuals died suddenly. When a history could be obtained, they were found to be on a normal therapeutic regimen of 0.125 to 0.25 mg of digoxin per day. In this group the hemolysis factor was not as prominent as in the preceding group, so the blood concentrations can be regarded as more accurate. The vitreous to blood ratios in this group were higher than in the first group and averaged 1.32.

Discussion

Digitalis toxicity is extremely common with up to 20% of hospitalized patients receiving digitalis showing some evidence of toxicity [2]. The daily maintenance dose for an average adult is between 0.25 and 0.50 ng daily. Renal failure with resultant decreased excretion of digoxin, however, may result in maintenance doses as low as 0.125 mg on alternate days. In the absence of renal failure, dosage is largely determined by body muscle mass and the level of metabolism [3]. Thus, older, smaller individuals require less digoxin. Variation in the potency and rate of absorption of digoxin tablets produced by different manufacturers may also result in different blood concentrations [4]. Some patients will take more than the prescribed dose in the mistaken belief that if one pill is good, two are better. Accidental overdosage may be more common than realized in elderly patients as they may easily take too many pills because of this latter reasoning or because of forgetfulness associated with senility.

The most striking aspect of our study was the fact that 37% of the individuals in the random study had blood digoxin concentrations in the toxic range. In 2 of these 13 cases, renal failure appears to have caused the elevated blood concentrations. The actual plasma digoxin values in the group with toxic concentrations (Group 1) were probably even higher than the reported values, since all but 2 of these 13 cases showed hemolysis.

It can be assumed that the concentration of digoxin in the vitreous humor at the time of complete distribution and equilibrium in the body should be roughly equivalent to that found in the blood, that is, a vitreous to blood ratio of 1.0, as digoxin is not strongly protein bound and would be readily diffusible into the vitreous humor. The observation that the concentration in the vitreous is lower than that in plasma in some cases and higher in others indicates that a considerable lag period exists before equilibrium is reached. During the absorptive period while the digoxin concentration is increasing in the blood, the vitreous humor concentration would be lower, thus making the vitreous to blood ratio less than one. Those patients dying during this period would show vitreous concentrations lower than that in the blood, leading one to assume that they had died within a few hours of having taken the last dose. It is of interest that in the 39 cases studied, none of the original 4 cases of overdose or the 13 cases with toxic digoxin concentrations (Table 2) showed exact equilibration of vitreous humor with blood. In fact, only 3 of these cases showed a vitreous blood ratio of over 0.5. In the 22 cases with therapeutic concentrations of digoxin at the time of death, 9 (45%) had vitreous to blood ratios of one or greater, while 8 (40%) had ratios between 0.5 and 0.9. This indicates the blood concentrations of those in the toxic ranges were rising at the time of death, while those with therapeutic concentrations had reached equilibration in distribution with other body compartments.

TABLE 3—*Cases with therapeutic concentrations of digoxin in the blood.*

Age, Race, Sex	Digoxin, ng/ml					Circumstances
	Blood	Hemolysis	Vitreous	Vitreous:Blood Ratio[a]	Urine	
55 n/f	1.5	0	1.8	1.20	...	digoxin 0.10 mg[b]; found dead on commode, 2: a.m.
80 n/m	1.1	0	0.8	0.73	...	digoxin; found dead, 7:20 p.m.
	<0.2	+++				
42 w/f	<0.2	+++	0.51	>2.50	...	found dead, 10 a.m.
73 w/f	<0.2	+++	1.18	>5.90	129.8	died suddenly, 7 p.m.
85 n/f	1.20	0	0.80	0.67	...	digoxin 0.125 mg; died suddenly, 11:55 p.m.
32 n/m	1.2	0	1.4	1.20	...	died suddenly, 4:30 p.m.
84 n/f	1.9	0	1.3	0.68	...	digoxin 0.50 mg; died between 1 and 3 p.m.
55 n/f	1.1	+	1.2	1.10	...	digoxin 0.25 mg; died between 4 and 8 a.m.
80 n/f	0.4	0	0.5	1.25	...	found dead, 9:15 a.m.
50 n/m	1.0	+	1.3	1.30	...	digoxin 0.25 mg; collapsed and died, 4:30 p.m.
37 n/m	1.6	+	0.4	0.25	58.5	digoxin 0.125 mg; collapsed and died, 5:55 p.m.
58 w/m	<0.2	++	0.5	2.50	<0.2	collapsed and died, 9:45 a.m.
72 w/f	2.0	++	1.5	0.75	2.5	found dead, 11:30 p.m.
58 w/m	0.5	+	...	...	0.8	blood diazepam 0.02 mg/100 ml
77 w/f	0	0	0.3	>3.00	0	found dead, 10:15 a.m.
62 n/f	1.3	+	0.7	0.54	...	collapsed and died, 6:30 a.m.
73 w/m	1.3	0	1.0	0.77	36.6	collapsed and died, 5:40 p.m.
56 n/f	1.5	++	1.2	0.80	36.0	died sitting in chair after eating, 11 a.m.
58 n/m	2.1	0	1.8	0.86	41.1	burns; in county hospital several days
49 w/m	1.5	++	0.5	0.33	...	collapsed while walking; died 8:30 a.m.
86 w/m	1.5	0	0.6	0.40	...	choked to death, 4:30 p.m.
52 n/m	1.0	0	1.0	1.00	<0.2	collapsed in cafe, died in emergency room

[a] Mean vitreous:blood ratios: therapeutic 1.32 and toxic 0.37.
[b] Daily dose given when known.

Those individuals dying with vitreous concentrations equivalent to or higher than those in the blood could be assumed to be in the postabsorptive phase. The vitreous concentrations then would indicate the blood level at some period several hours earlier when equilibration was reached. This determination may, therefore, be of value in estimating how much time has elapsed between dosing and death.

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