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Effect of Digoxin-like Immunoreactive Substances on the "Dac-Cel" Digoxin Assay

To the Editor:

Digoxin-like immunoreactive substances (DLIS) are found in certain groups of subjects, including patients in late pregnancy and those with renal failure. The amount of apparent digoxin in the serum of these patients when they are not receiving digoxin varies with the method of analysis (1). Graves et al. (2) reported that DLIS were decreased by increasing the incubation time in the five methods they studied. The "Dac-Cel" RIA serum digoxin kit (Wellcome Diagnostics, Dartford, England) has detected the presence of DLIS in three patients with combined hepatic and renal failure (3). According to the kit manufacturer, DLIS are <0.5 nmol/L in women taking oral contraceptives or who are in the last trimester of pregnancy.

In the present study I used the Dac-Cel method to measure apparent digoxin in the serum of 80 patients, none of whom was receiving digoxin. Forty patients were in the third trimester of pregnancy and 40 were undergoing maintenance hemodialysis for end-stage renal failure.

In the DacCel assay, 1 mL of solid-phase antibody is added to 0.1 mL of sample or standard, followed by 0.1 mL of 125I-labeled digoxin. These are mixed, incubated at room temperature for 0.5 h, and centrifuged. The supernatant fluid is decanted and the radioactivity in the deposit is measured. I modified the assay in two ways: Method 1 was according to the manufacturer's instructions, but using half quantities throughout; Method 2 was like Method 1, but with the incubation prolonged from 0.5 to 3 h. For Method 2 only, sample, solid-phase antibody, and label were mixed continuously on a rotary mixer during the incubation period. All 80 samples were assayed in duplicate by both methods.

By Method 1 all but five samples gave digoxin values below the limit of detection: four pregnancy sera gave results of 0.19, 0.22, 0.25, and 0.28 nmol/L, and one sample from a patient with renal disease gave a value of 0.21 nmol/L. By Method 2, all the results were below the limit of detection.

To compare the results obtained by the two methods in routine use, I also assayed, by both methods, sera from 50 patients receiving digoxin. The regression equation for the comparison was found to be Method 2 = 0.935 (Method 1) + 0.110 nmol/L (r = 0.994). The values for the slope and intercept are statistically significantly different from 1 and 0, respectively (P <0.01 in both cases), but the numerical difference between the values by the two methods would be <0.1 nmol/L for concentrations in the range 0.2 to 3.2 nmol/L.

Other characteristics of the two methods are summarized in Table 1. Values for drift are the mean decreases in apparent concentration from beginning to end of run. I determined the detection limits by assaying 20 replicates of the zero standard and using the method of Rodbard (4). I performed recovery studies, using the five sera with detectable DLIS, five of the sera with no detectable DLIS, and one normal serum. Equilibrium is reached in Method 2 but not in Method 1, so variations in timing have a smaller effect in Method 2. This would account for the better precision and smaller drift of Method 2. The larger detection limit of Method 2 is due to the smaller slope of this method's calibration graph at low digoxin concentrations.

In summary: the amount of DLIS measured by the Wellcome Dac-Cel digoxin kit in patients in late pregnancy or with renal disease was <0.3 nmol/L; moreover, prolonging the incubation decreased the DLIS to undetectable concentrations as well as improving the method precision and decreasing drift, but the limit of detection increased slightly. However, these assay modifications do not clinically significantly affect results for patients receiving digoxin. I recommend Method 2 if the longer assay interval is clinically acceptable.

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References

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Monoclonal Gammopathies in Asymptomatic HIV-Seropositive Patients

To the Editor:

A high prevalence of monoclonal gammopathies (MG) was found in serum from patients with AIDS or lymphadenopathy syndrome (LAS) (1, 2). To our knowledge, MG have not been reported in asymptomatic patients seropositive for human immunodeficiency virus (HIV). We have studied 243 HIV seropositive subjects, detected by systematic screening of blood donors (ELISA, with confirmation by Western blot) and followed up every six months: 114 were seen once, 62 twice, 52 three times, 15 four times.