The discrepancy in theophylline results was noted to be greater than 20%--50% in light of earlier reports of difficulties with immunoassays in uremic patients. This report should again emphasize the importance of obtaining clinical information on a patient as well as an appropriate sample.

References


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Charcoal Stripping Has No Effect on Prostate Androgen Receptor

To the Editor:

In attempting to develop a reliable method for the assay of androgen receptors in human prostate, Traish et al. (1) reported that "stripping" (treating) the cytosol with dextran-coated charcoal (DCC) before incubating it with labeled ligand revealed the existence of a new steroid-binding protein distinct from the cytosol androgen receptor. This conclusion was based on seven- to 31-fold more receptor in eight DCC-treated cytosols than in nonstripped cytosols, as determined by use of Scatchard plots.

We wondered whether such a discrepancy could be detected by using our method (2) for the receptor, which is similar to the method of Shain et al. (3). Dextran- and γ-globulin-coated charcoal (DGCC) are used to remove endogenous dihydrotestosterone and other steroids from cytosol. The stripping methods are compared in the following tabulation:

<table>
<thead>
<tr>
<th></th>
<th>Shain et al.</th>
<th>Traish et al.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Dextran, g/L</td>
<td>5.0</td>
<td>0.5</td>
</tr>
<tr>
<td>Human γ-globulin, g/L</td>
<td>10</td>
<td>—</td>
</tr>
<tr>
<td>Charcoal, g/L</td>
<td>50</td>
<td>5.0</td>
</tr>
<tr>
<td>Vol of coated charcoal</td>
<td>1/10</td>
<td>1/1</td>
</tr>
<tr>
<td>Incubation time, min</td>
<td>10</td>
<td>30</td>
</tr>
</tbody>
</table>

We saw no significant differences in receptor as measured in terms of milligrams of protein or of DNA in DGC Stripped vs nonstripped cytosols in parallel experiments with nine different prostate samples. Results were compared by paired Student's t-test.

Theoretically, removal of endogenous free steroid is desirable, because its presence would decrease the binding of labeled ligand to receptor. Actually, stripping made no difference in the quantity of receptor with our method. Perhaps, the difference between our results and those of Traish et al. (1) is related to the interval the cytosol is exposed to charcoal; we used 10 min, one-third as long as in Traish et al., who found a very significant increase in apparent receptor with stripping. All cytosols were apparently exposed to the same concentrations of DCC. The only other difference is our 1% coating of charcoal with human γ-globulin, which Shain et al. claim improves the specificity of the method.

We conclude that charcoal stripping of endogenous steroid, as described here at least, does not unmask other binding proteins from the methyltrienolone ligand, as was reported by Traish et al. (1).

This work was supported by the National Cancer Institute (NIH) Grant CA 18008 through the National Prostatic Cancer Project.

References


Unusually High Proportion of Creatine Kinase MB Isoenzyme, Evidently of Nonmyocardial Origin, in Serum of a Patient with Cancer of the Prostate

To the Editor:

Creatine kinase (EC 2.7.3.2) isoenzyme MB (CK-MB) in serum is widely regarded as a specific marker for myocardial necrosis (1–3). There are exceptions, however: CK-MB bands may be observed in electrophoretograms of serum in noncardiac disorders such as muscular dystrophies, polymyositis, Reye's syndrome, and alcoholic rhabdomyolysis (4–7). Its source in such cases is presumably skeletal muscle. In addition, CK-MB bands were reported to be persistently increased in the sera of two patients with malignant disorders (8, 9).

Here, I report a case of prostatic carcinoma in which the serum CK-MB was persistently detectable in high proportion relative to CK-MM. This patient, a 79-year-old man, was admitted to the hospital because of the sudden onset of chest discomfort. Measurements of lactate dehydrogenase (LDH), aspartate aminotransferase, γ-glutamyltransferase, and alkaline phosphatase in his serum were above normal: 469 (normal, 90–200), 39 (5–35), 166 (<65), and 176 (30–115) U/L, respectively. Total serum CK activity, however, was within the normal range: 139 U/L (normal, 35–232). Concurrently, CK and LDH isoenzyme patterns were quantified. All LDH isoenzyme fractions were abnormally increased, but no "flip" of the LDH-1 and LDH-2 ratio was observed. CK isoenzyme quantification by agarose gel electrophoresis showed CK-MM, CK-MB, and CK-BB isoenzymes to all be present (Table 1). During the next week the patient's total serum CK activities progressively declined, but CK-MB bands were still repeatedly detected in the electrophoretograms.

I considered the possibility that the fluorescent band migrating electrophoretically at the position of MB represented an atypical variant of CK isoenzyme. To rule this possibility in or out, I treated the patient's serum with a solid-phase anti-Ck-B subunit antibody (International Immunoadsay Labs, Inc., Santa Clara, CA 95050), then treated the resulting precipitate with radiolabeled antibody to CK-M subunit. After a brief centrifugation, the radioactivity recovered from the precipitate served as an indicator of the presence of CK-MB. As indicated in Table 1, CK-MB isoenzyme was detected in four consecutive determinations on different sera.

Had this patient had an episode of myocardial infarction? I think not. First of all, the patient's total CK activities showed persistently low values. Secondly, the LDH-1/LDH-2 ratio was not flipped. Thirdly, the persistence of detectable CK-MB after 72 h in this patient's sera is not typical of myocardial infarction.

Furthermore, the presence of the CK-MB isoenzyme in this patient's serum probably is unrelated to the damaged prostate, because this organ reportedly is devoid of CK-MB isoenzyme (10).

Thus the origin of the CK-MB isoenzyme in the sera of this patient is obscure. Can the CK-MB isoenzyme be produced by a tumor ectopically?

References


Enhancing the Sensitivity of an Immunoradiometric Assay for Thyrotropin

To the Editor:

We have modified the manual calculation procedure for the Bio-Rad Immunoradiometric Assay for human thrytropin (Bio-Rad Laboratories, Richmond, CA 94804). Bio-Rad (1) recommends plotting counting rate (cpm) or B/B₀₀ in percent vs thyrotropin concentration in micro-internation units per ml(μU/mL) on linear graph paper. We find that both of these techniques compromise the readable sensitivity inherent in the method for concentrations between 0 and 5 μU of thyrotropin per milliliter. By plotting %B/B₀₀ vs thyrotropin concentration on three-cycle semilog paper, utilizing

Table 1. Summary of Patient's Creatine Kinase Data

<table>
<thead>
<tr>
<th>Day after admission</th>
<th>Total CK acyt, U/L</th>
<th>% by electrophoresis</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>MM</td>
<td>MB</td>
</tr>
<tr>
<td>1</td>
<td>139</td>
<td>53</td>
</tr>
<tr>
<td>3</td>
<td>87</td>
<td>42</td>
</tr>
<tr>
<td>4</td>
<td>79</td>
<td>48</td>
</tr>
<tr>
<td>8</td>
<td>52</td>
<td>50</td>
</tr>
</tbody>
</table>

* Calculated in arbitrary units based on the immunological activity of a calibrator prepared from a CK-MB human tissue extract (International Immunoadsay Labs, Inc.); negative results are <2.5.

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