Table 2. Acceptable Limits for Precision and Accuracy (Our Selected Best Results)

<table>
<thead>
<tr>
<th>Test</th>
<th>Precision (day-to-day CV, %) from &quot;correct&quot; value</th>
<th>Accuracy (value)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Glucose</td>
<td>3.0</td>
<td>0.8</td>
</tr>
<tr>
<td>Urea</td>
<td>4.0</td>
<td>0.7</td>
</tr>
<tr>
<td>Uric acid</td>
<td>3.1</td>
<td>0.5</td>
</tr>
<tr>
<td>Creatinine</td>
<td>2.9</td>
<td>0.8</td>
</tr>
<tr>
<td>Chloride</td>
<td>2.6</td>
<td>1.6</td>
</tr>
<tr>
<td>Calcium</td>
<td>2.8</td>
<td>0.7</td>
</tr>
<tr>
<td>Phosphate</td>
<td>3.7</td>
<td>1.4</td>
</tr>
<tr>
<td>Total protein</td>
<td>2.1</td>
<td>1.1</td>
</tr>
<tr>
<td>Albumin</td>
<td>2.9</td>
<td>0.4</td>
</tr>
<tr>
<td>Total bilirubin</td>
<td>3.4</td>
<td>0.9</td>
</tr>
<tr>
<td>Cholesterol</td>
<td>2.6</td>
<td>0.7</td>
</tr>
</tbody>
</table>

*Only results from the higher-concentration control serum used here.*

correct values was calculated from the difference between the observed values and the correct values, related to the latter, and expressed as the average.

Our results for internal accuracy were poorer than the external ones, perhaps because of the leveling effect of calculating mean values from a large number of laboratories.

To look for limit values, we selected our best results obtained reiteratively over at least 15 months of the 30 months in the study. We used a score system similar to that of CAP (4) and we took the three best group results; if the total number of cases was 15, we considered the test as representative. In this way, limit values were obtained for the 11 different tests.

We consider these selected results (Table 2) as the acceptable limits for accuracy and precision for laboratories using the discrete automated system and analytical methods described in this report (Table 1).

One laboratory alone cannot provide guidelines, but we believe that if clinical chemists first solve the problems within their own laboratories, they could then together with other laboratorians and interlaboratory organizations develop a unified set of criteria, and provide general patterns for guidelines.

References

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Is Commercial Serum Suitable for Quality Control of Serum Total Iron-Binding Capacity?

To the Editor:
The Letter of Devgun et al. (1) prompts me to report my experience with use of lyophilized or liquid control sera for quality control of serum total iron-binding-capacity (TIBC).

I cannot agree that "different methods of measurement can consistently lead to higher or lower results from the target values." It must be borne in mind that one molecule of transferrin binds two atoms of iron; in other words, only one expected value (± allowable limits) of TIBC is possible for a known concentration of transferrin.

In a previous study (2) I assayed 12 human lyophilized control sera. Of these, only five were suitable for TIBC quality control: transferrin concentration, measured TIBC, manufacturer's value, and expected value agreed well. In the other sera the measured TIBC was consistently higher than the manufacturer's value, with poor reproducibility.

More recently I checked the performances of "Decision" liquid controls (Beckman). TIBC was determined with an AutoAnalyzer I (Technicon) by saturation of transferrin with a ferric chloride solution, removal of the excess by adsorption onto MgCO₃, and colorimetric measurement of Fe²⁺ by a ferrozine-neocuproine reagent. For immunochromatographic assay of transferrin I used an automated nephelometric system (AIP, Technicon). In agreement with Sheehan et al. (3), results were excellent; I found good agreement between transferrin concentration and TIBC values (mean of 12 different assays):

<table>
<thead>
<tr>
<th>Level 1</th>
<th>Level 3</th>
</tr>
</thead>
<tbody>
<tr>
<td>Transferrin, g/L</td>
<td>1.9</td>
</tr>
<tr>
<td>Expected TIBC, μmol/L</td>
<td>47.5</td>
</tr>
<tr>
<td>Measured TIBC, μmol/L</td>
<td>48.2</td>
</tr>
</tbody>
</table>

Nevertheless, I have been using lyophilized sera in internal quality control for several years, namely, "Q-PAK" serum (Travenol), then "QAP Serum Level 1" (Merz-Dade). Each of these was chosen after I found agreement between immunochromatography and chemical data.

After many assays, I am sure that the pH of the reconstituted serum plays a major role in the accuracy of TIBC determination. The pH must be less than 8.5; alkaline control sera exhibit poor reproducibility and accuracy. Moreover, I observed (2) that reconstituted lyophilized control sera perform differently from native human serum. The iron-binding kinetics is slower, and complete saturation requires between 5 and 15 min, whereas it is quite immediate for human native serum. I think that lyophilization somewhat alters the iron-binding capacity.

For an adequate comparison of results in external quality control, the control serum must be suitable, as determined by a preliminary assay to check agreement with results of immunochromatographic determination of transferrin. By this criterion, only human sera are appropriate, and they must have a pH lower than 8.5 after reconstitution.

To achieve complete saturation, I recommend a 10-min wait before adding magnesium carbonate, and this step should be carefully standardized.

References

Michèle Vernet-Nyssen
The authors of the Letter cited comment:

To the Editor:

Although Vernet-Nyssen does not agree, in principle, that different methods of measurement should give different results, in practice, however, such differences are often observed, either as a consequence of specimen preparation or as a result of analytical technique. In the previous Letter (Clin Chem 25: 544, 1982) we communicated our results based on external quality control schemes involving lyophilized samples only. As we have no experience with liquid controls, we cannot comment on these.

Vernet-Nyssen agrees with us that pH plays a major role in reproducibility and accuracy of TIBC determinations, and furthermore, calls to our attention the fact that lyophilized sera have a slower binding kinetics than native human serum. As a consequence, she suggests a 10-min incubation step to achieve complete saturation. Such a step would not be appropriate for quality-control purposes, as it will draw a special attention to quality control material but not other specimens. However, such a step would be appropriate if all samples were treated similarly. But is it a valid modification to change a routine methodology to accommodate external quality control material? If liquid controls are found satisfactory for internal quality control schemes, the problem of handling samples from external quality control schemes would remain.

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Separation of the High-Molecular-Mass “Immunoreactive ACTH” Measured with the CIS-Sorin Kit by Ultracentrifugation of Plasma

To the Editor:

Inappropriately high values for plasma corticotropin (ACTH), found in approximately 5% of normal subjects when the CIS-Sorin radioimmunoassay kit (1) is used, have been attributed to an “interfering activity” of high molecular mass (2). We discovered a high concentration of immunoreactive ACTH (280 ng/L) in the plasma of a normal volunteer, resolvable on G-50 chromatography into a high-molecular-mass (>30 000 daltons) immunoreactive peak and a peak (24% of total) co-eluting with 125I-labeled ACTH. To facilitate the separation of this high-molecular-mass component and hence to distinguish false from authentic increases in ACTH obtained with the CIS-Sorin kit, we analyzed fractions after ultracentrifugation of plasma in the Beckman Airfuge.

Plasma samples from the normal volunteer, a patient with Cushing’s disease, and a patient with Addison’s disease were centrifuged at 100 000 × g for 4 h, in the A100 rotor. These conditions were selected to sediment species with molecular mass >20 000 daltons into the lower half of the tube. The centrifuged plasma in each tube was then divided into a 75-μL upper and a 75-μL lower fraction, and ACTH was measured in the pooled upper and lower fractions (Table 1).

With plasma from the normal volunteer, the concentration of immunoreactive ACTH in the upper (low molecular mass) fraction decreased markedly into our “plasma normal range” (<70 ng/L), suggesting separation of the high-molecular-mass interfering activity. In the case of plasma from the patients with Cushing’s or Addison’s disease, there was some decrease in immunoreactive ACTH in the upper fraction, but the values remained markedly high. The decrease in immunoreactive ACTH in these fractions could be due to the sedimentation of high-molecular-mass precursors of ACTH present in the plasma of patients with pathological causes of increased ACTH.

These data indicate that ultracentrifugation in the Beckman Airfuge may be a convenient way to distinguish authentic from inappropriately high values for ACTH obtained with the CIS-Sorin ACTH kit.

References
2. Ristuccia RM, Sharp AM, Baxter RC.

<table>
<thead>
<tr>
<th>Table 1. Values for Immunoreactive ACTH, ng/L</th>
</tr>
</thead>
<tbody>
<tr>
<td>Utracent.</td>
</tr>
<tr>
<td>-----------</td>
</tr>
<tr>
<td>Sample</td>
</tr>
<tr>
<td>Normal volunteer</td>
</tr>
<tr>
<td>Cushing’s disease</td>
</tr>
<tr>
<td>Addison’s disease</td>
</tr>
</tbody>
</table>


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A Proposal for the Abandonment of Activity Units for Most Plasma Enzymes

To the Editor:

I have previously suggested (1) that measuring the concentration of most analytes per litre of plasma is physiologically and pathologically incorrect, and that plasma water should be the reference base. We use the former because with current instrumentation it is generally available and cheaper, and because it is familiar.

We should further consider the idea that plasma enzymes should not be measured in terms of activity (in U/L or nkat/L) but in terms of their concentration—and substance concentration (mmol/L) should be preferred to mass concentration (μg/L). With some exceptions—principally the coagulation enzymes (factors) and certain enzymes of lipid metabolism—enzymes in plasma have no function there. Their presence is an accident: they are passing from healthy or damaged cells, and are being transported in the plasma to disposal. We approach them as protein cell-markers, to investigate mainly either the extent of cell damage causing increased leakage, or the extent of cellular overproduction of enzyme or of obstruction of normal secretion into extracellular or transcellular fluid causing overflow. In these circumstances the measure that relates most closely to the cell pathology that we are investigating, ideally the rate of enzyme release from cells, is the number of enzyme molecules in plasma, for which the appropriate unit is substance concentration and not an activity unit based on a catalytic reaction. Here, too, we use the latter because with current instrumentation it is generally available and cheaper, and because it is familiar. Activity units should be used only when there is need to consider enzyme function. Their general use for plasma, which has given so much useful diagnostic information, should now be regarded as historical—to be replaced when possible.

Coagulation hematologists, who at present use arbitrary units for their