has no extraction requirements, and preserves most of the advantages of automation.

In summary, we report the development of a highly sensitive and precise semiautomated immunoassay that is capable of measuring estradiol-17β concentrations in women during the perimenopausal and postmenopausal intervals and in healthy men and children.

References
7. Norjavaara E, Ankarberg C, Albertsson-Wikland K. Diurnal rhythm of 17
6.
7. Norjavaara E, Ankarberg C, Albertsson-Wikland K. Diurnal rhythm of 17
6.
7. Norjavaara E, Ankarberg C, Albertsson-Wikland K. Diurnal rhythm of 17
6.
7. Norjavaara E, Ankarberg C, Albertsson-Wikland K. Diurnal rhythm of 17
6.
7. Norjavaara E, Ankarberg C, Albertsson-Wikland K. Diurnal rhythm of 17
6.
7. Norjavaara E, Ankarberg C, Albertsson-Wikland K. Diurnal rhythm of 17
6.
7. Norjavaara E, Ankarberg C, Albertsson-Wikland K. Diurnal rhythm of 17
6.
7. Norjavaara E, Ankarberg C, Albertsson-Wikland K. Diurnal rhythm of 17
6.
7. Norjavaara E, Ankarberg C, Albertsson-Wikland K. Diurnal rhythm of 17
6.
7. Norjavaara E, Ankarberg C, Albertsson-Wikland K. Diurnal rhythm of 17
6.
7. Norjavaara E, Ankarberg C, Albertsson-Wikland K. Diurnal rhythm of 17
6.
7. Norjavaara E, Ankarberg C, Albertsson-Wikland K. Diurnal rhythm of 17
6.
7. Norjavaara E, Ankarberg C, Albertsson-Wikland K. Diurnal rhythm of 17
6.
7. Norjavaara E, Ankarberg C, Albertsson-Wikland K. Diurnal rhythm of 17
6.
7. Norjavaara E, Ankarberg C, Albertsson-Wikland K. Diurnal rhythm of 17
6.
7. Norjavaara E, Ankarberg C, Albertsson-Wikland K. Diurnal rhythm of 17
6.
7. Norjavaara E, Ankarberg C, Albertsson-Wikland K. Diurnal rhythm of 17
6.
Table 1. Medians, 95% RIs, and the 90% confidence intervals for the 2.5% and 97.5% reference limits in the elderly (65 years or older). a

<table>
<thead>
<tr>
<th>Index</th>
<th>n (outliers)b</th>
<th>Median</th>
<th>RI</th>
<th>CI of the 2.5% reference limit</th>
<th>CI of the 97.5% reference limit</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hb, g/L</td>
<td>220</td>
<td>145</td>
<td>121–165</td>
<td>115–128</td>
<td>163–167</td>
</tr>
<tr>
<td>Men (iron replete)</td>
<td>207</td>
<td>145</td>
<td>124–165</td>
<td>118–129</td>
<td>163–167</td>
</tr>
<tr>
<td>Women</td>
<td>327</td>
<td>133</td>
<td>113–150</td>
<td>109–116</td>
<td>147–153</td>
</tr>
<tr>
<td>Women (iron replete)</td>
<td>258</td>
<td>133</td>
<td>115–151</td>
<td>113–118</td>
<td>148–154</td>
</tr>
<tr>
<td>Men (iron replete)</td>
<td>207</td>
<td>4.68</td>
<td>3.82–5.32</td>
<td>3.70–3.95</td>
<td>5.20–5.44</td>
</tr>
<tr>
<td>Women (iron replete)</td>
<td>258</td>
<td>4.34</td>
<td>3.64–5.03</td>
<td>3.55–3.74</td>
<td>4.94–5.12</td>
</tr>
<tr>
<td>Hematocrit, L/L</td>
<td>220</td>
<td>0.430</td>
<td>0.360–0.490</td>
<td>0.337–0.382</td>
<td>0.487–0.493</td>
</tr>
<tr>
<td>Men (iron replete)</td>
<td>207</td>
<td>0.430</td>
<td>0.366–0.490</td>
<td>0.344–0.388</td>
<td>0.489–0.491</td>
</tr>
<tr>
<td>Women</td>
<td>327</td>
<td>0.400</td>
<td>0.335–0.449</td>
<td>0.326–0.343</td>
<td>0.443–0.455</td>
</tr>
<tr>
<td>Women (iron replete)</td>
<td>258</td>
<td>0.400</td>
<td>0.339–0.450</td>
<td>0.332–0.345</td>
<td>0.443–0.456</td>
</tr>
<tr>
<td>Mean corpuscular volume, fl</td>
<td>220</td>
<td>93.0</td>
<td>85.1–102.0</td>
<td>83.6–86.7</td>
<td>100.3–103.8</td>
</tr>
<tr>
<td>Men (iron replete)</td>
<td>207</td>
<td>93.0</td>
<td>86.0–102.2</td>
<td>84.6–87.4</td>
<td>100.4–104.0</td>
</tr>
<tr>
<td>Women</td>
<td>327</td>
<td>91.0</td>
<td>83.2–99.4</td>
<td>81.5–84.8</td>
<td>98.4–100.4</td>
</tr>
<tr>
<td>Women (iron replete)</td>
<td>258</td>
<td>92.0</td>
<td>84.9–99.2</td>
<td>84.3–85.4</td>
<td>98.0–100.4</td>
</tr>
<tr>
<td>Mean corpuscular Hb, pg</td>
<td>220</td>
<td>31.0</td>
<td>28.7–34.5</td>
<td>27.9–29.5</td>
<td>33.7–35.3</td>
</tr>
<tr>
<td>Men (iron replete)</td>
<td>207</td>
<td>31.0</td>
<td>29.0–34.5</td>
<td>28.6–29.3</td>
<td>33.6–35.5</td>
</tr>
<tr>
<td>Women</td>
<td>327</td>
<td>31.0</td>
<td>27.5–33.4</td>
<td>26.6–28.3</td>
<td>32.6–34.2</td>
</tr>
<tr>
<td>Women (iron replete)</td>
<td>258</td>
<td>31.0</td>
<td>28.5–33.5</td>
<td>27.8–29.3</td>
<td>32.7–34.2</td>
</tr>
<tr>
<td>sTfR, mg/L</td>
<td>547</td>
<td>1.45</td>
<td>0.96–3.03</td>
<td>0.92–1.00</td>
<td>2.37–3.70</td>
</tr>
<tr>
<td>All (iron replete)</td>
<td>465</td>
<td>1.41</td>
<td>0.95–2.41</td>
<td>0.91–0.99</td>
<td>2.00–2.82</td>
</tr>
<tr>
<td>Serum ferritin, µg/L</td>
<td>220</td>
<td>91.5</td>
<td>15.4–342.1</td>
<td>11.9–18.9</td>
<td>296.8–387.3</td>
</tr>
<tr>
<td>Women</td>
<td>326 (1)</td>
<td>49.0</td>
<td>7.1–238.0</td>
<td>5.5–8.8</td>
<td>189.3–286.7</td>
</tr>
<tr>
<td>TR-F indexd</td>
<td>219 (1)</td>
<td>0.75</td>
<td>0.45–1.88</td>
<td>0.41–0.50</td>
<td>1.44–2.31</td>
</tr>
<tr>
<td>Women</td>
<td>326 (1)</td>
<td>0.84</td>
<td>0.46–3.29</td>
<td>0.42–0.50</td>
<td>2.69–3.88</td>
</tr>
<tr>
<td>Serum iron, µmol/L</td>
<td>220</td>
<td>19.0</td>
<td>9.1–32.7</td>
<td>8.3–9.9</td>
<td>29.2–36.2</td>
</tr>
<tr>
<td>Men (iron replete)</td>
<td>207</td>
<td>19.0</td>
<td>9.4–32.9</td>
<td>8.4–10.5</td>
<td>29.4–36.4</td>
</tr>
<tr>
<td>Women</td>
<td>327</td>
<td>17.0</td>
<td>7.7–27.1</td>
<td>6.8–8.6</td>
<td>26.1–28.1</td>
</tr>
<tr>
<td>Women (iron replete)</td>
<td>258</td>
<td>17.0</td>
<td>9.4–27.1</td>
<td>8.3–10.6</td>
<td>26.1–28.2</td>
</tr>
<tr>
<td>Serum transferrin, g/L</td>
<td>220</td>
<td>2.40</td>
<td>1.61–3.39</td>
<td>1.45–1.77</td>
<td>3.11–3.66</td>
</tr>
<tr>
<td>Men (iron replete)</td>
<td>207</td>
<td>2.40</td>
<td>1.64–3.18</td>
<td>1.49–1.79</td>
<td>2.92–3.43</td>
</tr>
<tr>
<td>Women</td>
<td>327</td>
<td>2.50</td>
<td>1.72–3.53</td>
<td>1.59–1.85</td>
<td>3.33–3.73</td>
</tr>
<tr>
<td>Women (iron replete)</td>
<td>258</td>
<td>2.40</td>
<td>1.66–3.15</td>
<td>1.53–1.80</td>
<td>3.02–3.29</td>
</tr>
<tr>
<td>Transferrin saturation, %</td>
<td>220</td>
<td>32.0</td>
<td>13.0–58.3</td>
<td>11.6–14.4</td>
<td>53.4–63.1</td>
</tr>
<tr>
<td>Men (iron replete)</td>
<td>207</td>
<td>32.4</td>
<td>14.2–58.4</td>
<td>12.5–15.8</td>
<td>53.9–62.9</td>
</tr>
<tr>
<td>Women</td>
<td>327</td>
<td>26.7</td>
<td>11.0–47.7</td>
<td>9.2–12.8</td>
<td>44.6–50.8</td>
</tr>
<tr>
<td>Women (iron replete)</td>
<td>258</td>
<td>27.8</td>
<td>15.2–49.3</td>
<td>13.3–17.1</td>
<td>44.6–54.0</td>
</tr>
</tbody>
</table>

a Values were determined from the general reference group and from the iron-replete subgroup, consisting of all the individuals from the general reference group with serum ferritin concentration ≥ 22 µg/L (8).

b Outliers were identified with the Dixon test.

c CI, confidence interval.

d TR-F index = sTfR/log serum ferritin.
were included in the general reference group. Of these, 465 (207 men and 258 women) were included in the iron-replete subgroup. The median ages were 71.6 years in the general reference group and 71.1 years in the iron-replete subgroup. The median ages were 71.6 years in the iron-replete subgroup and the general reference group. Despite of these slight trends, we think that the use of single reference limits in populations 65 years or older is justified to assure the sensitivity of these tests to detect ID and anemia.

ID has been reported to be quite common in the elderly (27, 28). Advancing ID can be divided into three stages according to concentrations of serum ferritin, sTfR, and Hb (4, 8). In our study, 15% of the whole initial population (n = 1260) was deemed to be storage iron-deficient (stage I, ferritin <22 mg/L). This prevalence of storage ID may be underestimated because the whole initial population included patients with diseases that may increase ferritin concentrations irrespective of current iron status. As evaluated according to iron-replete reference values for the elderly (Table 1), 7.0% presented with increased ferritin concentrations as a signal for iron-deficient erythropoiesis (stage II). Altogether, 7.1% were anemic, and 26% of these individuals (1.8% of the whole initial population) had IDA (stage III) according to increased sTfR and low Hb (<128 mg/L in men and <117 mg/L in women, i.e., lower reference limits of our hospital). These prevalences were lower than we expected. On the other hand, in a recent study, Fleming et al. (29) found ID only in 2.7% of the elderly participants and IDA in 1.2%, albeit with different criteria for ID. Furthermore, there is no evidence of a physiologic age-related decrease in erythropoietic potential (30).

Our findings support the idea that current RIs are often

In the general reference group, a slight statistically significant (P <0.05) positive correlation between age and sTfR was found, whereas statistically significant (P <0.05) negative correlations were observed between age and Hb, erythrocyte count, hematocrit, TIR-F index, serum ferritin (men), and serum iron (men). These trends seemed to be partly attributable to subclinical ID because correlations were diminished as the population was narrowed down to the iron-replete subgroup. Fig. 1 demonstrates how the median values for Hb and sTfR changed with age in the general reference group. Despite of these slight trends, we think that the use of single reference limits in populations 65 years or older is justified to assure the sensitivity of these tests to detect ID and anemia.

ID has been reported to be quite common in the elderly (27, 28). Advancing ID can be divided into three stages according to concentrations of serum ferritin, sTfR, and Hb (4, 8). In our study, 15% of the whole initial population (n = 1260) was deemed to be storage iron-deficient (stage I, ferritin <22 mg/L). This prevalence of storage ID may be underestimated because the whole initial population included patients with diseases that may increase ferritin concentrations irrespective of current iron status. As evaluated according to iron-replete reference values for the elderly (Table 1), 7.0% presented with increased ferritin concentrations as a signal for iron-deficient erythropoiesis (stage II). Altogether, 7.1% were anemic, and 26% of these individuals (1.8% of the whole initial population) had IDA (stage III) according to increased sTfR and low Hb (<128 mg/L in men and <117 mg/L in women, i.e., lower reference limits of our hospital). These prevalences were lower than we expected. On the other hand, in a recent study, Fleming et al. (29) found ID only in 2.7% of the elderly participants and IDA in 1.2%, albeit with different criteria for ID. Furthermore, there is no evidence of a physiologic age-related decrease in erythropoietic potential (30).

Our findings support the idea that current RIs are often

In the general reference group, a slight statistically significant (P <0.05) positive correlation between age and sTfR was found, whereas statistically significant (P <0.05) negative correlations were observed between age and Hb, erythrocyte count, hematocrit, TIR-F index, serum ferritin (men), and serum iron (men). These trends seemed to be partly attributable to subclinical ID because correlations were diminished as the population was narrowed down to the iron-replete subgroup. Fig. 1 demonstrates how the median values for Hb and sTfR changed with age in the general reference group. Despite of these slight trends, we think that the use of single reference limits in populations 65 years or older is justified to assure the sensitivity of these tests to detect ID and anemia.

Our findings support the idea that current RIs are often

In the general reference group, a slight statistically significant (P <0.05) positive correlation between age and sTfR was found, whereas statistically significant (P <0.05) negative correlations were observed between age and Hb, erythrocyte count, hematocrit, TIR-F index, serum ferritin (men), and serum iron (men). These trends seemed to be partly attributable to subclinical ID because correlations were diminished as the population was narrowed down to the iron-replete subgroup. Fig. 1 demonstrates how the median values for Hb and sTfR changed with age in the general reference group. Despite of these slight trends, we think that the use of single reference limits in populations 65 years or older is justified to assure the sensitivity of these tests to detect ID and anemia.

Our findings support the idea that current RIs are often

In the general reference group, a slight statistically significant (P <0.05) positive correlation between age and sTfR was found, whereas statistically significant (P <0.05) negative correlations were observed between age and Hb, erythrocyte count, hematocrit, TIR-F index, serum ferritin (men), and serum iron (men). These trends seemed to be partly attributable to subclinical ID because correlations were diminished as the population was narrowed down to the iron-replete subgroup. Fig. 1 demonstrates how the median values for Hb and sTfR changed with age in the general reference group. Despite of these slight trends, we think that the use of single reference limits in populations 65 years or older is justified to assure the sensitivity of these tests to detect ID and anemia.

Our findings support the idea that current RIs are often

In the general reference group, a slight statistically significant (P <0.05) positive correlation between age and sTfR was found, whereas statistically significant (P <0.05) negative correlations were observed between age and Hb, erythrocyte count, hematocrit, TIR-F index, serum ferritin (men), and serum iron (men). These trends seemed to be partly attributable to subclinical ID because correlations were diminished as the population was narrowed down to the iron-replete subgroup. Fig. 1 demonstrates how the median values for Hb and sTfR changed with age in the general reference group. Despite of these slight trends, we think that the use of single reference limits in populations 65 years or older is justified to assure the sensitivity of these tests to detect ID and anemia.

Our findings support the idea that current RIs are often

In the general reference group, a slight statistically significant (P <0.05) positive correlation between age and sTfR was found, whereas statistically significant (P <0.05) negative correlations were observed between age and Hb, erythrocyte count, hematocrit, TIR-F index, serum ferritin (men), and serum iron (men). These trends seemed to be partly attributable to subclinical ID because correlations were diminished as the population was narrowed down to the iron-replete subgroup. Fig. 1 demonstrates how the median values for Hb and sTfR changed with age in the general reference group. Despite of these slight trends, we think that the use of single reference limits in populations 65 years or older is justified to assure the sensitivity of these tests to detect ID and anemia.
derived from population samples containing individuals with ID. These findings together with earlier reports imply that there are grounds as well as means for improving the sensitivity of laboratory tests to diagnose ID. This is of clinical importance in the elderly, in whom ID often heralds severe underlying diseases, such as ulcers or malignancies. In practice, this should motivate the production and use of iron-replete reference values of sTfR and other markers of iron status.

This study was funded by grants from Turku University, the Medical Research Foundation of the Turku University Central Hospital, and the Finnish Association of Haematology. One of the authors, P. Suominen, has been employed part-time as a medical consultant for Orion Diagnostica, manufacturer of the sTfR assay used in this study.

References


Analytical Requirements for Measuring Monocyctic Human Lymphocyte Antigen DR by Flow Cytometry: Application to the Monitoring of Patients with Septic Shock, Guillaume Monnere, Nadia Elmenkouri, Julien Bohe, Anne-Lise Debar, Marie-Claude Gutovezu, Jacques Biewenn, and Alain Lepape

The concept of immunoparalysis has recently been proposed for explaining the failure of 20 years of clinical trials using antiinflammatory drugs in sepsis (1–3). Immunoparalysis is characterized mainly by the paralysis of monocyctic functions. In particular, because of decreased expression of HLA-DR, antigen-presenting capacity is severely depressed (4, 5). Recent clinical studies have confirmed that among a large panel of activation markers expressed on different leukocyte populations (i.e., neutrophils, lymphocytes, and monocytes), decreased HLA-DR expression on monocytes constitutes a reliable marker of immunoparalysis and seems to correlate with an increased risk for fatal outcome (6–8). Nevertheless, little work has been devoted to analytical aspects related to its measurement by flow cytometry. Indeed, flow cytometry cannot yet be considered a standardized tool, and many variables must be taken into account for ensuring the technical quality of results (9). This is especially required.