drew blood from 10 healthy in-house volunteers (CDT, 8–29 units/L) four times during a 24-h period, at 0800–0900, 1200–1300, 1600–1700, and 2100–2200. No restrictions regarding food intake were given. The serum samples were stored at −20 °C until analysis. The CDT concentrations from the different sampling times were calculated as percentages of the daily mean for each person. For the whole group, the mean values were 97%, 106%, 95%, and 102%, respectively. The variation between the samples was not statistically significant when analyzed using a linear model.

The effects of freezing and repeated freezing and thawing were investigated. Serum samples were collected from 26 subjects (9 females and 17 males; CDT, 11–62 units/L). Each sample was divided into two tubes, one stored at −20 °C and the other at 2–8 °C until analysis 1 day later. The mean ratio (frozen/unfrozen) was 98% (range, 86–112%). For the study of repeated freezing and thawing, serum samples from 17 subjects (CDT, 9–74 units/L) were included and stored at −20 °C. The stability was followed for 32 months (Table 1). Three of the samples were still available after 8 years. CDT concentrations at that time were, on average, 97% of the initial values.

In view of these results, we conclude that the diurnal variability of CDT is low. Thus, it does not seem to be necessary to restrict sample collection to a special time of the day.

We have also found a very good stability of CDT in most serum samples. Freezing and repeated freezing and thawing did not significantly influence the results. In general, CDT also showed high stability in thawed serum when stored both in the refrigerator at 2–8 °C and at high room temperature, 32 °C. However, one cannot exclude the possibility that individual samples may exhibit poor stability because of endogenous enzymes or bacterial contamination.

The finding of practically no change in CDTect values for sera stored >8 years at −20 °C points to a very good stability of the analyte in frozen serum. This may be of great importance for many clinical studies where the samples are stored for long periods of time before analysis.

<table>
<thead>
<tr>
<th>Percentage of initial value</th>
<th>Time of storage</th>
<th>Samples, n</th>
<th>Mean</th>
</tr>
</thead>
<tbody>
<tr>
<td>98</td>
<td>0.5 years</td>
<td>4</td>
<td></td>
</tr>
<tr>
<td>102</td>
<td>0.9 years</td>
<td>4</td>
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</tr>
<tr>
<td>95</td>
<td>1.4 years</td>
<td>4</td>
<td></td>
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<tr>
<td>95</td>
<td>1.9 years</td>
<td>4</td>
<td></td>
</tr>
<tr>
<td>94</td>
<td>2.6 years</td>
<td>4</td>
<td></td>
</tr>
<tr>
<td>97</td>
<td>8.4 years</td>
<td>3</td>
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</tr>
</tbody>
</table>

To the Editor:

The measurement of fecal fat excretion (1) remains an important test in the diagnosis of fat malabsorption despite the development of alternative biochemical investigations, such as the butterfat test (2), steatocrit (3), and triolein breath tests (4), and alternative diagnostic strategies in gastroenterology. However, the measurement of fecal fat excretion has suffered from poor reproducibility because of both preanalytical and analytical factors. Although the former may be minimized by the use of standardized study protocols, there is no universally employed method of internal quality control of the analytical phase (5). Although modern analytical techniques such as near infrared analysis (6) offer improved analytical performance, most laboratories performing fecal fat measurements still use chemical methods described in the 1950s. These methods involve the hydrolysis of fecal fat, followed by extraction of the liberated fatty acids and measurement by titrimetric or gravimetric methods (1).

The ideal quality-control material
for fecal fat assays would assess performance of the assay as a whole (hydrolytic, extraction, and titration steps), would be stable on storage, would be free of major health and safety problems, and would be easily and conveniently handled by laboratory staff. Anecdotal reports suggest that approaches to internal quality control are varied and range from control of the titration step alone to the use of single fats such as triolein, pure fat solutions such as oils, or fat-rich materials such as peanut butter. Although repeat analysis of a feces sample with the next patient sample may also be performed, health and safety considerations limit the use of stored feces as a method of monitoring long-term assay performance.

We have explored the use of flaked desiccated coconut as a suitable quality-control material. Desiccated coconut is rich in fat (~53 g/100 g) comprising mainly saturated fat (C12:0, C14:0, C8:0, and C16:0) together with C18:1 and C18:2 unsaturated fatty acids. Flaked desiccated coconut for culinary use was obtained from a commercial source. The coconut was mixed well, divided into 0.2-g aliquots, and frozen at −30 °C until analysis. This sample was added to a boiling tube, and the fat content was measured by the method of van de Kamer (1), which is our standard fecal fat method. Over a 4-month period this material gave, in the final alkali titration step of the assay, a titration volume of 1.92 mL (0.16) [mean (SD); n = 12], i.e., a coefficient of variation of 8.4%. There was no evidence of deterioration in the material over this period.

Flaked desiccated coconut is easy to handle, poses no health and safety problems, and is suitable for long-term storage. It allows quality control of all three analytical phases of the assay, and because it involves the extraction of a range of fatty acids from a biological matrix, albeit a somewhat different matrix than feces, it is nevertheless a more biologically suitable control material than single fat or pure fat materials. We therefore propose that desiccated coconut is a simple, cheap, and convenient material for monitoring the analytical performance of fecal fat assays. Furthermore, this material might prove suitable for use in external proficiency testing schemes.

References


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