were confirmed by a recombinant immunoblot assay (Chiron RIBA HCV 3.0 SIA, Chiron Corporation). Additional testing for viral RNA in confirmed HCV antibody-positive samples was performed by a commercially available PCR test kit (Ampli
cor HCV monitor, Hoffmann-La Roche AG).

HCV antibody positivity was detected in 328 (0.60%) donors (all negative for HIV antibody, hepatitis B surface antigen, and TPHA); 39 (0.072% of all donations) were positive in the RIBA assay, and 19 (0.035% of all donations) were reactive with PCR. Among the 4251 donations with increased neopterin concentrations (>10 nmol/L), 44 of 328 were HCV antibody seropositive (odds ratio = 1.83, \( \chi^2 = 14.37, P < 0.001 \)), 8 of 39 were RIBA positive (odds ratio = 3.04, \( \chi^2 = 8.74, P = 0.003 \)), and 7 of 19 were HCV PCR positive (odds ratio = 6.88, \( \chi^2 = 22.11, P < 0.001 \)). Similarly, 7 of 44 HCV antibody-positive donations with increased neopterin were positive by PCR; this was true for only 12 of 284 HCV antibody-positive donations with neopterin within reference values (odds ratio = 3.76, \( \chi^2 = 9.53, P = 0.002 \)). We conclude that there is a significant association between increased neopterin concentrations and a positive PCR result in the HCV antibody-seropositive donations.

From the data it becomes obvious that not only acute and symptomatic HCV infections are associated with increased serum neopterin values, as could have been expected from earlier studies (4, 5), but also that the frequency of asymptomatic chronic HCV carriers with RNA positivity as a sign for infectivity was approximately sevenfold higher in donors with increased concentrations of neopterin compared with those with concentrations within reference values. The data of this study further demonstrate that neopterin screening of blood donations contributes to reduce infectious risk of transfusion. In cases of HCV this is of limited relevance because antibody testing has been introduced, although some HCV antibody seronegative infections will certainly exist. The residual value of neopterin screening to further reduce HCV transmission by transfusion cannot be deduced from this study. For ethical reasons, it is impossible to transfuse blood donations with increased neopterin concentrations in Austria. Therefore, the benefit of additional neopterin screening of blood donations cannot be demonstrated by comparing the outcome of recipients; evaluation is only possible in a retrospective way when new specific tests become available. Our study further supports the view that increased neopterin concentrations in seronegative blood donations may be a marker for other currently unknown infections or infections not screened for and that transmission of these infections maybe reduced if they are transmissible by blood transfusion.

References

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Diurnal Variability and in Vitro Stability of Carbohydrate-deficient Transferrin

To the Editor:

Transferrin isoforms with pl values ≥5.7, known as carbohydrate-deficient transferrin (CDT), are present in increased concentrations in the serum of patients with current alcohol abuse. CDT has been found to be a specific and sensitive marker for detection and monitoring of high and continual alcohol consumption (1). The clinical performance of CDT has been studied using various techniques for analysis but also using various conditions for serum sample collection and sample storage. The biological (non-alcohol-influenced) variation over time has been found to be low (2). We have studied the diurnal variability of CDT and the effect of sample storage conditions in the commercial CDTect assays.

Blood was collected by venipuncture and allowed to clot at room temperature. The serum samples were separated by centrifugation, and 1-mL portions were aliquoted into polypropylene tubes (Sarstedt). CDT was measured by duplicate determinations in CDTect® RIA or CDTect EIA (Pharmacia & Upjohn Diagnostics AB). Results are expressed as units per liter. In an evaluation performed on 76 serum samples the assays gave comparable results and showed good correlation, with a correlation coefficient of 0.99 and a regression line of EIA = 0.72 + 0.90 × RIA. The within-assay and between-assay coefficients of variation for the RIA were 7.2% and 9.3%, respectively; for the EIA, they were 6.5% and 9.3%, respectively.

To study the diurnal variation, we...
drawd 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 sampling times 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 1. Effect of storage at −20 °C.

<table>
<thead>
<tr>
<th>Samples, n</th>
<th>Time of storage</th>
<th>Percentage of initial value</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Mean</td>
<td>Range</td>
</tr>
<tr>
<td>4</td>
<td>0.5 years</td>
<td>98 94–103</td>
</tr>
<tr>
<td>4</td>
<td>0.9 years</td>
<td>102 100–104</td>
</tr>
<tr>
<td>4</td>
<td>1.4 years</td>
<td>95 89–101</td>
</tr>
<tr>
<td>4</td>
<td>1.9 years</td>
<td>95 89–104</td>
</tr>
<tr>
<td>4</td>
<td>2.6 years</td>
<td>94 79–103</td>
</tr>
<tr>
<td>3</td>
<td>8.4 years</td>
<td>97 90–105</td>
</tr>
</tbody>
</table>

### References


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Desiccated Coconut as a Quality-Control Material in Fecal Fat Measurements

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