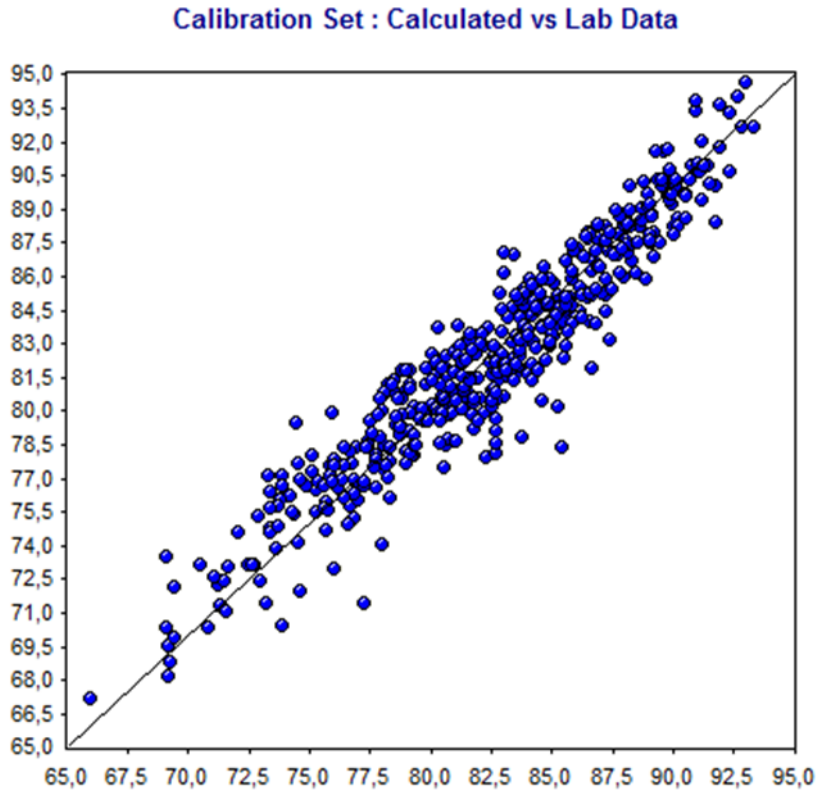
Near-Infrared Spectroscopy Application Note NIR–19

Determination of moisture, fat, and nitrogen in human feces by NIR spectroscopy



This application note describes the quantitative analysis of moisture, nitrogen, and fat content in human feces samples, which are important parameters in medical diagnostics, by NIRS.

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Method description

Introduction

Several diseases like pancreatic insufficiency or hepatic disorders produce malabsorption or maldigestion. The resulting changes in stool composition, e.g., in moisture, nitrogen content, or fat content, provide important information for medical diagnostics. Time-consuming lab methods like titration for the determination of fat content [1] can be replaced by rapid Near-Infrared Spectroscopy (NIRS).
This application describes the prediction of moisture, fat, and nitrogen content in human feces with a NIRS DS2500 Analyzer.

Experimental

522 samples of human feces were analyzed on a NIRS DS2500 Analyzer (**Tab. 1**). Three different parameters were analyzed (**Tab. 2**).

Table 1: Equipment

Analyzer	NIRS DS2500
Accessories	Customer ring cup (modified) Mini ISI ring cup (FOSS)
Software	Vision 4.01

Table 2: Analysis parameters

Moisture content	(Humidity)
Fat content	(STOT)
Nitrogen content	(CRTOT)

Method development:

During sample selection, which is the first step in method development with Vision software, 57 samples were detected as outliers. To detect the outliers, the Maximum Distance in Wavelength Space was used. This means that the mean spectrum of all 522 samples was calculated by the software, and all spectra that were outside the 5-standard-deviation acceptance range were identified as outliers. The remaining 465 spectra were suitable for quantitative method development.

The samples were separated into 2 subsets – one for calibration (75%) and one for cross validation (25%). An additional 15 samples were used for external validation to reduce the risk of overfitting the model. The reference values[1] are listed in **Tab. 3**.

Table 3: Humidity, STOT and CRTOT content of the 522 samples split into 23 classes

STOT reference value	CRTOT reference value	Humidity reference value
0.24	0.19	64.66
1.28	0.26	65.96
2.33	0.32	67.26
3.37	0.39	68.57
4.41	0.46	69.87
5.46	0.52	71.17
6.50	0.59	72.47
7.54	0.66	73.78
8.59	0.72	75.08
9.63	0.79	76.38
10.67	0.86	77.68
11.72	0.93	78.98
12.76	0.99	80.29
13.80	1.06	81.59
14.84	1.13	82.89
15.89	1.19	84.19
16.93	1.26	85.50
17.97	1.33	86.80
19.02	1.39	88.10
20.06	1.46	89.40
21.10	1.53	90.71
22.15	1.59	92.01

As math pretreatment, the 2nd derivative (gap: 0 nm; segment: 10 nm) was calculated in the Vision software in combination with an N-point smooth (segment: 10 nm). Smoothing is needed because human feces have a complex matrix, resulting in a baseline shift and high variance in spectra, which leads to random noise.

[1] Measured using the customer's old FeNIR Analyzer.

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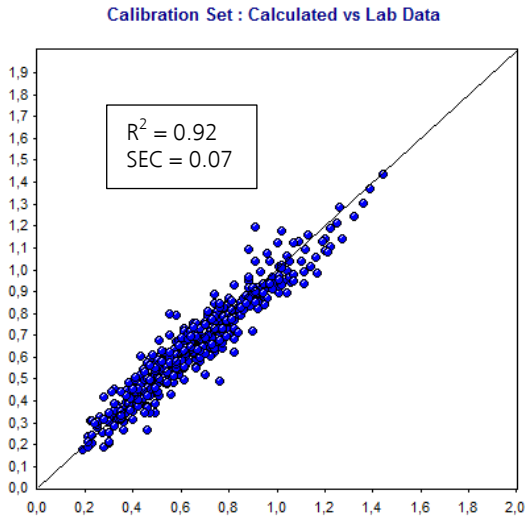
The regression was built using a Partial Least Square (PLS) model with five factors (CRTOT, SEV = 0.07) or four factors (STOT, SEV = 0.38; humidity, SEV = 1.06) over the wavelength ranges of 1200–2300 nm (CRTOT) or 1200–1500 nm plus 1600–2000 nm (TOT and humidity).

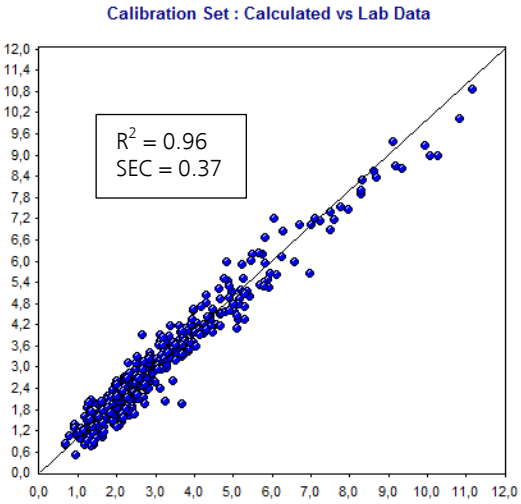
Results for CRTOT:

PLS method development for CRTOT revealed a correlation between the calculated (predicted) and the lab (reference) values. For the calibration set, this correlation was characterized by R^2 = 0.92 and a SEC = 0.07, and by R^2 = 0.95 and SEP = 0.11 for the validation set.



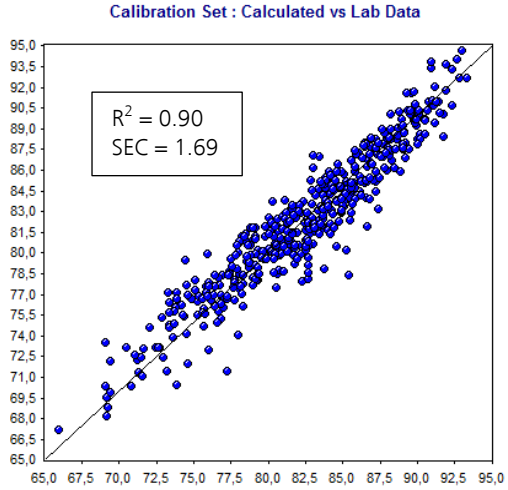
Results for STOT:

PLS method development for STOT revealed a correlation between the calculated and the lab values. For the calibration set, this correlation was characterized by R^2 = 0.96 and a SEC = 0.37, and by R^2 = 0.91 and SEP = 0.42 for the validation set.



Results for Humidity:

PLS method development for humidity revealed a correlation between the calculated and the lab values. For the calibration set, this correlation was characterized by R^2 = 0.90 and a SEC = 1.69, and by R^2 = 0.97 and SEP = 1.81 for the validation set.



Conclusions

With the NIRS DS2500 Analyzer, moisture, fat, and nitrogen content of human feces can be quantified in one run.

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Method description

References

[1] J.H. van de Kamer, H.te Bokkel Huinink, H.A. Weyers: “Rapid method for the determination of fat in feces”, J.Biol.Chem. 1949, 177:347-355,

Literature:

- Fecal-NIRS-Review (JNIRS)
- Comparison of near infrared reflectance analysis of fecal fat, nitrogen, and water with conventional methods, and fecal energy content (Clinical Biochemistry)
- Quantification of fecal carbohydrates by NIR reflectance analysis (Clinical Chemistry)

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