

# Experience with certified reference material (wheat) series for near infrared reflectance/near infrared transmittance equipment

**Judit Budai and Judit Fükö**

*National Office of Measures (OMH), 1535 Budapest, 144, Box 919, Hungary.*

## Certified reference material

For legal certification of the value of a quality or quantity parameter the traceability to the national or international standards should be assured. That means that the measuring equipment used in the laboratory should be compared with a working standard. There is a special type of working standards: the certified reference material, which is able to validate a measuring process and to calibrate an instrument. The Hungarian National Office of Measures (OMH) produces different type of certified reference materials, however, the production of certified plant (wheat)-based CRM has only recently become necessary.

## Necessity

Wheat is the most widely grown plant in Hungary, therefore wheat production is an important determining factor in the Hungarian economy. This is why its objective and unified qualification is of great importance. In wheat trading the price determining parameter is its inner composition. Both the buyer and the seller are interested in having an accurate standardised measurement. A laboratory network with sufficient experience is available for investigations. Proficiency testings organised regularly by OMH in recent years showed that the results of the testings were greatly different from each other and from the average of the measurements: they exceeded the allowed reproducibility. It has been observed that the grading in proficiency testings of the laboratories varies from time to time.

This year they have to acquire a great number of instruments (compared to the previous state)—first of all the fast testing near infrared (NIR) reflectance/NIR transmittance. The new instruments are the Tecator Infracore 1226 and 1275; the Minifra-5—developed and made by the Hungarian Focus GmK, and a large number of Infracore-61 instruments (the first generation of NIR instrument, developed and produced in Hungary) are installed as well.

The calibration of the NIR reflectance/NIR transmittance instruments is performed with properly prepared samples by expert laboratories which belong to different organisations. A recurring problem is whether the instruments should be calibrated according to the results of the central laboratory or they should be calibrated according to the laboratory's own results. What happened in practice, without exception, was that the central calibration was modified to conform to local results. However, this custom is against the trend towards the establishment of unified measurement and makes the test uncertain.

**Table 1. Certified values of the wheat.**

	Samples		
Certified value	CRM 1.7.1./1.	RM 1.7.1./2.	CRM 1.7.1./3.
Protein % (m/m) dry mat. ( $f = 6.25$ )	$16.14 \pm 0.18$	$14.33 \pm 0.16$	$11.75 \pm 0.12$
Wet gluten % (m/m)	$34.37 \pm 0.48$	$29.33 \pm 0.43$	$22.38 \pm 0.32$
Indicative value			
Moisture content % (m/m)	12.38	11.72	11.25

According to our proficiency testings, the situation prevailing is quite contradictory, since there is great variance between the results of both the chemical and the instrumental measurements (in case of wet gluten content with classical methods the value of variance was 1.6%, while that with the NIR reflectance/NIR transmittance method was 2.26%). The results of NIR reflectance/NIR transmittance measurements differed in many cases from the given laboratory's own chemical results, as well as from the "real value" obtained with the classical method in the course of proficiency testing: in case of crude protein content the deviation was 3.08% and in case of measurement of wet gluten it was 6.36%.

Measurement settlement—measurement based on certified reference material (calibration, checking during application)—is considered necessary to make measurements reliable and unified: this is also the precondition of legal measurement.

Among the laboratories in question (because of the increasing importance of the results of measurements) the need for regular proficiency testing—which ensures objective checking for a certain period of time—has increased, as well as for an always available reference material which provides "pilotage", guidance and which can ensure the possibility of continuous control. This is what the "series of wheat-based certified reference material", developed by OMH in the course of recent years' research, can offer to the users (Table 1).

## The role of reference material

Reference material is a measuring means which is suitable for the realisation of the unit and/or the accurate value of a quantity and for its further derivation to other measuring instruments. The accuracy of the measuring instruments used for performing legal measurements, whose verification is not obligatory, must be regularly checked by a certified reference material. Its validity is certified by a competent body which is the National Office of Measures (OMH) in our country.

## Production of the certified reference material

From 1994's crops we selected three pedigreed wheat samples whose difference in protein and wet gluten content exceeds the threefold value of the permitted variance among the laboratories. After appropriate preparation, cleaning, homogenisation and sterilisation, the 600 g samples were put into moisture-proof triplex foil.

We sent the prepared samples to 35 Hungarian and foreign laboratories accredited for the investigation of cereals. They performed the following examinations:

**Table 2. Wet gluten test results using certified reference material (%).**

	Infratec			Minifra-5			Infrapid-61		
1	22.4	29.1	32.9	21.8	28.8	32	21.4	29.5	32.9
2	22	29	33.7	21.5	29	31.9	21.7	29.2	32.4
3	22.8	29	33.7	22.8	28.5	32.3	22.1	30.1	33.6
4	21.7	29.2	34.1	21.8	29.9	33	23.2	30.2	31.6
5	22.3	29	33.9	22.6	30	32.2	22.6	30.3	33.2
6	21.5	29.3	34.1	22.8	29.9	31.7	21.9	28.3	32.8
7				22	28.5	31.8	22.1	29.0	35.3
Average	22.1	29.1	33.7	22.2	29.2	32.1	22.1	29.5	33.1
Std dev.	0.48	0.13	0.45	0.54	0.68	0.44	0.60	0.70	1.15
CRM	22.38	29.33	34.37	22.38	29.33	34.37	22.38	29.33	34.37
Std dev.	0.32	0.43	0.48	0.32	0.43	0.48	0.32	0.43	0.48

- measurement of moisture content by drying the samples at 130–133°C for two hours with five parallels
- determination of crude protein content by measuring the Kjeldahl nitrogen after digestion with five parallels
- wet gluten determination by manual or mechanical gluten washing.

Evaluation of the results was carried out on the basis of ISO Guide 35 Certification of reference materials—general and statistical principles.  $\chi^2$  test shows that measurement data follow the model of normal distribution. Grubb's test indicates that the surveyed database does not contain any outlying value.

In the case of protein and wet gluten the homogeneity of the material and the number of measurements made possible the determination of the consensus value and its variance. We accepted the consensus value as the certified value.

The uncertainty of the certified value concerns the probability of a 95% confidence interval. The validity of the certified value is one year.

## Test results using the certified reference material

In the course of preparation for the 1995 harvest and putting new instruments into action, the calibrating laboratories have completed the calibration of the new and the recalibration of the old instruments with the aid of the aforesaid experiences. The checking of the calibration on various instruments was carried out with certified reference materials at the measurement of wet gluten, which is the worst measured by NIR reflectance/NIR transmittance technique (Table 2).

The results of the measurement show that the application of a certified reference material can greatly improve the reliability of measurement results, to making measurements more exact and to establishing continuous control.