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# Soil Testing

## The benefits of external reference materials for soil analysis

**The agriculture of the near future must be reasoned, profitable, regulated at the production chain level, and clean for its environment and food quality. This requires diagnosis, monitoring and controls, particularly at the soil level. The soil, which is both a support and a source of food for plants, must be analysed to better understand its potential and to make the best choice of crops, maintenance, amendment and fertilisation.**

In this framework, analytical laboratories are subject to an increasing number of requirements regarding the reliability of the results they provide to test prescribers. The response of laboratories has been to adopt recognised quality systems, such as accreditation or approval by Ministries of Agriculture. Thus, the provision of simple results, without any information on their quality or traceability, is no longer considered satisfactory and laboratories must also demonstrate the traceability and reliability of their measurements, from the sample preparation to the final analysis result. Among the tools allowing laboratories to demonstrate and/or control the reliability of their results, participation in proficiency tests and use of reference materials play a major role since they concern accuracy and traceability.

Proficiency testing provides an independent assessment of the laboratory performance comparing its results to those of other laboratories. Participation to proficiency tests is a mandatory requirement of ISO 17025:2017 accreditation[1].

Reference materials are useful for calibration of measuring equipment, quality control of obtained results, development of the method with the study of linearity, specificity, accuracy, fidelity and determination of the limits of detection and quantification, but also for evaluation of measurement uncertainties.

Different definitions of reference materials can be found in literature and normative documents.

The ISO/Guide 30:2015[2] defined and distinguished between reference materials and certified reference materials as follows:

- Reference materials (RM): materials sufficiently homogeneous and stable with respect to one or more specified properties, which has been established to be fit for its intended use in a measurement process.
- Certified reference materials (CRM): samples characterised by a metrologically valid procedure for one or more specified properties, accompanied by a certificate that provides the value of the specified property, its associated uncertainty, and a statement of metrological traceability.

With regards to reference materials (RM), the AFNOR/Guide FD V 03-115[3], specific to the agri-food sector, introduced the concept of external reference materials and internal reference materials, depending on how the reference values were established:

- External reference materials (ERM): reference materials whose consensus values have been determined as a result of inter-laboratory studies, such as inter-comparison tests organised to assess laboratory performance.
- Internal reference materials (IRM): reference materials whose reference values are assigned by the user by comparison with the certified values of a reference material, or by addition of a known quantity of the analyte to the matrix free of that analyte. >

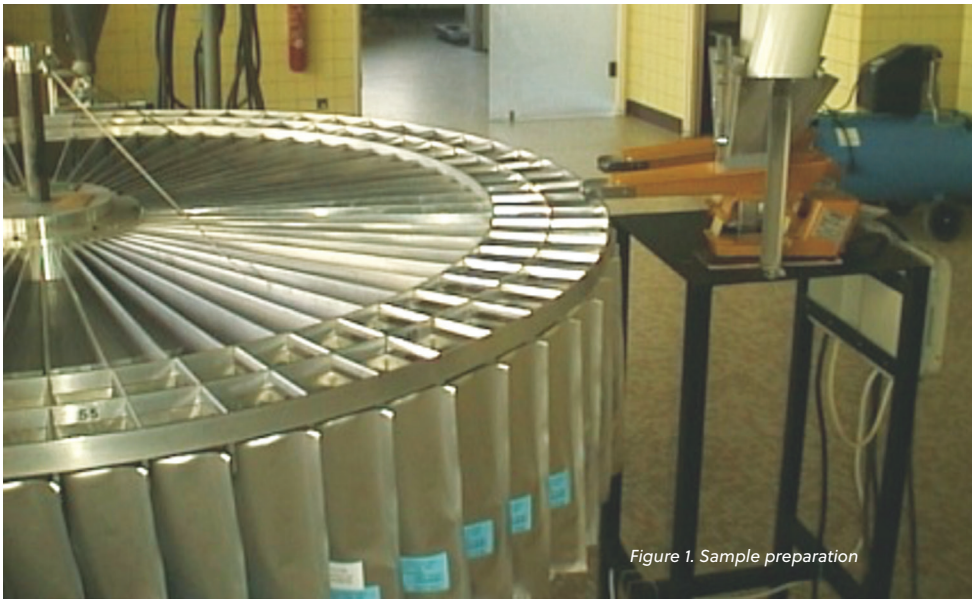

*Figure 1. Sample preparation*

The best-known reference materials are certified reference materials, but their preparation and certification process are time-consuming and require the mobilisation of important means and investment. Due to high price and limited availability of certified reference materials, an increasing number of laboratories prefer to order external reference materials, surplus samples from proficiency testing. However, the use of any reference material, even if it is not certified, must be time-limited, whereas the assigned reference values may change with time, and it is under the responsibility of the material producer to indicate the date until this can be used for calibration purpose with some guarantee.

The stability of a reference material can be demonstrated monitoring over time one or more analytical parameters. It can be the concentration of selected analytes, assuming it is representative of the global contents or sensitive to specific matrix degradation reactions. Reference materials are declared to be suitable if the monitoring analytes concentration remains within a specified interval.

Based on this concept and by reference to ISO 13528:2015[4], BIPEA set up studies to confirm the stability of surplus samples from proficiency testing for exchangeable cations (Calcium, $Ca_2$+, Magnesium, $Mg_2$+, Potassium, K+ and Sodium, Na+) in dried agricultural soil over a period of one year. The determination of exchangeable cations is in fact widely used by agronomists to characterise a soil, along with texture, organic matter content, soil pH and cation exchange capacity.

The levels of these four cations are determined in 95% of the soil tests requested by farmers and interpreted by agronomists. The target period of one year was chosen as a consequence of the availability of surplus samples from proficiency tests. In fact, BIPEA organises 10 proficiency tests per year for physico-chemical analyses of agricultural soil, which correspond to a test every month, except for July and August.

During this period, samples with different profiles are offered to cover a wide spectrum of concentrations of the tested analytes, giving a vast choice to laboratories. An example of production process, homogeneity and stability checks and estimation of reference (assigned) value on sandy soil external reference materials is described just below.

## Production process of external reference materials

### Materials and methods

A batch of sandy soil from Haut-Rhin (France) was air-dried, sieved at 2 mm in accordance with the ISO 11464:2006 standard[5], homogenised and then divided into series of 128 samples of 750g using a carousel. This last is a specific equipment based on the principle of turning spreading, which involves a progressive filling that ensures the homogeneity of the product between all the samples (Figure 1). 80 samples were sent to laboratories participating in the regular proficiency test programme on agricultural soil matrices. Assigned reference values for selected analytes were determined according to the guidelines of ISO 13528:20154, i.e., by taking the robust average of the results reported by a selected group of laboratories participating in the proficiency test, and calculated using algorithm A.

Surplus samples were stored at an ambient temperature (20°C ± 5°C) in thermally sealed packaging to limit water and gas exchanges and to keep them out of direct light.

Thirteen samples were analysed to check the stability of the target analytes. Analyses at $t_0$ were executed on 10 samples before the proficiency test took place (at the same time of the homogeneity check of the prepared batch) and those at $t_1$ on three samples after 13 months, the 27th of November 2018 and the 10th of February 2020, respectively. All analyses were performed in duplicate by an external accredited laboratory, following the technical requirements of the NF X 31-108 standard[6] (determination of cationic bases $Ca_2$+, $Mg_2$+, K+, Na+, that can be exchanged with ammonium acetate - agitation method). The choice of an external laboratory also allows the stability of the samples to be tested under shipping conditions. >

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*Table 1. Reference values and related data for exchangeable cations obtained from PT in sandy soil from Haut-Rhin*

| Analyte | $x_{pt}$ (1) g/kg | $u(x_{pt})$ (2) g/kg | $\sigma_{pt}$ (3) g/kg | $p(x_{pt})$ (4) | CV (5) % |
|---|---|---|---|---|---|
| CaO | 9.046 | 0.158 | 0.716 | 32 | 8 |
| MgO | 0.114 | 0.002 | 0.010 | 33 | 9 |
| K2O | 0.152 | 0.002 | 0.009 | 32 | 6 |
| Na2O | 0.026 | 0.001 | 0.002 | 25 | 8 |

(1). $x_{pt}$: Reference (assigned) value
(2). $u(x_{pt})$: Standard uncertainty of the reference value: $u(x_{pt})=1.25*s*/\sqrt{(p(x_{pt})}$
(3). $\sigma_{pt}$: Standard deviation for proficiency assessment: $\sigma_{pt}=s*$
(4). $p(x_{pt})$: Number of results considered for the estimation of the reference value
(5). CV: Coefficient of variation, standard deviation/reference value, in per cent

As these samples and the associated statistics, inter alia the assigned reference values and their uncertainties, came from a proficiency test, data obtained from the analyses were processed according to the recommendations of the Annex B of ISO 13528:20154. According to this standard, the means of the analyses are compared to each other, and the samples are considered stable if:

$$| \bar{y}_0 - \bar{y}_1 | \leq 0.3\ \sigma_{pt}$$

Where:

- $y_0$: mean of the analyses results at $t_0$,
- $y_1$: mean of the analyses results at $t_1$,
- $\sigma_{pt}$: Standard deviations for proficiency assessment.

Standard deviations for proficiency assessment ($\sigma_{pt}$) of each tested cation were those of the corresponding proficiency test and detailed in Table 1.

### Results and discussion

Reference values of each studied analytes and related statistical data obtained from the results of laboratories are summarised in Table 1. Standard uncertainties, $u(x_{pt})$, that allow quantification of the confidence that can be given to reference values, were calculated as indicated in paragraph 7.7 of the ISO 13528:2015 standard[4]. Coefficients of Variation, CV%, which are often used as standards to describe the precision of the results, are all below 10%, which is satisfactory.

Concerning stability checks, data obtained from the experimental studies are reported in Table 2 and all analytical results of each exchangeable cations (CaO, MgO, $K_2O$ and $Na_2O$) are showed graphically in Fig. 2. In these graphs, means of each analysis and the interval $\pm$ 0.3 $\sigma_{pt}$ , are also traced.

The treatment of experimental data confirmed a sufficient stability of the samples over the tested period and under defined storage and shipping conditions, as the difference between means at $t_0$ and at $t_1$ are all equal or inferior to the allowed ones for each tested parameter.

Results obtained in this study enabled to prove the interest of external reference materials from proficiency testing, both in terms of reliability of reference values and sample stability.

## Conclusions

Reference materials are reliable quality assurance tools that improve confidence in test results obtained by laboratories. They play a key role in calibration of laboratory instruments by providing precise reference values and data. However, reference materials must satisfy a specific task and so must be selected with great care. The main prerequisite of a reference material is that it should be as representative of the real sample as possible. This requirement of representativeness means that reference material should be similar to real routine samples in matrix composition and concentration level of measured substances.

> "more laboratories are moving towards external reference materials"

Laboratories often have difficulties finding real reference samples as certified reference materials are often synthetic materials and produced in small series. External reference materials on agricultural soil matrix answer to this request. Laboratories can choose between a wide range of soil samples with different concentrations of analytes within one year as proficiency tests are proposed regularly. This is particularly useful for equipment calibration or for development and validation of new methods.

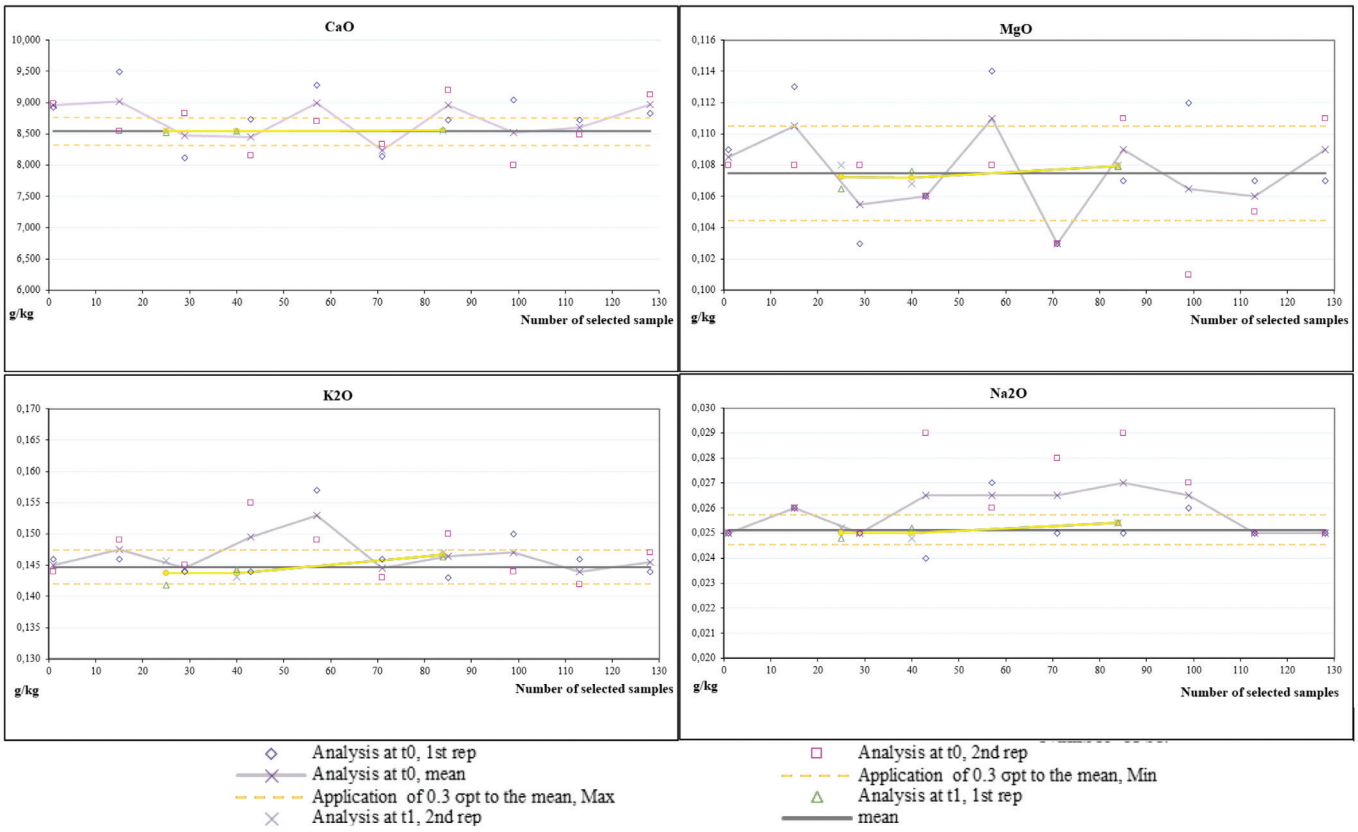
As a result, more and more laboratories are moving towards external reference materials even if their quality guarantees are different from those of certified ones. ■

*Table 2. Results of the stability studies on exchangeable cations in soil over a period of 13 months*

| Analyte (g/kg) | CaO | MgO | K2O | Na2O |
|---|---|---|---|---|
| Mean of the analyses results at $t_0$[1], $y_0$ | 8.718 | 0.108 | 0.147 | 0.026 |
| Mean of the analyses results at $t_1$[2], $y_1$ | 8.544 | 0.107 | 0.145 | 0.025 |
| Absolute value of the difference allowed, 0.3 $\sigma_{pt}$ | 0.215 | 0.003 | 0.003 | 0.001 |
| Absolute value of the observed difference, $\| y_0 - y_1 \|$ | 0.174 | 0.001 | 0.002 | 0.001 |

1. $t_0$: November 27, 2018
2. $t_1$: February 10, 2020

*Figure 2. Results of the stability checks of exchangeable cations in soil as a function of the production order*




*References*

1. *International standard: ISO/IEC 17025:2017 – General requirements for the competence of testing and calibration laboratories.*
2. *International standard: ISO/Guide 30:2015 – Reference materials – Selected terms and definitions.*
3. *French national standard: AFNOR/Guide FD V 03-115:1996 Analysis of agricultural and food products. Guide for the use of reference materials.*
4. *International standard: ISO 13528:2015 – Statistical methods for use in proficiency testing by interlaboratory comparisons.*
5. *International standard: ISO 11464:2006 – Soil quality – Pretreatment of samples for physico-chemical analysis.*
6. *French national standard: NF X 31-108:2002 – Soil quality - Determination of ammonium acetate extractable Ca++, Mg++, K+ and Na+ cations - Agitation method.*

*Author*

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