Uncertainty Assessment of Kjeldahl-measured Total Nitrogen in Black Soil, Corn and Soybean Grains: Implications for Agricultural Remote Sensing Ground Truth Validation

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Abstract. Ground truth values for total nitrogen content in farmland soil and crops are critical for agricultural remote sensing inversion, spatial nutrient mapping, and product validation. While laboratory measurement results are typically treated as unbiased references, the uncertainties inherent in the analytical process remain overlooked. These uncertainties are coupled with model inversion biases and pixel-scale spatial representativeness errors, making it difficult to effectively isolate them from the root mean square error (RMSE) during validation, thereby limiting the evaluation of high-precision remote sensing products. In this study, soil, corn, and soybean samples from farmland in Northeast China were selected. Total nitrogen was determined by the Kjeldahl method, and a bottom-up uncertainty assessment was conducted following the "Guide to the Expression of Measurement Uncertainty". The results indicate that titration endpoint judgment, incomplete sample digestion, and repeated measurements represent the dominant sources of uncertainty during analysis, with soil showing higher relative uncertainty than crops. Although the measurement uncertainty is lower than the RMSE typically observed in conventional remote sensing inversions, its constraining effect on the quality of ground truth validation cannot be overlooked. As agricultural remote sensing products evolve toward higher precision inversions, the constraining effect of laboratory analytical uncertainty will become increasingly significant; therefore, it should be archived as key metadata alongside the ground truth data. This study provides a reusable geochemical testing quality reference for conducting space-ground collaborative remote sensing observations in black soil agricultural regions, supporting enhanced reliability of ground truth datasets and greater rigor in the authenticity verification of remote sensing products.

Keywords: Total nitrogen, Measurement uncertainty, Ground truth data, Kjeldahl method, Black soil.

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1 Introduction

Total nitrogen content in soil and crops is a key indicator that characterizes soil fertility levels and plant nutrient conditions. Accurate determination provides fundamental support for farmland nutrient management, fertilization strategies, and sustainable agricultural development [1,2]. With the advancement of precision agriculture, comprehensive and high-frequency monitoring of nitrogen in both soil and crops is becoming increasingly critical. Hyperspectral and multispectral remote sensing data combined with machine learning and physical model inversion methods have enabled quantitative estimates of total nitrogen in soil and crops at a regional scale [3].

However, the essence of validation is to evaluate the uncertainty and applicability of remote sensing products by using ground measurement data as a reference [4]. However, all chemical testing procedures introduce unavoidable analytical uncertainty [5], which is often coupled with factors such as model inversion bias at the pixel scale in the error budget of remote sensing validation, making it difficult to effectively isolate it from the total root mean square error (RMSE) [6,7]. In areas with highly heterogeneous agricultural landscapes, the spatial mismatch between observations at the point scale on the ground and data at the pixel scale from satellites is particularly pronounced [8]. As agricultural remote sensing products evolve toward higher precision inversions, the constraining effect of laboratory analysis uncertainties will become increasingly significant; neglecting their contribution may lead to an overestimation of product accuracy or incorrect attribution of error sources.

Previous uncertainty assessments have primarily focused on validating the methods or reported measurement uncertainties for individual substrates [9, 10]. There remains a lack of uncertainty reference schemes designed for comparisons across multiple substrates that can support remote sensing validation requirements. This study focused on soil, corn, and soybeans from the black soil in Northeast China. Total nitrogen content was determined using the Kjeldahl method, and uncertainty components at each stage of the measurement process were systematically assessed in accordance with the "Guide to the Expression of Measurement Uncertainty (GUM)" guidelines. This study aims to clarify the distribution characteristics and primary sources of uncertainty across different matrices and across different nitrogen content levels, thereby providing a quantifiable reference for chemical testing uncertainty to support the quality archiving of ground truth datasets for agricultural remote sensing.

2 Methodology

2.1 Sample Collection and Treatment

Ten samples each of corn grain (C1–C10), soybean grain (SO1–SO10), and soil (S1–S10) were collected separately from a black soil farmland at the agricultural practice base of Jilin University. The fresh plants were washed and placed in an oven at 105 °C for 30 min, then dried at 80 °C to constant weight. Samples were then passed through a sieve with a 0.147 mm mesh and collected for later use.

2.2 Determination of Nitrogen

Plants (0.5 g) or soil (1.0 g) were weighed and transferred into a digestion tube, followed by addition of 5.0 g copper sulfate catalyst and 10 mL of concentrated sulfuric acid. Samples were digested in a digestion furnace at 480 °C for 1 h. Anhydrous sodium carbonate was used to calibrate the standard hydrochloric acid titrant according to "GB/T 601-2016". Three certified reference materials and replicate subsamples were analyzed alongside experimental samples under identical conditions to assess measurement accuracy and precision.

For distillation titration, 50 mL of 2% boric acid solution and three drops of the indicator were transferred into a conical flask, which was placed under the distillation outlet of the Kjeldahl apparatus to absorb released ammonia gas until the collected distillate reached approximately 150 mL. Afterwards, the solution was titrated with calibrated hydrochloric acid on a magnetic stirrer. The titration endpoint was defined as a stable color change from blue to pink that persisted for at least 30 s. The consumed volume of hydrochloric acid was recorded for subsequent total nitrogen calculation.

2.3 Creation of Uncertainty Model

The mathematical model for the determination of nitrogen content in plants and soil by the Kjeldahl method is as follows:

$$\omega_N = ((c \times (V - V_0) \times 14)/m) \times 1000 \quad (1)$$

where ω_N is the nitrogen content of samples (mg/kg), c is the concentration of hydrochloric acid (0.1 mol/L), V is the volume of hydrochloric acid consumed by samples (mL), V_0 is the volume of hydrochloric acid consumed by blank sample (mL), m is the sample mass (g), and 14 is the molar mass of nitrogen atom (g/mol).

3 Results and Discussion

3.1 Characteristics of Experimental Samples

The total nitrogen concentrations of soil, corn, and soybean grains are summarized in Table 1. The total nitrogen content ranged from 0.179 % to 0.275 % in soil. The total nitrogen content in corn grain varied between 1.211 % and 1.391 %, while that in soybean grain was much higher, ranging from 5.895 % to 6.273 %. The wide range of nitrogen concentrations across soil and crop specimens enables comparison of measurement uncertainty performance under low and high nitrogen matrix conditions.

3.2 Identification of Uncertainty Sources

The sources of uncertainty during sample digestion and testing mainly include the following five aspects: sample weighing, hydrochloric acid calibration, titration endpoint, sample digestion, and repetitive measurement (Fig. 1).

1. Sample weighing uncertainty $u_{rel(1)}$ originates from balance tolerance and weighing repeatability. The balance tolerance is treated as a type-B uncertainty component based on the manufacturer's instrument specification, assuming an appropriate probability distribution to convert the given tolerance into standard uncertainty. Weighing repeatability is derived from repeated weighing operations and evaluated as a type-A statistical component.

Table 1. Total nitrogen content of experimental samples.

Sample ID	N (%)	Sample ID	N (%)	Sample ID	N (%)
S1	0.179	C1	1.211	SO1	5.895
S2	0.183	C2	1.321	SO2	5.914
S3	0.190	C3	1.267	SO3	6.230
S4	0.195	C4	1.354	SO4	6.147
S5	0.205	C5	1.288	SO5	6.080
S6	0.212	C6	1.264	SO6	5.989
S7	0.225	C7	1.347	SO7	6.248
S8	0.238	C8	1.314	SO8	6.134
S9	0.262	C9	1.383	SO9	6.252
S10	0.275	C10	1.391	SO10	6.273

2. Uncertainty during HCl calibration $u_{rel(2)}$ comes from reference reagent purity, weighing of sodium carbonate, burette volume error, and visual endpoint judgment in titration, as well as repeatability of calibration operations. The purity of the reference reagent and nominal burette volume error are treated as type-B components according to reagent certificates and instrument metering specifications. The uncertainty from sodium carbonate weighing follows the same evaluation logic described above for sample weighing. Visual endpoint judgment is estimated from practical titration experience. The repeatability of calibration operation is obtained through multiple independent calibration runs and evaluated by type-A statistics.

3. Visual judgment of titration endpoint is an important error source $u_{rel(3)}$, which is affected by burette precision and human color discrimination. Burette precision is considered a type-B component according to instrument tolerance. Human color recognition bias is estimated from practical laboratory observation of color transition ranges during titration experiments.

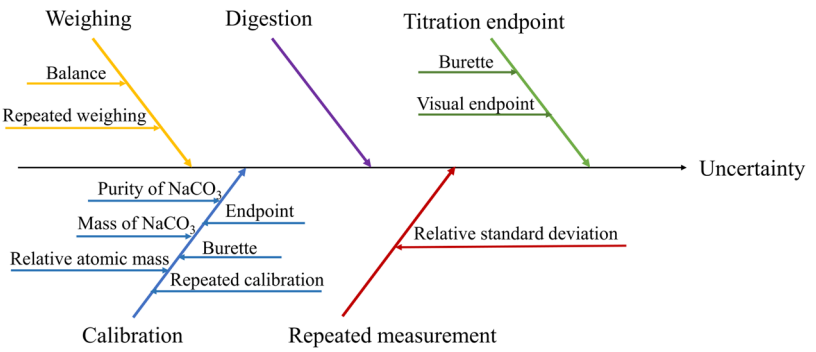


Fig. 1. Fishbone diagram depicting sources of uncertainty.

4. Incomplete digestion $u_{rel(4)}$ may lead to low nitrogen recovery and introduce additional bias, especially for complex soil matrices. This component is estimated from the recovery performance obtained in spiked or reference material tests, reflecting potential systematic deviation introduced by the digestion procedure.

5. Repetitive measurement uncertainty $u_{rel(5)}$ reflects random experimental variation across parallel replicates. This random uncertainty is calculated as a type-A uncertainty based on experimental results of multiple parallel sample digestions and measurements. All individual components were combined using the root-sum-of-squares propagation rule to obtain the relative standard uncertainty (u_{rel}), the combined standard uncertainty (u_c), and the extended uncertainty (U):

$$u_{rel} = \sqrt{u_{rel(1)}^2 + u_{rel(2)}^2 + u_{rel(3)}^2 + u_{rel(4)}^2 + u_{rel(5)}^2} \quad (2)$$

$$u_c = u_{rel} \times \omega_N \quad (3)$$

$$U = 2u_c \quad (4)$$

3.3 Analysis of Dominant Uncertainty Sources and Inter-Matrix Comparison

Through evaluating the uncertainty in nitrogen content determination for grain and soil samples, it was concluded from comparing five uncertainty sources that the main factors affecting the measurement uncertainty are titration endpoint, incomplete digestion, and repeated measurement (Fig. 2; Table 2). It is also clear from Fig. 2 that the overall relative standard uncertainty differed highly significantly among matrices (ANOVA, $p<0.0001$); soil samples presented the highest relative uncertainty, followed by soybean grain and corn grain.

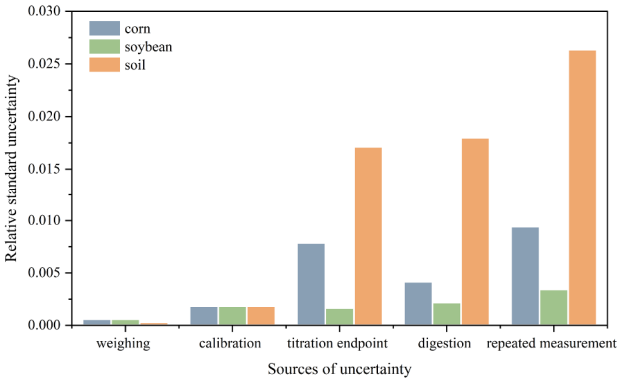


Fig. 2. Contributions of five uncertainty sources to total nitrogen measurement in corn, soybean, and soil samples.

In terms of matrix type, intrinsic material differences between soil and grain partly produce the observed gap in measurement uncertainty. From the perspective of sample heterogeneity, black-soil consists of complex mixtures of mineral and organic

components, which increases the risk of incomplete digestion and amplifies measurement uncertainty; in contrast, crop grain matrices are chemically more homogeneous and suffer less digestion-related uncertainty.

For nitrogen-concentration effects, soil possesses inherently lower baseline nitrogen content compared with plant grains. Within the soil matrix, higher total-nitrogen concentration corresponds to slightly lower relative combined uncertainty, although absolute expanded uncertainty increases with nitrogen level. These systematic effects of nitrogen concentration, sample heterogeneity, and matrix type should be considered when interpreting ground-truth measurement results for agricultural remote-sensing validation.

Table 2. Uncertainty of experimental samples.

Sample	u_{rel}			U (%)		
	Corn	Soybean	Soil	Corn	Soybean	Soil
1	0.0104	0.0037	0.0364	0.0253	0.0435	0.0130
2	0.0098	0.0037	0.0359	0.0258	0.0436	0.0132
3	0.0102	0.0036	0.0356	0.0258	0.0452	0.0135
4	0.0098	0.0036	0.0354	0.0264	0.0446	0.0138
5	0.0101	0.0036	0.0328	0.0259	0.0441	0.0134
6	0.0102	0.0037	0.0324	0.0257	0.0437	0.0137
7	0.0098	0.0037	0.0312	0.0263	0.0456	0.0141
8	0.0101	0.0036	0.0296	0.0264	0.0444	0.0141
9	0.0094	0.0036	0.0288	0.0260	0.0452	0.0151
10	0.0093	0.0036	0.0277	0.0259	0.0452	0.0153

3.4 Implications for Agricultural Remote Sensing Ground Truth Validation

Although the expanded uncertainty obtained in this study is generally lower than the typical RMSE reported in current remote sensing inversions (0.03%–0.12%), it represents an irreducible error floor for ground-truth validation. When inversion RMSE remains much larger than U, laboratory-derived uncertainty contributes a minor fraction of total validation error and exerts limited influence on accuracy assessment. However, when the inversion RMSE decreases to values comparable to U, further improvements in model algorithms or spectral band selection would yield diminishing returns in validation metrics, because validation performance can no longer exceed the intrinsic uncertainty of ground-reference measurements, as model-derived errors become obscured by ground-measurement noise, preventing reliable discrimination of genuine model improvements. For low-nitrogen black soil matrices, although the absolute U appears minor, its relative proportion reaches 4.7%–7.2% of the measured value. This level of analytical fuzziness has direct implications for pixel-based fertility classification. For instance, when a laboratory reports nitrogen content of 0.195% with $U = 0.014\%$, the true value may statistically range from 0.181% to 0.209%. This overlap covers the lower to medium fertility thresholds commonly used in agricultural zoning. Consequently, remote sensing inversions that achieve high spatial consistency with the measured point value might actually be coincidentally matching a random fluctuation of the wet-chemistry process rather than capturing the true spatial variability of the field.

In the context of satellite pixel-scale validation, laboratory measurement uncertainty is often aggregated with spatial representativeness errors arising from sub-pixel heterogeneity. This work suggests that when conducting ground-truth archiving, the laboratory uncertainty should be recorded separately from the spatial standard deviation of field replicates, thereby preventing overestimation of model bias caused by ground-reference fluctuations. Especially for homogeneous black soil croplands, where spatial variance is relatively low, the analytical uncertainty becomes a dominant confounder that must be explicitly accounted for in high-precision nutrient mapping.

4 Conclusions

This study implemented a workflow compliant with the bottom-up GUM evaluation for Kjeldahl-measured total nitrogen in black soil, corn grain and soybean grain samples from farmland in Northeast China. Titration endpoint judgment, incomplete sample digestion, and repeated measurements constitute the dominant uncertainty contributors, while weighing and hydrochloric acid calibration contribute marginally. Low-nitrogen soil exhibits higher relative expanded uncertainty compared with high-nitrogen crop grains, even though its absolute uncertainty value remains moderate. Strict control over experimental operations can reduce analytical uncertainty. In addition to laboratory operation optimization, this study highlights that measurement uncertainty derived from laboratory analysis should be archived as essential metadata for ground truth datasets used in agricultural remote sensing validation. The presented assessment workflow offers a reusable geochemical quality control reference for collaborative observation across space and ground over black soil agricultural regions.

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Disclosure of Interests

The authors have no competing interests.

References

1. H. Yang, J. Yan, G. Li, W. Ma, X. Yao, J. Li, Q. Da, X. Li, K. Cheng, Inversion Model for Total Nitrogen in Rhizosphere Soil of Silage Corn Based on UAV Multispectral Imagery, Drones 2025, 9(4), 270.
2. M. Yang, A. Mouazen, X. Zhao, X. Guo, Assessment of a soil fertility index using visible and near-infrared spectroscopy in the rice paddy region of southern China, European Journal of Soil Science 2020, 71(4), 615-626.

3. Z. Xu, S. Chen, B. Zhu, L. Chen, Y. Ye, P. Lu, Evaluating the Capability of Satellite Hyperspectral Imager, the ZY1–02D, for Topsoil Nitrogen Content Estimation and Mapping of Farmlands in Black Soil Area, China, Remote Sensing 2022, 14(4), 1008.
4. X. Wu, Q. Xiao, J. Wen, D. You, A. Hueni, Advances in quantitative remote sensing product validation: Overview and current status, Earth-Science Reviews 2019, 196, 102875.
5. E. Trikilidou, G. Samiotis, L. Tsikritzis, E. Amanatidou, Performance of Semi-Micro-Kjeldahl Nitrogen Method – uncertainty and nitrate interference, International Journal of Environmental Analytical Chemistry 2022, 102(18), 6204-6214.
6. X. Li, S. Liu, J. Ding, L. Song, T. Xu, Y. Ma, Z. Xu, X. Yang, Y. Zhang, J. Wang, A Framework for Quantifying the Uncertainty in Upscaling Evapotranspiration from Homogeneous to Heterogeneous Underlying Surface, IEEE Transactions on Geoscience and Remote Sensing 2024, 62, 1-24.
7. J. Wen, X. Wu, J. Wang, R. Tang, D. Ma, Q. Zeng, B. Gong, Q. Xiao, Characterizing the Effect of Spatial Heterogeneity and the Deployment of Sampled Plots on the Uncertainty of Ground "Truth" on a Coarse Grid Scale: Case Study for Near-Infrared (NIR) Surface Reflectance, Journal of Geophysical Research: Atmospheres 2022, 127(11), e2022JD036779.
8. J. Chen, R. Huang, L. Luo, S. Yi, Y. Qin, W. Qi, H. You, X. Han, G. Zhou, A multi-tiered representativeness framework mitigating spatial scale effects in FVC validation: UAV-based assessment of global products in Qinghai-Tibetan Plateau alpine grasslands, International Journal of Applied Earth Observation and Geoinformation 2025, 143, 104794.
9. W. Bich, M.G. Cox, P.M. Harris, Evolution of the 'Guide to the Expression of Uncertainty in Measurement', Metrologia 2006, 43(4), S161.
10. T. Anglov, I.M. Petersen, J. Kristiansen, Uncertainty of nitrogen determination by the Kjeldahl method, in: P. De Bièvre, H. Günzler (Eds.), Measurement Uncertainty in Chemical Analysis, Springer Berlin Heidelberg, Berlin, Heidelberg, 2003, pp. 273-279.

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