

LETTER

Accuracy and precision of no instrument is guaranteed: a reply to Rosenstock *et al.*

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Photoacoustic infrared spectroscopy (PAS) is increasingly used for measurement of N₂O and CO₂ fluxes at the soil surface. However, PAS calibration is complex. Water vapor, CO₂, and temperature interfere with accurate N₂O measurement. To accurately measure N₂O, PAS calibrations must compensate for these interferences. Our article, 'Evaluation of photoacoustic infrared spectroscopy for the simultaneous measurement of N₂O and CO₂ gas concentrations and fluxes at the soil surface' (Iqbal *et al.*, 2013), compared PAS and gas chromatography (GC) analytical procedures. Results demonstrated that PAS can measure N₂O concentrations (ca. 0.5–3.0 ppm) and fluxes (ca. 0.5–5.0 ppm min⁻¹) with accuracy and precision similar to GC without interferences from H₂O vapor or CO₂ concentrations typically encountered in static flux chambers at the soil surface.

In response, Rosenstock *et al.* (2013) initiated a valuable dialogue by reminding readers that analytical instrument accuracy and precision are not guaranteed. Although we agree with this primary assertion, several other assertions in the authors' letter are flawed. Based on two PAS evaluations that produced inaccurate N₂O measurements at low concentrations (ca. 0.0–0.7 ppm), Rosenstock *et al.* (2013) suggested that the results of Iqbal *et al.* (2013) were limited to unique experimental conditions including relatively high N₂O concentrations and fluxes. Ultimately, the authors questioned the ability of PAS to be calibrated for accurate measurement of N₂O concentrations and fluxes across the ranges encountered during standard operating conditions.

We highlight published data, new experimental results, and differences in calibration procedures that indicate Rosenstock *et al.* (2013) evaluated improperly calibrated instruments. We extend the experiments of Iqbal *et al.* (2013) to low N₂O concentrations and clarify the importance of evaluating high N₂O concentrations and fluxes. Finally, we suggest procedures to avoid faulty PAS calibrations.

The PAS evaluations conducted by Rosenstock *et al.* (2013) demonstrated: (i) two PAS units did not

accurately measure three N₂O concentrations in analytical standards (hereafter PAS 1 and PAS 2); and (ii) measurements of N₂O concentrations with a third PAS unit experienced significant interferences from H₂O vapor and/or temperature (hereafter PAS 3). Relative to our evaluations, they stated:

'The most parsimonious explanations for variation among instrument performance are differences in experimental conditions or calibration algorithms that account for interference among gas and water vapor absorption spectra and for cross-interferences among the targeted molecules. However, differences may also be attributable to the gas concentrations tested.'

Although the authors identified three potential causes for the differences among instrument performance, they devoted discussion to two of the three possibilities: experimental conditions and tested N₂O concentrations. They did not describe calibration procedures or discuss the potential for improper calibration. Nevertheless, the authors extended results from one experiment with one PAS unit to suggest PAS cannot be calibrated to produce accurate N₂O measurements due to uncorrectable interferences from nontarget gases and temperature.

We present three lines of evidence that indicate the authors evaluated improperly calibrated instruments:

First, Rosenstock *et al.* (2013) incorrectly asserted that 'Data from our experiments suggest PAS is less accurate and precise than Iqbal *et al.* (2013) suggest.' Both assertions in this statement are false: Although a quantum cascade laser was used to verify N₂O concentrations used in evaluations of PAS 3, the accuracy of 'known gas concentrations' used in evaluations of PAS 1 and PAS 2 is not reported nor independently verified with a different analytical technique; thus it is impossible to determine accuracy. Communications with the authors indicated that the 0.331 PPM and 0.649 ppm N₂O standards used in evaluations of PAS 1 and PAS 2 were accurate at ±10% (T. Rosenstock, personal communication). Also, PAS calibration is typically accurate only within ±5% of an

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independent standard (subject to ± 5 –10% error) used by the vendor to test the calibration. Regardless, the most parsimonious explanation for inaccurate standard measurement with an analytical instrument that has been demonstrated to be accurate is: *calibration error*.

Moreover, PAS precision reported by Rosenstock *et al.* (2013) and Iqbal *et al.* (2013) did not differ. Precision reported by Iqbal *et al.* (2013) ranged from 1.2 to 2.5% while precision reported by Rosenstock *et al.* (2013) ranged from 1.2 to 5.4%. Both ranges favorably compare to GC (Iqbal *et al.*, 2013; Tirol-Padre *et al.*, 2013).

Second, we conducted a new experiment similar to the authors' evaluation of PAS 3. This experiment tested for interactive effects of H₂O vapor and temperature on low-concentration N₂O measurements. In contrast to data presented by Rosenstock *et al.* (2013), we observed no H₂O vapor or temperature interferences with measurements of atmospheric and NIST-certified N₂O concentrations (Figure 1).

Third, Zhao *et al.* (2012) demonstrated that internal or external PAS calibration algorithms can be developed to produce accurate N₂O measurements at concentrations >0.03 PPM if cross-interfering gases (i.e. H₂O vapor and CO₂) are measured and accounted for in calibration algorithms. If environmental conditions produce inaccurate measurements, it is not because calibrations cannot account for interferences (assuming H₂O vapor and CO₂ are the only interfering gases); it is because the calibration algorithms are insufficient or

cross-interfering gases are unaccounted for. Other than H₂O vapor and CO₂, we know of no gases that interfere with N₂O.

Rosenstock *et al.* (2013) further suggested that our evaluations were limited because we worked at high N₂O fluxes:

'Iqbal *et al.* (2013) report changes in headspace concentrations of 50–600 ppb N₂O min⁻¹, roughly equivalent to 600–7000 g N₂O-N m⁻² h⁻¹ (or 50–600 kg N ha⁻¹ yr⁻¹) when assuming a 15 cm high chamber. Soil fluxes of that magnitude are rarely found and only occur under high emission conditions.'

Although evaluations in Iqbal *et al.* (2013) were conducted at high fluxes, the minimum flux detection limit was calculated and reported as 0.0033 ppm min⁻¹ (assuming five gas concentration measurements during an 8 min chamber closure). This was compared to the GC N₂O minimum flux detection limit calculated as 0.0007 ppm min⁻¹ (assuming three gas concentration measurements during a 45 min chamber closure). Additionally, Iqbal *et al.* (2013) explicitly identified two reasons for working at high N₂O concentrations: First, Ambus & Robertson (1998) previously determined that PAS and GC produce 'statistically identical' N₂O flux estimates at extremely low fluxes. Given that we stated 'Ambus & Robertson (1998) demonstrated similar N₂O

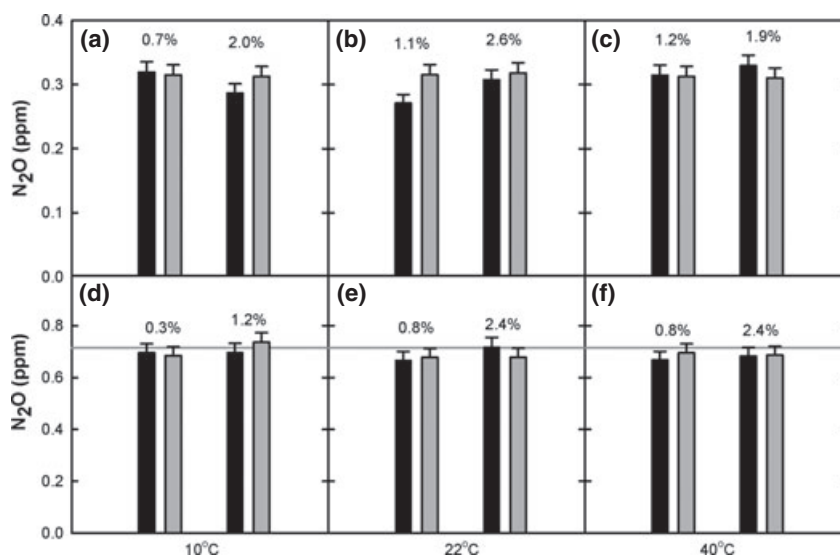


Fig. 1 Photoacoustic infrared spectroscopy (PAS; black bars) and gas chromatography (GC; gray bars) measurements of ambient air (a, b, c) and a National Institute of Science and Technology-certified (NIST) concentration of N₂O (0.718 ± 0.0017 ppm) (d, e, f) at three temperatures (10 °C, 22 °C, 40 °C) at variable humidity. The reference line (panels d, e, f) is the NIST-certified N₂O concentration. Percent values displayed above paired PAS and GC bars are the humidity values in each gas bag at each run. Each PAS bar represents the average of three consecutive measurements. Each GC bar indicates average of three samples withdrawn from each gas bag during PAS measurement. To facilitate comparison among methods, error bars indicate $\pm 5\%$ of each column (i.e. the average of replicate measurements with each instrument).

flux measurements among PAS and GC at low fluxes from 0.0005 to 0.0202 ppm min⁻¹ (ca. 0.46–18 kg N₂O-N ha⁻¹ yr⁻¹) we are surprised that the authors did not address this report. Second, the primary objective of Iqbal *et al.* (2013) was to test for H₂O vapor and CO₂ interferences with N₂O measurements because such interferences have been suggested to explain inaccurate N₂O measurements with PAS (Akdeniz *et al.* 2009). Water vapor and CO₂ concentrations are expected to produce the most interference with N₂O detection at high concentrations (Zhao *et al.*, 2012) and concentrations of these gases in static chambers are positively correlated (Xu *et al.*, 2008).

Further, the idea that N₂O fluxes of 0.05 PPM min⁻¹ are 'rarely found' is ambiguous. Such fluxes are not uncommon in N-fertilized agroecosystems, the largest anthropogenic source of N₂O and the focus of most N₂O measurements. Short periods of high emissions account for a disproportionate amount of cumulative annual emissions. In our data collected with GC from conventionally managed corn-soybean systems in Iowa, USA fluxes ≥ 0.5 PPM min⁻¹ accounted for ca. 40% of annual emissions, and a recent comparison of PAS and GC demonstrated that accurate measurement of high N₂O fluxes is the most important factor affecting variations in annual flux estimations (Tirol-Padre *et al.*, 2013).

We maintain that PAS *can* be calibrated to provide accurate N₂O measurements across ranges of CO₂, H₂O vapor and temperature that are encountered in static chambers at the soil surface. In summary: (i) four independent studies have reported positive evaluations of PAS across the range of N₂O fluxes encountered at the soil surface (<0.01 to >5.7 mg N₂O-N m⁻² h⁻¹) without interferences from H₂O vapor or CO₂ (Ambus & Robertson, 1998; Yamulki & Jarvis, 1999; Iqbal *et al.*, 2013; Tirol-Padre *et al.*, 2013); (ii) analytical simulations and experimental evaluations demonstrated that internal PAS calibration algorithms can eliminate cross-interferences from H₂O vapor and CO₂ (Zhao *et al.*, 2012); and (iii) the only data Rosenstock *et al.* (2013) presented regarding temperature and H₂O interferences are derived from a single experiment with $N = 1$.

Our PAS units were calibrated by California Analytical Instruments (Orange, CA, USA) whereas the authors' were calibrated by Lumasense Technologies (Santa Clara, CA, USA). A major difference in calibration procedures was the number of calibration points for CO₂; our machines used a two-point CO₂ calibration whereas the authors' used a one-point CO₂ calibration (T. Rosenstock, personal communication). This may be one factor contributing to inaccurate N₂O measurements (improper CO₂ interference calibrations will produce inaccurate calibration algorithms for N₂O; Zhao

et al., 2012). To avoid performance issues, PAS users should request confirmation of accurate measurement of a low N₂O concentration at high H₂O vapor and CO₂ concentrations. Users can also remove H₂O vapor and/or CO₂ during gas intake. As with all analytical equipment that is calibrated by a vendor, users must independently verify performance. Tests of PAS calibration accuracy should not be limited to gas standards in a N₂ balance with high potential error (e.g., ± 5 –10%).

Several manufacturers and manufacturer representatives market PAS units. Significant variation in calibration accuracy is probable. We highlight that Iqbal *et al.* (2013), and other reports cited herein, demonstrated that PAS *can* accurately measure a range of N₂O concentrations encountered in static chambers at the soil surface. This does not guarantee accuracy of PAS calibration.

We agree with Rosenstock *et al.* (2013): 'a better understanding of the mechanisms driving variation in PAS performance' is needed, and 'one should be careful when considering using PAS for measurements of greenhouse gases'. However, the data presented by Rosenstock *et al.* (2013) fall far short of suggesting that PAS cannot be calibrated for accurate measurement of N₂O concentrations encountered in static chambers at the soil surface.

Acknowledgement

This research was funded by USDA-NIFA, Award No. 2011-68002-30190.

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