NOTES

AUTOMATED ANALYSIS OF NITROUS OXIDE

T.B. PARKIN

Abstract

An autosampler was developed to interface with a gas chromatograph for N$_2$O analyses. The sampler was comprised of a modified fraction collector and pneumatic valves all controlled by a microprocessor unit. Cost of the autosampler system including the fraction collector was estimated to be less than $3500. The sampler was designed to analyze gas samples stored in rubber-stoppered, glass vials. Use of vials resulted in a slight loss of sensitivity due to dilution and absorption of N$_2$O; however, accuracy was the same as could be obtained by manual syringe injections.

Additional Index Words: gas chromatography, autosampler, denitrification, laboratory-automation

Materials and Methods

Autosampler

Gas samples (0.005 L) were collected with a syringe from incubation vessels (either stoppered flasks containing soil slurries or stoppered plexiglass tubes containing intact soil cores). Gas samples were injected into 0.003 L evacuated vials (sterile, no additive, silicone coated; Becton Dickinson Co., Rutherford, NJ), resulting in an overpressure of gas in the vials. Actual volumes of the vials were determined to be in the range of 0.00358 to 0.00368 L. Vials were stored for later analysis of O$_2$, CO$_2$ and N$_2$O which were performed using a $^{63}$Ni electron capture detector gas chromatograph (Tracer Model 222). To prevent interference of H$_2$O and C$_2$H$_2$ with detector operation, dual columns (2m long x 0.0032 m O.D.; packed with Porapak-QS 50/80 mesh) were installed. An 8-port switching valve allowed for one column to be on line to the detector while the second column was backflushed to remove interfering compounds. Gas samples were injected onto column with a 6-port sample valve fitted with a 0.0005 L sample loop. Both the 8-port switching valve

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3 Mention of a product by company or name is not an expressed or implied endorsement by the United States Department of Agriculture.
and the 6-port sample valve were driven by solenoid triggered pneumatic actuators (Valco Instruments Co., Houston, TX). Automatic transfer of sample from the vials to the sample loop was achieved by modifying a fraction collector (LKB 7000, L.K.B. Instruments, Gaithersburg, MD). A pneumatic cylinder (0.019 m bore × 0.0254 m stroke; Control Line Equipment Inc., Cleveland, Ohio) was secured to the fraction collector in a position over the sample vials. The push rod of the pneumatic cylinder was fitted with a side arm needle fitting (Popper & Sons Inc., New Hyde Park, NJ) and a 22 ga needle. An aluminum plate (0.003m thick) with a small hole was mounted between the pneumatic cylinder and the sample vials to serve as a needle guide. A 0.5 m length of stainless steel tube (0.0016 m O.D. × 0.00025 m I.D.) connected the side arm of this fitting to the inlet port of the 6-port sample valve. A 3-port stream select valve (Whitey Co., Highland Heights, OH) was attached to the outlet port of the 6-port sample valve. The stream select valve was also fitted with a pneumatic actuator and served to flush the sample loop and connector tubing between injections. Dead space in the tubing between the side-arm needle fitting and sample valve was less than 0.00003 L.

A pneumatic cylinder (0.019 m bore X 0.0254 m stroke; Control Line Equipment Inc., Cleveland, Ohio) was secured to the fraction collector in a position over the sample vials. The push rod of the pneumatic cylinder was fitted with a side arm needle fitting (Popper & Sons Inc., New Hyde Park, NJ) and a 22 ga needle. An aluminum plate (0.003m thick) with a small hole was mounted between the pneumatic cylinder and the sample vials to serve as a needle guide. A 0.5 m length of stainless steel tube (0.0016 m O.D. × 0.00025 m I.D.) connected the side arm of this fitting to the inlet port of the 6-port sample valve. A 3-port stream select valve (Whitey Co., Highland Heights, OH) was attached to the outlet port of the 6-port sample valve. The stream select valve was also fitted with a pneumatic actuator and served to flush the sample loop and connector tubing between injections. Dead space in the tubing between the side-arm needle fitting and sample valve was less than 0.00003 L. Disposable plastic syringe barrels (5cc) were used as spacers in the fraction collector racks to hold the sample vials securely in place.

Figure 1 shows a schematic of the valving required to interface the autosampler with the gas chromatograph. At a signal from the controller unit (described below) a solenoid is triggered and the pneumatic cylinder punctures a sample vial with the 22 ga needle. The overpressure of gas in the vial flows through the side arm connector tubing and fills the sample loop on the 6-port valve (Fig. 1A). The sample valve is then actuated bringing the sample loop in line with column 1 (Fig. 1B). After N₂O elutes (ca 3.5 min) the 8-port stream select valve is actuated placing column 2 on line to the detector and backflushing slow chromatographing components out of column 1 (Fig. 1C). At this point the 6-port sample valve is reset and the 3-port flush valve is actuated, flushing the sample loop and connecting tubing. After a 15 s flush, the 3-port valve is reset to the vent position. The fraction collector, operating under a timing mode, advances the next sample vial to a position under the pneumatic cylinder. When the base-line on column 2 stabilizes (2 min.) the next sample is injected. Complete automation of this procedure was accomplished by controlling the pneumatic valve actuators and the pneumatic cylinder with a microprocessor unit.

The microprocessor controller unit consists of an inexpensive microcomputer (Sinclair 1000, Timex Computer Corp., Waterbury, Conn.) and a relay board (Byte Back Co., Leesville, SC). The relay board plugs into the expansion slot of the microcomputer and has eight independent relays. Each of the solenoids controlling the valves and the pneumatic cylinder is connected to one of the eight relays. The relays can be opened or closed using a single BASIC command statement; thus, a timed events program (BASIC) is used to automatically control the sequence of valve operations. The computer and relay board are contained in a plexiglass box equipped with a cooling fan. As an added convenience the solenoid were also wired with toggle switches so individual events could be controlled manually.

Output from the EC detector is directed to an integrator (Shimadzu, Scientific Inst., Columbia, MD) which performs the peak area analyses.

**Evaluation of Automatic Sampler**

Sensitivity and accuracy of N₂O detection with the automatic samples were evaluated by analyzing evacuated vials prepared with .005 L N₂O standard gas (Air Products, Inc., Hyattsville, MD). Three N₂O standard gas concentrations were used; 3.36, 38.8 and 368 μg N₂O-N/L in Argon. Analyses of these standards using the automatic sampler was compared to N₂O analyses by manual syringe injection (0.0005 L) of N₂O standards.

Storage effects of samples in vials were evaluated by preparing vials with N₂O standard gas and analyzing them after 1, 2, 7, 14, and 28 d storage (room temperature). Nitrous oxide determination of the stored standards were compared to freshly prepared standards at each time point.

Absorption of N₂O by the vials was investigated by preparing vials with the 3.36 μg-N/L and 368 μg-N/L standards. After exposure to these N₂O concentrations for 24 hours, stoppers were removed from one half of the vials from each set and boiled in dH₂O for 30 min.

Stoppers were then replaced and all the evacuated vials were flushed with Ar and evacuated to −0.090 MPa using a vacuum pump. These vials were then injected with 0.005 L of 3.36 μg N₂O-N/L standard gas and analyzed on the autosampler. Unused evacuated vials were also prepared with 0.005 L of the 3.36 μg N₂O-N/L standard.

**Results**

Variability of N₂O standard determinations using the vial automated system was low, with coefficients of variation ranging from 1.90% to 4.90% (Table 1). This variability was the same as observed from direct
syring injection of the standards into the gas chromatograph. There was a slight loss of sensitivity when using the vials (ca. 82% recovery). This level of recovery is due to a dilution of sample in the vial. The pressure in the vials is —0.08 MPa which corresponds to a 15% dilution for a 0.005 L gas sample. This dilution was the same over the range of standards tested. It was also noted that the vials contained a relatively high amount of background N₂O (ca. 2 µg N₂O-N/L ± 4%), which is approximately six times ambient N₂O concentrations. If sample N₂O concentrations at or near ambient levels are to be measured it is important that background N₂O concentrations in the vials be the same. If greater sensitivity is required, vials can be flushed with argon and reevacuated to remove contaminating N₂O prior to use.

The effect of storage of samples on N₂O detection is presented in Table 2. Vials prepared with the 3.38 µg N/L N₂O standard showed no significant drop in recovery over the 28-d storage period. Lack of significant storage effect is likely due to the variability, which was higher for the low standards. Variability increased slightly with time. Significant decreases in recovery were noted for the medium and high N₂O standards. An initial rapid drop in N₂O levels at day 1 was followed by a slower decrease. After 28 d of storage recovery levels of 87% and 91% were obtained for the medium and high standards, respectively.

Absorption of N₂O by sample vials was investigated by comparing N₂O concentration in vials with a variety of preexposure conditions (Table 3). There was no significant difference in N₂O detection between vials with no previous exposure to N₂O, vials previously exposed to the low N₂O standard, and vials previously exposed to the low standard with stoppers boiled afterwards. Vials exposed to the high N₂O standard showed a significant increase in N₂O concentration over vials with no preexposure. Boiling the stoppers of vials previously exposed to the high standard reduced these N₂O concentrations somewhat, but the increased levels were still significantly higher than the N₂O levels in vials with no preexposure. It was also noted that variability was greater in vials exposed to the high N₂O standard. Thus, unless the higher variability associated with reused vials can be tolerated, vials should only be used once.

**Discussion**

Storage of gas samples in evacuated vials has several advantages, primarily that many soil incubations can be run simultaneously and gas samples can be analyzed at a later time. This feature is a necessity in field studies where remote sites are sampled. In addition, storage of samples in vials lends itself well to automated analyses of the gas.

Data presented here indicate that variability is not substantially increased when samples are stored in vials. However, there is a slight loss of sensitivity due to dilution in the vial and absorption of N₂O by the rubber stopper. This effect appeared to be constant at 82% recovery over the 100 fold concentration range tested here. Typical N₂O levels of soil core incubations, for which this system is used, fall into the low to medium N₂O standard range (ca. 10 to 100 times ambient N₂O levels). However given the variability of the autosampler system, changes in N₂O concentrations as low as 0.05 µg N₂O-N/L can be detected (ca. 0.4 times ambient levels). To account for slight differences in storage times of N₂O samples collected during soil incubations, N₂O standards should be routinely prepared at each time soil incubations are sampled and these standards stored with the unknown samples. This procedure provides a way of accounting for storage and dilution effects as well as for detector drift which may occur during long automated analyses runs.

Concurrent with the development of this automated sampler, an autosampler for analyzing gas stored in vials was developed by Robertson and Tiedje (Plant and Soil, in press). Their sampler operates using the same basic principles as the unit described here but has several different features. They use a commercially available external events controller for a Hewlett Packard integrator to control the sample valves and flush the sample loop by overpressing with carrier gas, with a pair of samplers for two detectors to increase capacity for analysis, and He as an internal standard as well as for pressurizing gas in the vials.

Automatic analyses of samples is relatively easily accomplished. Virtually any fraction collector can be modified to accommodate the sample vials and the pneumatic plunger. Construction of the microcomputer control unit is simple and inexpensive with cost

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**Table 1. Comparison of automatic analysis of N₂O in vials with manual syringe injections.**†

<table>
<thead>
<tr>
<th>N₂O-N injected, µg</th>
<th>Integrator counts (peak area)</th>
<th>% Recovery‡</th>
<th>Syringe</th>
<th>Vial</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>0</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>0.63</td>
<td>3 763 (8.43)</td>
<td>5 453 (4.50)</td>
<td>82.0</td>
<td></td>
</tr>
<tr>
<td>1.94</td>
<td>53 717 (5.88)</td>
<td>46 759 (1.90)</td>
<td>85.6</td>
<td></td>
</tr>
<tr>
<td>184</td>
<td>524 400 (5.32)</td>
<td>429 700 (4.36)</td>
<td>81.5</td>
<td></td>
</tr>
</tbody>
</table>

† Values presented are means of 12 replicates.
‡ Numbers in parentheses are coefficients of variation (%).
§ 100 × (Vial Counts — 2367)/Syringe Counts.

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**Table 2. Recovery of N₂O stored in vials.**

<table>
<thead>
<tr>
<th>Days storage</th>
<th>Percent recovery</th>
</tr>
</thead>
<tbody>
<tr>
<td>Low†</td>
<td>Medium‡</td>
</tr>
<tr>
<td>1</td>
<td>97.7 (2.56)</td>
</tr>
<tr>
<td>2</td>
<td>101 (3.52)</td>
</tr>
<tr>
<td>7</td>
<td>105 (4.74)</td>
</tr>
<tr>
<td>14</td>
<td>103 (6.32)</td>
</tr>
<tr>
<td>28</td>
<td>98.2 (6.14)</td>
</tr>
</tbody>
</table>

† Low, Medium, and High N₂O standards correspond to 3.36, 38.8 and 368 µg N₂O-N/L. Recovery calculated by comparing stored vials to vials freshly prepared on day of analysis.
‡ Numbers in parentheses are coefficients of variation (%) for stored set of vials, 12 determinations were made for each set.

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**Table 3. Absorption of N₂O by rubber stoppers.**

<table>
<thead>
<tr>
<th>Preexposure condition</th>
<th>Integrator counts†</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fresh vials</td>
<td>3 636 (4.46)</td>
</tr>
<tr>
<td>Low standard</td>
<td>3 702 (6.60)</td>
</tr>
<tr>
<td>Low standard, stoppers boiled</td>
<td>3 865 (5.43)</td>
</tr>
<tr>
<td>High standard</td>
<td>13 503 (17.3)</td>
</tr>
<tr>
<td>High standard, stoppers boiled</td>
<td>4 718 (8.84)</td>
</tr>
</tbody>
</table>

† All vials prepared with 0.005 L of low N₂O standard. Value presented are means of 12 replicates.
‡ Numbers in parentheses are coefficients of variation (%).
A SPOT TEST WITH TOLUIDINE BLUE FOR ALLOPHANE AND IMOGOLITE

KOJI WADA AND YASUKO KAKUTO

Abstract

Toluidine blue is adsorbed on negatively charged colloids and exhibits a characteristic color change from blue to purplish red (metachromasia). A procedure to test the metachromasia for the soil was developed and tested for Chile and Ecuador soil samples containing allophane and imogolite. The results show that the presence of the metachromasia can be used for the test of allophane and imogolite.

Additional Index Words: Andepts, amorphous material, metachromasia.


Toluidine blue \((\text{CH}_3)_2\text{N}^+\text{C}_6\text{H}_3\text{NSC}_6\text{H}_5(\text{CH}_3)\text{NH}_2\) is adsorbed from aqueous solution on negatively charged colloids and exhibits a characteristic color change from blue to purplish red (metachromasia). This reaction is used for staining particular components in thin-sectioned biological specimens for microscopic observation and for determining the end point in the colloidal titration. The metachromasia of toluidine blue is a very sensitive reaction and is known to occur at the presence of \(10^{-4}\) mmol (e\(^{-}\))/mL polyvinyl sulfate \([\text{CH}_3\text{CHO}_{\text{SO}_3}\text{H}]^\text{+}\). (Terayama, 1952).

The metachromasia of toluidine blue was found for nearly all soil samples derived from granite, andesite and sedimentary rocks, whereas it was found for 20 to 44 soil samples derived from volcanic ash (Nomoto et al., 1955). The difference between the two groups of soil samples may be related to the presence of allophane and imogolite particular to some of the latter group of soil samples. Recent studies showed that Dystrandepts in which allophane and imogolite predominate in the exchange complex have only a small amount of negative charge under the condition as they occur in the field (Okamura and Wada, 1983) and that large organic cations such as tetramethyl- or tetraethyl-ammonium ion are rather "excluded" even from those negative charge sites (Wada and Tange, 1984). These observations suggest a possibility that the absence of the metachromasia of toluidine blue, a large organic cation, can be used for the test of allophane and imogolite. We developed a procedure for this test and tested it for the Chile and Ecuador soil samples which are derived from volcanic ash and analyzed for clay minerals.

Procedure

Take 30 to 50 mg of an air-dry or undried soil (<2 mm) on a white spot plate using a spatula and add 0.4 ml of 0.02 \% toluidine blue \((\text{C}_9\text{H}_4\text{N}_3\text{SCl})\) aqueous solution. Stir the mixture with the spatula for 15 s and allow it to stand for the soil to sediment. If the color of the supernatant remains blue, it suggests the presence of allophane and imogolite. If the supernatant becomes colorless and the color of the sedimented soil turns to purple or purplish red, it indicates the presence of negatively charged humus and/or layer silicates. It is expected from the reaction mechanism that the toluidine blue test is affected by the soil sample/solution ratio, more specifically the amount of soil colloid in the sample/solution ratio. The soil sample/solution ratio of 30 to 50 mg to 0.4 mL was adopted on the basis of the test for the six samples which are different in the species and content of clay minerals (Table 1). A larger amount of sample, 50 to 100 mg, can be used for the coarse-texture soil.

Results and Discussion

Table 2 shows the test for the Chile and Ecuador soil samples and relevant mineralogical and chemical data. The metachromasia did not occur for any samples in which allophane and imogolite coexisted. The Si/Al molar ratio of the acid oxalate soluble fraction of the clay suggested that allophane has the Si/Al molar ratio of \(\leq 1/2\). In most of the samples containing allophane and imogolite, humus (several Al horizon samples were included for the test) and/or 2:1 and 1:1 layer silicates were also present, but they did not show the metachromasia. This is probably due to the blocking of their cation-exchange sites by polymer hydroxy-Al and Fe ions as suggested from the studies on the electric charge characteristics of Andepts (Wada and Okamura, 1980; Okamura and Wada, 1983).

Ten Ecuador samples containing allophane but not imogolite did not show the metachromasia, whereas nine Ecuador samples containing unidentified acid-oxalate soluble aluminosilicates \([\Lambda(\text{?})]\) showed the metachromasia or the discoloration of toluidine blue (Table 2). There was no particular difference in the Si/Al molar ratio of the acid oxalate soluble fraction of the clay, but the unidentified aluminosilicates were dissolved in hot 1/3 \(M\) sodium citrate, gave the Si/Al
Table 1. Effect of soil sample/solution ratio on the reaction for toluidine blue.

<table>
<thead>
<tr>
<th>Sample</th>
<th>Main species</th>
<th>Content, %</th>
<th>Reaction for toluidine blue†</th>
<th>Weight of air-dry soil sample (mg)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>10</td>
<td>30</td>
</tr>
<tr>
<td>Chile</td>
<td>A, Im &gt;</td>
<td>49</td>
<td>Blue (+ + +)</td>
<td>Blue (+ +)</td>
</tr>
<tr>
<td>2/2</td>
<td>L.S.</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Chile</td>
<td>A, Im &gt;</td>
<td>50</td>
<td>Blue (+ + +)</td>
<td>Blue (+ +)</td>
</tr>
<tr>
<td>4-3</td>
<td>L.S., Ht</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Chile</td>
<td>L.S., Ht</td>
<td>56</td>
<td>Blue (+)</td>
<td>Tinged with purple</td>
</tr>
<tr>
<td>13-1</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ecuador</td>
<td>A (lm) &gt;</td>
<td>9</td>
<td>Blue (+ + +)</td>
<td>Blue (+ +)</td>
</tr>
<tr>
<td>4-2</td>
<td>L.S.</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Ecuador</td>
<td>L.S.</td>
<td>4</td>
<td>Colorless</td>
<td>Colorless</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
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</tr>
<tr>
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<td>L.S., Ht</td>
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<td>Colorless</td>
<td>Colorless</td>
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<tr>
<td>2/2</td>
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</tr>
</tbody>
</table>

† Data from Tour Guides (Sixth International Soil Classification Workshop, 1984). A = allophane, Ht = halloysite, Im = imogolite, L.S. = 2:1 and 2:1:1 layer silicates and their intergrades and mixed-layer minerals. The analyses were made by the present authors.

‡ Top: the color of supernatant. Bottom: the color of sedimented soil. The volume of 0.02% toluidine blue solution was fixed at 0.4 mL.

§ For abbreviation see footnote † of Table 1.

Table 2. Toluidine blue test for Chile and Ecuador soil samples.

<table>
<thead>
<tr>
<th>Metachromasis†</th>
<th>Number of sample</th>
<th>Mineral§ species</th>
<th>Si/Al molar ratio (acid oxalate soluble fraction)</th>
<th>pH NaF (1)</th>
<th>pH NaF (2)</th>
<th>Acid oxalate extractable Al, %</th>
</tr>
</thead>
<tbody>
<tr>
<td>Chile</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>-</td>
<td>A, Im</td>
<td>0.35</td>
<td>9.0</td>
<td>10.3</td>
<td>0.3</td>
</tr>
<tr>
<td></td>
<td>-</td>
<td>A, Im &gt; L.S.</td>
<td>0.32 ± 0.05</td>
<td>10.8 ± 0.4</td>
<td>11.3 ± 0.2</td>
<td>4.1 ± 1.7</td>
</tr>
<tr>
<td></td>
<td>-</td>
<td>A, Im &gt; L.S.</td>
<td>0.42 ± 0.06</td>
<td>10.4 ± 0.1</td>
<td>11.1 ± 0.2</td>
<td>5.3 ± 0.6</td>
</tr>
<tr>
<td></td>
<td>-</td>
<td>L.S., Ht</td>
<td>0.27 ± 0.07</td>
<td>8.8 ± 0.8</td>
<td>10.3 ± 0.4</td>
<td>1.1 ± 0.3</td>
</tr>
<tr>
<td></td>
<td>+</td>
<td>L.S., Ht</td>
<td>0.34 ± 0.06</td>
<td>10.2 ± 1.0</td>
<td>10.8 ± 0.4</td>
<td>1.0 ± 0.5</td>
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<tr>
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<td>-</td>
<td>A ≥ L.S.</td>
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<td>10.2 ± 0.7</td>
<td>10.8 ± 0.5</td>
<td>0.9 ± 0.4</td>
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<td>+ (?)</td>
<td>L.S., Ht</td>
<td>0.33 ± 0.08</td>
<td>9.5 ± 0.5</td>
<td>10.0 ± 0.4</td>
<td>0.5 ± 0.2</td>
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<td>+</td>
<td>L.S., Ht</td>
<td>0.39 ± 0.07</td>
<td>9.0 ± 0.4</td>
<td>9.8 ± 0.2</td>
<td>0.9 ± 1.4</td>
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<td>+</td>
<td>L.S., Ht</td>
<td>0.34 ± 0.08</td>
<td>9.9 ± 0.4</td>
<td>9.8 ± 0.6</td>
<td>0.5 ± 0.4</td>
</tr>
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<td>Ecuador</td>
<td></td>
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<td></td>
<td>- (?)</td>
<td>L.S., Ht</td>
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<td>8.8 ± 0.8</td>
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<td>+ (?)</td>
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<td>L.S., Ht</td>
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<td>9.8 ± 0.6</td>
<td>0.5 ± 0.4</td>
</tr>
</tbody>
</table>

† Metachromasis: Supernatant, Sedimented soil

‡ Data from Tour Guides (Sixth International Soil Classification Workshop, 1984). The values show the average and the standard deviation. The analyses of clay minerals and pH NaF (1) (Fields and Perrott, 1966) were made by the present authors and the analyses of pH NaF (2) (Soil Survey Staff, 1972) and acid oxalate extractable Al were made by Dr. J. Kimble.

§ For abbreviation see footnote † of Table 1.

ratio $\geq 1/1$ and showed features of "embryonic halloysites" on the difference infrared spectra (Wada and Kakuto, 1985). The metachromasis was found for all soil samples containing humus and/or layer silicates but not imogolite and/or allophane (Table 2), and the purplish red color was particularly conspicuous for the samples containing halloysite.

In Table 2, the pH NaF and the acid oxalate extractable Al values of soil samples are listed as relevant chemical data. The pH NaF was proposed by Fieldes and Perrott (1966) as a rapid test for allophane. The pH NaF $>9.4$ is now used as a criterion for the exchange complex being dominated by amorphous material (Soil Survey Staff, 1975). The acid oxalate extractable Al (Soil Survey Staff, 1972) $>0.4$ is used as a feature characterizing andic soil properties (Leamy, 1983). Table 2 shows that all soil samples containing imogolite or allophane and some soil samples not containing imogolite or allophane had the pH NaF $>9.4$ and the acid oxalate extractable Al $>0.4$ %, though there were some differences between the two groups of soil samples. The result indicates that the acid oxalate extractable Al includes "active aluminum" from sources other than allophane and imogolite and the pH NaF denotes also the activity of such aluminum.

Conclusions

The absence of metachromosis of toluidine blue can be used for the test whether the exchange complex of a soil is dominated by imogolite and/or allophane, though it is not a specific test for allophane and imogolite but rather a test for the absence or near-absence of negative charge sites in the soil, or for the negative charge sites with no access to a large organic cation. Though further tests are required, when used along with the pH NaF or the acid oxalate extractable Al, the toluidine blue test can provide key information on the soil-forming processes from volcanic ash and is useful for classification of Andepts and related soils.
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References

A REFINED SOIL CORING SYSTEM
GREGORY A. RUARK

Abstract
Materials and construction specifications are detailed for a soil sampling system comprised of a slide hammer driver, an open probe, a core probe, and a probe puller. Intact soil cores, contained in clear plastic tubing, can be extracted from a site with minimal disturbance. Replaceable hardened steel tips facilitate sampling soils with rock fragments and high concentrations of roots. This soil sampling system is portable, versatile, and durable

Additional Index Words: soil sampler, soil core.


A VARIETY of hand-held soil samplers have been reported in the literature and an assortment of samplers is commercially available. Veihmeyer (1929) first proposed using a sliding hammer to drive a soil sampler. The idea was further developed by Uhland (1947) who devised what is now commonly known as a double-cylinder, hammer driven core sampler. This sampler effectively takes large diameter, shallow surface samples (Blake 1965). Lutz (1947) used a push sampler which directly filled a sample can. The edge of the can was sharpened to serve as a cutting tip. The Lutz sampler was further modified by Jamison et al. (1950) with the incorporation of a sliding hammer. This idea was reworked again by Jurgensen et al. (1977) who modified a hammer driven corer for sampling soils on rocky sites. In addition to specialized core samplers, bucket and screw augers, and open probes of the Oakfield types are commercially available.

Often it is desirable to extract an intact soil core under a variety of site conditions while inducing minimal site disturbance. To avoid sampling bias, the sampler should permit extraction of soil from a randomly located point regardless of course fragments or root obstructions. The sampling equipment must be versatile, dependable, and easily repaired in the field as the integrity of an experimental design is often contingent upon continuous equipment operation. The soil sampling system detailed in this paper fills the need for a hand-carried, durable, and versatile soil sampler.

This sampler represents a refinement of the above mentioned samplers with the incorporation of some new concepts. Oil hardened and drawn steel is used in the fabrication of replaceable cutting tips and coupler components. Clear plastic core sleeves, compatible with the inside diameter of the core probe, can be used to retain cores in a relatively undisturbed state. Pickering and Veneman (1984) used a binocular polarizing microscope to examine thin sections of soil cores that were taken with this core probe. Sample disturbance was found to be confined to a 0.5 to 1.5 mm zone adjacent to the outer edge of the core.

Materials and Construction

Slide Hammer

The slide hammer is shown in Fig. 1. The aluminum extraction plunger (A) was attached to the steel rod (F) with a countersunk hex-head cap screw and a lock washer. The stainless steel set plate (D) was inset into the shatterproof nylon handle (B) and held into place by two countersunk hex-head set screws (C). A large rubber washer (E) was used to protect the cutting tip from the top of the weight when the core probe was slid over the rod for storage or transportation. The 4 kg weight (after drilling) was machined with a light knurled finish for improved grip. The top and bottom edges of the hole through the weight were beveled where they abut the steel rod. The weight should move freely along the length of the rod. The weight was fabricated from American Industrial Steel Institute (AISI) 1020 stock with dimensions adjusted to achieve the proper weight.

The thumb guard (H) is made from aluminum and three hex head set screws fasten it flush with the bottom of the weight. A hardened steel coupler (I) is fashioned from AISI 4340 stock. (Note that the 6.5 cm hole for the rod is not drilled completely through the bottom of the coupler.) It is oil hardened and drawn back to 45 Rockwell units for optimum strength.
Fig. 1—Slide hammer components: A. Aluminum plunger, B. Nylon handle, C. Hex-head set screw, D. Stainless steel set plate, E. Rubber washer, F. Steel rod, G. Steel weight, H. Aluminum thumb guard, I. Hardened steel coupler, and J. Shear pin. All units in cm unless specified.

The shear pin (J) attaches the coupler to the steel rod and can be easily replaced in the field. Alternatively, a 40-D common nail will serve well as a shear pin.

Open Probe

The open probe is viewed in Fig. 2. The spanner wrench hole (A) is used for removal of the hardened steel coupler (C) from the stainless steel tube (D). A spanner wrench (not shown) can be machined to the necessary dimensions for coupler and tip (G) removal. Hex-head cap screws (B) are threaded into the coupler and are used to attach the probe puller (Fig. 4.). The top, inside edge of the coupler is slightly beveled. The hardening and beveling prevent flare in from the slide hammer impact. The coupler is right-hand threaded into the stainless steel tube to preclude unscrewing the coupler during removal form the soil with the probe puller. The bottom-most portion of the coupler fits flush against the top of the cut-out trough (E) when the coupler is fully threaded into the tube.

The width of the cutout trough (3.5 cm) matches the inside diameter of the tip (G). This is large enough to facilitate sample removal, while preventing the sample from falling out. The inside diameter of the tube is 3.8 cm, allowing for a 1.5mm radial expansion of the soil sample.

AISI 1020 steel may be substituted for the stainless steel tube. If this is done the tube should be either zinc plated or maintained with a lubricant to prevent corrosion. If a probe of a different inside diameter is desired the specifications can be modified. The critical dimensions are for tolerances between couplers and expansion room between the inside diameters of the probe and the tip.

All couplers and tips were constructed from AISI 4340 stock which was oil hardened and drawn to 45 Rockwell units. Tips can be made slightly more rigid by only drawing
them back to 48 to 46 Rockwells but above this point the steel becomes brittle and is likely to fracture. Tips are right-hand threaded into the tube. Several replacement tips should be made and carried into the field.

Core Probe

The core probe is illustrated in Fig. 3. The steel coupler (B) was beveled, hardened, and threaded in the same manner as the open probe coupler. Hex-head cap screws (C) were threaded into the tube body for attachment of the probe puller. A small air hole (E) was drilled to relieve air pressure that may build-up when the probe is driven into the ground. The stainless steel tube may range from 35 to a maximum of 50 cm in effective sampling length without significant alteration in performance. The hardened steel tip was fabricated as previously described. Clear plastic core sleeves (6.35 cm O.D., 0.089 cm wall thickness) can be ordered in any length. The core sleeves are made from cellulose acetate butyrate (CAB), manufactured by Eastman Chemical. The

CAB tubing and reusable plastic end caps were supplied by Petro Packaging Co., Inc. of Cranford, New Jersey.

Probe Puller

The probe puller, diagrammed in Fig. 4, is made from AISI 1020 steel. It fits over both the open probe and the core probe. The hex-head spring plungers keep the puller attached to the probe during transport. For an improved grip, rubber tubing of the appropriate inside diameter can be fitted over the puller handles.

Operation

The procedure for taking bulk soil samples with the core probe is as follows. Locate the probe tip over the sample point. Fit the coupler end of the slide hammer into the coupler on the probe. Place one hand on the slide hammer handle and the other on the weight and drive the probe into the ground. Avoid raising the weight up to the steel set plate on the upward stroke as this could unseat the coupling. Continue driving the probe to the desired depth (max 50 cm).

Remove the slide hammer from the probe and attach the probe puller by placing the puller over the hex-head cap screws and locking the cap screws behind the spring plungers. Turn the core a half turn clockwise (it is designed to operate only in this direction) to shear the soil core from the soil below; then lift while continuing to rotate as needed. Stand the slide hammer upright and set the core probe (tip down) on top of the plunger (A in Fig. 1). The soil core can now be pushed out of the top of the core probe. If a root or rock is wedged in the core probe, the probe can be set on the ground, coupler end down. Place the plunger into the bottom of the probe and gently tap the weight down onto the steel set plate (D in Fig. 1) to loosen the sample. This will usually facilitate further removal of the sample in the conventional manner.

If an undisturbed soil sample is desired, inset a plastic sleeve into the core probe (prior to sampling) until the sleeve rests on the inside edge of the cutting tip. A steel spacer ring, of the appropriate length, should be placed on top of the plastic sleeve. The spacer clearance between the top of the plastic and the bottom of the slide hammer coupler (I in Fig. 1) should be minimal to preclude upward movement of the plastic sleeve during sampling. This prevents soil grains from lodging between the plastic sleeve and the inside wall of the core probe. A plastic sleeve should always be used when sampling wet soil as deformation of the soil core can make extraction from an unlined core probe difficult. Soil samples in plastic sleeves can be retained by plastic end caps.

If a short undisturbed core is desired it is best to just start the bottom edge of a short plastic sleeve (capped at the top) into the top of the core probe (after sampling but prior to
COMMENTS AND LETTERS TO THE EDITOR

Comments on The DTPA-Extractable Iron, Manganese, Copper and Zinc from Neutral and Calcareous Soils Dried Under Different Conditions.

Leggett and Argyle (1983) berated standard texts for devoting little attention to sample preparation. In fact Jackson (1958) stated that many types of analyses must be carried out on moist samples immediately after collection, such as exchangeable Fe, Mn and K; acid-extractable P; NO$_3$-N, NH$_4$-N and pH. To these I would add electrical conductivity, and Bartlett (1979) has added Cr. Uehara and Gillman (1981) reported changes in cation exchange capacity and rheological properties of variable-charge soils: "in the tropics the rule of thumb is not to air-dry or oven-dry samples prior to analysis". In summary it emerges that drying is unsuitable for all analyses on some soils, and some analyses on all soils. Drying can have the same chemical effects in the field. In Northern Ireland, for example, cereals are regularly Mn-deficient in spring and early summer, symptoms disappearing in July or (in unusually wet years) Aug., some while after the soils begin to dry out in the field. Exchangeable Mn increases during this period, and so does Mn concentration in lysimeters (McAllister and Benians, unpublished). The process culminates in massive leaching of Mn with the fall, and so Mn deficiency in the following spring.

The inevitable conclusion is that soils really do change significantly when they dry, whether in the lab or in the field. Leggett and Argyle (1983) criticised the diethylenetriaminepentaacetic acid (DTPA) test for Fe and Mn on the ground that the results change after drying. In my view any test for Fe and Mn that did not change if the soil was dried would be next to worthless, as it would not reflect the true situation in the soil. Lindsay (1979) explained the mathematics of the DTPA extraction, but its basis is very simple: the concentration of the chelate ligand-metal complex is directly proportional to the activity of the metal in the aqueous phase. In effect it is like a water extraction made more sensitive (at least until the reserves of metal to replenish the aqueous phase is substantially reduced). Changing the solubility of the reserves of metal changes the activity of its ions in the aqueous phase, which in turn changes the concentration of the metal in the chelated phase. This is how a good soil test should work.

Finally, I should like to ask my colleagues at large, as well as Drs. Leggett and Argyle, why they dry soil samples. The procedure is time-consuming, when farmers everywhere are grumbling about slowness in soils labs; it is also expensive. Most important, it gives spurious results — except where field soils air-dry anyway, e.g. Colorado. And why store the samples before testing them?

I value Leggett and Argyle's paper as a useful addition to our knowledge of the effects of drying and storage, but their quest for correction factors to relate products of different laboratories' drying rooms (= "Lab Dirt"; Bartlett, 1979) is a wrong response to the situation.

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References


