

**GRACEnet**  
**Chamber-based Trace Gas Flux Measurement Protocol**  
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**Trace Gas Protocol Development Committee<sup>1</sup>**

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**Scope:**

1. This protocol only addresses N<sub>2</sub>O and CH<sub>4</sub> flux measurement methodology. The reactivities of other gasses of interest such as NO<sub>x</sub>, O<sub>3</sub>, CO, and NH<sub>3</sub> will likely dictate that separate chambers and associated instrumentation be employed. CO<sub>2</sub> can also be included as an analyte with this protocol, however, when plants are present, interpretation of CO<sub>2</sub> data is complicated.
2. This protocol adopted chamber-based flux methodology (the least expensive option available) in order to allow inclusion of as many sites as possible. Since micromet techniques are expensive, they will be used at only locations with current micromet capability (Minnesota, Iowa, others?).
3. In deciding on a chamber design, our goal was the adoption of methodology which is sensitive, unbiased, has low associated variance, and allows accurate interpolation/extrapolation over time and space. Because of our inability, at this time, to *precisely* assess the extent of bias associated with a given chamber design and sampling protocol under the range of conditions which might exist, we have adopted our 'best guess' protocol. Assessment, refinement and/or modifications of the protocol may continue in the future. At some sites this may include evaluation of chambers against micromet fluxes or performing comparisons of alternate chamber designs. Recognizing that any measurement technique will have disadvantages, the best we can do at this time is to select a technique which minimizes potential problems. To facilitate the adoption of a common technique, it is important to attain a common understanding of the potential shortcomings associated with chamber-based flux measurement techniques. The following section discusses some of these issues.

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<sup>1</sup>Questions or comments on the protocol can be directed to Tim Parkin ([parkin@nstl.gov](mailto:parkin@nstl.gov)).

## Background

Mosier (1989) reviewed the key issues related to chamber techniques for gas flux measurement. These are summarized below along with recommendations to minimize potential problems.

1. Soil Disturbance:
  - Soil disturbance upon installation
  - Longer term microclimate effects
  - Recommendations:*
    - Use temporary/portable chambers.
    - Install permanent chamber anchors at least 24 h prior to flux determinations.
    - Anchors or collars should be as short as possible to minimize micro environment perturbations.
    - Move chamber anchors if soil microclimate effects are observed.
  
2. Temperature perturbations:
  - Influence biological activity
  - May cause physical absorption or dissolution of dissolved gasses.
  - Recommendations:*
    - Use insulated, reflective chambers.
    - Keep deployment time as short as possible.
  
3. Pressure perturbations:
  - Wind may cause pressure-induced mass flow over chamber collar
  - Closed chamber may reduce natural mass flux.
  - Sampling effects may induce mass flow
  - Recommendations:*
    - Use vented chamber.
    - Use skirted chambers
  
4. Humidity perturbations:
  - Gas solubility changes (probably a minor effect)
  - Humidity increases in the chamber may result in dilution of the gas of interest and resulting underestimate of the flux
  - Changes in humidity may impact biological activity (minor)
  - Recommendations:*
    - Keep chamber deployment short
    - Measure relative humidity changes inside chamber to correct for dilution effects from water vapor.
  
5. Temporal Variability:
  - Diurnal variations. There is some evidence in the literature that diurnal variations (up to a factor of 10) in soil gas flux follow diurnal temperature fluctuations, however, this characterization is not consistent.
  - Daily variation. Day-to-day variation may be highly dependant upon rainfall, fertility, tillage or freeze thaw events.
  - Seasonal variation. Spring and Winter fluxes can be substantial and need to be considered.
  - Recommendations:*
    - Measure flux at times of the day that more closely correspond to daily average temperature (mid morning, early evening).
    - Apply a temperature correction algorithm to measured fluxes when time-of-day temperature induced biases might be present.

- Measure fluxes 3 to 4 times/week, all year long.
- Stratify sampling to account for episodic events.

6. Spatial Variability:

-Can be extremely high. Coefficients of Variation associated with chamber-based fluxes commonly exceed 100%.

*Recommendations:*

- Use chambers with larger footprint to minimize small scale variability.
- Use as many chambers as possible.

7. Gas Mixing :

-It is generally assumed that molecular diffusion is sufficiently rapid within the chamber headspace such that homogeneous gas concentrations exist when sampling. However, this may not necessarily be true if large amounts of vegetation are present or the chamber volume:surface area is large (Livingston and Hutchinson, 1995).

*Recommendations:*

- If it is deemed that mixing of the headspace gas is necessary, there are a couple of options.
  - 1. Chambers can be fit with small fans. A 12 VDC computer fan will run on a 9 volt transistor radio battery and is a cost effective way of incorporating a fan into a chamber design. Computer fans can be obtained from Action Electronics, Santa Anna, CA. Phone: (800) 563-9405, [www.action-electronics.com](http://www.action-electronics.com). Example of a 12vdc fan from this company is part # 108idc12vdc1b. Cost: ~ \$7.00
  - 2. A gas manifold within the chamber attached to the sampling port can be used. The manifold has a single port on one end (which extends out the top of the chamber) and multiple ports on the other end which accept narrow teflon tubing (e.g., 1/16") that extend into the chamber. The narrow tubing from each of the multiple inner ports is extended to different points inside the chamber, so that when the sample is collected, gas is pulled from multiple points in the chamber. Manifolds can be purchased from Small Parts, Inc. 800-220-4242, [www.smallparts.com](http://www.smallparts.com). An example part no. is TCM-13-20/4-10 (description = Tubing Manifold 13G inlet 20G outlet).

Given these considerations, there have been a number of different chamber-based methods proposed in the literature. Below are provided our best recommendations. See referenced literature for additional details.

## Recommended Protocol

### General:

Gas flux will be measured by static chambers deployed on the soil surface for a period of no more than 60 min. During chamber deployment, samples of the chamber headspace gas will be removed at regular intervals, and stored for later analysis by gas chromatography. Specific recommendations on chamber design, gas sampling and analysis, and flux calculations are provided below. Investigators are encouraged to examine the referenced literature underlying these recommendations.

### Chamber Design

#### Minimum Requirements:

1. Flux chambers should be fabricated of non-reactive materials (stainless steel, aluminum, PVC, polypropylene, polyethylene, or plexiglass.)
2. Material should be white or coated with reflective material, (Mylar, or painted).
3. Chambers should be large enough to cover at least 175 cm<sup>2</sup> of the soil surface, and have a target height of 15 cm (height can be decreased to increase sensitivity or increased to accommodate plants).
4. Chambers should contain a vent tube, at least 10 cm long and 4.8 mm in diameter (e.g. 1/4" stainless steel tubing). See Fig. 1 for details.
5. Chambers should have a sampling port to enable the removal of gas samples. Possible options include: butyl rubber stopper (Alltech # 95256), or nylon/polyethylene stopcock (ColeParmer # A-30600-000 : Qosina, #99705 or #99717).

#### Recommended Design:

Two part chamber consisting of a permanent anchor, driven at least 8 cm into the soil and extending no more than 5 cm above the soil surface, and a cap which contains the vent tube and sampling port. Anchors are fabricated so that they can accommodate the flux chamber during measurement phase. Anchors and chambers made of 8" (or larger) diameter PVC. Alternatively, anchors can be made of thin-walled stainless steel or aluminum to minimize physical disturbance upon insertion. The vent tube is necessary to avoid pressure perturbations (and subsequent mass flow) when chambers are installed and when gas samples are collected. Schematics of two potential chamber designs are presented and photographs of a variety of

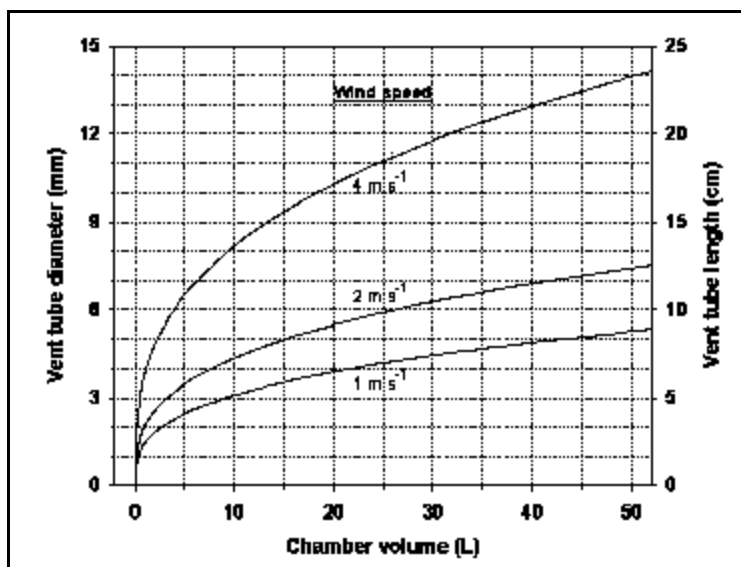


Figure 1. Optimum vent tube diameter and length for selected wind speeds and enclosure volumes as described by Hutchinson & Mosier (1981).

chambers in operation are provided in Appendices 3 and 4.

### **Chamber Deployment**

*Anchors* : As indicated above, anchors should be installed at least 8 cm into the ground and extend no more than 5 cm above the surface. Permanent anchors should be installed at least 24 h prior to first flux measurement. There are no fixed guidelines regarding how long anchors can (or should) be left in place. In cultivated systems, chamber anchors are typically removed prior to cultivation, planting, or fertilizer application, then replaced. In grassland studies anchors have been left for over 10 years with no apparent deleterious effects. One advantage of leaving anchors in place is that soil disturbance and root damage are minimized. However, there have been reported problems with microclimate effects within the anchors left in place for extended periods. For example changes in humidity or shading can cause algal growth, and in heavy or compacted soils ponding of rainwater can occur. This is not a desirable situation. It will be up to the investigator to determine how often chambers should be moved.

### *Plants:*

If the goal of this project is to quantify ecosystem contributions to net trace gas flux, then ideally, plants should be included inside chambers during flux determinations. There is some information indicating that N<sub>2</sub>O emissions may be facilitated by living plants (Chang et al., 1998; Chen et al, 1999; Smart and Bloom, 2001). However, inclusion of plants presents an interesting problem. With regard to sensitivity, inclusion of plants would likely dictate that chamber height be increased, but an increase in chamber height results in a corresponding decrease in sensitivity (i.e. increase in minimum detectable limit, see below). Significant reductions in sensitivity might, in some cases, result in all the flux measurements being below the detection limit. In such cases, it is advisable to also measure bare soil fluxes (i.e. between rows in row-crop agriculture) using shorter chambers which have higher sensitivity. Results could then be reported as fluxes within a range of the bounds established by the two measurements. If it is not feasible to include plants (at all growth stages) at least chambers should be deployed both within and between rows (in row crop agriculture). Alternatively, chambers with a larger foot print and therefore providing more representative coverage of the ecosystem under study, can be used.

### *Sample numbers:*

Trace gas fluxes exhibit a high degree of spatial variability. Thus, the more chambers, the better. Variability may also be a function of chamber size, and may be reduced by using larger chambers. Recommendation for minimum number is 2 chambers per treatment in plot scale studies. In landscape or field scale studies it is recommended that 'similar' landscape elements be identified and a stratified sampling design employed, whereby samples are stratified by landscape element, soil type, or vegetation (Livingston and Hutchinson, 1995). In situations where identifiable hot spots may occur (e.g. urine patches in a grazed system) a stratified sampling may have to be developed to account for this. Gilbert (1987) gives some sampling guidelines when hot spots exist.

### *Sampling frequency:*

Trace gas fluxes exhibit a high degree of temporal variability. Thus, the more frequently measurements are made, the better. There are several elements to temporal variability that must

be considered: diel or diurnal variations, seasonal variations, and variations induced by perturbation (e.g. tillage, fertility, irrigation/rainfall, thawing). Flux measurements should be made mid-morning of each sampling day to minimize biases associated with diurnal variations. However, a  $Q_{10}$  temperature correction procedure may be applicable to adjust rates determined at different times. The temperature correction procedure assumes that temperature variations are the primary factor driving diurnal flux variations, thus the temperature correction adjusts the measured flux to the average daily soil temperature. To account for perturbation effects it is recommended that fluxes be measured as soon as possible after the perturbation (such as rainfall, tillage, or fertility event), then daily for the next several days during and following the specific event. During the remainder of the year gas flux measurements should be made at regular time intervals (1, 2 or 3 week intervals) as resources allow.

### Gas sampling

Fluxes are measured by determining the rate of change of trace gas concentration in the chamber headspace. In most cases trace gas concentrations are determined by physically removing a gas sample from the chamber headspace for analysis in the laboratory. Gas samples should be withdrawn at regular intervals during the chamber deployment. Chambers should be in place no longer than 60 minutes. The shorter the deployment time, the better, but deployment must be long enough so that sensitivity is not compromised. At least 3 time points are required for flux calculation: time 0, and two additional points, equally spaced in time (e.g. 0, 30 60 min. or 0, 20, 40 min). *[Note: Sampling is performed at regular intervals to facilitate flux calculation by Eq. 1 (below). However, more samples can be collected, and sampling does not have to be at regular intervals if the stochastic model of Petersen et al., (2001) is used.]* Sampling is performed by inserting a polypropylene syringe into the chamber septa and slowly removing a gas sample. Mixing of headspace gas by pumping the syringe before sampling is not recommended as pumping may cause pressure perturbations and/or excess dilution of headspace gas by entry of outside air through the vent tube. The gas volume removed at each time point is dictated by the specific gas analysis technique to be used. Typically, from 5 to 30 ml are removed. If the syringe is equipped with a stopcock, the sample can be stored directly in the syringe. Alternatively, the gas sample can be transferred to a previously evacuated glass vial sealed with a grey butyl rubber septum. If this option is selected, excess gas is usually injected into the evacuated vial (relative to the vial volume) to produce an overpressure. This overpressure facilitates the subsequent removal of a gas sample for analysis. Brooks (1993) evaluated several storage protocols and found that red rubber stoppers such as found on commercially available evacuated blood vials were the worst. Parkin has observed that red rubber stoppers are reactive to methane. However, others report no problems with coated red

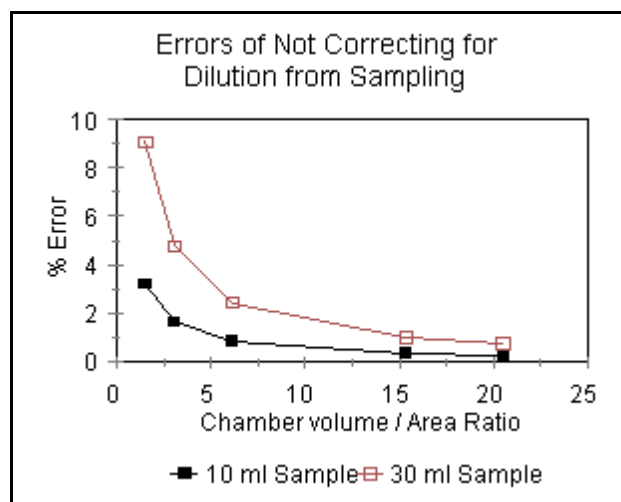


Figure 2. Percentage underestimation of flux rate due to headspace dilution as a result of sampling, presented as a function of chamber geometry and gas sample size.

rubber stoppers. Details of gas sampling and analyses are noted in Mosier et al. (1991, 1996). It should be noted that each time a headspace gas sample is removed from the chamber outside air flows into the chamber through the vent tube. This results in a dilution of the analyte in the chamber headspace. The error associated with this dilution effect is a function of both the sample volume withdrawn and the chamber Volume/Surface Area ratio (Figure 2). Correction for this dilution effect should not be necessary for chamber Volume/Surface Area ratios >10 and sample volumes < 30 ml. An example of a gas sampling protocol is presented in Appendix 2.

### **Gas Analysis**

Samples should be run as soon as possible after collection. Gas chromatography will be used for analysis of N<sub>2</sub>O and CH<sub>4</sub> (Electron capture detector for N<sub>2</sub>O and Flame ionization detector for CH<sub>4</sub>). Specific method of gas sample injection into the GC will depend upon the specific instrumentation available at each location. However, it is recommended that the GC be fit with a sample valve to minimize injection error. To account for problems associated with GC drift it is recommended that: 1) samples from individual chambers be run in sequence (e.g. t<sub>0</sub>, t<sub>1</sub>, t<sub>2</sub>,) rather than segregating all the samples by time (e.g. all the to samples run together) and ii) standards be run periodically throughout the sample run (e.g. every 10 to 20 samples).

#### *Standards:*

Standards should be prepared each sampling time. Standards should be handled in a manner similar to samples with regard to collection and storage. Preferably samples should be prepared in the field (i.e. injected into glass vials, or collected in syringes). Several different standard concentrations should be run, as detector response may be nonlinear. The range of standards should bracket the concentrations found in samples. Examples: N<sub>2</sub>O; 0.1, 1.0 and 10 ppm. CH<sub>4</sub>; 0.5, 1, 2, 10 ppm. Standard curves are then used to convert the GC output of the samples into units of ppm. Certified standard gasses can be obtained from Scott Specialty Gas ([www.scottgas.com](http://www.scottgas.com)) or Scott Marian.

### **Data Analyses:**

#### *Flux Calculation:*

Fluxes are calculated from the rate of change of the concentration of the analyte of interest in the chamber headspace. Since the units associated with the gas standards are typically ppm(v), when the standard curve relationship is applied to calculate gas concentrations of the samples, the resulting unit of the analyte is also ppm(v). Volumetric parts per million (ppm(v)) has units of uL trace gas L<sup>-1</sup> total gas.

If the rate of change of headspace trace gas concentration is constant (ppm(v) vs. time data is linear) then linear regression can be used to calculate the slope of the concentration vs. time data. The slope of the line is the trace gas flux. Thus, a regression of ppm(v) vs. minutes will result in a slope with units of ppm(v) min<sup>-1</sup>. Multiplying the slope by the chamber volume (L) and dividing by the chamber surface area (m<sup>2</sup>) will result in a flux with units of uL trace gas m<sup>-2</sup> min<sup>-1</sup>

If the rate of change of headspace trace gas concentration is not constant (ppm(v) vs. time data is curvilinear) then linear regression is not appropriate. Curvilinear concentration data with time is

attributed to a build up of the analyte concentration in the chamber headspace, which alters the diffusion gradient and the resulting flux. To account for this effect, Hutchinson and Mosier (1981) proposed an algorithm as an alternative to linear regression (Eq. 1).

$$f_0 = V(C_1 - C_0)^2 / [A * t_1 * (2 * C_1 - C_2 - C_0)] * \ln[(C_1 - C_0)/(C_2 - C_1)] \quad \text{Eq. [1]}$$

where  $f_0$  is the flux at time 0,  $V$  is the chamber headspace volume (L),  $A$  is the soil surface area ( $m^2$ ),  $C_0$ ,  $C_1$ , and  $C_2$  are the chamber headspace gas concentrations (ppm(v)) at time 0, 1, and 2, respectively, and  $t_1$  is the interval between gas sampling points (min). The resulting units of  $f_0$  are:  $\mu\text{L trace gas } m^{-2} \text{ min}^{-1}$

It should be noted that this correction algorithm only works if  $[(C_1 - C_0)/(C_2 - C_1)] > 1$  and if time points are equally spaced.

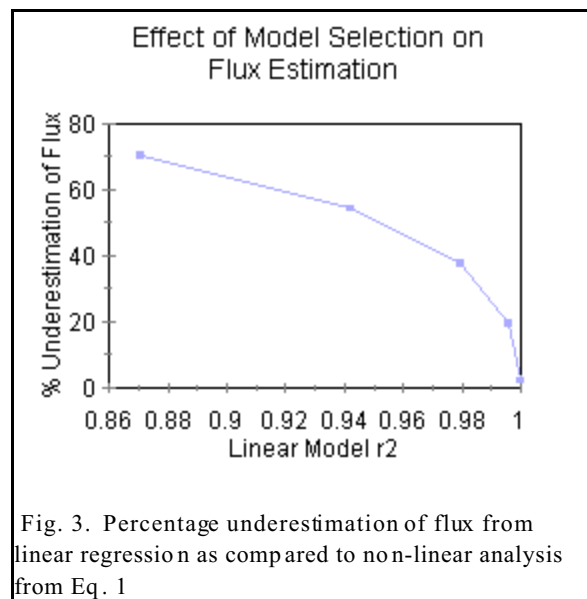
As an alternative to Eq. 1 for calculating a flux from curvilinear data, Pedersen et al. (2001) has proposed a stochastic diffusion model. The reported advantages of the Pedersen model are: i) a more rigorous treatment of gas diffusion theory, ii) there is no requirement for equi-spaced data points, and iii) it can accomodate more than 3 data points, iv) it provides an assessment of goodness of fit, and v) it has a lower failure rate than Eq. 1. This technique will not be described in detail here, however, the computer model can be obtained from S.O. Petersen at [Soren.O.Petersen@agrsci.dk](mailto:Soren.O.Petersen@agrsci.dk).

Regarding linear regression, it should be realized, that in deciding whether to use linear regression or a non-linear model, a strict criteria for goodness of fit should be established for the linear model. Simulation data shows that even slight deviations from linearity can have a dramatic influence on the calculated flux (Fig. 3).

Flux calculations from linear regression or the non linear models described above produce values with units of  $\mu\text{L trace gas } m^{-2} \text{ min}^{-1}$ . An additional calculation has to be performed in order to covert flux values from a volumetric basis to a mass basis. To perform this conversion the ideal gas law must be invoked (Eq. 2)

$$PV = nRT \quad \text{Eq. [2]}$$

where  $P$  = pressure,  $V$  = volume,  $n$  = the number of moles of gas,  $R$  = the gas law constant, and  $T$  = temperature.



The ideal gas law quantifies the relationship between pressure, volume, mass and temperature of a gas.

When the value of  $R = 0.08206 \text{ L atm Mol}^{-1} \text{ }^\circ\text{K}^{-1}$  is used, units of P, V, n and T have corresponding units of Atm, Liters, Moles, and  $^\circ\text{K}$ ., respectively. The goal of applying Eq. 2 is to convert uL trace

$$1 \text{ uL trace gas} * 0.965 \text{ atm} / ((0.08206 \text{ L atm Mol}^{-1} \text{ }^\circ\text{K}^{-1}) * (273 + 20)^\circ\text{K}) * 1 \text{ L}/10^6 \text{ uL} * 10^6 \text{ uMol/Mol}$$

Sample calculation to convert uL gas to uMol. (Note: conversion from  $^\circ\text{C}$  to  $^\circ\text{K}$  by adding 273)

gas to uMol trace gas. To do this, one must have knowledge of both the air temperature and atmospheric pressure. A table relating elevation and atmospheric pressure is provided. For example, at an altitude of 1000 ft., and at an air temperature of  $20^\circ\text{C}$ , we can calculated from Eq. 2 that 1 uL of trace gas contains 0.0401 uMol of trace gas (see calculation box above). Thus, multiplying the calculated flux with units of  $\text{uL trace gas m}^{-2} \text{ min}^{-1}$ , by 0.0401 gives flux units of  $\text{uMol trace gas m}^{-2} \text{ min}^{-1}$ . (Note above that  $^\circ\text{K}=(273+^\circ\text{C})$ ).

### Noisy Data

The change in chamber headspace trace gas concentration over time typically will be linear or curvilinear as shown in Figs. 3 and 4. In these situations linear regression or the non-linear diffusion based models can be used to calculate the flux. However, often concentration with time data are noisy and time course data are obtained similar to those shown in Figs. 5 and 6 (Anthony et al., 1995). Determination of a flux from noisy data often requires investigator judgement. Several possibilities exist for flux estimation from noisy data including: 1) linear regression using all the points, 2) calculation of the slope from points 1 and 2, 3) slope calculation from points 1 and 3, or 4) slope calculation from points 2 and 3. If the investigator cannot discount outliers based on experience and judgement of past performance of the site or chamber, the most conservative

Relationship between altitude and atmospheric pressure.

Alt (ft)	mm Hg	psi	atm
0	29.92	14.7	1.000335
1000	28.86	14.18	0.964949
1320	28.54	14.02	0.954061
2000	27.82	13.67	0.930244
2640	27.14	13.33	0.907107
3000	26.81	13.17	0.896219
3960	25.77	12.66	0.861513
4000	25.84	12.69	0.863555
5000	24.89	12.22	0.831571
5280	24.47	12.02	0.817961
6000	23.98	11.78	0.801629
6600	23.25	11.42	0.777131
7000	23.09	11.34	0.771687
7920	22.15	10.88	0.740384
8000	22.22	10.91	0.742426
10560	20.11	9.88	0.672334

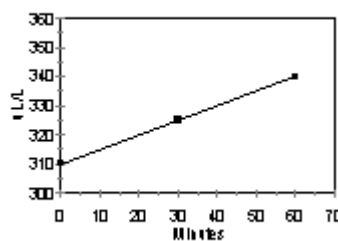


Fig. 3

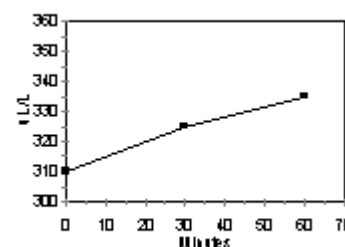


Fig 4

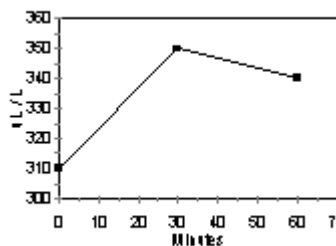


Fig. 5

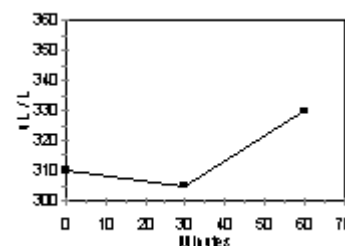


Fig. 6

approach would be to adopt option 1. If noisy data proves to be a persistent problem, evaluation of GC precision, chamber design, and/or sampling protocols should be performed. Also, collection of more points during chamber deployment may help in discriminating outliers and may also yield improved estimates if the Pedersen stochastic model is applied.

### **Minimum Detection Limit**

Often field fluxes are low, thus it is important to have an idea of the minimum detection limit (MDL). The MDL is a function of the sampling and analytical precision as well as the chamber volume and surface area. Sampling + analytical precision is determined by calculating the standard deviation of many standards on the gas chromatograph ( $n > 20$ ). Because instrument precision is usually a function of concentration, the standards used should contain trace gas concentrations at or near ambient levels. From analysis of large numbers of standards, precision is determined to be  $\pm 2$  standard deviations of the mean. This  $\Delta$  ppm ( $2 \times \text{std dev}$ ), along with specific information on the chamber volume, surface area, and chamber deployment time is used to compute the MDL as described below.

$$\text{MDL} = 2 \times \text{std.dev uL/L} \times \text{Chamber Volume (L)} / \text{Chamber Footprint (m}^2\text{)} / \text{total deployment time (min)}.$$

Units for the above computation of the MDL are  $\text{uL trace gas m}^{-2} \text{ min}^{-1}$ . To convert to  $\text{uMol m}^{-2} \text{ min}^{-1}$  the universal gas law must be used.

### **Quality Assurance /Quality Control:**

#### *Standards and standardization:*

It has been reported that Scott Standard Gases may differ substantially from their stated concentrations. An alternative source of certified standard gasses is Scott Marian (these are still only  $\pm 2\%$  at best). If a network of ARS sites is going to be established, it is suggested two tanks of very high quality standards containing  $\text{CO}_2$ ,  $\text{CH}_4$  and  $\text{N}_2\text{O}$  be purchased from NOAA at the cost of about \$3500 + new regulator (assuming that ARS will come up with some funds). These tanks should be shipped around for people to check their GC calibrations and their standard tanks. In the interim, Ft. Collins is arranging to have one of these standard tanks made, and there may be a possibility to distribute samples of this standard in vials to different locations on a limited basis. This known standard gas would then be used to standardize gas tanks at each location. Alternatively, it has been suggested that ARS fund a trace gas analysis lab where all samples are analyzed. At this point in time agency funds do not exist to support this proposal. Details of these activities will be worked out at a future date.

#### *Stopper Reactivity:*

Currently, gray butyl rubber septa or stoppers appear to be the least reactive to  $\text{N}_2\text{O}$  and  $\text{CH}_4$ , however, there have been reports that different batches of gray butyl rubber may differ regarding their reactivity. It is recommended that individual investigators perform their own assessment of trace gas reactivity with each new batch of stoppers, regardless of the type of stoppers used. A suggested protocol for this is:

- 1: Prepare 60 vials with standard gas. This will be the test set.
2. Immediately after these vials are prepared run 20 of these samples.

3. After 1 day of storage (at room temperature and pressure) run 20 vials from the test set prepared on day 0, and prepare and run 20 newly prepared vials with the same standard used to prepare the test set.
4. After 1 week of storage, run the final 20 vials from the test set along with 20 vials freshly prepared.
5. Evaluate: 1) Changes in average concentration as a function of storage. 2) Changes in precision (i.e. standard deviations) as a function of storage.

*Syringe Reactivity/Carryover:*

Plastic syringes will leak over time. If gases are stored at any length of time in syringes equipped with stopcocks, a similar test of storage efficacy should be performed with each new batch of syringes. Polypropylene syringes are not inert, however, cross-contamination due to carryover is usually not a problem unless high concentrations are sampled, and if syringes are flushed with air between use. Similarly, if syringes are reused, the investigator might want to perform an assessment of trace gas carryover.

**Ancillary Measurements**

In addition to the measurements prescribed by soil sampling protocol additional measurements are recommended.

*At time flux is measured:*

Air temperature

5 cm Soil Temperature

Soil Water content (0-6 cm) gravimetric, capacitance (Theta Probe), or TDR.

*At time of chamber installation:*

Bulk density, texture, organic C and N .

Chamber headspace volume (average chamber height at several locations within the chamber multiplied by the chamber surface area)

Soil Nitrate and Ammonium (0-10 cm). **Note:** It is desirable that soil nitrate and ammonium be determined throughout the year at time intervals deemed appropriate by the individual investigator as dictated by resource availability and plot constraints.

*Weather data* - rainfall, air temperature, relative humidity, solar radiation.

### **Advice and Consultation**

Several investigators involved in GRACEnet have experience in trace gas analysis and flux measurement. These people have agreed to serve as resource contacts for investigators with questions on GC set up, soils chambers, gas sampling, flux calculation, field variability, and data interpretation.

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**Appendix 1.** Example of Trace gas Flux Sampling Procedure  
- Set of 12 Anchors placed in pairs (in-row and inter-row) -

For each set of 12 Chambers:

1. Lay out Chambers, Vials, Syringes by each anchor
2. Install 5 cm temperature Probes (1 in each plot). Air temperature and chamber temperature probes in first plot only.
3. Take ambient Gas Sample
4. Start Measurement (t 0) - Start Stop Watch
  - a. Record Temperatures
    1. Place chamber on anchor #1 (vent facing downwind )
    2. Remove 10 ml gas sample
    3. Inject sample into vial
    4. Flush syringe with Air 2x
    5. Place chamber on anchor #2
    6. Remove 10 ml gas sample
    7. Inject sample into vial
    8. Flush syringe with air 2x
  - b. Move to next pair of chambers in plot
    1. Record time on stop watch
    2. Place chamber 3 on anchor
    3. Remove 10 ml gas sample
    4. Inject into vial
    5. Flush syringe with Air 2x
    6. Place chamber 4 on anchor
    7. Remove 10 ml gas sample
    8. Inject into vial
    9. Flush syringe with air 2x
  - c. Move to next plot
    1. Record Temperatures
    2. Repeat steps 4b.1 through 4b.8 (above)
  - d. Repeat step 4c until all 12 chambers are in place and have been sampled for time 0
5. First Time Point ( t 1)
  - a. Move to position 1 (chamber 1)
    1. Record Soil Temperatures, record chamber temperature and air temperature.
    2. Insert syringe into chamber septa
    3. When stopwatch shows t-1 time (e.g. 20 minutes), remove 10 ml Gas sample
    4. Inject gas sample into appropriate vial
    5. Flush syringe 2x
    6. Move to next chamber, repeat steps 5a.2 - 5a.5, above.
    7. Continue until all chambers have been sampled for time 1
5. Second and third time points (t 2 and t-3)
  - a. same as step 5 above.
6. Remove all chambers, Move to next set of 12 anchors. Repeat steps 1-5
7. When all plots have been done, one person collect all chambers and place in truck other person take soil moisture readings in each plot (4 measurements/plot)

## Appendix 2: Suppliers

### *Sample Vials and Stoppers:*

Option 1: Glass serum vials 6.0 ml (22 x 38 mm) and butyl rubber stoppers and aluminum crimps: Alltech, 2051 Waukegan Rd, Deerfield, IL 60015 (vial stock # 98768, butyl rubber stoppers stock # 95256). These vials fit in the custom autosampler described by Arnold et al., 2001.

Option 2. Exetainers, screw cap 12 ml vials that have a butyl rubber septa-same idea as the serum vials and butyl rubber stoppers-just cheaper and more or less disposable-can buy new screw caps and septa relatively cheaply. Exetainers are purchased through Labco Limited (Brow Works, Copyground Land, High Wycombe, Buckinghamshire. HP123HE, United Kingdom (phone 44-1494-459741) (fax: 44-1494-465101) (Email: sales@labco.co.uk or enquiries@labco.co.uk) The cost is about \$275/1000 vials. Our new CombiPal autosampler (purchased through Varian with a new GC and data system uses these vials. Exetainer vials recommended by Reynald Lemke at Swift Current. The Canadians have 4 of these instruments running-the autosampler has the capacity for 200 samples per batch.:

### *Standard gases*

Scott Speciality Gas <http://www.scottgas.com/>. Standards come certified at +/- 5%, however, actual concentrations may be suspect.

Scott Marian.

*Syringes:* Beckton-Dickenson (obtained from most laboratory supply companies)

*Syringe stopcocks:* (ColeParmer # A-30600-000 : Qosina, #99705 or #99717).

### *Reflective Tape:*

Industrial Tape Connection: <http://www.tapeconnection.com/>

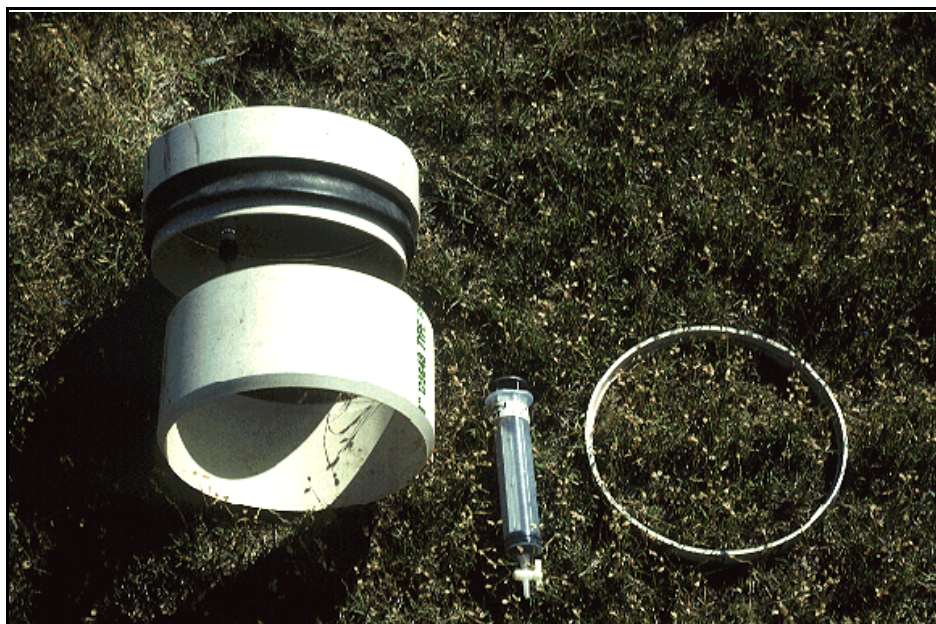
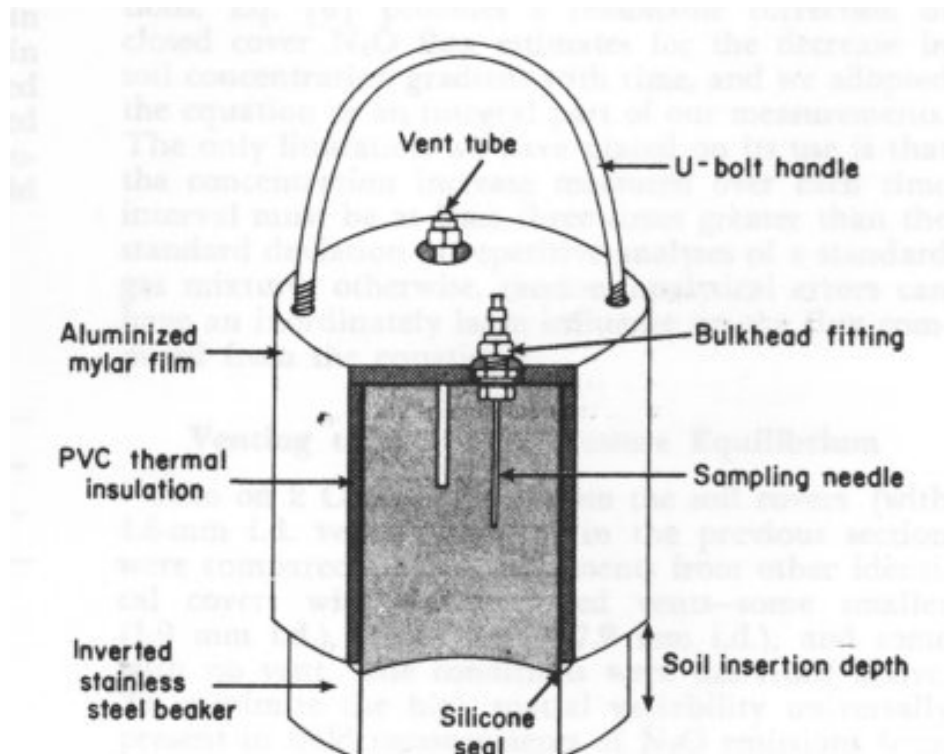
Silver 0.9 mil Metalized Mylar Polyester Film with a brilliant, vibrant mirror-like finish; coated with an aggressive long lasting acrylic adhesive system. 2"x72yards Mylar Film Tape  
Alternative to 3M #850; Ideal #505; Tesa #4137; TLC #CT941M; Venture #1555CW  
PRICE: \$32.70/roll

### *Gas Manifolds:*

Small Parts, Inc. 800-220-4242, [www.smallparts.com](http://www.smallparts.com). An example part no. is TCM-13-20/4-10 (description = Tubing Manifold 13G inlet 20G outlet).

### *Recirculating fans:*

Computer fans can be obtained from Action Electronics, Santa Anna, CA. Phone: (800) 563-9405, [www.action-electronics.com](http://www.action-electronics.com). Example of a 12vdc fan from this company is part # 108idc12vdcslb. This fan is 25 mm x 25 mm x 10 mm and can be run on a 9 volt transistor radio battery.



PVC soil anchor and chamber used by Mosier.



Rectangular chambers used by Mosier



Example of temporary/portable chamber used by Parkin. Chamber has an attached polyethylene skirt held in place on the soil surface with a length of chain. As shown, the chamber is monitoring soil CO<sub>2</sub> flux by recirculating gas through an infrared analyzer. Gas samples can be withdrawn through septum in top of chamber for N<sub>2</sub> and CH<sub>4</sub> analyses.



Large skirted chamber used for CO<sub>2</sub> flux from corn/soil system.  
Applicability of chamber for N<sub>2</sub>O and CH<sub>4</sub> flux measurements has not been tested.

## Appendix 4. Schematic Drawings of Chambers

### **Round PVC Chamber Description:**

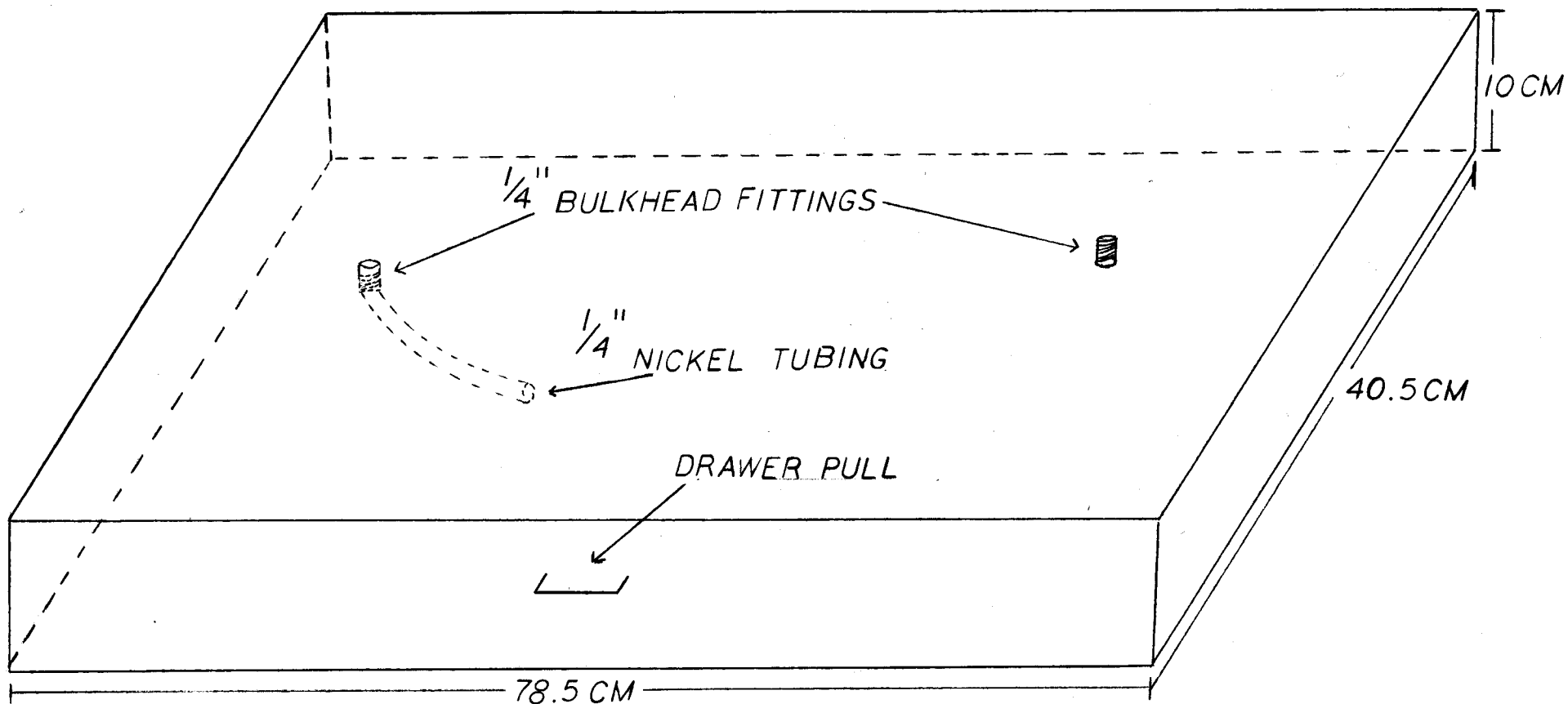
**Anchor:** Made from PVC pipe, 15 – 30 cm diameter. It can be tapered on the bottom for easier insertion into the soil. We typically insert the anchor 8-9 cm into the soil. The chamber can fit onto the anchor, either flush (resting on the anchor), inserted into the anchor, or if an end cap is used, fit over the anchor. A seal is made using an approximately 5 cm wide tire inner tube.

**Chamber:** The chamber can be made from a PVC pipe end cap of the appropriate size or a piece of PVC pipe with a top made from sheet PVC or plexiglass that is cut to fit and cemented into place. Two holes, to accommodate swagelock fittings are drilled and tapped in each chamber top.

**Rectangular aluminum Chambers:** Made from sheet aluminum. Can be made any size to fit the field situation.

**Anchors:** Made from sheet aluminum with a trough to hold water welded on top. The anchors are inserted 10 cm into the soil.

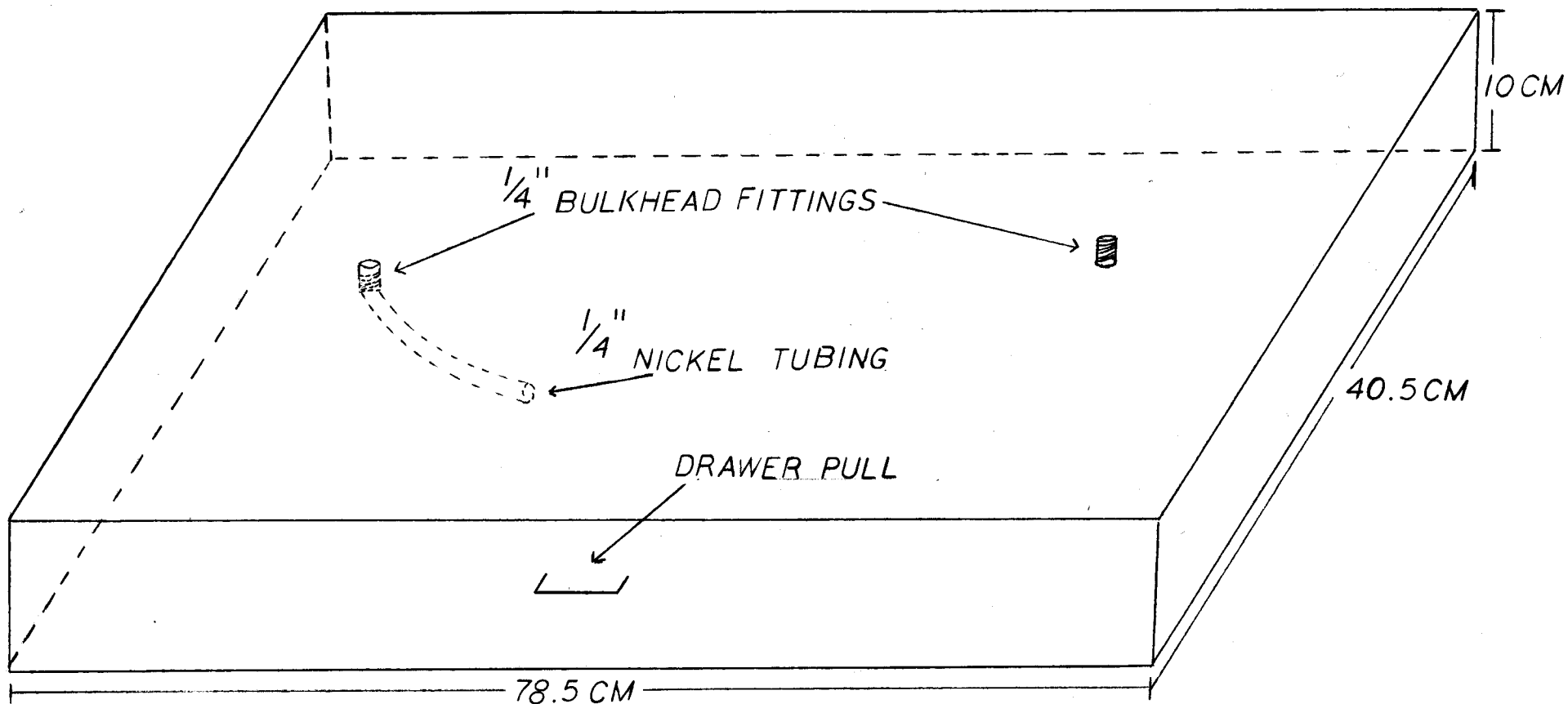
**Chamber:** Made from sheet aluminum to desired dimensions. Two holes, to accommodate swagelock fittings for vent tube and gas collection septum are drilled and tapped in each chamber top.



CHAMBER MADE OF 1 SHEET 1/8" ALUMINUM  
 INSULATED WITH CORKBOARD AND MYLAR TAPE

DRAWING 1 OF 2

TRACE GAS CHAMBER	3/32" = 1 CM
USDA-ARS-SPNR	3-2003

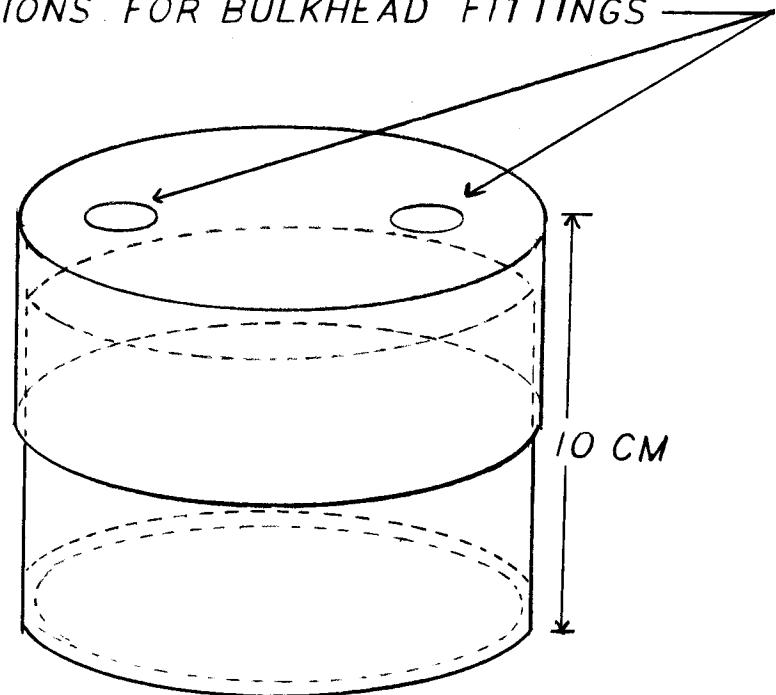


CHAMBER MADE OF 1 SHEET  $\frac{1}{8}$ " ALUMINUM  
 INSULATED WITH CORKBOARD AND MYLAR TAPE

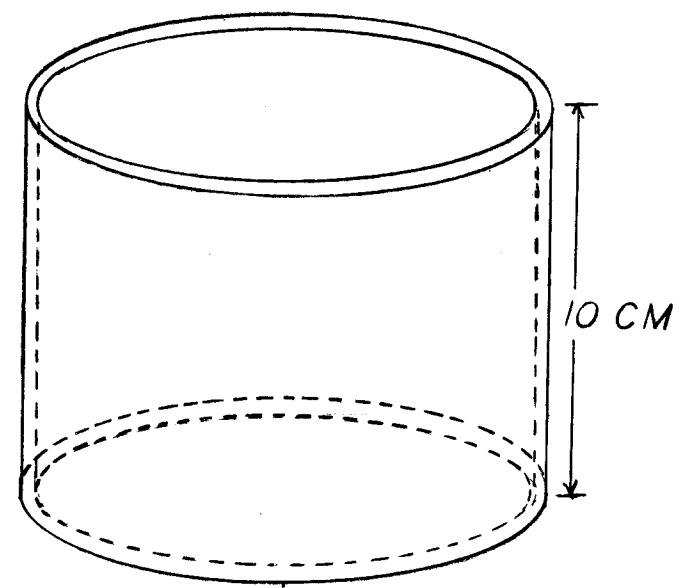
DRAWING 1 OF 2

TRACE GAS CHAMBER	$\frac{3}{32}$ " = 1 CM
USDA-ARS-SPNR	3-2003

LOCATIONS FOR BULKHEAD FITTINGS



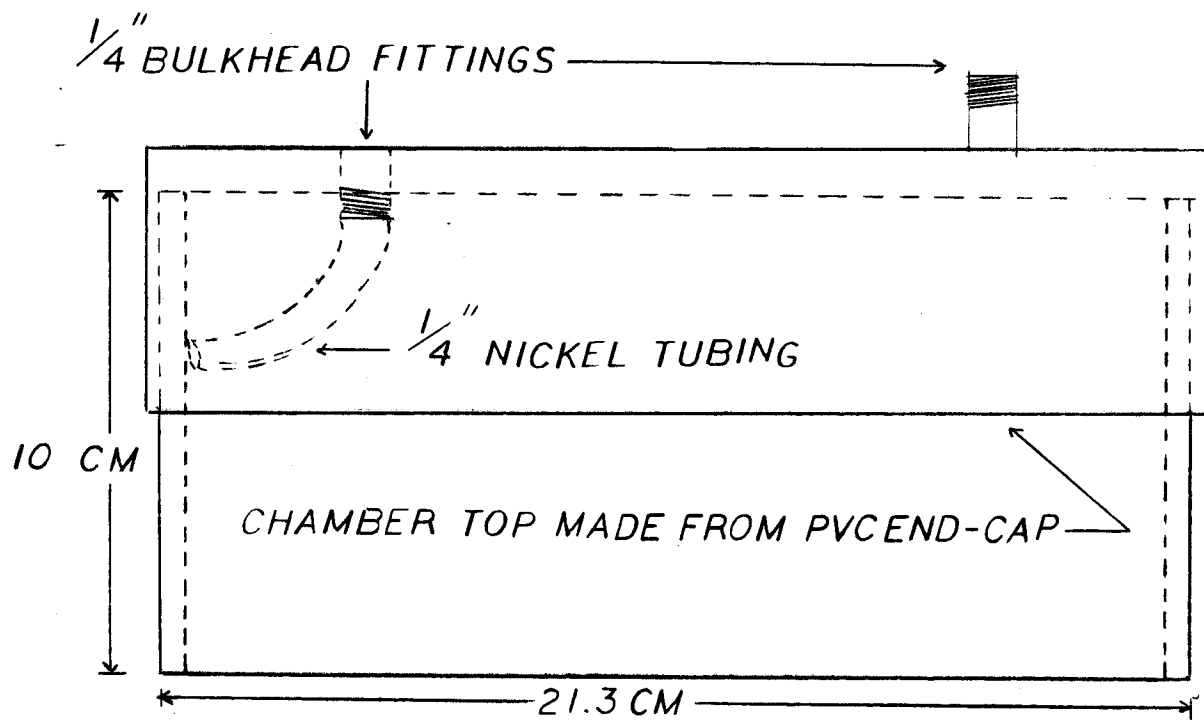
CHAMBER MADE FROM PVC PIPE  
AND MATCHING END CAP



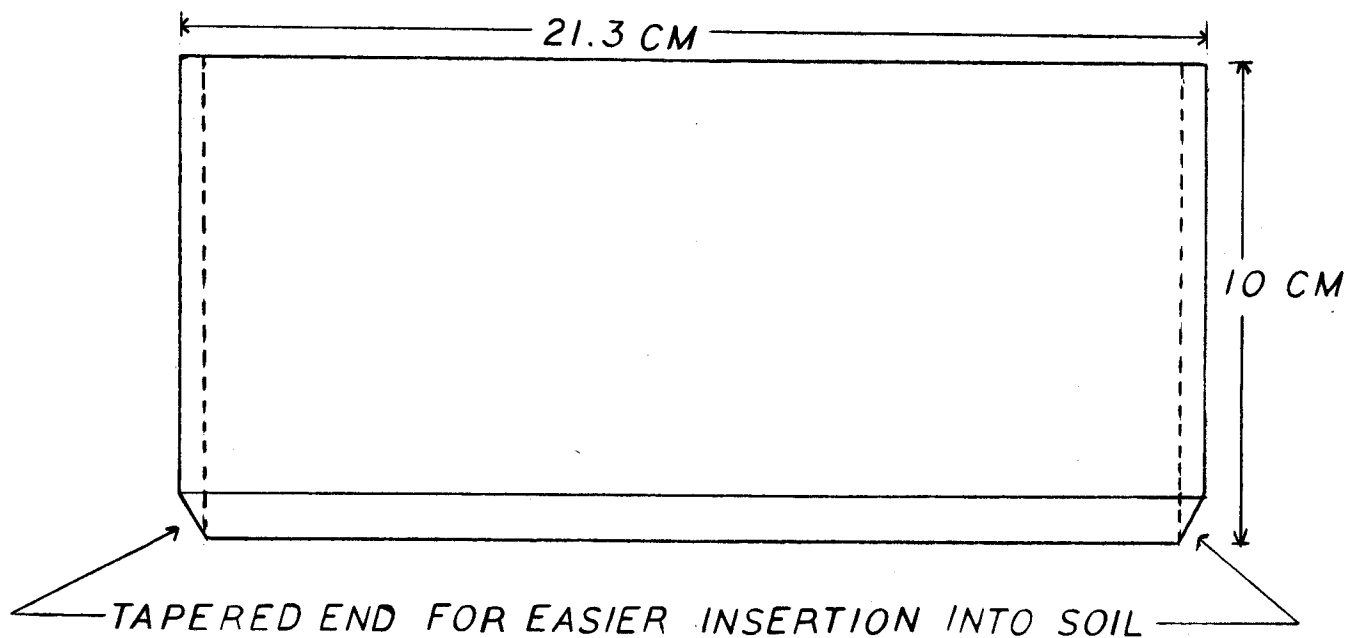
PVC RING MADE FROM SAME  
PIPE AS CHAMBER

DRAWING 1 OF 2

CHAMBER AND ANCHOR	NOT TO SCALE
USDA ARS SPNR	3 - 2003



- PVC CHAMBER AND RING MADE FROM SCHEDULE 40 PIPE
- COVER TOP AND SIDES WITH MYLAR TAPE
- SEAL CHAMBER AND RING WITH AN INNER TUBE GASKET



DRAWING 2 OF 2

PVC CHAMBER AND RING SET	$\frac{1}{4}$ " = 1 CM
USDA - ARS - SPNR	3 - 2003