Annex A – Standardised method for the determination of compost stability by measurement of evolved carbon dioxide

A method to determine the aerobic stability of composted organic materials
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Safety warning

Care should be taken when handling samples that may contain sharp fragments, chemical contaminants or possible pathogenic organisms.

1.0 Scope and field of application

A method for the determination of dynamic aerobic stability of composted materials. The sample shall be obtained in accordance with SOIL IMPROVERS AND GROWING MEDIA - SAMPLING (EN 12579). The procedures described herein are not necessarily applicable to or suitable for all types of composted materials, e.g. very coarse materials with a low percentage of particles passing a 20 mm screen.

2.0 Normative references

This method incorporates by dated or undated reference, provisions from other publications. These normative references are cited at the appropriate places in the text and the publications are listed hereafter. For dated references, subsequent amendments to or revisions of any of these publications apply to this method only when incorporated in it by amendment or revision. For undated references the latest edition of the publications referred to apply.

ISO 5725:1994 Precision of test methods - determination of repeatability and reproducibility for a standard test method by inter-laboratory tests
EN 12579:2000 Soil improvers and growing media - Sampling
EN 13040:2007 Soil improvers and growing media - Sample preparation for chemical and physical test, determination of dry matter content, moisture content and laboratory compacted bulk density
EN 13039:2011 Soil improvers and growing media - Determination of organic matter content and ash
PAS 100:2011 Specification for composted material
PD CR 13456:1999 Soil improvers and growing media - Labelling, specifications and product schedules

3.0 Principle

Moisture-adjusted compost is incubated at 30°C with continuous replacement of carbon dioxide-free air. Carbon dioxide evolved from the compost is collected in a sodium hydroxide solution as sodium carbonate. The collected carbonate is precipitated as barium carbonate by the addition of excess barium chloride. The concentration of carbon dioxide evolved by the compost is measured by titration of the residual sodium hydroxide with standard acid. The method can be adapted to use other CO₂ measuring devices connected directly to the outlet of the incubation vessels.

NOTE 1: Barium carbonate is not decomposed by the action of the acid when phenolphthalein is used as an indicator, colour change occurs at pH 8.5.
4.0 Definitions

For the purpose of this standard the definitions given in PD CR 13456, EN 12579, EN13040 and PAS 100 apply.

5.0 Reagents

5.1 General
All reagents used shall be of recognised analytical quality. Use water of Grade 2 complying with EN ISO 3696.

5.1.1 24% barium chloride solution
Dissolve 244g of BaCl$_2$.2H$_2$O in 1 litre of water to obtain a 24% solution. Filter if necessary.

5.1.2 Hydrochloric acid (HCl)
= 1mol/l; purchase this solution ready prepared.

5.1.3 Phenolphthalein indicator solution
Dissolve 1g of phenolphthalein in 100 ml of ethyl or isopropyl alcohol. Add 100 ml water. The indicator may be purchased ready prepared.

5.1.4 Sodium hydroxide (NaOH)
= 1mol/l; purchase this solution ready prepared and standardised in a collapsible airtight container. Discard when blanks turn cloudy after addition of barium chloride.

6.0 Apparatus

6.1 Constant temperature room or incubator
Capable of maintaining a temperature of 30 ± 1 °C.

6.2 Carbon dioxide scrubbing vessel
1000 ml Drechsel bottle design or similar fitted with a sintered disc e.g. aquarium air diffuser.

6.3 Carbon dioxide collecting vessel
150 ml Drechsel bottle design or similar fitted with a sintered disc, e.g. aquarium air diffuser. A simple 150 ml test tube with rubber bung fitted with inlet and outlet tube connections is sufficient.

6.4 Incubation vessels
1000 ml polyethylene jars with airtight screw top lids incorporating internal and external inlet and outlet tube connections. A flexible narrow bore plastic tube long enough to reach the bottom of the jar should be connected to the air inlet on the inside of the lid.

6.5 Flexible tubing
Narrow bore plastic.

6.6 Air pump
Small aquarium type. Ability to adjust airflow is advantageous but not essential. Must be capable of supplying a minimum of 1000 ml per minute.

6.7 Dispensing pipette
50 ml capacity, Grade A.
6.8 Burette
50 ml capacity, Grade A.

6.9 Titration flask
Erlenmeyer type 500 ml.

6.10 Magnetic stirrer
Optional.

6.11 Sieve
20 mm square apertures.

6.12 Balance
Capable of weighing 120 g with an accuracy of 0.1 g.

6.13 Diffusers
Aquarium sinter type or similar.

6.14 Flow restrictor or bleed valve to adjust air flow
Only needed if pump is not adjustable.

6.15 Air flow-rate meter
Digital or bubble types are acceptable.

7.0 Procedure

7.1 Apparatus assembly
Sequentially connect together with the flexible tubing (6.5) the air pump (6.6), the carbon dioxide scrubbing vessel (6.2), the incubation vessel (6.4) and the carbon dioxide trapping vessel (6.3). See Figure 1 and Figure 2.

7.2 Sample storage
Samples should be tested as soon as possible after receipt. The time between sample receipt at the laboratory and the start of the 72 hour equilibration period shall not exceed one week.

Samples that undergo this stability test shall not be frozen and shall be stored in refrigerator or similar contained space suitable for storing sample under cool conditions (temperature control settings: 4 to 5°C), in the dark, to minimise any changes in sample characteristics over time.

Test materials should always be kept refrigerated when not being prepared for testing or undergoing testing. Given that temperatures in the refrigerator may not stay between 4 to 5°C, actual refrigeration temperatures shall be checked and recorded daily by the lab using a min-max thermometer.
7.3 Sample preparation

7.3.1 Thoroughly mix the submitted sample by spreading on a clean impermeable surface and gently breaking any lump or agglomerate likely to have been caused by compression during transportation. Care must be taken to avoid breaking the sample's intrinsic parts (such as twigs and dried leaves). A 1 litre representative sub-sample shall be taken from the prepared sample. This sub-sample shall be used as the test sample.

7.3.2 Gently sieve the test sample through a 20 mm aperture sieve. The test sample shall not be forced through the sieve by application of any form of pressure. Separately weigh the sieved fraction and fraction retained in the sieve. If more than 10% of the test sample by weight is retained on the sieve then the test procedure is inappropriate to the material under test and this shall be reported. If less than 10% of the test sample by weight is retained on the sieve, this material shall be broken down in equal parts and as few times as necessary to permit the entire test sample to pass through the sieve apertures.

7.3.3 Take two representative sub-samples of the <20mm (sieved) material (7.3.2) and determine, in accordance with EN 13039, their volatile solids content. Calculate the mean of these two results and record this as VS.

7.3.4 Take two representative sub-samples of the <20mm (sieved) material (7.3.2) and determine, in accordance with EN 13040 clause 10, their dry matter. Calculate the mean of these two results and record this as DM.

7.3.5 Assess the moisture content of approximately 500g of sample (7.3.1) using a “fist test”: A fistful of compost is squeezed very hard, for about 10 seconds. If water beads escape between the fingers then the sample is too wet and should be partially air dried by spreading it out on a plastic tray and allowed to lose moisture at ambient room temperature. An unheated fan blowing across the surface and frequent mixing will assist air drying. If the sample crumbles without further action when the fist is opened, then the sample is too dry and should be carefully moistened. Moisten by adding small additions of water with frequent mixing until moisture can just be seen glistening on the surface when a fistful is squeezed hard. The moistened compost must remain friable with plenty of air porosity. This is the analytical sample.

7.3.6 Take two representative sub-samples of the sample adjusted for moisture (7.3.5) and determine, in accordance with EN 13040 clause 10, their dry matter. Calculate the mean of these two results and record this as ADM.

NOTE 1: The fist test should aim to adjust the sample so that its moisture content is within the range of 40 – 60%, although it is acknowledged that for relatively coarse samples this may be difficult to achieve.

NOTE 2: Any lab technician carrying out the fist test should periodically ‘calibrate his/her hands’ by checking his/her fist test evaluation results against the actual dry matter content of the sample, measured in accordance with EN 13040 clause 10.

NOTE 3: For a four-day test it is very convenient to adjust the moisture on a Friday afternoon, equilibrate over the weekend, and start the incubations on the Monday afternoon.
7.4 Determination of carbon dioxide evolution rate

Three samples shall be tested in triplicate.

For each of the three replicates, transfer 100 g ± 2 g representative subsample of the analytical sample (7.3.5) weighed to the nearest 0.1 g into the incubation vessel (6.4). Record the weight of material used as SW.

Transfer approximately 250 ml of sodium hydroxide solution (5.1.4) to the carbon dioxide scrubbing vessel (6.2) and add 50.0 ml of water into the carbon dioxide collecting vessel (6.3). Attach and seal all lids and stoppers.

Ensure the air inlet diffuser of the carbon dioxide scrubbing vessel (6.2) reaches the bottom of the vessel. Ensure the air inlet tube of the incubation vessel (6.4) reaches the bottom of the vessel. Ensure that the air inlet diffuser of the carbon dioxide collecting vessel (6.3) reaches the bottom of the vessel.

Switch on the air pump and adjust the airflow rate (6.14) to approximately 25-75 ml/min measured using the air flow-rate meter (6.15) at the outlet of each carbon dioxide collecting vessel. Equilibrate at 30°C for 72 hours.

After 72 hrs equilibration remove the collecting vessel (6.3) containing water and connect a collecting tube containing 50.0 ml of 1 M sodium hydroxide (5.1.4). Change the collecting tube with another containing a fresh 50.0 ml of 1 M sodium hydroxide every 24 hours over a 4-day period. Do not turn off the air pump at any time or back-pressure may cause NaOH to siphon back to the pump. Maintain the temperature of the incubation units at 30°C at all times.

Transfer the contents of the carbon dioxide trapping vessel (6.3) into the titration flask (6.9) with water washing. Add 20 ml of barium chloride solution (5.1.1) to precipitate any carbon dioxide. Add two to three drops of phenolphthalein solution (5.1.3) and titrate with 1M hydrochloric acid (5.1.2) with vigorous stirring until the pink colour just changes to white (colourless in the case of blanks) with one drop of the acid.

**NOTE 1:** In the presence of strong alkali it is better to use rubber stoppers than glass stoppers.

**NOTE 2:** The method can be adapted to use other CO₂ measuring devices connected directly to the outlet of the incubation vessels. However, any laboratory intending to make an adaptation of this kind must first check that the measured and calculated total CO₂ evolved is equivalent to that achieved by the titration method described here.

**NOTE 3:** It is preferable to set up a series of parallel tests using the same pump to facilitate running replicates and blanks simultaneously with the same batch of reagents. A bank of 10 units from one air pump is convenient for simultaneously testing three samples in triplicate plus one blank.
7.5 Determination of blank value

An apparatus and reagent blank test shall be carried out in parallel with the determination, by the same procedure using the same quantities of all reagents but omitting the test portion.

**NOTE 1** If the apparatus has been set up correctly and the reagents are correctly and freshly prepared the blank titration value should be 48-50 ml. Replace the 1M NaOH when blank titrations are below 47 ml.

8.0 Calculations and expression of results

The rate of carbon dioxide evolution over 4 days is given by the following equations:

\[
\text{mg CO}_2 \text{ evolved per 24 h time period} = \frac{\{ \text{B}_{\text{vol}} - \text{S}_{\text{vol}} \} \times 44.2}{2}
\]

where

- \( \text{B}_{\text{vol}} \) is the volume in ml M HCl for the blank titre
- \( \text{S}_{\text{vol}} \) is the volume in ml M HCl for the sample titre

Total mg CO\(_2\) = sum of mg CO\(_2\) evolved over 4 days

\[
\text{mg CO}_2 / \text{g VS/d} = \frac{[\text{Total mg CO}_2]}{[\text{SW} \times \text{DM} \times \text{VS} \times 4]}
\]

dry weight of compost is DM (as determined in 7.3.4, where the sample did not require wetting or drying to bring it within the required dry matter range) or ADM (as determined in 7.3.6, where the sample did require wetting or drying to bring it within the required dry matter range) in grammes, multiplied by the weight of sample tested (SW).

VS (as % m/m dry matter) is the sample content of organic matter, as determined in 7.3.3

The final result shall be reported as the average value of the sample's three replicates, expressed in mg CO\(_2\) / g VS / d.

9.0 Precision

The laboratory that developed this method, testing a range of composts of different stabilities, demonstrated a pooled in-house standard deviation of 0.906 mg CO\(_2\)/g VS/day. This equates to an in-house repeatability (r) of 2.56 mg CO\(_2\)/g VS/day. Sample homogeneity and the measurements of dry matter and volatile solids are important factors influencing the precision of this method.

10.0 Test Report

The test report shall include the following information:

- a reference to this method of test;
- a complete identification of the sample;
- the results of the determination expressed as mg CO2 /g VS / d (milligrammes carbon dioxide per gramme of volatile solids per day); and
- any further details not specified in the method of test or which are optional, as well as any other factor, which may have affected the results.
11.0 Quality control record

The laboratory’s quality control record shall include the following information:

- sample identity;
- sample receipt date;
- date on which the 72 hour equilibration period started;
- temperature settings of refrigerator (or similar contained space) in which the sample has been stored prior to commencing sample preparation and the 72 hour equilibration period;
- actual minimum and maximum temperatures in refrigerator (or similar contained space), in which the sample has been stored prior to commencing sample preparation and the 72 hour equilibration period;
- Dry matter (DM) of the sample as received, expressed as mass/mass on a fresh matter basis;
- Where drying or wetting was necessary, the amended dry matter (ADM) of the sample after the procedures described in 7.3.5 expressed as mass/mass on a fresh matter basis;
- Volatile Solids (VS) of the sample, expressed as mass/mass on a dry matter basis;
- quantity of CO₂ evolved in each 24 hour period after the 72 hour equilibration period, per sample replicate;
- CO₂ evolution rate per sample replicate and average value of all the sample’s replicates, expressed in mg CO₂ / g VS / d; and
- volume of the blank titre, expressed in ml.
12.0 Picture and schematic diagram of principal components of a single test unit

**Figure 1** Principal components of a single test unit

**Figure 2** Schematic diagram