Designation: D6866 – 12

StandardTest Methods for Determining the Biobased Content of Solid, Liquid, and Gaseous Samples Using Radiocarbon Analysis[1]

This standard is issued under the fixed designation D6866; the number immediately following the designation indicates the year of original adoption or, in the case of revision, the year of last revision. A number in parentheses indicates the year of last reapproval. A superscript epsilon (ε) indicates an editorial change since the last revision or reapproval.

1. Scope*

1.1 These test methods do not address environmental impact, product performance and functionality, determination of geographical origin, or assignment of required amounts of biobased carbon necessary for compliance with federal laws.

1.2 These test methods are applicable to any product containing carbon-based components that can be combusted in the presence of oxygen to produce carbon dioxide (CO_2) gas. The overall analytical method is also applicable to gaseous samples, including flue gases from electrical utility boilers and waste incinerators.

1.3 These test methods make no attempt to teach the basic principles of the instrumentation used although minimum requirements for instrument selection are referenced in the References section. However, the preparation of samples for the above test methods is described. No details of instrument operation are included here. These are best obtained from the manufacturer of the specific instrument in use.

1.4 Currently, there are no ISO test methods that are equivalent to the test methods outlined in this standard.

1.5 *This standard does not purport to address all of the safety concerns, if any, associated with its use. It is the responsibility of the user of this standard to establish appropriate safety and health practices and determine the applicability of regulatory limitations prior to use.*

2. Referenced Documents

2.1 *ASTM Standards:*[2]

D883 Terminology Relating to Plastics

3. Terminology

3.1 The definitions of terms used in these test methods are referenced in order that the practitioner may require further information regarding the practice of the art of isotope analysis and to facilitate performance of these test methods.

3.2 Terminology D883 should be referenced for terminology relating to plastics. Although an attempt to list terms in a logical manner (alphabetically) will be made as some terms require definition of other terms to make sense.

3.3 *Definitions:*

3.3.1 *dpm*—disintegrations per minute. This is the quantity of radioactivity. The measure dpm is derived from cpm or counts per minute (dpm = cpm – bkgd / counting efficiency). There are 2.2 by 10^6 dpm / uCi (**14,17**).[3]

3.3.2 *dps*—disintegrations per second (rather than minute as above) (**14,17**).

3.3.3 *scintillation*—the sum of all photons produced by a radioactive decay event. Counters used to measure this as described in these test methods are Liquid Scintillation Counters (LSC) (**14,17**).

3.3.4 *specific activity (SA)*—refers to the quantity of radioactivity per mass unit of product, that is, dpm per gram (**14,17**).

3.3.5 *automated efficiency control (AEC)*—a method used by scintillation counters to compensate for the effect of quenching on the sample spectrum (**14**).

3.3.6 *AMS facility*—a facility performing Accelerator Mass Spectrometry.

3.3.7 *accelerator mass spectrometry (AMS)*—an ultrasensitive technique that can be used for measuring naturally occurring radio nuclides, in which sample atoms are ionized, accelerated to high energies, separated on basis of momentum, charge, and mass, and individually counted in Faraday collectors. This high energy separation is extremely effective in filtering out isobaric interferences, such that AMS may be used to measure accurately the $^{14}C/^{12}C$ abundance to a level of 1 in 10^{15}. At these levels, uncertainties are based on counting statistics through the Poisson distribution (**8,9**).

3.3.8 *background radiation*—the radiation in the natural environment; including cosmic radiation and radionuclides

[1] These test methods are under the jurisdiction of ASTM Committee D20 on Plastics and are the direct responsibility of Subcommittee D20.96 on Environmentally Degradable Plastics and Biobased Products.

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[2] For referenced ASTM standards, visit the ASTM website, www.astm.org, or contact ASTM Customer Service at service@astm.org. For *Annual Book of ASTM Standards* volume information, refer to the standard's Document Summary page on the ASTM website.

[3] The boldface numbers in parentheses refer to the list of references at the end of this standard.

***A Summary of Changes section appears at the end of this standard**

present in the local environment, for example, materials of construction, metals, glass, concrete (2,4,7,8,14-19).

3.3.9 *biobased content*—the amount of biobased carbon in the material or product as a percent of the weight (mass) of the total organic carbon in the product (1).

3.3.10 *coincidence circuit*—a portion of the electronic analysis system of a Liquid Scintillation Counter which acts to reject pulses which are not received from the two Photomultiplier Tubes (that count the photons) within a given period of time and are necessary to rule out background interference and required for any LSC used in these test methods (7,14,17).

3.3.11 *coincidence threshold*—the minimum decay energy required for a Liquid Scintillation Counter to detect a radioactive event. The ability to set that threshold is a requirement of any LSC used in these test methods (14,17).

3.3.12 *contemporary carbon*—a direct indication of the relative contributions of fossil carbon and "living" biospheric carbon can be expressed as the fraction (or percentage) of contemporary carbon, symbol f_C. This is derived from f_M through the use of the observed input function for atmospheric ^{14}C over recent decades, representing the combined effects of fossil dilution of ^{14}C (minor) and nuclear testing enhancement (major). The relation between f_C and f_M is necessarily a function of time. By 1985, when the particulate sampling discussed in the cited reference the f_M ratio had decreased to ca. 1.2 (8,9).

3.3.13 *chemical quenching*—a reduction in the scintillation intensity (a significant interference with these test methods) seen by the Photomultiplier Tubes (PMT, pmt) due to the materials present in the scintillation solution that interfere with the processes leading to the production of light. The result is fewer photons counted and a lower efficiency (4,7,17).

3.3.14 *chi-square test*—a statistical tool used in radioactive counting in order to compare the observed variations in repeat counts of a radioactive sample with the variation predicted by statistical theory. This determines whether two different distributions of photon measurements originate from the same photonic events. LSC instruments used in this measurement should include this capability (14,17,27).

3.3.15 *cocktail*—the solution in which samples are placed for measurement in a LSC. Solvents and Scintillators (chemicals that absorbs decay energy transferred from the solvent and emits light (photons) proportional in intensity to the deposited energy) (4,7,14,17).

3.3.16 *decay (radioactive)*—the spontaneous transformation of one nuclide into a different nuclide or into a different energy state of the same nuclide. The process results in a decrease, with time, of the number of original radioactive atoms in a sample, according to the half-life of the radionuclide (8,14,17).

3.3.17 *discriminator*—an electronic circuit which distinguishes signal pulses according to their pulse height or energy; used to exclude extraneous radiation, background radiation, and extraneous noise from the desired signal (14,17,18,32).

3.3.18 *efficiency*—the ratio of measured observations or counts compared to the number of decay events which occurred during the measurement time; expressed as a percentage (14,17).

3.3.19 *external standard*—a radioactive source placed adjacent to the liquid sample in to produce scintillations in the sample for the purpose of monitoring the sample's level of quenching (14,17).

3.3.20 *figure of merit*—a term applied to a numerical value used to characterize the performance of a system. In liquid scintillation counting, specific formulas have been derived for quantitatively comparing certain aspects of instrument and cocktail performance and the term is frequently used to compare efficiency and background measures (14,17,20).

3.3.21 *fluorescence*—the emission of light resulting from the absorption of incident radiation and persisting only as long as the stimulation radiation is continued (14,17,25).

3.3.22 *fossil carbon*—carbon that contains essentially no radiocarbon because its age is very much greater than the 5730 year half-life of ^{14}C (8,9).

3.3.23 *half-life*—the time in which one half the atoms of a particular radioactive substance disintegrate to another nuclear form. The half-life of ^{14}C is 5730 years (8,14,25).

3.3.24 *intensity*—the amount of energy, the number of photons, or the numbers of particles of any radiation incident upon a unit area per unit time (14,17).

3.3.25 *internal standard*—a known amount of radioactivity which is added to a sample in order to determine the counting efficiency of that sample. The radionuclide used must be the same as that in the sample to be measured, the cocktail should be the same as the sample, and the Internal Standard must be of certified activity (14,17).

3.3.26 *modern carbon*—explicitly, 0.95 times the specific activity of SRM 4990b (the original oxalic acid radiocarbon standard), normalized to $\delta^{13}C = -19$ ‰ (Currie, et al., 1989). Functionally, the fraction of modern carbon equals 0.95 times the concentration of ^{14}C contemporaneous with 1950 wood (that is, pre-atmospheric nuclear testing). To correct for the post 1950 bomb ^{14}C injection into the atmosphere (9), the fraction of modern carbon is multiplied by 0.95 (as of the year 2010).

3.3.27 *noise pulse*—a spurious signal arising from the electronics and electrical supply of the instrument (14,17,21,29).

3.3.28 *phase contact*—the degree of contact between two phases of heterogeneous samples. In liquid scintillation counting, better phase contact usually means higher counting efficiency (14,17).

3.3.29 *photomultiplier tube (PMT, pmt)*—the device in the LSC that counts the photons of light simultaneously at two separate detectors (29,32).

3.3.30 *pulse*—the electrical signal resulting when photons are detected by the Photomultiplier tubes (14,17,18,32).

3.3.31 *pulse height analyzer (PHA)*—an electronic circuit which sorts and records pulses according to height or voltage (14,17,18,32).

3.3.32 *pulse index*—the number of afterpulses following a detected coincidence pulse (used in three dimensional or pulse height discrimination) to compensate for the background of a liquid scintillation counter performing (**14**,**18**,**29**,**32**).

3.3.33 *quenching*—any material that interferes with the accurate conversion of decay energy to photons captured by the PMT of the LSC (**2**,**4**,**7**,**14**,**15**,**17**,**20**).

3.3.34 *region*—regions of interest, also called window and/or channel in regard to liquid scintillation counters. Refers to an energy level or subset specific to a particular isotope (**4**,**14**,**18**,**21**,**29**).

3.3.35 *scintillation reagent*—chemicals that absorbs decay energy transferred from the solvent and emits light (photons) proportional in intensity to the decay energy (**4**,**14**,**29**).

3.3.36 *solvent—in scintillation reagent*, chemical(s) which act as both a vehicle for dissolving the sample and scintillator and the location of the initial kinetic energy transfer from the decay products to the scintillator; that is, into excitation energy that can be converted by the scintillator into photons (**4.14**,**17**,**29**).

3.3.37 *standard count conditions (STDCT)*—LSC conditions under which reference standards and samples are counted.

3.3.38 *three dimensional spectrum analysis*—the analysis of the pulse energy distribution in function of energy, counts per energy, and pulse index. It allows for auto-optimization of a liquid scintillation analyzer allowing maximum performance. Although different Manufacturers of LSC instruments call Three Dimensional Analysis by different names, the actual function is a necessary part of these test methods (**14**,**17**,**18**).

3.3.39 *true beta event*—an actual count which represents atomic decay rather than spurious interference (**10**,**11**).

3.3.40 *flexible tube cracker*—the apparatus in which the sample tube (Break Seal Tube) is placed (**5**,**6**,**10**,**11**).

3.3.41 *break seal tube*—the sample tube within which the sample, copper oxide, and silver wire is placed.

4. Significance and Use

4.1 Presidential (Executive) Orders 13101, 13123, 13134, Public Laws (106-224), AG ACT 2003 and other Legislative Actions all require Federal Agencies to develop procedures to identify, encourage and produce products derived from biobased, renewable, sustainable and low environmental impact resources so as to promote the Market Development Infrastructure necessary to induce greater use of such resources in commercial, non food, products. Section 1501 of the Energy Policy Act of 2005 (Public Law 109–58) and EPA 40 CFR Part 80 (Regulation of Fuels and Fuel Additives: Renewable Fuel Standard Requirements for 2006) require petroleum distributors to add renewable ethanol to domestically sold gasoline to promote the nation's growing renewable economy, with requirements to identify and trace origin.

4.2 Method B utilizes Accelerator Mass Spectrometry (AMS) along with Isotope Ratio Mass Spectrometry (IRMS) techniques to quantify the biobased content of a given product. Intsrumental error can be within 0.1-0.5 % (1 rsd), but empirical studies identify a total uncertainty up to ±3 % (absolute) inclusive of indeterminant sources of error in the final biobased content result. Sample preparation methods include the production of CO2 within a vacuum manifold system where it is ultimately distilled, quantified in a calibrated volume, transferred to a quartz tube, and torch sealed. Details are given in 8.7-8.10. The stored CO_2 is then delivered to an AMS facility for final processing and analysis.

4.3 Method C uses LSC techniques to quantify the biobased content of a product using sample carbon that has been converted to benzene. This test method determines the biobased content of a sample with a maximum total error of ±3 % (absolute), as does Method B.

4.4 The test methods described here directly discriminate between product carbon resulting from contemporary carbon input and that derived from fossil-based input. A measurement of a product's $^{14}C/^{12}C$ content is determined relative to the modern carbon-based oxalic acid radiocarbon Standard Reference Material (SRM) 4990c, (referred to as HOxII). It is compositionally related directly to the original oxalic acid radiocarbon standard SRM 4990b (referred to as HOxI), and is denoted in terms of f_M, that is, the sample's fraction of modern carbon. (See Terminology, Section 3.)

4.5 Reference standards, available to all laboratories practicing these test methods, must be used properly in order that traceability to the primary carbon isotope standards are established, and that stated uncertainties are valid. The primary standards are SRM 4990c (oxalic acid) for ^{14}C and RM 8544 (NBS 19 calcite) for ^{13}C. These materials are available for distribution in North America from The National Institute of Standards and Technology (NIST), and outside North America from the International Atomic Energy Agency (IAEA), Vienna, Austria.

4.6 Acceptable SI unit deviations (tolerance) for the practice of these test methods is ±5 % from the stated instructions unless otherwise noted.

5. Safety

5.1 The specific safety and regulatory requirements associated with radioactivity, sample preparation, and instrument operation are not addressed in these test methods. It is the responsibility of the user of these test methods to establish appropriate safety and health practices. It is also incumbent on the user to conform to all the Federal and State regulatory requirements, especially those that relate to the use of open radioactive source, in the performance of these test methods. Although ^{14}C is one of the safest isotopes to work with, State and Federal regulations must be followed in the performance of these test methods.

5.2 The use of glass and metal, in particular with closed systems containing oxygen that are subjected to 700°C temperatures pose their own safety concerns and care should be taken to protect the operators from implosion/explosion of the glass tube. *Material Safety Data Sheets* should always be followed with special concern for eye, respiratory, and skin protection. Radioactive ^{14}C compounds should be handled and disposed of in accordance with State and Federal Regulations.

NOTE 1—Prior to D6866 - 11, this standard contained a Method A, which utilized LSC and CO_2 absorption into a cocktail vial. Error was cited as ±15 % absolute due to technical challenges and low radiocarbon counts. Empirical evidence now indicates error may be ±20 % or higher in routine use. This method was removed in this revision due to the inapplicability of this low precision method to biobased analysis.

5.3 In Method C, benzene is generated from the sample carbon. Benzene is highly toxic and is an EPA-listed carcinogen. It must be handled accordingly, using all appropriate eye, skin, and respiratory protection. Samples must be handled and disposed of in accordance with State and Federal regulations. Other hazardous chemicals are also used, and must be handled appropriately (see Material Safety Data Sheets for proper handling procedures).

METHOD B

6. Apparatus and Reagents

6.1 *AMS and IRMS Apparatus:*

6.1.1 A vacuum manifold system for with capabilities for air and non-condensable gas evacuation, sample introduction, water distillation, cryogenic gas transfer, and temperature and pressure monitoring. The following equipment is required:

6.1.2 Manifold tubing that is composed of clean stainless steel and/or glass.

6.1.3 Vacuum pump(s) capable of achieving a vacuum of 101 Pa or less within the vacuum region.

6.1.4 Calibrated pressure transducers with coupled or integrated signal response controllers.

6.1.5 A calibrated sample collection volume with associated temperature readout.

6.1.6 Clean quartz tubing for sample combustion and subsequent gas transfer, quantification and storage.

6.1.7 A hydrogen/oxygen torch or other heating device and/or gas for sealing quartz tubing.

7. AMS and IRMS Reagents

7.1 A stoichiometric excess oxygen for sample combustion; introduced into sample tube as either a pure gas or as solid copper (II) oxide.

7.2 A stoichiometric excess of silver, nominally 30 mg, introduced into sample tube for the removal of halogenated species.

7.3 A −76°C slurry mixture of dry ice (frozen CO_2) and alcohol distillation and removal of sample water.

7.4 Liquid nitrogen.

8. Sample Preparation

8.1 Method B is a commonly used procedure to quantitatively combust the carbon fraction within product matrices of varying degrees of complexity. The procedure described here for Method B is recommended based on its affordability and extensive worldwide use. Nevertheless, laboratories with alternative instrumentation such as continuous flow interfaces and associated CO_2 trapping capabilities are equally suitable provided that the recovery of CO_2 is quantitative, 100 ± 5 %.

8.2 Based on the stoichiometry of the product material, sufficient sample mass shall be weighed such that 1-10 mg of carbon is quantitatively recovered as CO_2. Weighed sample material shall be contained within a pre-cleaned quartz sample container, furnace-baked at 900°C for ≥2 h, and torch sealed at one end. Typically 2 mm OD/1 mm ID quartz tubing is sufficient, however any tubing configuration needed to accommodate large sample volumes is acceptable.

8.3 The weighed sample shall then be transferred into an appropriately sized quartz tube, typically 6 mm OD/4 mm ID.

8.4 The sample, thus configured shall then be adapted to a vacuum manifold for evacuation of ambient air to a pressure 101 Pa or less.

8.5 If the material is known to be volatile or contains volatile components, the sample material within the tube shall be frozen with liquid nitrogen to –196°C prior to evacuation. The evacuated tube shall be torch sealed then combusted in a temperature controlled furnace at 900°C for 2 to 4 h.

8.6 After combustion, the quartz sample tube shall be scored to facilitate a clean break within a flexible hose portion of a "tube cracker" assembly adapted to the manifold. One example configuration of a tube cracker is shown in the photo below. The materials are composed of stainless steel. Compression fittings with appropriate welds are used to assemble the individual parts. This and alternative assemblies are given in the References section (**5**,**6**,**10**,**11**).

8.7 With the manifold closed to the vacuum pump, the quartz tubing is cracked, the sample CO_2 is liberated and immediately cryogenically (with liquid nitrogen) transferred to a sample collection bulb attached to a separate port on the manifold.

8.8 The contents of the sample collection bulb shall be distilled to remove residual water using a dry ice/alcohol slurry maintained at ca. −76°C. Simultaneously the sample CO_2 gas is released and immediately condensed in a calibrated volume.

8.9 The calibrated volume is then closed and the CO_2 shall equilibrate to room temperature.

8.10 Recovery shall be determined using the ideal gas law relationship.

8.11 The sample shall be transferred to a borosilicate break seal tube for storage and delivery to an AMS facility for analysis of $^{14}C/^{12}C$ and $^{13}C/^{12}$ C isotopic ratios.

9. Analysis, Interpretation, and Reporting

9.1 $^{14}C/^{12}C$ and $^{13}C/^{12}C$ isotopic ratios are measured using accelerator mass spectrometry. The isotopic ratios of $^{14}C/^{12}C$ and $^{13}C/^{12}C$ are determined relative to the appropriate primary reference material, that is, SRM 4990c and RM 8544, for ^{14}C and ^{13}C, respectively. Zero percent ^{14}C represents the entire lack of ^{14}C atoms in a material thus indicating a fossil (for example, petroleum based) carbon source. One hundred percent ^{14}C, after correction for the post-1950 bomb injection of ^{14}C into the atmosphere, likewise indicates an entirely modern carbon source. The percent modern carbon can be slightly greater than 100 % due to the continuing, but diminishing, effects of the 1950s nuclear testing programs.

Because all sample ^{14}C activities are referenced to a "pre-bomb" standard, all percent modern carbon values must be multiplied by 0.95 to correct for the bomb carbon and to subsequently obtain the true biobased content of the sample. References for reporting carbon isotopic ratio data are given in Refs (**27**,**3**) for ^{14}C and ^{13}C, respectively.

9.2 All percent modern carbon (pMC) values obtained from the radiocarbon analyses must be corrected for isotopic fractionation (**28**) using stable isotope data ($^{13}C/^{12}C$ ratios) obtained on CO_2 derived from combustion of the sample. Do not determine $^{13}C/^{12}C$ ratios on the raw product material itself, since that approach can lead to erroneous results in some cases.

9.3 Biobased content values that exceed 100 % should be reported as having a biobased content of 100 %. A biobased content that is greater than 103 % suggests that either an analytical error occurred, or that the source of biobased carbon is more than several years old. In such cases, the analyst should discuss the product with the manufacturer to determine if re-analysis of the product is warranted. Re-analysis would be warranted if the biobased carbon is less than two or three years old, in which case an analytical error would be suspected.

METHOD C

10. Detailed Requirements

NOTE 2—Acceptable tolerance levels of ±5 % are standard to this method unless otherwise stated.

10.1 Low level liquid scintillation analyzers with active shielding that can produce consistent background counts of less than 5 dpm.

10.2 Anticoincidence systems such as two and three photomultiplier tubes (multidetector systems).

10.3 Coincidence circuits.

10.4 Software and hardware that include thresholds and statistics, pulse rise and shape discrimination, and three-dimensional spectrum analysis.

10.5 Use of external and internal standards must be used in LSC operation.

10.6 Optimized counting regions to provide very low background counts while maintaining counting efficiency greater than 60 % of samples 0.7 to 1.5 g in clean, 3-mL, 7-mL or 20-mL low potassium glass counting vials. Alternatively, clean PTFE or quartz counting vials may be used in this method.

10.7 No single LSC is specified for this method. However, minimum counting efficiency and control of background interference is specified. Like all analytical instruments, LS counters require study as to their specific components and counting optimization.

10.8 Standardization of sample preparation is required.

10.9 Standardization and optimization of clean sample vials, which must be made of either PTFE, quartz, or low-potassium glass with PTFE tops. Sample vials may be either 3-mL, 7-mL or 20-mL in volume. Plastic vials must not be used for this method.

10.10 Counting efficiency and background optimization should be performed using a suitable reference standard (for example, NIST oxalic acid or Australian National University (ANU) sucrose) using the same reagents and counting parameters as the samples.

10.11 Counting efficiency (E) shall be determined by dividing the measured cpm by the known dpm, and multiplying this by 100 to obtain the counting efficiency as a percentage. For example, for the Oxalic Acid I standard, E = (cpm/g Oxalic Acid/ 14.27 dpm/g) × 100, where E = counting efficiency in %, cpm/g Oxalic Acid is the net activity per gram measured for the oxalic acid after subtracting background, and 14.27 dpm/g is the absolute value of the NIST "OxI" reference standard. The NIST "OxII" standard (SRM 4990C) has a slightly different ^{14}C activity level. ANU sucrose (NIST SRM 8542) can be used as a suitable standard in place of oxalic acid.

10.12 Counting interference concerns that must be addressed as part of specific instrument calibration and normalization include luminance, chemical or color quench, static electricity, random noise, temperature, and humidity variability (**13**).

10.13 Alternate regions of interest parameters may be used based upon testing of 20, or more, 6-h counts of the same reference (STDCT) standard that record the raw data and spectrum for keV regions of interest 4 through 96. Optimal counting conditions should be established by maximizing the Figure of Merit (E^2/bkg) values to obtain the highest count efficiency and the lowest background and other interference. Counting efficiency of less that 60 % is unacceptable and can be improved by LSC instrument optimization and sample/reagent compatibility or shielding improvements.

10.14 Samples will be equilibrated with reference standards under identical conditions of time and temperature.

10.15 Samples will be counted for a minimum of 10 h with region of interest (ROI) channels including ROI energy levels of 0-155 keV such that E^2/B is 1000 or higher in 20 to 120-min subsets with raw data saved to disk for later statistical analysis and documentation of stable counting conditions.

10.16 Before commercial testing, laboratories that intend to implement this method must participate in an interlaboratory comparison study to assess between laboratory reproducibility.

11. Apparatus and Reagents

11.1 *Benzene Synthesis Apparatus*

11.1.1 A benzene synthesis unit will be required to convert sample carbon to benzene. These units are commercially available, but can also be home-made if desired. Examples of benzene synthesis units are discussed in (**12**) and (**30**).

11.2 *LSC Apparatus*

11.2.1 Liquid scintillation analyzer as described in Section 10.

11.2.2 Clean low potassium scintillation vials with a volume of 3 mL, 7 mL, or 20 mL.

11.3 *LSC and Benzene Synthesis Reagents*

11.3.1 High purity oxygen for converting sample carbon to CO_2. Alternatively, technical grade oxygen can be used if scrubbed with a suitable material such as Ascarite.

11.3.2 High purity nitrogen for combining with the oxygen when combusting highly volatile samples. Alternatively, technical grade nitrogen can be used is scrubbed with a suitable material such as Ascarite.

11.3.3 Cupric oxide wire for conversion of CO to CO_2 when combusting highly volatile samples with oxygen/nitrogen blends.

11.3.4 Reagent grade powdered lithium or lithium rod (each packed in argon) for converting CO_2 to lithium carbide (Li_2C_2).

11.3.5 Reagent grade potassium chromate (in sulfuric acid) or phosphoric acid for purifying acetylene gas.

11.3.6 Suitable catalyst material such as a Si_2O_3/Al_2O_3 substrate activated with either chromium (as Cr_2O_3) or vanadium (as V_2O_5) for converting acetylene gas to benzene (**23**).

11.3.7 Scintillation cocktail.

11.3.8 De-ionized or distilled water for hydrolysis of Li_2C_2 to acetylene gas.

12. Sample Preparation and Analysis

12.1 Tolerance of ±5 % is to be assumed unless otherwise stated.

12.2 Standard procedures are to be employed for the conversion of original sample material to benzene using the liquid scintillation dating technique (**12**).

12.3 Based on the stoichiometry of the product material, sufficient sample mass shall be weighed such that quantitative recovery of the carbon would theoretically yield 1.00 to 4.00 g of carbon for conversion to benzene.

12.4 The carbon within each sample shall first be combusted to CO_2 by placing the sample in a closed system which is purged or evacuated of air.

12.5 The system is then purged several times with pure nitrogen. After verifying the integrity of the closed system, the sample is bathed in 100 % oxygen (non-volatile samples) or a mixture of nitrogen and oxygen (volatile samples) and ignited. Samples ignited using a nitrogen/oxygen mix must pass through a cupric oxide furnace at 850°C to avoid carbon loss to CO. The generated sample CO_2 is collected using liquid nitrogen cold traps. If desired, the CO_2 can be passed through a series of chemical traps to remove various contaminants prior to cryogenic collection of the CO_2 (**12**).

12.6 As an alternative combustion approach for volatile materials, the samples can be combusted in a bomb that is pressurized with oxygen to 300-400 psi. The CO_2 generated in the bomb is subsequently released to a dry ice trap for moisture removal, followed by a liquid nitrogen cold trap for CO_2 collection.

12.7 The collected CO_2 is reacted with a stoichiometric excess (3:1 lithium:carbon ratio) of molten lithium which has been preheated to 700°C. Li_2C_2 is produced by slowly bleeding the CO_2 onto the molten lithium in a stainless steel vessel (or equivalent) while under a vacuum of ≤135 mPa.

12.8 The Li_2C_2 is heated to about 900°C and placed under vacuum for 15-30 minutes to remove any unreacted gases and to complete the Li_2C_2 synthesis reactions (**27**).

12.9 The Li_2C_2 is cooled to room temperature and gently hydrolyzed with distilled or de-ionized water to generate acetylene gas (C_2H_2) by applying the water in a drop-wise fashion to the carbide. The evolved acetylene is dried by passing it through dry ice traps, and the dried acetylene in subsequently collected in liquid nitrogen traps.

12.10 The acetylene gas is purified by passing it through a phosphoric acid or potassium chromate (in sulfuric acid) trap to remove trace impurities, and by using dry ice traps to remove water.

12.11 The C_2H_2 gas is catalyzed to benzene (C_6H_6) by bleeding the acetylene onto a chromium catalyst which has been preheated to ≥90°C, or onto a vanadium catalyst (the later activates at ambient temperature). In the former case, the reaction is cooled with a water jacket to avoid decomposition from excessive heat generated during the exothermic reaction.

12.12 The benzene is thermally evolved from the catalyst at 70-110°C and then collected under vacuum at roughly -78°C. The benzene is then frozen until it is counted. Radon can be removed by pumping on the benzene while it is at dry ice temperatures.

12.13 The ^{14}C content shall be determined in a LSC with optimization of the instrument as described in Section 10. Either single vial counting or "chain" counting is acceptable.

12.14 Radiocarbon activity in the sample is to be determined by "benzene cocktail" analysis, consisting of a scintillator plus sample benzene, in constant volume and proportion. A recommended scintillator is butyl-PBD or PPO/POPOP dissolved in toluene or equivalent (**13**) and(**16**). Alternatively, some scintillators (including butyl-PBD) can be added to the benzene as a solid with good results.

12.15 Standard methods consist of counting a cocktail containing sample benzene plus a scintillation solution. For example, a cocktail might contain 4-mL sample benzene plus 0.5-mL scintillation solution. In this example, if 4 mL of sample benzene is not available, reagent grade (99.999% pure) thiophene-free benzene can be added to bring the sample volume to 4 mL. Larger or smaller volumes may be utilized depending upon the configuration of the specific laboratories counting protocols.

12.16 Scintillation counters are to be monitored for background and stability with traceable documentation.

12.17 Should anomalies appear during sample counting, the benzene is to be re-measured in another counter to verify the activity, or the sample must be completely re-analyzed.

12.18 Traceable quench detection should be performed on each sample to ensure benzene purity. In the event the sample is substantially quenched, the data should be discarded and the sample should be re-analyzed.

12.19 Measurements are to be made on an interval basis (usually 50 or 100 minutes) to allow statistical analysis of the measurement.