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CHEMICAL OXYGEN DEMAND

Thermo Orion Method CODL00 and CODH00

Revision 5, 03/25/2005

Accepted by U.S. EPA on March 31, 2005 for wastewater compliance monitoring.

1.0 Scope and Applications

1.1 This method is a convenient packaging of the technology in Standard Methods "Chemical Oxygen Demand", Method 5220 D, "Closed Reflux, Colorimetric Method".(See *Ref. 16.1*). It is based on the use of Thermo Orion Test Kits Cat. No. CODL00, or CODH00, or equivalent. The method determines chemical oxygen demand (See *Definitions, Section 3.1).* Chemical oxygen demand is an aggregate property of water, and no CAS Registry Number is appropriate.

1.2 The method consists of two procedures: the first ("Low Range") is applicable to natural and treated waters at concentrations from 15 to 150 mg/L COD; and the second ("Mid-Range") is applicable from 100 to1500 mg/L COD . Where sample results exceed the applicable range of the method, the sample must be diluted to within the applicable range and reanalyzed.

1.3 This method is for use in the Environmental Protection Agency's (EPA's) data gathering and monitoring programs under the Clean Water Act, the Resource Conservation and Recovery Act, and the Comprehensive Environmental Response, Compensation and Liability Act.

1.4 The MDL for the low range procedure in this method was determined to be 4.6 mg/L COD using an AQUAfast AQ2040 Colorimeter and an COD L00 Test Kit. This value was derived from analysis of 9 aliquots with a concentration of 7 mg/L COD. The MDL for the mid-range procedure was determined to be 16 mg/L, using 9 aliquots with a concentration of 70 mg/L COD.

1.5 Each laboratory that uses this method must demonstrate the ability to generate acceptable results using the quality control procedure in Section 9.2.

2.0 Summary of Method

2.1 Samples, standards, and blanks are heated at 150 +/- 2 ^{0}C in a closed reactor for two hours in the presence of acid dichromate solution. The tubes are then cooled and measured either at 420 nm (low range) or 610 nm (mid-range). When a sample is digested, the dichromate ion oxidizes COD material in the sample. This results in the change of chromium from the hexavalent (chromate) state to the trivalent (chromic) state. Both of these chromium species are colored and absorb in the visible region of the spectrum. For the low level, the decrease in chromate is used for the analysis, while for the higher values the chromic ion is measured directly.

3.0 Definitions

3.1 Chemical oxygen demand (COD) is defined as the amount of a specified oxidant that reacts with the sample under controlled conditions. The quantity of oxidant consumed is expressed in terms of its oxygen equivalence. Because of its unique chemical properties, the dichromate ion ($Cr_2O_7^{=}$) is the specified oxidant, and is reduced to the chromic ion (Cr^{+3}). Both organic and inorganic components of a sample are subject to oxidation, but in most cases the organic component predominates and is of the greater interest.

3.2 Material Safety Data Sheet (MSDS)- Written information provided for each chemical reagent or standard about a chemical's toxicity, health hazards, physical properties, flammability, and reactivity. It also includes storage, spill, and handling precautions.

3.3 Method Detection Limit (MDL)-The minimum concentration of an analyte that can be identified, measured and reported with 99% confidence that the analyte concentration is greater than zero.

3.4 Calibration Blank (CB)- A volume of reagent water fortified with the same matrix as the calibration standard, but without the analyte, internal standards or surrogate analytes.

3.5 Calibration Standard (CAL)- A solution prepared from the primary dilution standard or stock standards, and the internal standards and surrogate analytes. Used to calibrate the instrument response with respect to analyte concentration.

3.6 Instrument Performance Check Solution (IPC)- A solution of one or more method analytes, surrogates, internal standards, or other test substances

used to evaluate the performance of the instrument system with respect to a defined set of criteria.

3.7 Laboratory Fortified Blank (LFB)- An aliquot of reagent water or other blank matrices to which known quantities of the method analytes are added in the laboratory. The LFB is analyzed exactly like the sample and is used to determine whether the methodology is in control, and if the laboratory is capable of making accurate and precise measurement.

3.8 Laboratory Reagent Blank (LRB)- An aliquot of reagent water or other blank matrices that are treated as a sample including exposure to all glassware, equipment, solvents, reagents, internal standards, and surrogates that are used with other samples. Use the LRB to determine if method analytes or other interferences are present in the laboratory environment, reagents or apparatus.

3.9 Quality Control Sample (QCS)- A solution of method analytes of known concentration that is used to fortify an aliquot of LRB or sample matrix. Obtain the QCS from a source external to the laboratory that is different from the source of calibration standards. It is used to check laboratory performance with externally prepared test materials.

3.10 Matrix Spike (MS)- An aliquot of an environmental sample, to which a known quantity of the method analyte is added in the laboratory. The MS is analyzed exactly like a environmental sample to determine whether the sample matrix contributes bias to the analytical results. The background concentrations of the analyte in the sample matrix must be determined in a separate aliquot and the measured values in the MS corrected for background concentrations.

3.11 Matrix Spike Duplicate (MSD)- An aliquot of the same sample used for the MS, to which the exact same known quantity of the method analyte has been added as for the MS, and which is analyzed separately with the identical procedure. Analysis of the MS and MSD give a measure of the precision associated with the laboratory procedures, but not with sample collection, preservation, or storage procedures.

3.12 Calibration Range- The range of concentration of analyte for which the use of the method has been approved and for which the instrument has been pre-programmed with internal calibration as given in the Method Scope, Section 1

4.0 Interferences

4.1 The most common interferent is chloride ion. Under the rigorous digestion procedures for COD analyses, chloride, bromide, or iodide can react with

dichromate to produce the elemental form of the halogen and chromic ion. Results then are in error on the high side. The difficulties caused by the presence of the chloride can be overcome largely, though not completely, by complexing with mercuric sulfate, which is used in this procedure. This method is suitable for chloride levels up to 1000 mg/L.

5.0 Safety

5.1 Follow the test procedure carefully and observe all precautionary measures.

5.2 This method uses small amounts of mercury salts and contains significant amounts of strong acids. Always wear safety glasses, and handle samples with extreme caution.

5.3 The heating plate may reach temperatures of 150°C during the heating and cooling phases, and reacted samples will be hot. Use good laboratory practices throughout the test procedure.

5.4 An updated Material Safety Data Sheet (MSDS) is available for the reagents used in this method, which contains all known toxicological information. Detailed information is available on the Internet, as well as by toll-free telephone: 1-800-225-1480 or 1-978-232-6000. Go to www.thermo.com/waterapps. Search on "CODL00" and "CODH00" or follow links.

5.5 The COD thermoreactor is equipped to operate at either 115 or 230 volts. Before plugging in the unit, verify the correct voltage setting on the rear panel selector switch.

5.6 This method does not address all safety issues associated with its use. The laboratory is responsible for maintaining a safe work environment and a current awareness file of OSHA regulations regarding the safe handling of all chemicals. Additional information on laboratory safety can be found in *Ref. 16.2.*

6.0 Equipment and Supplies

6.1 Thermo Orion Cat. No. CODL00 or CODH00, or equivalent. These kits contain the necessary reagents.

6.2 Thermo Orion thermoreactor (heater block), Cat. No. COD125, or equivalent

6.3 Photometer, Thermo Orion AQ4000, AQ2040, or equivalent.

6.4 Pipettes, 2 mL, Lint free wipes.

7.0 Reagents and Standards

7.1 COD Standard, 1,000 ppm, Thermo Orion Cat. No. CODS01, or equivalent

7.2 COD Standard, 10,000 ppm, Thermo Orion Cat. No. CODS10, or equivalent.

8.0 Sample Collection and Storage

8.1 Preferably collect samples in glass bottles. Use of plastic containers is permissible if it is known that no organic contaminants are present in the containers.

8.2 Samples to be analyzed and reported for wastewater compliance monitoring under the Clean Water Act must be preserved by acidification to pH < 2 using concentrated H_2SO_4 and cooled to 4 deg. C at the time of collection.

8.3 Samples should be analyzed as soon as possible after collection. If storage is required, preserved samples maintained at 4 deg. C may be held for up to 28 days prior to analysis.

8.4 Blend (homogenize) all samples containing suspended solids before analysis.

8.5 Details on sampling techniques from conduits may be found in *Ref.16.3.*

9.0 Quality Control

9.1 Each laboratory using this method for compliance reporting is required to operate a formal quality control (QC) program *(see Reference 16.4).* The minimum requirements of this program are initial demonstration of laboratory capability, and the periodic analysis of laboratory reagent blanks, fortified blanks, fortified samples, duplicates and other laboratory solutions as a continuing performance check. The laboratory must maintain performance records that define the quality of the data that are generated. See Section 17.1 for QC Performance Criteria.

9.2 Initial Demonstration Of Performance

9.2.1 The initial demonstration of performance is used to characterize instrument performance (determination of calibration ranges and analysis of QCS) and laboratory performance (determination of MDLs) prior to performing analyses by this method.

9.2.2 Calibration Range Verification - The calibration range must be verified at least every six months or whenever a significant change in instrument response is observed. The verification must use a minimum of a blank and three standards that cover the entire calibration range. Any verification data must verify that the instrument is within ±15% of calibration for each concentration used. If any verification data exceeds ±15% of calibration, recalibrate the instrument and repeat the procedure. If the problem is not resolved, call the kit manufacturer for assistance.

9.2.3 Quality Control Sample (QCS)- Obtain an independent quality control standard for COD available from Thermo Orion or other commercial sources (preferably verified against American Association for Laboratory Accreditation or National Institute of Standards and Technology reference materials, if available). If needed, dilute the standard according to the directions supplied with the standard to obtain standard concentrations within the calibration range.

9.2.4 When beginning the use of this method, on a quarterly basis, or as required to meet data-quality needs, verify the calibration standards and acceptable instrument performance with the preparation and analyses of a QCS. If the determined concentrations are not within 15% of the stated values, performance is unacceptable. Identify and correct the source of the problem before proceeding with the initial determination of MDLs or continuing with on-going analyses.

9.2.5 Method Detection Limit (MDL)- MDLs must be established for all analytes, using reagent water (blank) fortified at a concentration of two to three times the estimated instrument detection limit. To determine MDL values (*40 CFR, Part 136, Appendix B*), take seven replicate aliquots of the fortified reagent water and process through the entire analytical method. Perform all calculations defined in the method and report the concentration values in the appropriate units. Calculate the MDL as follows:

$$MDL = t \times S$$

Where:

t = Student's t value for a 99% confidence level and a standard deviation estimate with n-1 degrees of freedom. S is the standard deviation of the replicate analyses. [t= 3.14 for seven replicates]

MDLs should be determined every six months, when a new operator begins work, or whenever there is a significant change in the background or instrument response.

9.3 Assessing Laboratory Performance

9.3.1 Laboratory Reagent Blank (LRB)- The laboratory must analyze at least one LRB with each batch of samples. Perform a reagent blank determination according to Section 11.0, *Procedure,* substituting DI water for the sample. Data produced are used to assess contamination from the laboratory environment. Values that exceed the MDL indicate laboratory or reagent contamination may be present and corrective actions must be taken before continuing the analysis.

9.3.2 Laboratory Fortified Blank (LFB)- The laboratory must analyze at least one LFB fortified with 50 mg/L COD with each batch of samples in the low range, and with 500 mg/L in the intermediate range. Calculate accuracy as percent recovery (see Section 9.4.2). If the recovery of any analyte falls outside the required control limits of 85-115%, that analyte is out of control. Identify and resolve the problem before continuing analyses.

9.3.3 The laboratory must use LFB analyses data to assess laboratory performance against the required control limits of 85-115%. When sufficient internal performance data are available (at least 20-30 analyses), the analyst may develop optional control limits from the percent mean recovery (x) and the single analyst standard deviation (S) of the mean recovery. Use these data to establish the upper and lower control limits as follows:

Upper Control Limit = x + 3S Lower Control Limit = x - 3S

The optional control limits must be equal to or better than the required control limits of 85-115%.

After each 5-10 new recovery measurements, calculate new control limits using only the most recent 20-30 data points. Also, use the standard deviation (S) data to establish an on-going precision statement for the level of concentrations included in the LFB. These data must be kept on file and be available for review.

9.3.4 Instrument Performance Check Solution (IPC)

For all determinations the laboratory must analyze the IPC (a midrange check standard) and a calibration blank immediately

following daily calibration, after every tenth sample (or more frequently, if required), and at the end of the sample run. Analysis of the IPC solution and calibration blank immediately following calibration must verify that the instrument is within ± 10% of calibration. Subsequent analyses of the IPC solution must verify the calibration is still within + or - 15%.

If the calibration is not within the specified limits, reanalyze the IPC solution. If the second analysis of the IPC solution confirms the calibration is outside the limits, halt sample analysis, and determine the cause. In the case of drift, recalibrate the instrument, or call the manufacturer for assistance. Reanalyze all samples following the last acceptable IPC solution. The analysis data of the calibration blank and IPC solution must be kept on file with the sample analyses data.

9.4 Assessing Analyte Recovery And Data Quality

9.4.1 Matrix Spike and Matrix Spike Duplicate (MS and MSD) The laboratory must add a known amount of analyte to an aliquot of at least 10% of the routine samples. The MS and MSD aliquots must be duplicates of the aliquot used for sample analysis. The analyte concentration must be high enough to be detected above the original sample level and should not be less than four times the MDL. The added analyte concentration should be the same as that used in the laboratory fortified blank.

9.4.2 Calculate the percent recovery for each analyte, corrected for concentrations measured in the unfortified sample, and compare these values to the designated MS/MSD recovery range of 85-115 %. In regulated monitoring, the percent relative range (RR) must be calculated using the following equation:

$$RR = 100 \frac{(|C1-C2|)}{(C1+C2)/2}$$

where:
C1 = Concentration of the analyte in the MS sample, and
C2 = Concentration of the analyte in the MSD sample.

The result obtained should be compared with the precision requirement listed in Section 17.1, “QC Performance Criteria”.

9.4.3 If the recovery of any analyte falls outside the designated LFM recovery range and the laboratory performance for that analyte is shown to be in control (see Section 9.3), the problem encountered with the LFM is matrix or solution related, not system related.

9.4.4 If reference materials are available, analyze them to provide additional performance data. Analyzing reference samples is valuable for demonstrating the ability to perform the method acceptably.

10.0 Calibration

10.1 General Precautions

10.1.1 The blank is stable when stored in the dark and can be used for future measurements with vials of the same batch.

10.1.2 Do not place hot vials in the colorimeter adaptor. Cool the vials to room temperature for final measurement.

10.1.3 Suspended solids in the vial lead to incorrect measurements. For this reason, it is important to place the vials carefully in the adaptor. Any precipitate at the bottom of the sample should not be suspended. Do not mix.

10.1.4 Clean the outside of the vials with a lint-free towel. Fingerprints or other marks should be removed.

10.1.5 Vials must always be positioned in the sample chamber so the graduation on the vials line up with the mark on the housing.

10.1.6 Avoid spilling water into the vial compartment. If water should leak into the photometer housing, it can damage electronic components cause corrosion.

10.1.7 Contamination of the optics (LED and photo-sensor) in the compartment will result in incorrect measurements. The windows in the vial compartment must be checked at regular intervals and cleaned as required. Use a moistened cloth or cotton balls.

10.1.8 Large temperature differences between the photometer and ambient conditions can lead to incorrect measurements, or build-up of condensate around the optics of the vial.

10.2 Zero Calibration

10.2.1 Place the 16 mm adapter into the AQ4000, and turn the power on. (*Note: Specific directions in Sections 10.2 -12 have been written for the AQ4000 COD meter, but other equivalent colorimeters may be used. Consult the instrument instruction manual or the manufacturer for any changes in instrument settings or operation needed to conform to this method).*

10.2.2 Take an empty 16 mm size vial and fill it with 5.0 mL distilled water or sample.

10.2.3 Screw the cap onto the vial and wipe the exterior of the vial with lint free wipes to ensure it is clean and dry.

10.2.4 Insert the vial into the AQ4000. Match the line on the vial to ♦ on the colorimeter. Cover the vial with the sample cover.

10.2.5 Press the ZERO key. The "zero" icon will light up on the upper right hand corner.

10.2.6 "WAIT" is then displayed. Result is displayed as "0.000".

10.2.7 The colorimeter is now zeroed and is ready for measurements.

11.0 Procedure

11.1 Sample Preparation

11.1.1 Choose test range appropriate for your sample. For low range (15 – 150 mg/L), a blank vial is required. Create a blank reagent vial using deionized water as an additional sample.

11.1.2 Add a 2.0 mL sample into COD vial, using the syringe provided in the kit or a 2 mL pipet.

11.1.3 Replace the cap on the sample vial and make sure it is tightly screwed on. Invert the vial several times to mix. Be sure to hold only the cap of the vial while mixing, as the vial may become very hot as contents are mixed.

11.1.4 Turn the unit on and insert prepared samples into the heating block, then press the START/TEMP key.

11.1.5 Use the default settings of a temperature of 150°C and a run time of 120 minutes. This choice will be shown by the corresponding LEDs.

11.1.6 Once the selected temperature has been reached, a beep will sound intermittently for 5 seconds. After this, the LED corresponding to the selected time will start to blink, indicating that the timer has started. The end of the cycle is indicated when the beeper sounds continuously for 5 seconds. At the completion of the cycle the heater automatically turns off and the LEDs indicating temperature and time also turn off.

11.1.7 Let the samples cool for at least 45 minutes to come to room temperature. Samples are now ready for measurement.

11.2 Blank setup

11.2.1 Press SETUP key.

11.2.2 Press “A” or “1” keys until "BLANK" is displayed.

11.2.3 Press YES key, "SET BLNK?" is displayed.

11.2.4 Press YES key, "SAMPLE?" is displayed.

11.2.5 Insert vial containing digested deionized water sample and reagent into sample chamber. Cover sample with sample cover. Press YES key and allow instrument to read the blank.

11.2.6 Blank value is displayed and unit will proceed to the next setup function.

11.2.7 Press MEAS key to proceed to the measure mode for sample measurement.

11.3 Measurement

11.3.1 Perform the Instrument Zero in Section 10. 2 above.

11.3.2 Choose the appropriate chemistries for your measurement range.

11.3.3 Select the corresponding program ID for your selected chemistry (#41 for low range, or #42 for midrange) by pressing the PRGM key, then the appropriate program number and the YES key.

11.3.4 Place cooled sample into the AQ4000 sample chamber.

11.3.5 Cover with sample cover and press the MEAS key.

11.3.6 The result will be displayed in mg/L (ppm) COD.

12.0 Data Analysis and Calculations

12.1 The meter will report the results in mg/L (ppm) COD.

12.2 Report results to the precision shown on the meter (1 mg/L).

13.0 Method Performance

13.1 The method detection limit (MDL) study was performed by one analyst, and was determined to be 4.6 mg/L COD for the low range procedure, and 16 mg/L for the medium range.

13.2 The minimum level (ML) for this method is 15 mg/L.

13.3 In a single laboratory, a single analyst performed replicate spiked sample analyses on a waste treatment effluent sample obtained from a local sewage treatment plant. The mean recovery for a 47.6 mg/L addition of COD to a diluted baseline effluent water sample containing 62 mg/L COD was 92.6% with a relative standard deviation of 7.9 % for n = 7 replicates. This experiment was run three times, using different blanks, and the results averaged. The mean recovery for a 476 mg/L addition of COD to the baseline effluent water sample containing 710 mg/L COD was 95.3% with a relative standard deviation of 1.4 % for n = 7 replicates. This experiment was run three times, using different blanks, and the results averaged.

14.0 Pollution Prevention

14.1 This method uses small amounts of inorganic mercury salts, and also contains strong acids. These materials must be disposed of by following applicable local, state and federal regulations.

15.0 Waste Management

15.1 It is the laboratory's responsibility to comply with all federal, state, and local regulations governing waste management, particularly the hazardous waste identification rules and land disposal restrictions, and to protect the air, water, and land by minimizing and controlling all releases from fume hoods and bench operations. Compliance with all sewage discharge permits and regulations is also required.

15.2 For further information on waste management, consult "*The Waste Management Manual for Laboratory Personnel*", and "*Less is Better: Laboratory Chemical Management for Waste Reduction*", both available from the American Chemical Society's Department of Government Relations and Science Policy, 1155 16th Street N.W., Washington, D.C. 20036.

16.0 References

16.1 "Standard Methods for the Examination of Water and Wastewater", 20th ed.,1998, Method 5220 D, pp. 5-17, 5-18.

16.2 "OSHA Safety and Health Standards, General Industry," (29 CFR 1910), Occupational Safety and Health Administration, OSHA 2206 (Revised, January 1976).

16.3 "Standard Practices for Sampling Water," ASTM Annual Book of Standards, Part 31, D3370-76, American Society for Testing and Materials, 100 Barr Harbor Drive, West Conshohocken, PA 19428.

16.4 40 CFR part 136, Appendix A, Methods 1624 and 1625. See also, "Handbook of Analytical Quality Control in Water and Wastewater Laboratories," USEPA, EMS-CI, Cincinnati, OH 45268, EPA-600/4-79-019, March 1979.

17.0 Tables and Validation Data

17.1 QC Performance Criteria

Type	Frequency	Acceptance Criteria
Laboratory Reagent Blank (LRB)	Daily	10 mg/L COD for the low range procedure, and 30 mg/L for the mid-range procedure.
Precision (duplicates MS and MSD)	5-10%	RR equal to, or less than, 15% at 50 mg/L COD for the low range, and at 500 mg/l for the mid-range procedure.
Accuracy (LFB or MS/MSD)	5-10%	85-115 % recovery at 50 mg/L COD.
Instrument Performance Check (IPC)	Immediately after any calibration; after every 10th sample and at the end of a sample run.	85-115% of the initial calibration by the analyst at 50 mg/L COD for the low range, and at 500 mg/ L for the mid-range, procedure
Independent Standard (QCS)	Initially, or quarterly, and as required to meet data quality needs	85-115% recovery at 50 mg/L COD for the low range, and at 500 mg/l for the mid-range, procedure.