NC DEQ/DWR WASTEWATER/GROUNDWATER LABORATORY CERTIFICATION

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| LABORATORY NAME: | | CERT #: | |
| PRIMARY ANALYST: | | DATE: | |
| NAME OF PERSON COMPLETING CHECKLIST (PRINT): | | | |
| SIGNATURE OF PERSON COMPLETING CHECKLIST: | | | |

Parameter: **Chemical Oxygen Demand (COD)**
**Closed Reflux, Colorimetric Method**
Method: **Standard Methods 5220 D – 2011 (Aqueous)**

**Chemical Oxygen Demand is considered a method-defined parameter per the definition in the Code of Federal Regulations, Part 136.6, Section (a) (5). This means that the method may not be modified per Part 136.6, Section (b) (3).**

Equipment:

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| | Digestion vessels:<br>□ Borosilicate culture tubes w/ TFE-lined screw caps [**Circle:** 16 x 100 mm, 20 x 150 mm, or 25 x 150 mm]<br>□ Borosilicate ampules, 10 mL<br>□ Commercially supplied digestion vessels with premixed reagents<br>o High range<br>o Low range | | Spectrophotometer, 600 nm and/or 420 nm. **Model:** | | Volumetric pipettes, Class A |
| | Block heater, 150 ± 2°C<br>**Model**: | | Cuvettes | | |

**PLEASE COMPLETE CHECKLIST IN INDELIBLE INK**
**Please mark Y, N or NA in the column labeled LAB to indicate the common lab practice and in the column labeled SOP to indicate whether it is addressed in the SOP.**

| | GENERAL | LAB | SOP | EXPLANATION |
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| 1 | Is the SOP reviewed at least every 2 years? What is the most recent review/revision date of the SOP? [15A NCAC 2H .0805 (a) (7)]<br>**ANSWER:** | | | Quality assurance, quality control, and Standard Operating Procedure documentation shall indicate the effective date of the document and be reviewed every two years and updated if changes in procedures are made.<br>Verify proper method reference. During review notate deviations from the approved method and SOP. |
| 2 | Are all revision dates and actions tracked and documented? [15A NCAC 2H .0805 (a) (7)] | | | Each laboratory shall have a formal process to track and document review dates and any revisions made in all quality assurance, quality control and SOP documents. |
| 3 | Is there North Carolina data available for review? | | | If not, review PT data. |
| | **PRESERVATION and STORAGE** | **LAB** | **SOP** | **EXPLANATION** |
| 4 | Are samples preserved at the time of collection with $H_2SO_4$ to pH of <2? [40 CFR Part 136.3, Table II] | | | |
| 5 | Are samples iced to above freezing but ≤ 6°C during shipment? [40 CFR Part 136.3, Table II and footnote 18] | | | 40 CFR footnote 2 allows 15 minutes for sample preservation, including thermal. This means that if a sample is received in the lab within 15 minutes, it is not required to be on ice. Document temperature downward trend for short transport samples. |
| 6 | Is pH checked to document pH <2 upon receipt? [40 CFR Part 136.3, Table II] | | | pH indicator strips may be used. |
| 7 | What action is taken if pH is >2? [15A NCAC 2H .0805 (a) (7) (M)]<br>**ANSWER:** | | | If another sample cannot be collected, analyze immediately or adjust pH to <2 and notify NC WW/GW Laboratory Certification that a non-compliant sample was received and analyzed. |

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| 8 | Are samples refrigerated above freezing to 6°C during storage? [40 CFR Part 136.3, Table II and footnote 18] | | | |
| 9 | Are samples analyzed within 28 days of collection? [40 CFR Part 136.3, Table II] | | | |
| | **PROCEDURE – Calibration** | **LAB** | **SOP** | **EXPLANATION** |
| 10 | What is your laboratory's reporting limit? [15A NCAC 2H .0805 (a) (7) (H)]<br><br>**ANSWER:** | | | Greater than or equal to the lowest calibration or calibration verification standard. |
| 11 | Does the laboratory construct their own curve or verify a factory set curve?<br><br>**ANSWER:** | | | |
| 12 | List the values of standards used for the lab generated calibration or factory-set curve verification [SM 5220 D-2011 (4) (c)]<br><br>**ANSWER:** | | | **SM 5220 D-2011 (4) (c) -** Prepare at least five standards from potassium hydrogen phthalate solution with COD equivalents to cover each concentration range. |
| 13 | Are calibration standards digested using the same procedure as for samples? [SM 5220 D-2011 (4) (c)] | | | Make up to volume with reagent water; use same reagent volumes, tube, or ampule size, and digestion procedure as for samples. |
| 14 | When using a factory-set curve, is the curve verified at the following times? [SM 5220 D-2011 (4) (c)]<br><br>o Every 12 months □YES □NO<br>o With each new lot of tubes or ampules □YES □NO<br>o When ICV differs by ≥5% □YES □NO | | | **SM 5220 D-2011 (4) (c):** Prepare calibration curve for each new lot of tubes or ampules or when standards prepared in ¶ *a* above differ by ≥5% from calibration curve.<br><br>**15A NCAC 2H .0805 (a) (7) (H) (v):** For colorimetric analyses, a series of five or more non-zero standards for a curve prepared every 12 months or three or more non-zero standards for curves established each day, or standards as set forth in the analytical procedure, shall be analyzed to establish a calibration curve.<br><br>**Note: Due to the method requirements, 3 standards daily cannot be used in place of 5 standards.** |
| 15 | When the laboratory generates a calibration curve, is a new calibration curve constructed at the following times? [SM 5220 D-2011 (4) (c) and 15A NCAC 2H .0805 (a) (7) (H) (v)]<br><br>o Every 12 months □YES □NO<br>o With each new lot of tubes or ampules □YES □NO<br>o When ICV differs by ≥5% □YES □NO | | | **SM 5220 D-2011 (4) (c):** Prepare at least five standards from potassium hydrogen phthalate solution with COD equivalents to cover each concentration range. Make up to volume with reagent water; use same reagent volumes, tube, or ampule size, and digestion procedure as for samples. Prepare calibration curve for each new lot of tubes or ampules or when standards prepared in ¶ *a* above differ by ≥5% from calibration curve.<br><br>**15A NCAC 2H .0805 (a) (7) (H) (v):** For colorimetric analyses, a series of five or more non-zero standards for a curve prepared every 12 months or three or more non-zero standards for curves established each day, or standards as set forth in the analytical procedure, shall be analyzed to establish a calibration curve.<br><br>**Note: Due to the method requirements, 3 standards daily cannot be used in place of 5 standards.** |

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| 16 | What acceptance criterion is used for the curve verification? [NC WW/GW LC Policy]<br><br>**ANSWER:** | | | Each calibration verification standard must be within ±10% of its expected value unless different criteria are specified in an individual method. At the lower limit of the operational range, acceptance criteria may be wider. Such criteria must be defined in the laboratory's standard operating procedure or quality assurance manual. If the measured concentrations vary by more than the stated acceptance criteria, the factory-set calibration curve must not be used for compliance monitoring until troubleshooting is carried out to determine and correct the source of error. |
| | **PROCEDURE – Sample Preparation** | **LAB** | **SOP** | **EXPLANATION** |
| 17 | If borosilicate culture tubes are used, are they and their caps washed with 20% $H_2SO_4$ before first use? [SM 5220 D-2011 (4) (a)] | | | **SM 5220 D-2011 (4) (a)** – Prepare, digest, and cool samples, blank, and one or more standards as directed in 5220 C. 4. Note safety precautions. Premixed reagents in digestion tubes are available commercially.<br>**SM 5220 C-2011 (4)** - Wash culture tubes and caps with 20% $H_2SO_4$ before first use to prevent contamination. |
| 18 | If commercially prepared digestion tubes with premixed reagents are used, is traceability documented on the laboratory benchsheets? [NC WW/GW LC Policy] | | | The laboratory shall have a documented system of traceability for the purchase, preparation, and use of all chemicals, reagents, standards, and consumables.<br><br>All chemicals, reagents, standards and consumables used by the laboratory must have the following information documented: Date received, Date Opened (in use), Vendor, Lot Number, and Expiration Date (where specified). This information as well as the vendor and/or manufacturer, lot number, and expiration date must be retained for primary standards, chemicals, reagents, and materials used for a period of five years. **Consumable materials such as pH buffers, lots of pre-made standards and/or media, solids and bacteria filters, etc. are included in this requirement**. |
| 19 | If commercially prepared digestion tubes with premixed reagents are used, do they contain mercury? [40 CFR Part 136.6 (b) (1)] | | | There are versions of mercury-free digestion vials available, but these are not approved under method modification due to changes in underlying chemistry. |
| 20 | What sample volume is analyzed? [SM 5220 D-2011 (4) (a)]<br><br>**ANSWER:** | | | **SM 5220 D-2011 (4) (a)** – Measure a suitable volume of sample and reagents into tube or ampule as indicated in Table 5220:I. |
| 21 | Is Class A volumetric ware used to measure sample and digestion solution volumes? [SM 5220 D-2011 (4) (a)] | | | **SM 5220 D-2011 (4) (a)** – Prepare, digest, and cool samples, blank, and one or more standards as directed in 5220 C. 4. Note safety precautions. It is critical that the volume of each component be known and that the total volume be the sample for each reaction vessel. If volumetric control is difficult, transfer digested samples, dilute to a known volume, and read. Premixed reagents in digestion tubes are available commercially.<br>**SM 5220 C-2011 (4)** – Make volumetric measurements as accurate as practical; use Class A volumetric ware. The most critical volumes are of the sample and digestion solution….Measure $H_2SO_4$ to ± 0.1 mL. The use of hand-held pipettors with non-wetting (polyethylene) pipet tips is practical and adequate. |

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| 22 | Are tubes placed in a block digester preheated to 150°C and refluxed for 2 h? [SM 5220 D-2011 (4) (a)] | | | **SM 5220 D-2011 (4) (a)** – Prepare, digest, and cool samples, blank, and one or more standards as directed in 5220 C. 4.<br>**SM 5220 C-2011 (4)** – Place tubes or ampules in block digester preheated to 150°C and reflux for 2 h behind a protective shield. |
| 23 | Is the temperature of the block digester checked and documented to be 150°C? [15A NCAC 2H .0805 (a) (7) (E)] | | | All analytical records, including original observations and information necessary to facilitate historical reconstruction of the calculated results, shall be maintained for five years. All analytical data and records pertinent to each certified analysis shall be available for inspection upon request. |
| 24 | Are samples slowly cooled to room temperature after digestion? [SM 5220 D-2011 (4) (b)] | | | Cool sample to room temperature slowly to avoid precipitate formation. |
| 25 | Once cooled, are samples vented to relieve pressure, and then mixed? [SM 5220 D-2011 (4) (b)] | | | Once samples are cooled, vent, if necessary, to relieve any pressure generated during digestion. Mix contents of reaction vessels to combine condensed water and dislodge insoluble matter. |
| 26 | Is suspended material allowed to settle prior to measuring absorption of each sample, blank and standard at the selected wavelength (420 nm or 600 nm)? [SM 5220 D-2011 (4) (b)] | | | Let suspended matter settle and ensure that optical path is clear. Measure absorption of each sample blank and standard at selected wavelength (420 nm or 600 nm). |
| | **PROCEDURE – Sample Analysis** | **LAB** | **SOP** | **EXPLANATION** |
| 27 | Is an <u>undigested blank</u> used as the reference solution for samples analyzed at 600 nm? [SM 5220 D-2011 (4) (b)] | | | **HR – SM 5220 D-2011 (4) (b) -** At <u>600 nm</u>, use an **undigested blank** as reference solution. Analyze a digested blank to confirm good analytical reagents and to determine the blank COD; subtract blank COD from sample COD. Alternately, use **digested blank** as the reference solution once it is established that the blank has a low COD.<br><br>In other words, for the high range (negligible dichromate contribution) – You would zero the instrument with an undigested blank and then analyze a digested blank. Subtract the value of the digested blank from the samples. In this scenario, you are really only looking at how much COD is attributed to the digestion reagents and subtracting it out. However, if the digested blank has a low value (e.g., <½ RL), you could then just zero the instrument with the digested blank and report samples as analyzed. No blank subtraction. |
| 28 | Is a digested blank analyzed and the value determined subtracted from the sample COD for samples analyzed at 600 nm? SM 5220 D-2011 (4) (b)] | | | See above |
| 29 | Is <u>reagent water</u> used as the reference solution for samples analyzed at 420 nm? [SM 5220 D-2011 (4) (b)] | | | **LR – SM 5220 D-2011 (4) (b) -** At <u>420 nm</u>, use **reagent water** as a reference solution. Measure all samples, blanks, and standards against this solution. The absorption measurement of an undigested blank containing dichromate with reagent water replacing sample, will give initial dichromate absorption. Any digested sample, blank, or standard that has a COD value will give lower absorbance because of the decrease in dichromate ion. Analyze a **digested blank** with reagent water replacing sample to ensure reagent quality and to determine the reagents' contribution to the decrease in absorbance during a given digestion. The difference between absorbances of a given digested sample and the digested blank is a measure of |

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| | | | | the sample COD. When standards are run, plot differences of digested blank absorbance and digested standard absorbance versus COD values for each standard.<br>In other words, For the low range (unknown dichromate contribution) – You would zero with plain reagent water (no dichromate present). Measure an undigested blank against the reagent water to determine initial dichromate level (this seems to be just a check and is not used in any calculation). The value of the digested blank will tell you how much COD is attributed to the digestion reagents. Then you subtract that from each digested standard and sample to get the COD value. |
| 30 | Is a digested blank analyzed and the value determined subtracted from the sample COD for samples analyzed at 420 nm? [SM 5220 D-2011 (4) (b)] | | | See above |
| 31 | Are over-range samples diluted to fall within the range of the calibration curve? [SM 5220 D-2011 (1) (a)] | | | Higher values can be obtained by sample dilution. |
| | **QUALITY ASSURANCE** | **LAB** | **SOP** | **EXPLANATION** |
| 32 | Is a method blank analyzed daily or with each batch of 20 or fewer samples, whichever is more frequent? [SM 5020 B-2010 (2) (d)] | | | When appropriate (Table 5020:I), include at least one MB daily or with each batch of 20 or fewer samples, whichever is more frequent. |
| 33 | Is the method blank concentration less than or equal to ½ the concentration of the lowest calibration standard? [15A NCAC 2H .0805 (a) (7) (H) (i)] | | | The concentration of reagent, method, and calibration blanks shall not exceed 50 percent of the lowest reporting concentration or as otherwise specified by the reference method. |
| 34 | What corrective action is taken if the method blank is not acceptable? [15A NCAC 2H .0805 (a) (7) (B) and SM 5020B-2010 (2) (d)]<br><br>**ANSWER:** | | | Our Rule requires corrective action any time quality control results indicate a problem.<br>**SM states:** If any MB measurements are at or above the reporting level, take immediate corrective action as outlined in Section 1020 B.5. This may include re-analyzing the sample batch or qualifying the reported data. Sample results less than the MRL are considered valid even if the MB has a positive result, but **should** be flagged. |
| 35 | Is the calibration verified by analyzing a **calibration verification standard** initially, after every 10th sample and at the end of the run? [15A NCAC 2H .0805 (a) (7) (H)]<br><br>**List acceptance criterion and value(s) of standard used:** | | | A calibration blank and calibration verification standard shall be analyzed prior to sample analysis, after every tenth sample, and at the end of each sample group, unless otherwise specified by the method, to check for carryover and calibration drift. |
| 36 | What corrective action does the laboratory take if the calibration verification standard result differs by ≥5% of the true value? [SM 5020 B-2010 (2) (b)]<br><br>**ANSWER:** | | | If a calibration verification fails, immediately cease analyzing samples and initiate corrective action. Then, re-analyze the calibration standard and blank. If the calibration verification passes, continue the analysis. Otherwise, repeat initial calibration and re-analyze samples run since the last acceptable calibration verification.<br><br>**SM 5220 D-2011 states:** Prepare calibration curve for each new lot of tubes or ampules or when standards prepared in ¶ *a* above **differ by ≥5% from calibration curve.** |
| 37 | Is the calibration verified by analyzing a **calibration blank** initially, after every 10th sample and at the end of the run? [15A NCAC 2H .0805 (a) (7) (H)]<br><br>**List acceptance criterion:** | | | A calibration blank and calibration verification standard shall be analyzed prior to sample analysis, after every tenth sample, and at the end of each sample group, unless otherwise specified by the method, to check for carryover and calibration drift. |

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| 38 | What corrective action is taken if the calibration blank results are greater than one-half the reporting level? [15A NCAC 2H .0805 (a) (7) (B)]<br><br>**ANSWER:** | | | If quality control results fall outside established limits or show an analytical problem, the laboratory shall identify the Root Cause of the failure. The problem shall be resolved through corrective action, the corrective action process documented, and any samples involved shall be reanalyzed, if possible. If the sample cannot be reanalyzed, or if the quality control results continue to fall outside established limits or show an analytical problem, the results shall be qualified as such. |
| 39 | Does the laboratory analyze a second source standard to verify standard preparation of a lab-generated curve? [SM 5020 B-2010 (2) (b) and (2) (e)]<br><br>**List value(s) and acceptance criterion of standard used.** | | | Only applies to lab generated curves since all standards are second source when verifying a factory set curve.<br>A second source standard must be analyzed at least initially to confirm the accuracy of the standard preparation. The Laboratory Fortified Blank (LFB) may serve as the second source standard (refer to question #39). If the LFB is second source, use control charts to establish limits.<br>If a purchased quality control standard is used, the manufacturer's limits may be used.<br>**SM 5020 B (2) (b) states:** If the LFB is **not** prepared from a second source to confirm method accuracy, (unless the method specifies otherwise) the lab must also verify the accuracy of its standard preparation by analyzing a mid-level second-source calibration standard **whenever a new initial calibration curve is prepared**. Results must agree within 15%, unless otherwise specified in a method.<br>**SM 5020 B (2) (e) states:** LFBs (and LFMs) do not have to be made from a second source (unless the method specifies otherwise) as long as each initial calibration solution is verified via a second source. |
| 40 | What corrective action is taken if the second source standard recovery does not meet the acceptance criterion? [15A NCAC 2H .0805 (a) (7) (B)]<br><br>**ANSWER:** | | | Our Rule requires corrective action any time quality control results indicate a problem.<br>The standard should be remade and analyzed again. If it still does not pass, a new calibration curve must be analyzed. |
| 41 | Does the laboratory analyze a laboratory-fortified blank (LFB) at least daily or per batch of 20 or fewer samples? [SM 5020 B-2010 (2) (e)]<br><br>**List value(s) and acceptance criterion of standard used.** | | | The LFB may be either a primary or secondary source standard so it may serve dual roles.<br>If the LFB is primary source, it may be equivalent to the CVS (refer to question #37).<br>If the LFB is secondary source, it may be equivalent to the second source standard (refer to question #37). Analyze one daily or per batch of 20 or fewer samples. Use control charts to establish limits.<br>**SM states:** LFBs (and LFMs) do not have to be made from a second source (unless the method specifies otherwise) as long as each initial calibration solution is verified via a second source. Ideally, vary LFB concentrations to cover the range from the midpoint to the lower part of calibration curve, including the reporting limit. Include one LFB daily or per each batch of 20 or fewer samples. |

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| 42 | What is the acceptance criterion for LFB recovery? [15A NCAC 2H .0805 (a) (7) (A)]<br><br>**ANSWER:** | | | Unless specified by the method or this Rule, each laboratory shall establish performance acceptance criteria for all quality control analyses. |
| 43 | What corrective action is taken if the LFB recovery does not meet the acceptance criterion? [15A NCAC 2H .0805 (a) (7) (B)]<br><br>**ANSWER:** | | | Our Rule requires corrective action any time quality control results indicate a problem. |
| 44 | Is a Laboratory Fortified Matrix (LFM)/Laboratory Fortified Matrix Duplicate (LFMD) analyzed with each batch of 20 or fewer samples? [SM 5020 B-2010 (2) (g)] | | | Laboratory fortified matrix is the same as a matrix spike; that is, a spiked sample. Note: **There is an option to perform an environmental sample duplicate and then spike separately – Table 5020 B indicates that duplicates or LFMD of the sample will be run.** |
| 45 | How is the LFM (spike) prepared? [SM 5020 B-2010 (2) (g)]<br><br>**ANSWER:** | | | See *Matrix Spike Technical Assistance* document. Use the same solution as the LFB for the LFM to evaluate bias attributed to matrix and accuracy of the LFM.<br>**SM states:** To prepare an LFM, add a known concentration of analytes (**ideally** from a second source) to a randomly selected routine sample without increasing its volume by more than 5%. Ideally the new concentration **should be** at or below the midpoint of the calibration curve, and for maximum accuracy, the spike **should** approximately double the sample's original concentration. If necessary, dilute the spiked sample to bring the measurement within the calibration curve. Also rotate the range of spike concentrations to verify performance at various levels. If the spike solution contribution to the fortified sample is kept to 1% or less, a spike dilution correction does not have to be calculated. |
| 46 | What is the acceptance criterion for LFM/LFMD **percent recovery**? [SM 5020 B-2010 (2) (g)]<br><br>**ANSWER:** | | | If analyzing LFMD instead of sample duplicate, there will be two % recovery calculations for accuracy from spike recoveries and one RPD calculation for precision from duplicate calculation.<br>**SM states:** Calculate percent recovery and RPD, plot control charts (unless the method specifies acceptance criteria), and determine control limits for spikes at different concentrations. |
| 47 | What corrective action does the laboratory take if the LFM/LFMD results are outside of established control limits for **accuracy**? [15A NCAC 2H .0805 (a) (7) (B)]<br><br>**ANSWER:** | | | Our Rule requires corrective action any time quality control results indicate a problem. Compare to LFB result and other QC. Reanalyze LFM. If it still fails, qualify the spiked sample result.<br>**SM states:** Ensure that the method's performance criteria are satisfied. |
| 48 | What is the acceptance criterion for LFM/LFMD or sample/duplicate **relative percent difference**? [SM 5020 B-2010 (2) (g)]<br><br>**ANSWER:** | | | **SM states:** Calculate percent recovery and relative percent difference, plot control charts (unless method specifies acceptance criteria) and determine control limits for spikes at different concentrations. Ensure the method's performance criteria are satisfied.<br>**Bottom line:** We are not requiring control charts but will instead accept a system of trend analysis. That is, the lab's monitoring of the trends in the data. 40 CFR part 136.7 (viii) states: Control charts (or other trend analysis of quality control results). |

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| 49 | What corrective action does the laboratory take if the duplicate results are outside of established control limits for **precision**? [15A NCAC 2H .0805 (a) (7) (B)]<br><br>**ANSWER:** | | | Our Rule requires corrective action any time quality control results indicate a problem.<br>**SM states:** Ensure the method's performance criteria are satisfied. |
| 50 | Is a lower reporting limit standard analyzed or back-calculated with each analysis? [15A NCAC 2H .0805 (a) (7) (H)] | | | Laboratories shall analyze or back-calculate a standard at the same concentration as the lowest reporting concentration each day samples are analyzed. |
| 51 | What is the acceptance criterion for the lower reporting limit standard? [15A NCAC 2H .0805 (a) (7) (A)]<br><br>**ANSWER:** | | | Unless specified by the method or this Rule, each laboratory shall establish performance acceptance criteria for all quality control analyses. |
| 52 | What corrective action does the laboratory take if the lower reporting limit standard does not meet the acceptance criterion? [15A NCAC 2H .0805 (a) (7) (B) and SM 5220 D-2011 (4) (c)]<br><br>**ANSWER:** | | | Prepare a new standard. If it still doesn't pass, recalibrate/re-verify the curve. |
| 53 | Is the data qualified on the Discharge Monitoring Report (DMR) or client report if Quality Control (QC) requirements are not met? [15A NCAC 2H .0805 (a) (7) (B) ] | | | If the sample cannot be reanalyzed, or if the quality control results continue to fall outside established limits or show an analytical problem, the results shall be qualified as such.<br><br>If data qualifiers are used to qualify samples not meeting QC requirements, the data may not be useable for the intended purposes. It is the responsibility of the laboratory to provide the client or end-user of the data with sufficient information to determine the usability of the qualified data. |

If samples, standards, and blanks are run under the same conditions of volume and optical path length, calculate COD as follows:

$$\text{COD as mg}O_2\text{/L} = \frac{\text{mg } O_2 \text{ in final volume x 1000}}{\text{mL sample}}$$

It is recommended that the laboratory rotate the position of samples, blanks and quality control samples in the block digester during TKN, COD and metals digestions. Random placement will help to identify when optimal performance is not achieved in an individual sample well. It is also recommended that the laboratory implement a temperature grid check of the block digester by alternating the well location of the thermometer and quality control samples each time samples are digested. This will document heating uniformity and consistency of all sample wells in the block.

Additional Comments:

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Inspector: _____ Date: _____