SIDE BY SIDE COMPARISON TABLE FOR HACH METHOD NUMBER 10265 COLORIMETRIC DETERMINATION OF CYANIDE

TOPIC	EPA METHOD NUMBER 335.2 (1980) Direct Photometric Method 0.04 – 0.80 mg cyanide/L	HACH METHOD NUMBER 10265 (1 January 2015) 0.01 - 0.60 mg cyanide/L
SCOPE AND APPLICATION	1. This method is applicable to the determination of cyanide in drinking, surface and saline waters, domestic and industrial wastes. 2. The colorimetric procedure is used for concentrations below 1 mg/L of cyanide and is sensitive to about 0.02 mg cyanide/L.	1. For wastewater, seawater, drinking water, surface water and process water. 2. The method is capable of measuring cyanide from 0.01 to 0.60 mg cyanide/L in the aqueous phase using cyanide as a standard. The method detection limit is 0.002 mg cyanide/L.
SUMMARY OF METHOD	1. The cyanide as hydrocyanic acid (HCN) is released from cyanide complexes by means of, a reflux-distillation operation and absorbed in a scrubber containing sodium hydroxide solution. The cyanide ion in the absorbing solution is then determined colorimetrically. 2. In the colorimetric measurement the cyanide is converted to cyanogen chloride, CNCl, by reaction with chloramine-T at a pH less than 8 without hydrolyzing to the cyanate. After the reaction is complete, color is formed on the addition of pyridine-pyrazolone or pyridine-barbituric acid reagent. The absorbance is read at 620 nm when using pyridine-pyrazolone or 578 nm for pyridine-barbituric acid. To obtain colors of comparable intensity, it is essential to have the same salt content in both the sample and the standards.	Cyanides react with chlorine to form cyanogen chloride. Cyanogen chloride reacts with pyridine when barbituric acid is in the sample and forms a violet color. The measurement wavelength is 588 nm.

COMMENTS	Cyanide is defined as cyanide ion and complex cyanides converted to hydrocyanic acid (HCN) by reaction in a reflux system of a mineral acid in the presence of magnesium ion.	Same
SAFETY	Reacted samples may contain cyanide and must be disposed of as a hazardous waste. It is imperative that these materials be handled safely to prevent the release of hydrogen cyanide gas (an extremely toxic material with the smell of almonds). Most cyanide compounds are stable and can be safely stored for disposal in highly alkaline solutions (pH >11) such as 2 N sodium hydroxide. Never mix these wastes with other laboratory wastes which may contain lower pH materials such as acids or even water. Dispose of reacted solutions according to local, state and federal regulations.	Same
INTERFERENCES	1. Interferences are eliminated or reduced by using the distillation procedure 2. Sulfides adversely affect the colorimetric and titration procedures. Samples that contain hydrogen sulfide, metal sulfides or other compounds that may produce hydrogen sulfide during the distillation should be distilled by the optional procedure. 3. Fatty acids will distill and form soaps under the alkaline titration conditions, making the end point almost impossible to detect. 3.1 Acidify the sample with acetic acid (1 + 9) to pH 6.0 to 7.0. Caution: This operation must be performed in the hood and the sample left there until it can be made alkaline again after the extraction has been performed. 3.2 Extract with iso-octane, hexane, or chloroform (preference in order	**Chlorine:** If chlorine or other oxidizing agents are known to be present, pretreat the sample before the test with the procedure in this table for oxidizing agents. **Metals:** Nickel or cobalt in concentrations up to 1 mg/L do not interfere. Eliminate the interference from up to 20 mg/L copper and 5 mg/L iron: add the contents of one HexaVer Chelating Reagent Powder Pillow to a fresh portion of sample and mix. Use this treated sample in the test procedure. Prepare a reagent blank of deionized water and reagents to zero the instrument. **Oxidizing agents:** 1. Adjust a 25-mL portion of the alkaline sample to pH 7–9 with 2.5 N Hydrochloric Acid Standard Solution. Count the number of drops of acid added. 2. Add two drops of Potassium

	named) with a solvent volume equal to 20% of the sample volume. One extraction is usually adequate to reduce the fatty acids below the interference level. Avoid multiple extractions or a long contact time at low pH in order to keep the loss of HCN at a minimum. When the extraction is completed, immediately raise the pH of the sample to above 12 with NaOH solution. 4. High results may be obtained for samples that contain nitrate and/or nitrite. During the distillation nitrate and nitrite will form nitrous acid which will react with some organic compounds to form oximes. These compounds formed will decompose under test conditions to generate HCN. The interference of nitrate and nitrite is eliminated by pretreatment with sulfamic acid.	Iodide Solution and two drops of Starch Indicator Solution to the sample. Swirl to mix. The sample will turn blue if oxidizing agents are present. 3. Add Sodium Arsenite Solution drop-wise until the sample turns colorless. Swirl the sample thoroughly after each drop. Count the number of drops. 4. Take another 25-mL sample and add the total number of drops of Hydrochloric Acid Standard Solution counted in step 1. 5. Subtract one drop from the amount of Sodium Arsenite Solution added in step 3. Add this amount to the sample and mix thoroughly. Use this treated sample in the cyanide test procedure. **Reducing agents:** 1. Adjust a 25-mL portion of the alkaline sample to pH 7–9 with 2.5 N Hydrochloric Acid Standard Solution. Count the number of drops added. 2. Add four drops of Potassium Iodide Solution and four drops of Starch Indicator Solution to the sample. Swirl to mix. The sample should be colorless. 3. Add Bromine Water drop-wise until a blue color shows. Swirl the sample thoroughly after each addition. Count the number of drops. 4. Take another 25-mL sample and add the total number of drops of Hydrochloric Acid Standard Solution counted in step 1. 5. Add the total number of drops of Bromine Water counted in step 3 to the sample and mix thoroughly. 6. Use this treated sample in the cyanide test procedure. **Turbidity**: Large amounts of

		turbidity will cause high readings. Use filter paper and a funnel to filter highly turbid water samples. Use the filtered sample for the blank and sample preparation in the test procedure. The test results should then be recorded as soluble cyanide.
APPARATUS	1. Reflux distillation apparatus. The boiling flask should be of 1 liter size with inlet tube and provision for condenser. The gas absorber may be a Fisher-Milligan scrubber. 2. Spectrophotometer suitable for measurements at 578 nm or 620 nm with a 1.0 cm cell or larger. 3. Reflux distillation apparatus for sulfide removal. The boiling flask same as in step 1. The sulfide scrubber may be a Wheaton Bubber #709682 with 29/42 joints, size 100 mL. The air inlet tube should not be fritted. The cyanide absorption vessel should be the same as the sulfide scrubber. The air inlet tube should be fritted. 4. Flow meter, such as Lab Crest with stainless steel float (Fisher 11-164-50).	1. Micro Dist distillation block or DRB200 reactor block with Micro Dist distillation tubes. 2. Cyanide TNTplus reagent set TNT 862. 3. Pipets with tips. 4. Spectrophotometer suitable for measurements at 588 nm with a 13-mm round vial sample cell.
REAGENTS AND STANDARD	Sodium hydroxide Lead acetate Sulfuric acid Hydrochloric acid Sodium dihydrogenphosphate Potassium cyanide Chloramine-T Barbituric acid Pyridine 3-Methyl-1-phenyl-2-pyrazolin-5-one 3,3'Dimethyl-1, 1'-diphenyl-[4,4'-bi-2 pyrazoline] -5,5'dione (bispyrazolone) Magnesium chloride Sulfamic acid	Sodium hydroxide Lead acetate Sulfuric acid Hydrochloric acid Sodium dihydrogenphosphate Monopotassium phosphate Potassium cyanide Chloramine-T Pyridine 1,3-Dimethyl Barbituric acid Magnesium chloride Sulfamic acid
SAMPLE COLLECTION PRESERVATION	The sample should be collected in plastic or glass bottles of 1 liter or larger size. All bottles must be	1. Collect samples in clean glass or plastic bottles. 2. The presence of oxidizing agents,

	thoroughly cleansed and thoroughly rinsed to remove soluble material from containers. 2. Oxidizing agents such as chlorine decompose most of the cyanides. Test a drop of the sample with potassium iodide-starch test paper (KI-starch paper); a blue color indicates the need for treatment. Add ascorbic acid, a few crystals at a time, until a drop of sample produces no color on the indicator paper. Then add an additional 0.06 g of ascorbic acid for each liter of sample volume. 3. Samples must be preserved with 2 mL of 10 N sodium hydroxide per liter of sample (pH 2 > or = 12) at the time of collection. 4. Samples should be analyzed as rapidly as possible after collection. If storage is required, the samples should be stored in a refrigerator or in an ice chest filled with water and ice to maintain temperature at 4°C.	sulfides and fatty acids can cause the loss of cyanide during sample storage. Samples that contain these substances must be pretreated as described in the sections that follow before preservation with sodium hydroxide. If the sample contains sulfide and is not pretreated, it must be analyzed within 24 hours. 3. To preserve samples for later analysis, adjust the sample pH to a minimum pH 12 with 5.0 N sodium hydroxide standard solution (about 4 mL per liter). Use a glass serological pipet and pipet filler. Measure the pH and add more sodium hydroxide if necessary. 4. Keep the preserved samples at or below 6°C (43 °F) for up to 14 days. 5. Before analysis, adjust the pH to 7 with 2.5 N hydrochloric acid standard solution. 6. Let the sample temperature increase to room temperature before analysis. 7. Correct the test result for the dilution caused by the volume additions. **Oxidizing agents** Oxidizing agents such as chlorine decompose cyanides during storage. To test for and remove oxidizing agents, pretreat the sample as follows: 1. Measure 25 mL of the sample and add one drop of 10-g/L m-Nitrophenol Indicator Solution. Swirl to mix. 2. Add 2.5 N Hydrochloric Acid Standard Solution by drops until the color changes from yellow to colorless. Swirl the sample thoroughly after the addition of each drop. 3. Add two drops of Potassium	

		Iodide Solution, 30-g/L and two drops of Starch Indicator Solution to the sample. Swirl to mix. The solution will turn blue if oxidizing agents are present. 4. If the color is blue, add two level, 1-g measuring spoonfuls of ascorbic acid per liter of sample. 5. Remove a 25-mL portion of the treated sample and repeat steps 1 to 3. If the sample turns blue, repeat steps 4 and 5. 6. If the 25-mL sample remains colorless, preserve the remaining sample to pH 12 for storage with 5 N Sodium Hydroxide Standard Solution. 7. Complete the procedure given under Interfering Substances and Levels, Reducing Agents, to eliminate the effect of excess ascorbic acid, before the cyanide procedure is started. **Sulfides** Sulfides will quickly convert cyanide to thiocyanate (SCN^-). To test for and remove sulfide, pretreat the sample as follows: 1. Put a drop of sample on a disc of Hydrogen Sulfide Test Paper that has been wetted with pH 4 Buffer Solution. 2. If the test paper darkens, add a 1-g measuring spoon of Lead Acetate to the sample. Repeat step 1. 3. If the test paper continues to turn dark, keep adding Lead Acetate until the sample tests negative for sulfide. 4. Filter the lead sulfide precipitate through Filter Paper and a Funnel. Preserve the sample for storage with 5 N Sodium Hydroxide Standard Solution or neutralize to a pH of 7 for analysis. **Fatty acids**	

		CAUTION Perform this operation under a ventilation hood and complete as quickly as possible. When distilled, fatty acids will pass over with cyanide and under the alkaline conditions of the absorber, will form soaps. If the presence of fatty acid is suspected, use the following pretreatment before preserving samples with sodium hydroxide. 1. Acidify 500 mL of sample to pH 6 or 7 with a 4:1 dilution of glacial Acetic Acid. 2. Pour the sample into a 1000-mL separation funnel and add 50 mL of Hexane. 3. Stopper the funnel and shake for 1 minute. Allow the layers to separate. 4. Drain off the lower sample layer into a 600-mL beaker. If the sample is to be stored, add enough 5 N Sodium Hydroxide Standard Solution to raise the pH to a minimum pH 12.	
CALIBRATION AND STANDARDIZATION	If the colorimetric procedure is used, calculate the cyanide, in Fg/L, in the original sample as follows: $CN,\ \mu g/L = \frac{A \times 1{,}000}{B} \times \frac{50}{C}$ where: A = Fg CN read from standard curve B = mL of original sample for distillation C = mL taken for colorimetric analysis 0.04 – 0.80 mg cyanide/L	Concentration range for this reagent set is 0.01 – 0.60 mg cyanide/L.	
PROCEDURE	Distillation procedure: Distill the samples and standards following the procedure listed in USEPA 335.2.	Distillation procedure: Distill all samples and standards with a Micro Dist distillation block. Refer to the Micro Dist documentation for the distillation procedure. Always distill the standards with the samples.	

DATA ANALYSIS AND CALCULATIONS	1. Prepare a standard curve by plotting the absorbance value of standards versus the corresponding cyanide concentrations. 2. Obtain concentration value of sample directly from standard curve.	Same
PRECISION AND ACCURACY	1. In a single laboratory (EMSL), using mixed industrial and domestic waste samples at concentrations of 0.06, 0.13, 0.28 and 0.62 mg/L CN, the standard deviations were ±0.005, ±0.007, ±0.031 and ±0.094, respectively. 2. In a single laboratory (EMSL), using mixed industrial and domestic waste samples at concentrations of 0.28 and 0.62 mg/L CN, recoveries were 85% and 102%, respectively.	<u>Precision, Single Lab:</u> x = 0.094 mg cyanide/L SD = 0.001 mg cyanide/L RSD = 1.02% x = 0.383 mg cyanide/L SD = 0.002 mg cyanide/L RSD = 0.54% <u>Method Detection Limit:</u> 0.016 mg cyanide/L SD = 0.0005 mg cyanide/L x = 0.002 mg cyanide/L <u>Spikes Recoveries:</u> Industrial wastewater recovery spike duplicate = 93.7% and 92.8%. Domestic wastewater recovery spike duplicate = 92.7% and 95.2%

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