Phenols

DOC316.53.01108

USEPA[1] 4-Aminoantipyrine Method[2] Method 8047

0.002 to 0.200 mg/L

Scope and application: For water and wastewater.

[1] USEPA accepted (distillation required); procedure is equivalent to USEPA method 420.1 for wastewater.
[2] Adapted from *Standard Methods for the Examination of Water and Wastewater.*

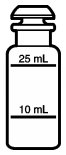
Test preparation

Instrument-specific information

Table 1 shows all of the instruments that have the program for this test. The table also shows sample cell and orientation requirements for reagent addition tests, such as powder pillow or bulk reagent tests.

To use the table, select an instrument, then read across to find the applicable information for this test.

Table 1 Instrument-specific information

Instrument	Sample cell orientation	Sample cell
DR6000 DR3800 DR2800 DR2700 DR1900	The fill line is to the right.	2612602 
DR5000 DR3900	The fill line is toward the user.	

Before starting

Analyze samples within 4 hours to prevent oxidation.
Spilled reagent can have an effect on the test results and is hazardous to skin and other materials.
Use chloroform only with proper ventilation.
The Phenol 2 Reagent Powder Pillows contain potassium nitroferricyanide. **Keep cyanide solutions at pH > 11 to prevent exposure to hydrogen cyanide gas.** Collect the reacted samples for proper disposal.
Do not pour chloroform solutions down the drain. Collect the water that is saturated with chloroform, chloroform solutions and the cotton plug used in the delivery tube of the separatory funnel for proper disposal. Refer to a current MSDS/SDS for safe handling and disposal instructions.
Make the cotton plug the size of a pea. A larger plug restricts the flow and a smaller plug can dislodge from the delivery tube of the funnel.
In bright light conditions (e.g., direct sunlight), close the cell compartment, if applicable, with the protective cover during measurements.
Review the Safety Data Sheets (MSDS/SDS) for the chemicals that are used. Use the recommended personal protective equipment.
Dispose of reacted solutions according to local, state and federal regulations. Refer to the Safety Data Sheets for disposal information for unused reagents. Refer to the environmental, health and safety staff for your facility and/or local regulatory agencies for further disposal information.

Items to collect

Description	Quantity
Chloroform, ACS	60 mL
Phenol Reagent Powder Pillow	2
Phenol 2 Reagent Powder Pillow	2
Clippers	1
Cotton balls	varies
Cylinder, graduated, 50-mL	1
Cylinder, graduated, 500-mL	1
Funnel, 500 mL separatory with stand and stopper	2
Hardness 1 Buffer Solution, pH 10.1	10 mL
Pipet, volumetric, Class A, 5.00-mL	1
Ring, support, 4-in.	2
Support for ring stand, 5 x 8 in. base	1
Sample cells (For information about sample cells, adapters or light shields, refer to Instrument-specific information on page 1.)	1
Water, deionized	varies

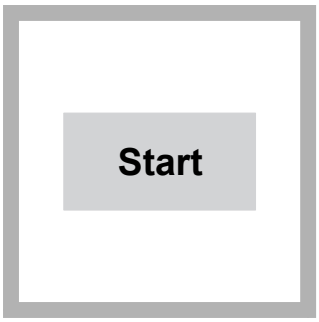
Refer to Consumables and replacement items on page 6 for order information.

Sample collection and storage

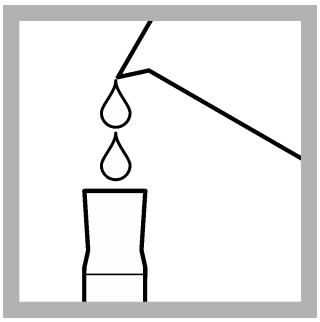
For the most reliable results, analyze the samples within 4 hours after collection. Use the storage instructions that follow if prompt analysis is not possible:

1. Collect 500 mL of the sample in a clean glass container.
2. Add the contents of two Copper Sulfate Powder Pillows.
3. Adjust the pH to 4 or less with 10% Phosphoric Acid Solution.
4. Keep the preserved samples at or below 6 °C (43 °F) for a maximum of 24 hours.
5. Analyze the preserved samples within 24 hours.

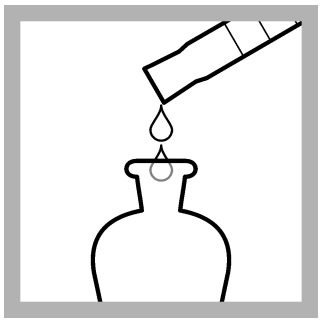
Test procedure



1. Start program **470 Phenol**. For information about sample cells, adapters or light shields, refer to Instrument-specific information on page 1.



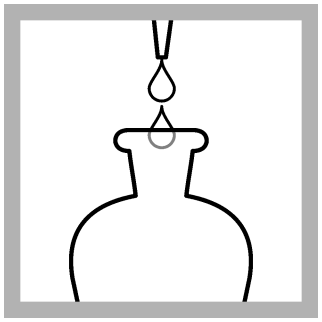
2. Measure 300 mL of deionized water in a 500-mL graduated cylinder.



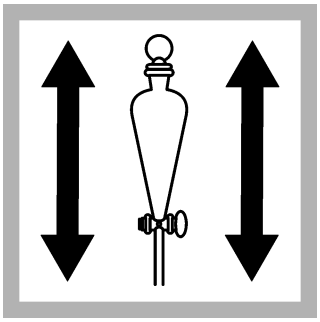
3. Prepare the blank: Pour the measured deionized water into a 500-mL separatory funnel.

4. Measure 300 mL of the sample in a 500-mL graduated cylinder.

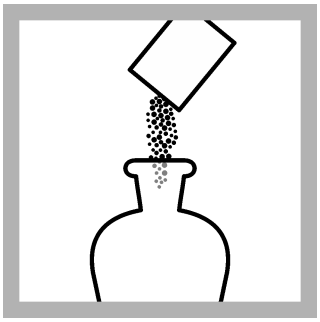
5. Prepare the sample: Pour the measured sample into another 500-mL separatory funnel.



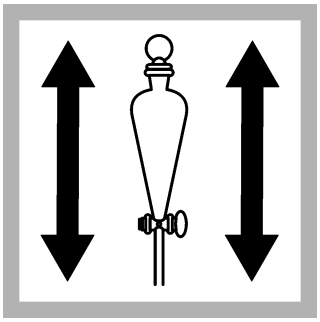
6. Add 5 mL of Hardness Buffer to each separatory funnel.



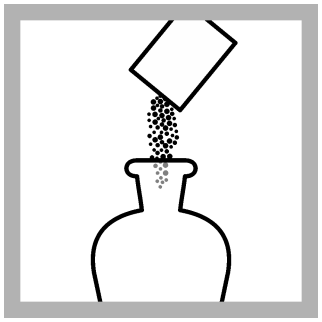
7. Put the stoppers on the funnels. Shake to mix.



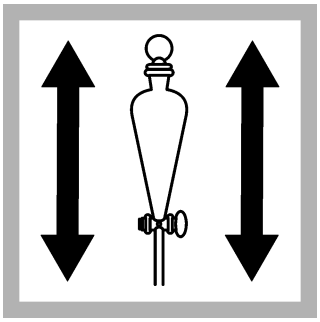
8. Add the contents of one Phenol Reagent Powder Pillow to each separatory funnel.



9. Put the stoppers on the funnels. Shake to dissolve the reagent.

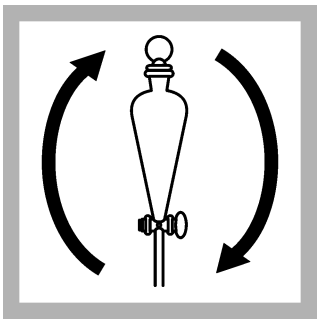


10. Add the contents of one Phenol 2 Reagent Powder Pillow to each separatory funnel.

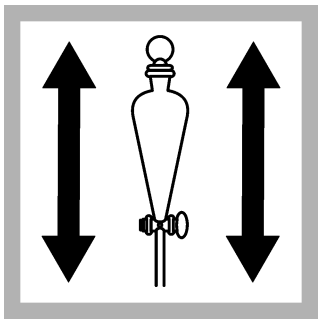


11. Put the stoppers on the funnels. Shake to dissolve the reagent.

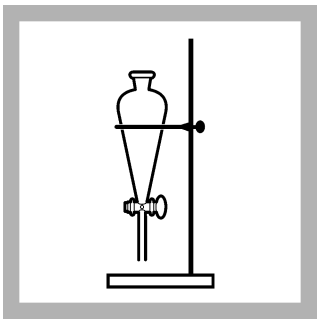
12. Add 30 mL of chloroform to each separatory funnel.



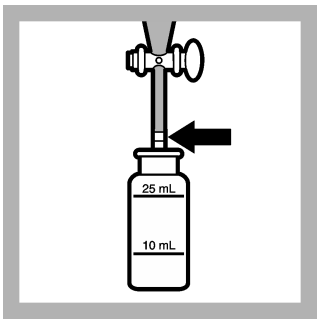
13. Put the stoppers on the funnels. Invert each funnel and open the stopcock to vent. Close the stopcocks, shake each funnel briefly, then invert and open the stopcock to vent.



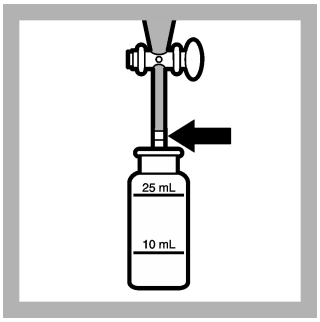
14. Vigorously shake each funnel for 30 seconds. Open the stopcock to vent if necessary.



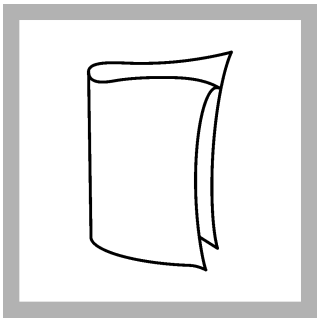
15. Remove the stoppers. Wait for the layers to separate. If there is phenol in the sample, the bottom chloroform layer will have a yellow to amber color.
Note: *Do the rest of the test procedure quickly because the chloroform will evaporate, which causes high readings.*



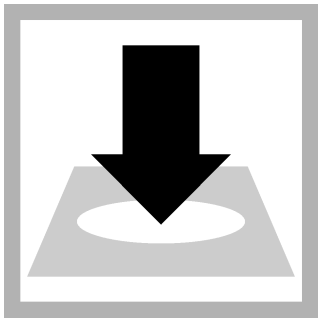
16. Put a cotton plug the size of a pea into the delivery tube of each funnel to remove suspended water or particles from the chloroform.



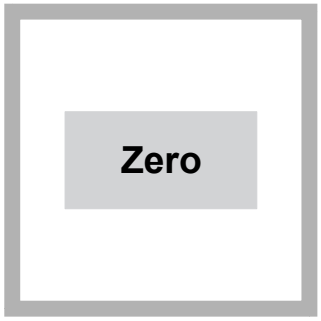
17. Drain the chloroform layers into separate sample cells (one for the blank and one for the sample). Put the stopper on the cells. The volume of chloroform extract is approximately 25 mL. The water phase in the funnel contains chloroform. Make sure to dispose of the solution properly.



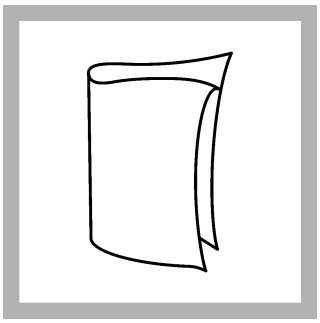
18. Clean the blank sample cell.



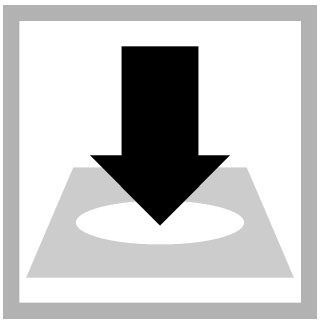
19. Insert the blank into the cell holder.



20. Push **ZERO**. The display shows 0.000 mg/L Phenol.



21. Clean the prepared sample cell.



22. Insert the prepared sample into the cell holder.

23. Push **READ**. Results show in mg/L Phenol.

Interferences

Interfering substance	Interference level
pH	The sample pH must be between 3 and 11.5 for best results.
Oxidizing or reducing agents	Can interfere. Distill the samples (Refer to Distill the sample on page 5).
Sulfides or suspended matter	Distillation or the following pretreatment is necessary: **1.** Measure 350 mL of sample with a clean 500-mL graduated cylinder. Pour the sample into a clean 500-mL Erlenmeyer flask. **2.** Add the contents of one Sulfide Inhibitor Reagent Powder Pillow. Swirl to mix. **3.** Filter 300 mL of the sample through a folded filter paper. Use this solution in step 4 of the procedure.

Distill the sample

To remove interferences, use the steps that follow to distill the sample. For the best results, make sure that the sample pH is between 3 and 11.5. Refer to Interferences on page 4 for pretreatment guidelines.

1. Set up the distillation apparatus for general purpose distillation. Refer to the Distillation Apparatus manual for proper assembly.
2. Set up a 500-mL Erlenmeyer flask to collect the distillate. It may be necessary to use a laboratory jack to elevate the flask.
3. Measure 300 mL of water sample in a clean 500-mL graduated cylinder. Pour the sample into the distillation flask.

   Note: *For proof of accuracy, use a 0.200-mg/L phenol standard in addition to the sample. Refer to* Accuracy check *on page 5.*
4. Add a magnetic stir bar and 5 glass beads.
5. Use a serological pipet to add 1 mL of Methyl Orange Indicator to the distillation flask.
6. Set the stirrer power to on. Set the stir control to 5.
7. Add 10% Phosphoric Acid Solution drop-wise until the indicator changes from yellow to orange.
8. Add one Copper Sulfate Powder Pillow to the distillation flask. Let the reagent dissolve. Do not do this step if copper sulfate is used to preserve the sample.
9. Put the cap on the distillation flask.
10. Turn on the water and adjust to maintain a steady flow through the condenser.
11. With the thermometer inserted, set the heat control to 10. The yellow pilot lamp is an indication that the heater is on.
12. Collect 275 mL of distillate in the Erlenmeyer flask, then set the heat to off.
13. Fill a 25-mL graduated cylinder to the 25-mL mark with deionized water.
14. Add the water to the distillation flask.
15. Set the heat control to 10. Heat until another 25-mL of distillate is collected.
16. Use a clean graduated cylinder to measure the distillate again to make sure that 300 mL was collected.
17. Use the distillate in the test procedure.

Accuracy check

Standard solution method

Items to collect:

- Phenol, ACS
- 1000-mL volumetric flasks (2), Class A
- 500-mL volumetric flask, Class A
- 10-mL volumetric pipet, Class A, with safety bulb
- Deionized water

1. Prepare a 1000-mg/L phenol stock solution as follows:
   a. Weigh 1.000 g of phenol.
   b. Add the phenol to a 1000-mL volumetric flask.
   c. Dilute to the mark with freshly boiled and cooled deionized water. Mix well to dissolve.
2. Prepare a 10-mg/L working phenol standard solution as follows:
   a. Use a pipet to add 10 mL of the stock phenol solution to a 1000-mL volumetric flask.
   b. Dilute to the mark with deionized water. Mix well.

3. Prepare a 0.200-mg/L standard solution as follows:
   a. Use a pipet to add 10 mL of the 10-mg/L working solution to a 500-mL volumetric flask.
   b. Dilute to the mark with deionized water. Mix well.
4. Use the test procedure to measure the concentration of the prepared standard solution.
5. Compare the expected result to the actual result.

   Note: *The factory calibration can be adjusted slightly with the standard calibration adjust option so that the instrument shows the expected value of the standard solution. The adjusted calibration is then used for all test results. This adjustment can increase the test accuracy when there are small variations in the reagents or instruments.*

Method performance

The method performance data that follows was derived from laboratory tests that were measured on a spectrophotometer during ideal test conditions. Users can get different results under different test conditions.

Program	Standard	Precision (95% confidence interval)	Sensitivity Concentration change per 0.010 Abs change
470	0.100 mg/L Phenol	0.093–0.107 mg/L Phenol	0.002 mg/L Phenol

Summary of Method

The 4-aminoantipyrine method measures all ortho- and meta-substituted phenols. These phenols react with 4-aminoantipyrine in the presence of potassium ferricyanide to form an antipyrine dye. The dye is removed from the aqueous phase by extraction with chloroform. The sensitivity of the method changes with the type of phenolic compound. Because water samples can contain various types of phenolic compounds, the procedure results are shown as the equivalent concentration of phenol. The measurement wavelength is 460 nm.

Consumables and replacement items

Required reagents

Description	Quantity/Test	Unit	Item no.
Phenols Reagent Set (100 Tests), includes:	—	—	2243900
Chloroform, ACS	60 mL	500 mL x 16	1445849
Hardness 1 Buffer Solution, pH 10.1	10 mL	500 mL	42449
Phenol Reagent Powder Pillows	2	100/pkg	87299
Phenol 2 Reagent Powder Pillows	2	100/pkg	183699
Water, deionized	varies	4 L	27256

Required apparatus

Description	Quantity/test	Unit	Item no.
Clippers for plastic powder pillows	1	each	93600
Cotton balls, absorbent	1	100/pkg	257201
Cylinder, graduated, 50 mL	1	each	50841
Funnel, separatory, 500 mL	1	each	52049
Pipet filler, safety bulb	1	each	1465100
Pipet, volumetric, Class A, 5.00 mL	1	each	1451537

Required apparatus (continued)

Description	Quantity/test	Unit	Item no.
Support Ring, 4-inch	1	each	58001
Support, Ring Stand, 5-inch x 8-inch base	1	each	56300

Distillation reagents and apparatus

Description	Unit	Item no.
Balance, analytical, 80 g x 0.1 mg 100–240 VAC	each	2936701
Copper Sulfate Powder Pillows	50/pkg	1481866
Distillation apparatus set, general purpose	each	2265300
Filter paper, folded, 12.5-cm	100/pkg	189457
Funnel, poly, 65 mm	each	108367
Methyl Orange Indicator Solution, 0.5-g/L	100 mL MDB	14832
Phosphoric Acid Solution, 10%	100 mL MDB	1476932
Sulfide Inhibitor Reagent Powder Pillows	100/pkg	241899
Paper, pH, 0–14 pH range	100/pkg	2601300
Thermometer, non-mercury, –10 to +225 °C	each	2635700
Pipet, TenSette®, 1.0–10.0 mL	each	1970010
Pipet tips for TenSette® Pipet, 1.0–10.0 mL	250/pkg	2199725
Pipet tips for TenSette® Pipet, 1.0–10.0 mL	50/pkg	2199796
Safety goggles, vented	each	2550700
Gloves, chemical resistant, size 9–9.5	pair	2410104[1]
Flask, volumetric, Class A, 500 mL, glass	each	1457449
Flask, volumetric, Class A, 1000 mL glass	each	1457453

[1] Other sizes available

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