**Scope and Application:** For screening THMs in drinking water.

**THM Plus™ Water Bath Method**

(0–200 ppb as Chloroform)

1. **Press the soft key under HACH PROGRAM.** Select the stored program number for Trihalomethane (THM) Plus by pressing 3560 on the numeric keys.

Press ENTER.

**Note:** For the most precise results, use matched cells. See page 6.

2. **The display will show:**

HACH PROGRAM: 3560
THM Plus

The wavelength (\(\lambda\)), 515 nm, is automatically selected.

3. **Prepare a hot water bath by adding 500 mL of water to an evaporating dish. Put the dish on a hot plate and turn heater on high.**

**Note:** If analyzing more than four samples, use 450 mL of water.

4. **Prepare a cooling bath by adding 500 mL of cold (18–25 °C) tap water to a second evaporating dish.**

**Note:** Maintain the water temperature between 18 and 25 °C.

**Note:** If analyzing more than four samples, use 450 mL of water.

5. **Fill two sample cells to the 10 mL mark with sample. Label one sample and the other blank.**

**Note:** Perform steps 5 through 9 rapidly so as not to lose volatile THMs from the sample. If you are testing more than one sample, complete steps 5 through 9 for one sample before going on to the next.

**Note:** If dispensing sample with a pipette, the pipette must dispense quickly without causing aeration or back pressure.

6. **Add three drops of THM Plus Reagent 1 to each cell.**

7. **Cap tightly and mix gently by swirling each cell three times.**

**Note:** Vigorous shaking can cause loss of THMs.

8. **Use a TenSette® pipette to add 3 mL of THM Plus Reagent 2 to each cell.**

**Note:** The liquid is viscous and a small amount may remain in the tip after dispensing. This will not affect the results.

**Note:** The THM Plus Reagent 2 must be at room temperature before use.
9. Cap tightly and mix by shaking ten times.
   **Note:** Thorough mixing ensures that all of the THM goes into the liquid and does not accumulate in the head space.

10. Place the sample cell in the cell holder assembly. Set the blank aside.

11. Place the basket in the hot-water bath when the water is boiling rapidly.
    **Note:** Do not allow water to rise above the white line near the top of the sample cells.

12. Press: **START TIMER** 1 to begin a five-minute reaction period.

13. At the end of the reaction period, remove the basket and sample cell from the hot-water bath and place in the cooling bath.

14. Press: **START TIMER 2**. Cool for three minutes. At the end of the cooling period, remove the cell from the cooling bath.

15. Use the Repipet Jr. to add 1 mL of THM Plus Reagent 3 to the sample cell and to the blank. Swirl to mix.
    **Note:** The sample and blank will become warm.
    **Note:** The liquid is viscous and may not be entirely dispensed if measured using any other pipetting method.

16. Replace the cooling water with fresh, cold tap water. Place the basket containing the sample and blank cells into the cooling bath.
17. Press **START TIMER** 3 to begin a three-minute cooling time. At the end of the cooling period, remove the cells from the cooling bath. 

*Note:* At the end of the cooling time, the temperature of the sample should be between 15 and 25 °C.

18. Add one THM Plus Reagent 4 Powder Pillow each to the sample cell and to the blank.

19. Cap each cell tightly and mix by shaking ten times. 

*Note:* All the powder should dissolve.

20. Press **START TIMER** 4 to begin a 15-minute color development time.

21. While the color is developing, insert the AccuVac® Ampul Adapter into instrument.

22. Wipe the reagent blank with a damp towel, followed by a dry one, to remove fingerprints or other marks.

23. At the end of the 15 minutes, place the blank into the cell holder and close the light shield.

24. Press the soft key under **ZERO**. The display will show:

    0 ppb CHCl₃

*Note:* For alternate concentration units, press the soft key under **OPTIONS**, and then the soft key under **UNITS**. Scroll through the available options. Press **ENTER** to return to the Read screen.
Sampling and Storage

Collect samples in 40-mL glass bottles sealed with Teflon®-lined septa caps. Use Cat. No. 27940-05 or equivalent for best results. Fill the bottles slowly to overflowing so that no air is included with the sample. Seal the bottles tightly and invert to check that no air has been trapped.

Because trihalomethane compounds (THMs) are extremely volatile, immediate analysis yields the greatest accuracy. If the samples cannot be analyzed immediately, cool samples to 4 °C. This will slow the formation of any additional THM compounds in chlorinated samples. Store the preserved samples at 4 °C in an atmosphere free of organic vapors. Samples should not be held more than 14 days. Allow the samples to equilibrate to 15–20 °C before analyzing.

Accuracy Check

Standard Additions Method

Prepare the standard additions sample at the same time as the unspiked water sample. Snap the neck off a THM Standard Ampule, 10 ppm as chloroform. Using a Wiretrol™ Pipet (Cat. No. 25689-05), add 0.050 mL of the standard to 10 mL of water sample. Immerse the tip of the pipet below the surface of the water sample and dispense the aliquot of chloroform standard. Cap the sample cell immediately and swirl three times to mix. Prepare the sample and the spiked sample according to the procedure steps 6–26.

a. Leave the unspiked sample in the sample compartment after completing step 26. Verify that the units displayed are in ppb. Select standard additions mode by pressing the soft keys under OPTIONS, (MORE) and the STD ADD.

b. Press ENTER to accept the default sample volume (mL), 10.0.

c. Use the keypad to enter 10000, then press ENTER to accept the standard concentration (ppb) 10,000.

d. Press the soft key under ENTRY DONE.
e. Read the standard additions sample prepared above. Accept the standard additions reading by pressing the soft key under READ. The addition should reflect 80–120% recovery.

f. See Section 1.4.1 Standard Additions for more information.

⚠️ Chloroform is extremely volatile! Do not shake it when mixing.

**Standard Solutions Method**

Prepare a 99 ppb chloroform standard by pipetting 10.0 mL of organic-free water into a sample cell. Snap the neck off a THM Standard Ampule, 10 ppm as chloroform. Using a Wiretrol Pipette (Cat. No. 25689-05), transfer 0.100 mL of the chloroform standard into the organic-free water. Immerse the end of the pipet tip under the water to dispense the chloroform. Cap the sample cell immediately and swirl three times to mix. Immediately perform steps 6–25 of the procedure. Do not make up the standard in advance. Use the standard immediately upon preparation.

**Method Performance**

**Precision**

Standard: 60 ppb CHCl₃

<table>
<thead>
<tr>
<th>Program</th>
<th>95% Confidence Limits</th>
</tr>
</thead>
<tbody>
<tr>
<td>3560</td>
<td>56–64 ppm CHCl₃</td>
</tr>
</tbody>
</table>

For more information on determining precision data and method detection limits, refer to Section 1.5.

**Estimated Detection Limit**

<table>
<thead>
<tr>
<th>Program</th>
<th>EDL</th>
</tr>
</thead>
<tbody>
<tr>
<td>3560</td>
<td>6 ppb CHCl₃</td>
</tr>
</tbody>
</table>

For more information on derivation and use of Hach’s estimated detection limit, see Section 1.5.2. To determine a method detection limit (MDL) as defined by the 40 CFR part 136, appendix B, see Section 1.5.1.

**Sensitivity**

Program Number: 3560

<table>
<thead>
<tr>
<th>Portion of Curve</th>
<th>∆Abs</th>
<th>∆Concentration</th>
</tr>
</thead>
<tbody>
<tr>
<td>Entire Range</td>
<td>0.010</td>
<td>21 ppb as CHCl₃</td>
</tr>
</tbody>
</table>

See Section 1.5.3, Sensitivity Explained for more information.
Sample Cell Matching

The THM Plus method requires that the 1" sample cells be optically matched for best performance. Although sample cells supplied by Hach Company are distortion-free, nicks and scratches from handling, fingerprints, and other foreign material on the glass surfaces may cause an optical mismatch between two sample cells and introduce error into the test results. This type of error may be avoided by optically matching the sample cells and following the cell precaution statements listed in the procedure.

Procedure:

1. Turn on your instrument and select the THM Plus method. Select the wavelength indicated in the procedure if your instrument has not automatically done so.

2. Change the instrument to the absorbance mode.

3. Pour at least 10 mL of deionized water into each of the samples cells to be matched.

4. Place one of the sample cells into the cell holder. Note and mark the orientation of the cell in the cell holder. Close the light shield. (Sample cells should be carefully wiped with a lint free cloth to remove any fingerprints or other foreign matter on the outside of the cell.)

5. Press: ZERO. The display will show: 0.000 Abs

6. Place the next sample cell into the cell holder. Close the light shield.

7. Wait for the absorbance value to stabilize and record the value.

8. Turn the cell 180 degrees and repeat steps 6–7. Try to achieve an absorbance value within ±0.001 Abs of the first cell. Note the orientation of the sample cell in the cell holder. This will allow the cells to be oriented consistently in the cell holder.

Reagent Storage

Refrigerate THM Plus Reagent 2 for maximum stability. Long-term exposure to temperatures above 35 °C may cause reagent degradation.

Interferences

The substances in the following table have been tested and found not to interfere up to the indicated levels (in ppm):
Interferences which have no effect up to the maximum level tested

<table>
<thead>
<tr>
<th>Interference</th>
<th>Maximum Level tested</th>
</tr>
</thead>
<tbody>
<tr>
<td>Chlorine</td>
<td>&lt;10 ppm</td>
</tr>
<tr>
<td>Copper</td>
<td>&lt;1000 ppm</td>
</tr>
<tr>
<td>Hardness, Ca</td>
<td>&lt;1000 ppm as CaCO₃</td>
</tr>
<tr>
<td></td>
<td>May have some turbidity until Reagent 3 is added</td>
</tr>
<tr>
<td>Hardness, Mg</td>
<td>&lt;4000 ppm as CaCO₃</td>
</tr>
<tr>
<td></td>
<td>May have some turbidity until Reagent 3 is added</td>
</tr>
<tr>
<td>Iron</td>
<td>&lt;10 ppm</td>
</tr>
<tr>
<td>Lead</td>
<td>&lt;2 ppm</td>
</tr>
<tr>
<td>Mercury</td>
<td>&lt;10 ppm</td>
</tr>
<tr>
<td>Monochloramine</td>
<td>&lt;20 ppm</td>
</tr>
<tr>
<td>Nickel</td>
<td>&lt;10 ppm</td>
</tr>
<tr>
<td>Sodium Bisulfite</td>
<td>&lt;100 ppm</td>
</tr>
<tr>
<td>EDTA</td>
<td>Interferes negatively at all levels</td>
</tr>
</tbody>
</table>

Additional disinfection by-products which react

<table>
<thead>
<tr>
<th>Compound</th>
<th>Effect</th>
</tr>
</thead>
<tbody>
<tr>
<td>1,1,1-trichloro-2-propanone</td>
<td>Interferes positively</td>
</tr>
<tr>
<td>1,1,1-trichloroacetonitrile</td>
<td>Interferes positively</td>
</tr>
<tr>
<td>Chloral hydrate</td>
<td>Interferes positively</td>
</tr>
<tr>
<td>Dibromochloroacetic acid</td>
<td>Interferes positively</td>
</tr>
<tr>
<td>Dichlorobromoacetic acid</td>
<td>Interferes positively</td>
</tr>
<tr>
<td>Tribromoacetic acid</td>
<td>Interferes positively</td>
</tr>
<tr>
<td>Trichloroacetic acid</td>
<td>Interferes positively</td>
</tr>
</tbody>
</table>

Summary of Method

The THM Plus method reacts with the trihalogenated disinfection by-products formed as the result of the disinfection of drinking water with chlorine in the presence of naturally occurring organic materials. These disinfection by-products (DBPs) may be produced in the treatment plant or the distribution system as long as the water is in contact with free chlorine residual. The formation of the DBPs is influenced by chlorine contact time, chlorine dose and residual, temperature, pH, precursor concentration, and bromide concentration.

The predominant DBPs formed by the chlorination of drinking water are the trihalomethanes or THMs. The four trihalogenated compounds that form are chloroform, bromoform, dichlorobromomethane, and dibromochloromethane. These four compounds comprise the Total Trihalomethanes (TTHMs) group which is regulated under the Safe Drinking Water Act. The combined concentration of the TTHMs, reported as chloroform, is regulated to be 100 ppb or less in drinking water samples. Other DBPs that may be present and react under the conditions of the THM Plus method are listed in Interferences.

In the THM Plus method, THM compounds present in the sample react with N, N-diethylnicotinamide under heated alkaline conditions to form a dialdehyde intermediate. The sample is then cooled and acidified to pH 2.5. The dialdehyde intermediate formed is then reacted with 7-napthylamine-1,3 disulfonic acid to form a colored Schiff base which absorbs at 515 nm. The color formed is directly
TRIHALOMETHANES, continued

proportional to the total amount of THM compounds present in the sample. The results are reported as ppb chloroform.

Safety

Good safety habits and laboratory techniques should be used throughout the procedure. Consult the Material Safety Data Sheet for information specific to the reagents used.

REQUIRED REAGENTS

Reagent Set (50 tests) .............................................................................................................................. 27908-00
Includes: (1) 27539-29, (1) 27540-48, (1) 27541-42, (1) 27566-99

<table>
<thead>
<tr>
<th>Description</th>
<th>Quantity Required</th>
<th>Unit</th>
<th>Cat. No.</th>
</tr>
</thead>
<tbody>
<tr>
<td>THM Plus Reagent 1</td>
<td>6 drops</td>
<td>15 mL/bottle</td>
<td>27539-29</td>
</tr>
<tr>
<td>THM Plus Reagent 2</td>
<td>6 mL</td>
<td>330 mL/bottle</td>
<td>27540-48</td>
</tr>
<tr>
<td>THM Plus Reagent 3</td>
<td>2 mL</td>
<td>110 mL/bottle</td>
<td>27541-42</td>
</tr>
<tr>
<td>THM Plus Reagent 4</td>
<td>2 pillows</td>
<td>100 pillows</td>
<td>27566-99</td>
</tr>
</tbody>
</table>

REQUIRED APPARATUS

Beaker, 600-mL .................................................................................................................. each 500-52
Cell Holder Assembly, TTHM ............................................................................................. each 47880-00
Evaporating Dish, 125 mm x 65 mm .................................................................................. each 27647-00
Hot Plate, 7 x 7 in., 120 V AC ................................................................................... each 23441-00
Hot Plate, 7 x 7 in., 240 V AC ................................................................................... each 23441-02
Repipetter, 1 mL .............................................................................................................. each 21113-02
Pipet, Tensette, 1–10 mL ................................................................................................. each 19700-10
Pipet tips, 1–10 mL (for 19700-10) ........................................................................... 50/pkg 25589-96
Sample cells, 10-mL, w/caps .................................................................................... 2/box 24276-06
Wipers, Disposable, KimWipes .................................................................................... 280/pkg 20970-00

OPTIONAL REAGENTS

Chloroform, 10-ppm ampoule ................................................................................................. each 27567-07
Water, Reagent, Organic-free ............................................................................................... 500 mL 26415-49

OPTIONAL APPARATUS

Flask, volumetric, 100 mL, class A .................................................................................. each 14574-42
Pipet, filler, safety bulb ................................................................................................. each 14651-00
Pipet, volumetric, class A, 10 mL .................................................................................. each 14515-38
Pipettes, Wiretrol™, 50–100 µL ..................................................................................... 250/pkg 25689-05
Timer, 5-channel, 1 sec. to 100 hrs. ................................................................................. each 26304-00
Vials, glass, 40-mL, with Septa cap ............................................................................... 5/pkg 27940-05