LCK555 $BOD_5/BOD_{[n]}$

DOC312.53.94393

Method

4–1650 mg/L $BOD_5/BOD_{[n]}$(5/[n]-day biochemical oxygen demand) LCK555

Scope and application: For municipal and industrial waste water.

Test preparation

Test storage

Storage temperature: 2–8 °C (35–46 °F)

pH/Temperature

The pH of the water sample must be between pH 4–10.

The temperature of the water sample and the dilution water must be between 18–24 °C (64–75 °F).

Before starting

Dilution water for LCK555
The dilution water set (LZC901) contains technical data on the preparation and handling of dilution water. Please take note of these data.
Analytical Quality Assurance
addista is an analytical quality assurance system with which you can check the accuracy and precision of your analysis results at any time. Regular checks ensure that your measurement system is functioning properly and is being correctly operated, and reveal sample-specific interferences.
On grounds of quality and reliability, the analysis should be carried out only with original accessories from the manufacturer.
Review safety information and expiration date on the package.
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.

1. Choice of dilution steps

Waste water matrix	Recommended measurement range (in mg/L)	Probable upper limits of measuring range
Sewage treatment plant outflow Biologically purified waste water Lightly contaminated industrial waste water	**A** 4–58	=> **A1** => 4–19 mg/L * => **A2** => 7–38 mg/L * => **A3** => 11–58 mg/L *
Municipal waste water after preliminary settling stage Contaminated industrial waste water Municipal raw sewage	**B** 25–413	=> **B1** => 25–138 mg/L * => **B2** => 50–275 mg/L * => **B3** => 75–413 mg/L *
Municipal raw sewage Strongly contaminated industrial waste water Landfill leachate	**C** 100–1650	=> **C1** => 100–550 mg/L * => **C2** => 200–1100 mg/L * => **C3** => 300–1650 mg/L *

* for probable upper limits of measuring range refer to For special attention on page 4 point 2.

The dilution level should be chosen so that the estimated BOD_5 is in the middle of the measurement range.

For example: estimated BOD_5 ≈ 200 mg/L chosen measurement range **B3** = 75–413 mg/L

For samples of unknown BOD_5, the most favorable dilution level is determined in advance with the help of an **estimated value R**.

Type of waste water	Estimated value R
Untreated industrial and municipal waste water	**35–65 %** of the COD of the sample = mg/L BOD_5 estimated
Biologically treated waste water	**25 %** of the COD of the sample = mg/L BOD_5 estimated

2. Sample preparation

a) Homogenization of the sample

Homogenize the sample (20 °C (68° F)) **5 minutes** at 700 to 900 rotations per minute (magnetic stirrer) or depending on the floc size, first of all use a homogenizer (**30 seconds** at 20000 rotations per minute).

Note: If large particles or flocs are still present after homogenization, carry out multiple determinations to determine the BOD_5.

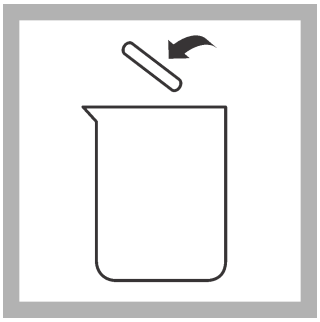
b) Preliminary dilution in reaction tube for the dilution steps

Pipet sample and dilution water into the reaction tube (in accordance with the table), close the tube, and shake vigorously for **1 minute** in order to enrich the sample with oxygen.

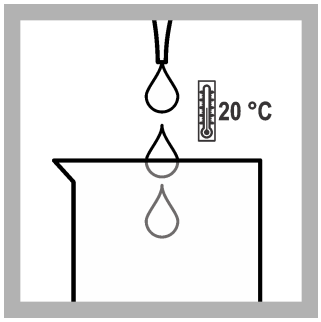
Measuring range (in mg/L)	Preliminary dilution in reaction tube Sample	Preliminary dilution in reaction tube Dilution water	Prepared sample pipet into the cuvette test:	Dilution factor
A => **4–58** *				
=> **A1** => 4–19 mg/L *	4 mL	—	1.8 mL	**3.5**
=> **A2** => 7–38 mg/L *	4 mL	—	0.9 mL	**7.0**
=> **A3** => 11–58 mg/L *	4 mL	—	0.6 mL	**10.5**
B => **25–413** *				
=> **B1** => 25–138 mg/L *	1 mL	1 mL	0.5 mL	**25**
=> **B2** => 50–275 mg/L *	1 mL	3 mL	0.5 mL	**50**
=> **B3** => 75–413 mg/L *	1 mL	5 mL	0.5 mL	**75**
C => **100–1650** *				
=> **C1** => 100–550 mg/L *	0.4 mL	2.8 mL	0.5 mL	**100**
=> **C2** => 200–1100 mg/L *	0.4 mL	6.0 mL	0.5 mL	**200**
=> **C3** => 300–1650 mg/L *	0.4 mL	9.2 mL	0.5 mL	**300**

* probable upper limits of measuring range refer to For special attention on page 4 point 2.

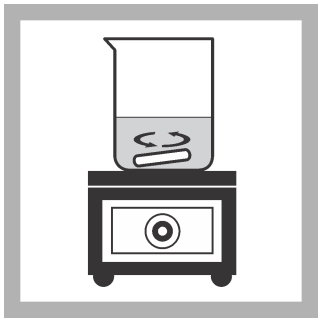
Procedure—Sample preparation



1. Homogenization of the sample: Introduce the magnetic stirring bar into a **beaker**.



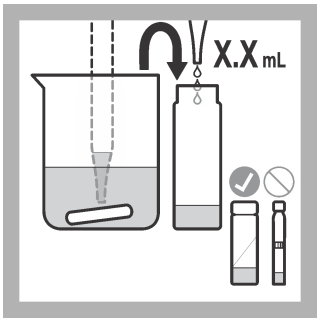
2. Add **sample** (20 °C) to the beaker.



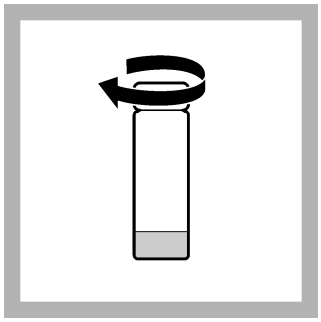
3. Place the beaker on the magnetic stirrer.



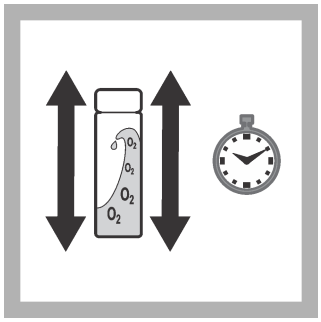
4. Stir for **5 minutes** at 700–900 rpm.



5. Pipet **sample** and **dilution water** prepared as described in 2. Sample preparation on page 2 into the reaction tube.

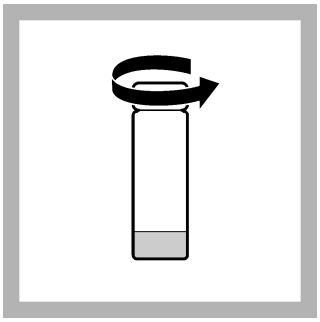


6. Close the reaction tube.

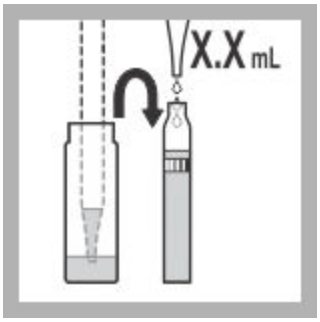


7. Shake **vigorously** for **1 minute** to enrich the sample with oxygen.

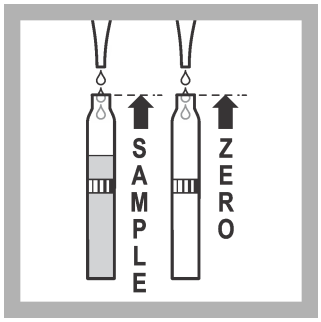
3. Procedure—Filling the cuvettes with sample and dilution water



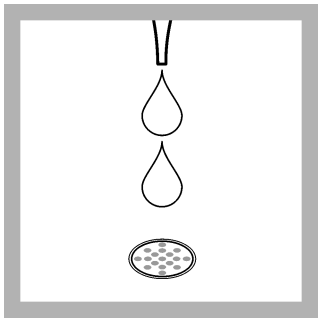
1. Open the reaction tube



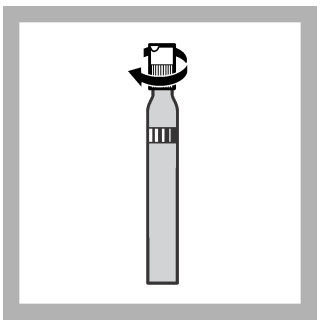
2. Pipet the **required sample volume** (X.X mL) prepared as described in 2. Sample preparation on page 2 into the **sample** cuvette.



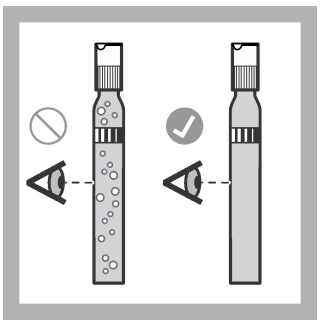
3. Use a transfer pipette to fill the **sample** cuvette and the **zero** cuvette to **the brim** with dilution water (ensure that **no air bubbles** remain in the cuvette). **Pipet** the dilution water **uniformly** into the cuvettes.
Each day that analyses are carried out, **one** dilution water cuvette (cuvette containing only dilution water) must be prepared as a blank value for all measured values. **Only one** dilution water cuvette is necessary for analysis series.



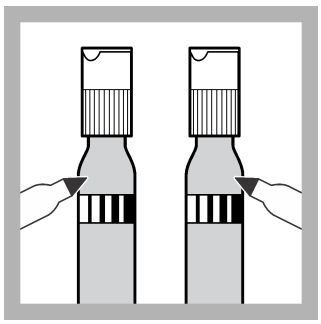
4. Discard excess dilution water remaining in the transfer pipette after a cuvette has been filled.



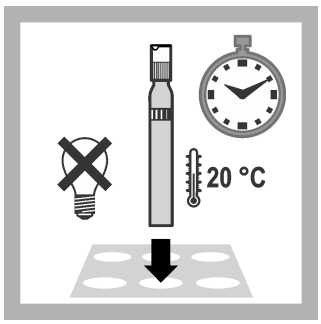
5. Close the cuvettes again with DosiCapZip.



6. Ensure that **no air bubbles** remain in the cuvettes.



7. Note the dilution step and sample type on the cuvette label to safeguard against any mixups after 5/[n] days.



8. Leave to stand for **5/[n] days in darkness** in the block thermostat at **20 °C** (68° F).

For special attention

1. Check the dilution water value after 5 /[n] days

Display: ***I : o***

Dilution water is oversaturated with oxygen.

Response: Do not use the dilution water until **1 hour** has elapsed after aeration.

Display: ***I : u***

Dilution water is low in oxygen.

Response: Check inoculation. Check aeration. Check the temperature of the dilution water and the location where it is kept.

2. Variable measuring range limits

Only the lower measuring limit is defined when the measuring range is selected. The upper limit serves as a guide value, because it is calculated from the previously measured dilution water value.

If the result lies outside the measuring range, this is shown on the display and the printer.

Conversion table

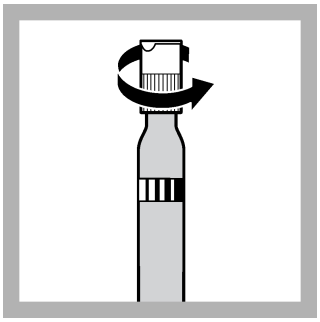
If it not possible to determine the BOD_5 at the correct time (e.g. during the weekend), the approximate BOD_5 can be calculated with the help of a factor.

Premature BOD measurement after 4 days
Result BOD_4 x 1.14 ≈ approximate BOD_5
Late measurement of BOD after 6 days
Result BOD_6 x 0.91 ≈ approximate BOD_5

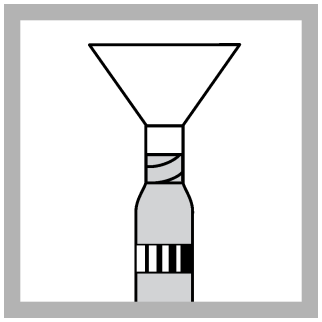
The results are approximate values for municipal waste water.

The factor for industrial waste water can vary. We advise determining this factor from a number of samples on a case by case basis.

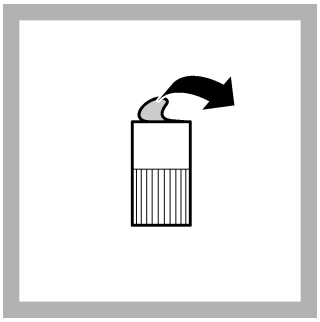
4. Procedure BOD Evaluation after 5/[n] days



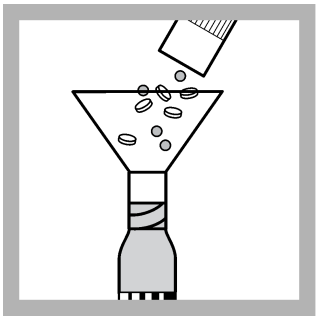
1. Unscrew the **DosiCapZip** from the **dilution water cuvette**.



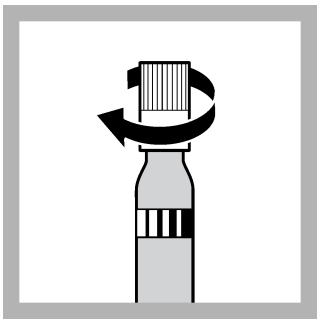
2. Place the funnel on the **dilution water cuvette**.



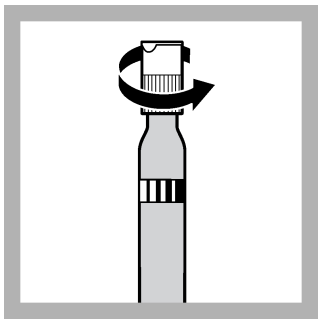
3. Carefully pull the foil off the **DosiCapZip**.



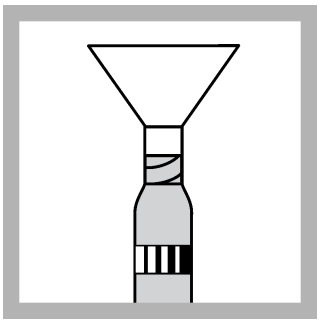
4. Pour the contents (tablets and glass beads) through the funnel into the **dilution water cuvette**.



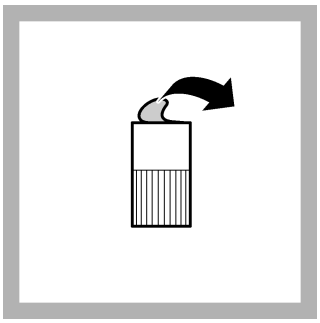
5. Remove the funnel and immediately seal the **dilution water cuvette** with **DosiCapZip**, taking care that the cuvette contains no air bubbles.
Note: *If the liquid meniscus falls below the cuvette opening when the funnel is removed, make up the volume by adding 2 to 4 glass beads.*



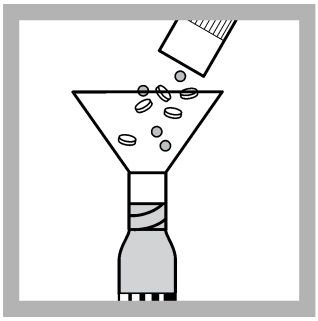
6. Unscrew the **DosiCapZip** from the **sample cuvette**.



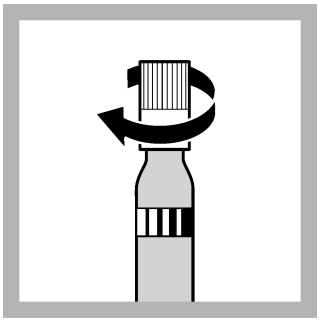
7. Place the funnel on the **sample cuvette**.



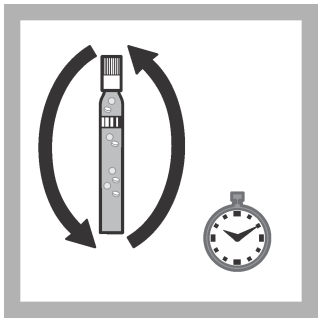
8. Carefully pull the foil off the **DosiCapZip**.



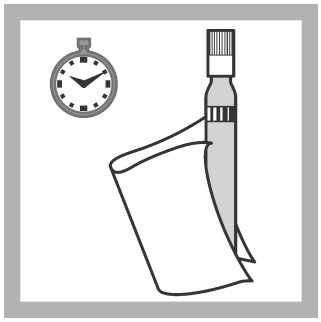
9. Pour the contents (tablets and glass beads) through the funnel into the **sample cuvette**.



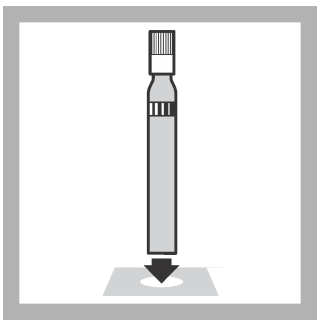
10. Remove the funnel and immediately seal the **sample cuvette** with **DosiCapZip**, taking care that the cuvette contains no air bubbles.
Note: *If the liquid meniscus falls below the cuvette opening when the funnel is removed, make up the volume by adding 2 to 4 glass beads.*



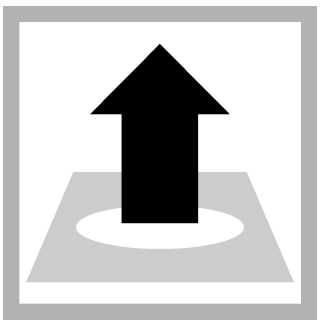
11. Repeatedly invert the **cuvettes** for **3 minutes** until the reagent tablets are completely dissolved.



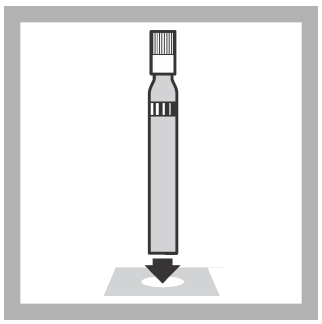
12. After another **3 minutes**, thoroughly clean the outside of the cuvettes and evaluate.



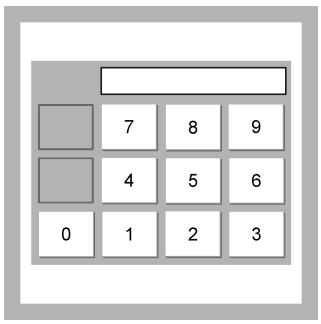
13. Insert the **dilution water cuvette** into the cell holder.
DR1900: Go to LCK/TNTplus methods. Select the test, push **ZERO**.



14. Remove the dilution water cuvette.



15. Insert the **sample cuvette** into the cell holder.
DR1900: push **READ**.



16. Insert dilution factor (refer to: 2. Sample preparation on page 2).
It is absolutely essential to input the exact dilution factor (refer to: Conversion table on page 5.

Interferences

The ions listed in the table have been individually checked against the given concentrations and do not cause interference. The cumulative effects and the influence of other ions have not been determined.

There is no interference from:

Interference level	Interfering substance
10000 mg/L	Cl^-
2000 mg/L	SO_4^{2-}
500 mg/L	PO_4^{3-}, CO_3^{2-}, NO_3^-
200 mg/L	NH_4^+, Ca^{2+}
100 mg/L	Mg^{2+}
50 mg/L	SCN^-
20 mg/L	S^{2-}, HCHO, Fe^{3+}

Interference level	Interfering substance
10 mg/L	SiO_2, EDTA, Pb^{2+}, CN^-, Zn^{2+}, Cu^{2+}, Cr^{6+}, Mn^{2+}, Ni^{2+}, Sn^{2+}, Ag^+, F^-
5 mg/L	Al^{3+}
2 mg/L	Cr^{3+}, NO_2^-
1 mg/L	Fe^{2+}

Peroxide compounds and other strong oxidants are also detected and cause low-bias results to be obtained. High chlorine concentrations can cause high or low-bias results to be obtained. If reducing agents are present the results will have a high-bias.

The measurement results must be subjected to plausibility checks (dilute and/or spike the sample).

This can be achieved with LCK555 BOD_5/$BOD_{[n]}$ through multiple determinations and further dilutions of the water sample.

Summary of method

Determination of **5/[n]-day biochemical oxygen demand** with inhibition of nitrification by **5 mg/L allylthiourea**. The dissolved oxygen is analyzed in an alkaline solution with a pyrocatechol derivative in the presence of Fe^{2+}, under which conditions a red dye is formed.

HACH LANGE GMBH
Willstätterstraße 11
D-40549 Düsseldorf
Tel. +49 (0) 2 11 52 88-0
Fax +49 (0) 2 11 52 88-143
info-de@hach.com
www.hach.com

© Hach Company, 2020, 2025. All rights reserved. Original instructions.

08/2025, Edition 3