$BOD_5/BOD_{[n]}$

Principle
Determination of ***5/[n]-day*** biochemical oxygen demand ($BOD_5/BOD_{[n]}$) with inhibition of nitrification by ***5 mg/L*** allylthiourea. The dissolved oxygen is analysed in an alkaline solution with a pyrocatechol derivative in the presence of Fe^{2+}, under which conditions a red dye is formed.

Range of Application
Municipal and industrial waste water

Dilution water for LCK 555
The dilution water set (LZC 901) contains technical data on the preparation and handling of dilution water. Please take note of these data. The ***HACH LANGE booklet A 122*** describes all the ways in which the dilution water can be prepared in combination with the LZC 555 BioKIT, LZC 901 dilution water set and LCK 555 BOD_5 cuvette test. You can order the booklet free of charge from from your HACH LANGE Agency.

Storage Information
The test reagents are stable at +2 to +8°C up to the expiry date given on the package.

Interferences
The ions listed in the table have been individually checked up to the given concentrations. Cumulative effects and the influence of other ions have not been determined by us. There is no interference from:

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+}
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 the water sample).
This can be achieved with LCK 555 $BOD_5/BOD_{[n]}$ through multiple determinations and further dilutions of the water sample.

pH/Temperature
The pH of the water sample must be between pH 4 and pH 10.
The temperature of the water sample and the dilution water must be between 18 and 24°C.

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.

Safety Advice
On grounds of quality and reliability, the analysis should be carried out only with original HACH LANGE accessories.

CADAS 100 (LPG 185 / ≥ LPG 210)
If this test is not already stored in your instrument please ask your HACH LANGE Agency for programming instructions.

LCK 555 GB

Data table LCK 555

LP2W	*04/1998*
$BOD_5/BOD_{[n]}$ (A1) • F_1 = 0 • F_2 = -36.44 • K = 0.65	
$BOD_5/BOD_{[n]}$ (B1) • F_1 = 0 • F_2 = -260.3 • K = 0.65	
$BOD_5/BOD_{[n]}$ (C1) • F_1 = 0 • F_2 = -1041 • K = 0.65	
CADAS 30/50 (Version 1.8/1.9)	***04/1998***
$BOD_5/BOD_{[n]}$ (A1) • λ: 620 nm • Pro.: 13 • F_1 = 34.48 • F_2 = -34.48 • K = 0.64	
$BOD_5/BOD_{[n]}$ (B1) • λ: 620 nm • Pro.: 13 • F_1 = 246.5 • F_2 = -246.4 • K = 0.652	
$BOD_5/BOD_{[n]}$ (C1) • λ: 620 nm • Pro.: 13 • F_1 = 986 • F_2 = -986 • K = 0.648	
CADAS 30/30S/50/50S	***04/1998***
$BOD_5/BOD_{[n]}$ (A) • λ: 620 nm • Pro.: 16 • F_1 = 9.855 • F_2 = 3.5 • K = 0.646	
$BOD_5/BOD_{[n]}$ (B) • λ: 620 nm • Pro.: 16 • F_1 = 9.855 • F_2 = 25 • K = 0.646	
$BOD_5/BOD_{[n]}$ (C) • λ: 620 nm • Pro.: 16 • F_1 = 9.855 • F_2 = 100 • K = 0.651	
ISIS 6000/9000	***04/1998***
$BOD_5/BOD_{[n]}$ (A) • λ: 610 nm • Pro.: 16 • F_1 = 8.016 • F_2 = 3.5 • K = 0.650	
$BOD_5/BOD_{[n]}$ (B) • λ: 610 nm • Pro.: 16 • F_1 = 8.016 • F_2 = 25 • K = 0.650	
$BOD_5/BOD_{[n]}$ (C) • λ: 610 nm • Pro.: 16 • F_1 = 8.012 • F_2 = 100 • K = 0.650	
CADAS 100 / LPG 185	***04/1998***
$BOD_5/BOD_{[n]}$ (A1) • λ: 620 nm • F_1 = -33.71 • F_2 = 0.65	
$BOD_5/BOD_{[n]}$ (B1) • λ: 620 nm • F_1 = -240.8 • F_2 = 0.65	
$BOD_5/BOD_{[n]}$ (C1) • λ: 620 nm • F_1 = -963.0 • F_2 = 0.65	
CADAS 100 / ≥ LPG 210	***04/1998***
$BOD_5/BOD_{[n]}$ (A1) • λ: 620 nm • F_1 = -33.71 • K = 0.65	
$BOD_5/BOD_{[n]}$ (B1) • λ: 620 nm • F_1 = -240.8 • K = 0.65	
$BOD_5/BOD_{[n]}$ (C1) • λ: 620 nm • F_1 = -963.0 • K = 0.65	

Applies to all types of photometer

Edition 04/1998

$BOD_5/BOD_{[n]}$

1. Choice of dilution steps

Wastewater matrix	Recommended measurement range *(in mg/L)*
sewage treatment plant outflow biologically purified waste water lightly contaminated industrial waste water	**A** ***4 – 58***
municipal waste water after preliminary settling stage contaminated industrial waste water municipal raw sewage	**B** ***25 – 413***
municipal raw sewage strongly contaminated industrial waste water landfill leachate	**C** ***100 – 1650***

A ⇒ **A1** ⇒ ***4 – 19 mg/L ****
⇒ **A2** ⇒ ***7 – 38 mg/L ****
⇒ **A3** ⇒ ***11 – 58 mg/L ****

B ⇒ **B1** ⇒ ***25 – 138 mg/L ****
⇒ **B2** ⇒ ***50 – 275 mg/L ****
⇒ **B3** ⇒ ***75 – 413 mg/L ****

C ⇒ **C1** ⇒ ***100 – 550 mg/L ****
⇒ **C2** ⇒ ***200 – 1100 mg/L ****
⇒ **C3** ⇒ ***300 – 1650 mg/L ****

**** probable upper limits of measuring range (see on page 1e "For special attention" point 2)***

For samples of unknown BOD_5, the most favourable 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

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

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

Applies to all types of photometer

Edition 04/1998

$BOD_5/BOD_{[n]}$

2. Sample preparation

a) Homogenization of the sample
Homogenize the sample (20°C) ***5 min*** at 700 to 900 rpm (magnetic stirrer) or depending on the floc size, first of all use a homogenizer (***30 sec*** at 20000 rpm).
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
Pipette sample and dilution water into the reaction tube (in accordance with the table), close the tube, and shake ***vigorously for 1 min*** in order to enrich the sample with oxygen.

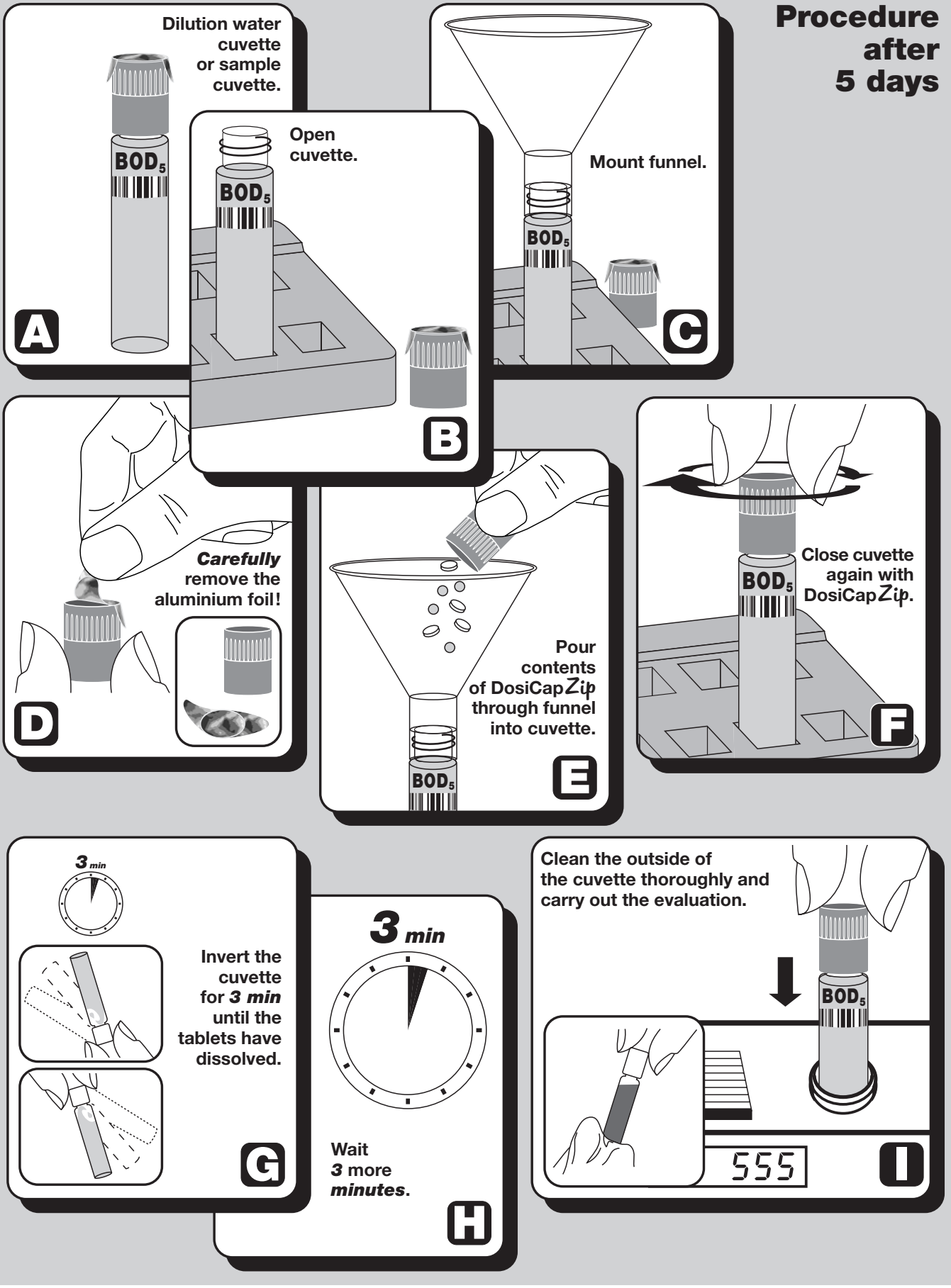
Measuring range *(in mg/L)*	Preliminary dilution in reaction tube: *sample*	*dilution water*	Prepared sample: *pipette into the cuvette test*	Dilution factor *for CADAS 200 / LASA 30/50/100 / XION 500*
A ⇒ 4 – 58 *				
A1 ⇒ 4 – 19 *	4 mL	—	1.8 mL	**3.5**
A2 ⇒ 7 – 38 *	4 mL	—	0.9 mL	**7.0**
A3 ⇒ 11 – 58 *	4 mL	—	0.6 mL	**10.5**
B ⇒ 25 – 413 *				
B1 ⇒ 25 – 138 *	1 mL	1 mL	0.5 mL	**25**
B2 ⇒ 50 – 275 *	1 mL	3 mL	0.5 mL	**50**
B3 ⇒ 75 – 413 *	1 mL	5 mL	0.5 mL	**75**
C ⇒ 100 – 1650 *				
C1 ⇒ 100 – 550 *	0.4 mL	2.8 mL	0.5 mL	**100**
C2 ⇒ 200 – 1100 *	0.4 mL	6.0 mL	0.5 mL	**200**
C3 ⇒ 300 – 1650 *	0.4 mL	9.2 mL	0.5 mL	**300**

**** probable upper limits of measuring range (see on page 1e "For special attention" point 2)***

Procedure
Applies to all types of photometer

Cuvette Test LCK 555

Procedure after 5 days



$BOD_5/BOD_{[n]}$

Edition 04/1998

Evaluation

1. Select »Barcode Programs«.
2. Select test number (see below).
3. Control number must be **4**.
4. Insert dilution water cuvette and press »Read 1«.
5. Insert sample cuvette and press »Read 2«.
6. Select dilution factor (see table on *page 1b* for sample preparation point 2).

If more than one sample is measured, start the next evaluation with point 5.

Parameter	Test-No.	Meas. range
$BOD_5/BOD_{[n]}$	555	4 – 1650 mg/L

$BOD_5/BOD_{[n]}$

Edition 04/1998

Evaluation

1. Press ”Mode“ key and check program control number: **__ : 40**
2. Insert program filter **623 nm**.
3. Select test with ”Mode“ key.
4. Insert dilution water cuvette. Display: »NULL« (zero). Remove dilution water cuvette.
5. Select measuring range with ✻ key.
6. Insert sample cuvette.

If more than one sample is measured, start the next evaluation with point 5 and select the measuring range again.

Parameter	Display	Meas. range
$BOD_5/BOD_{[n]}$ (A)	BOD 5 LCK 555	4 – 58 mg/L
$BOD_5/BOD_{[n]}$ (B)	BOD 5 LCK 555	25 – 413 mg/L
$BOD_5/BOD_{[n]}$ (C)	BOD 5 LCK 555	100 – 1650 mg/L

$BOD_5/BOD_{[n]}$

Edition 04/1998

Evaluation

1. Press any key.
2. Check program control number: **__ : 40**
3. Select test with ↑ or ↓ key.
4. Insert dilution water cuvette. Display: » I : «. Remove dilution water cuvette.
5. Select measuring range with ↓ key.
6. Insert sample cuvette.

If more than one sample is measured, start the next evaluation with point 5 and select the measuring range again.

Parameter	Display	Meas. range
$BOD_5/BOD_{[n]}$ (A)	BOD 5 LCK 555	4 – 58 mg/L
$BOD_5/BOD_{[n]}$ (B)	BOD 5 LCK 555	25 – 413 mg/L
$BOD_5/BOD_{[n]}$ (C)	BOD 5 LCK 555	100 – 1650 mg/L

$BOD_5/BOD_{[n]}$

Edition 04/1998

Evaluation

1. Insert filter **605 nm**.
2. Select »Dr. Lange« mode.
3. Select test number (see below).
4. Control number must be **4**.
5. Insert dilution water cuvette and press green key.
6. Insert sample cuvette and press green key.
7. Select dilution factor (see table on *page 1b* for sample preparation point 2).

If more than one sample is measured, start the next evaluation with point 6.

Parameter	Test-No.	Meas. range
$BOD_5/BOD_{[n]}$	555	4 – 1650 mg/L

$BOD_5/BOD_{[n]}$

Edition 04/1998

Evaluation

1. Insert program filter **620 nm**.
2. Press ”Tests“ key until display (see below) appears.
3. Control number must be:
   1 ($BOD_5/BOD_{[n]}$ A) or **4** ($BOD_5/BOD_{[n]}$ B) or
   8 ($BOD_5/BOD_{[n]}$ C).
4. Insert dilution water cuvette and press ”Null“ (zero) key.
5. Insert sample cuvette and press ”Ergebnis“ (result) key.

If more than one sample is measured, start the next evaluation with point 5.

Parameter	Display	Meas. range
$BOD_5/BOD_{[n]}$ (A1)	BOD 5 A LCK 555	A1 = 4 – 19 mg/L
$BOD_5/BOD_{[n]}$ (B1)	BOD 5 B LCK 555	B1 = 25 – 138 mg/L
$BOD_5/BOD_{[n]}$ (C1)	BOD 5 C LCK 555	C1 = 100 – 550 mg/L

Please note that the displayed measurement result for the dilution steps A2 / B2 / C2 must be multiplied by 2.

The displayed measurement result for the dilution steps A3 / B3 / C3 must be multiplied by 3.

$BOD_5/BOD_{[n]}$

Edition 04/1998

Evaluation

1. Insert dilution water cuvette.
2. Display: »BOD A BOD B BOD C«.
3. Press key under the required measuring range.
4. Insert sample cuvette.

If more than one sample is measured, start the next evaluation with point 2.

This evaluation is possible from the Eprom-version:
Version 1.8 (CADAS 30)
Version 1.9 (CADAS 50)

Parameter	Meas. range
$BOD_5/BOD_{[n]}$ (A1)	A1 = 4 – 19 mg/L
$BOD_5/BOD_{[n]}$ (B1)	B1 = 25 – 138 mg/L
$BOD_5/BOD_{[n]}$ (C1)	C1 = 100 – 550 mg/L

Please note that the displayed measurement result for the dilution steps A2 / B2 / C2 must be multiplied by 2.

The displayed measurement result for the dilution steps A3 / B3 / C3 must be multiplied by 3.

$BOD_5/BOD_{[n]}$

Edition 04/1998

Evaluation

1. Insert dilution water cuvette.
2. Select measuring range.
3. Select dilution steps.
4. Display: »BOD 5 [A1 – C3] NO. 4«.
5. Insert sample cuvette.

CADAS 30/30S/50/50S:
If more than one sample is measured, start the next evaluation with point 5 and press the key under the required measuring range again.

ISIS 9000:
If more than one sample is measured, insert the next sample cuvette and start the evaluation with point 2.

This evaluation is possible from the Eprom-version:
Version 2.0 (CADAS 30/50)
Program control no.: __ : 40 (CADAS 30S/50S/ISIS 9000)

Parameter	Meas. range
$BOD_5/BOD_{[n]}$ (A)	4 – 58 mg/L
$BOD_5/BOD_{[n]}$ (B)	25 – 413 mg/L
$BOD_5/BOD_{[n]}$ (C)	100 – 1650 mg/L

$BOD_5/BOD_{[n]}$

Edition 04/1998

Evaluation

1. Check program control number: **__ : 40**
2. Select »CUVETTE TEST« mode.
3. Select test number (see below).
4. Display: : »BOD A BOD B BOD C«.
   Press key under the required measuring range.
5. Display: »1 2 3«.
   Press key under the required dilution step.
6. Control number must be **4**.
7. Insert dilution water cuvette and press blue key.
8. Insert sample cuvette and press green key.

If more than one sample is measured, insert the next sample cuvette and start the evaluation with point 4, without inserting the dilution water cuvette again.

Parameter	Meas. range
$BOD_5/BOD_{[n]}$ (A)	4 – 58 mg/L
$BOD_5/BOD_{[n]}$ (B)	25 – 413 mg/L
$BOD_5/BOD_{[n]}$ (C)	100 – 1650 mg/L

$BOD_5/BOD_{[n]}$

Edition 04/1998

Evaluation

1. Insert dilution water cuvette.
2. Insert sample cuvette.
3. Insert dilution factor (see table on *page 1b* for sample preparation point 2).

It is absolutely essential to input the exact dilution factor.

Parameter	Meas. range
$BOD_5/BOD_{[n]}$	4 – 1650 mg/L

$BOD_5/BOD_{[n]}$

Edition 04/1998

Evaluation

1. Check program control number: **__ : 40**
2. Select test number (see below).
3. Control number must be **4**.
4. Insert dilution water cuvette and press green key.
5. Insert sample cuvette and press green key.
6. Insert dilution factor (see table on *page 1b* for sample preparation point 2).

It is absolutely essential to input the exact dilution factor.

Parameter	Test-No.	Meas. range
$BOD_5/BOD_{[n]}$	555	4 – 1650 mg/L

$BOD_5/BOD_{[n]}$

Edition 04/1998

Evaluation

1. Insert dilution water cuvette.
2. Insert sample cuvette.
3. Select dilution factor (see also table on *page 1b* for sample preparation point 2).

If more than one sample is measured, start the next evaluation with point 2.

Parameter	Meas. range
$BOD_5/BOD_{[n]}$	4 – 1650 mg/L

$BOD_5/BOD_{[n]}$

Edition 04/1998

Evaluation

1. Select »TEST« mode.
2. Select symbol (see below).
3. Check factors and measuring wavelength in memory »Mem«.
4. Insert dilution water cuvette and press ”NULL“ (zero) key.
5. Insert sample cuvette and press ”MESS“ (measure) key.

If more than one sample is measured, start the next evaluation with point 5.

Parameter	Symbol	Meas. range
$BOD_5/BOD_{[n]}$ (A1)	$ 555 A	A1 = 4 – 19 mg/L
$BOD_5/BOD_{[n]}$ (B1)	$ 555 B	B1 = 25 – 138 mg/L
$BOD_5/BOD_{[n]}$ (C1)	$ 555 C	C1 = 100 – 550 mg/L

Please note that the displayed measurement result for the dilution steps A2 / B2 / C2 must be multiplied by 2.

The displayed measurement result for the dilution steps A3 / B3 / C3 must be multiplied by 3.

$BOD_5/BOD_{[n]}$

Edition 04/1998

Evaluation

1. Select »TEST« mode.
2. Select symbol (see below).
3. Control number must be:
   7 [($BOD_5/BOD_{[n]}$ (A1)] or **8** [($BOD_5/BOD_{[n]}$ (B1)] or
   3 [($BOD_5/BOD_{[n]}$ (C1)].
4. Insert dilution water cuvette and press ”NULL“ (zero) key.
5. Insert sample cuvette and press ”MESS“ (measure) key.

If more than one sample is measured, start the next evaluation with point 5.

Parameter	Symbol	Meas. range
$BOD_5/BOD_{[n]}$ (A1)	555 A	A1 = 4 – 19 mg/L
$BOD_5/BOD_{[n]}$ (B1)	555 B	B1 = 25 – 138 mg/L
$BOD_5/BOD_{[n]}$ (C1)	555 C	C1 = 100 – 550 mg/L

Please note that the displayed measurement result for the dilution steps A2 / B2 / C2 must be multiplied by 2.

The displayed measurement result for the dilution steps A3 / B3 / C3 must be multiplied by 3.

Applies to all types of photometer

$BOD_5/BOD_{[n]}$

Edition 04/1998

3. Dilution water cuvette preparation

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. Filling the cuvette with sample and dilution water

For special attention:
Pipette the sample prepared as described in point 2 (see table for volume) into the cuvette test, then use a transfer pipette to ***fill the cuvette to the brim*** with dilution water (ensure that ***no air bubbles*** remain in the cuvette).
Pipette the dilution water ***uniformly*** into the cuvette.
Excess dilution water remaining in the transfer pipette after a cuvette has been filled should be discarded.

5. Closing the cuvette

Close the cuvette again with **DosiCap** *Zip*. ***Check*** each cuvette for ***air bubbles*** by inverting it. Leave to stand for ***5/[n] days in darkness*** in the block thermostat at ***20°C***.

6. Labelling the cuvette

The dilution step and sample type should be noted on the cuvette label to safeguard against any mix-ups after 5/[n] days.

Applies to all types of photometer

$BOD_5/BOD_{[n]}$

Edition 02/1999

Procedure II after 5/[n] days

1. Dilution water cuvette

Unscrew the **DosiCap** *Zip* from the ***dilution water cuvette***, then place the funnel on it. ***Carefully*** pull the aluminium foil off the **DosiCap** *Zip* and pour the contents (tablets and glass beads) through the funnel into the ***dilution water cuvette***. Remove the funnel and ***immediately seal the dilution water cuvette*** with **DosiCap** *Zip*, taking care that the cuvette contains ***no air bubbles***.
Attention! If the liquid meniscus falls below the cuvette opening when the funnel is removed, the missing volume can be made up by adding 2 to 4 glass beads.

2. Sample cuvette

Unscrew the **DosiCap** *Zip* from the ***sample cuvette***, then place the funnel on it. ***Carefully*** pull the aluminium foil off the **DosiCap** *Zip* and pour the contents (tablets and glass beads) through the funnel into the ***sample cuvette***. Remove the funnel and ***immediately seal the sample*** cuvette with **DosiCap** *Zip*, taking care that the cuvette contains ***no air bubbles***.
Attention! If the liquid meniscus falls below the cuvette opening when the funnel is removed, the missing volume can be made up by adding 2 to 4 glass beads.

Repeatedly invert the cuvettes prepared as described in points 1 and 2 for ***3 min*** until the reagent tablets have dissolved completely. ***Wait 3 min***, then thoroughly clean the outside of the cuvette again and evaluate (see evaluation).

$BOD_5/BOD_{[n]}$

Edition 04/1998

All photometers (except LP2W and CADAS 100) have a new type of variable BOD_5 analysis evaluation.

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

a) Display » I : o «
Dilution water is oversaturated with oxygen.
Response: Do not use the dilution water until ***1 hour*** has elapsed after aeration.

b) 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.

$BOD_5/BOD_{[n]}$

Edition 04/1998

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.