

# Bilirubin Auto Total FS\*

Diagnostic reagent for quantitative in vitro determination of total bilirubin in serum or plasma on Sysmex BX-Series

## Order information

Cat. No.	Kit size	Number of tests
1 0811 99 10 972	R1 3 x 15.8 mL	BX-3010 3 x 125 tests BX-4000 3 x 87 tests
	R2 3 x 6.5 mL	BX-3010 3 x 125 tests BX-4000 3 x 87 tests

## Method

Photometric test using 2,4-dichloroaniline (DCA)

## Principle

Direct bilirubin in presence of diazotized 2,4-dichloroaniline forms a red colored azocompound in acidic solution. A specific mixture of detergents enables a safe determination of the total bilirubin.

## Reagents

### Components and Concentrations

<b>R1:</b>	Phosphate buffer	50 mmol/L
	NaCl	150 mmol/L
<b>R2:</b>	2,4-Dichlorophenyl-diazonium salt	5 mmol/L
	HCl	130 mmol/L

### Storage Instructions and Reagent Stability

The reagents are stable up to the end of the indicated month of expiry, if stored at 2 – 8°C, protected from light and contamination is avoided. Do not freeze the reagents!

### Warnings and Precautions

- Reagent 1 and 2: Warning. H290 May be corrosive to metals. H319 Causes serious eye irritation. P234 Keep only in original packaging. P264 Wash hands and face thoroughly after handling. P280 Wear protective gloves/protective clothing/eye protection. P305+P351+P338 If in eyes: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing. P337+P313 If eye irritation persists: Get medical advice/attention. P390 Absorb spillage to prevent material damage.
- In very rare cases, samples of patients with gammopathy might give falsified results [6].
- Eltrombopag medication leads to falsely low or high results in patient samples.
- Please refer to the safety data sheets and take the necessary precautions for the use of laboratory reagents. For diagnostic purposes, the results should always be assessed with the patient's medical history, clinical examinations and other findings.
- For professional use only!

### Waste Management

Please refer to local legal requirements.

### Reagent Preparation

The reagents are ready to use. The bottles are placed directly into the reagent trays.

### Specimen

Serum or heparin plasma

It is very important to store the samples protected from light!

Stability [1]:

1 day	at	20 – 25°C
7 days	at	4 – 8°C
6 months	at	–20°C

in case of immediate freezing.

Freeze only once!

Discard contaminated specimens.

## Calibrators and Controls

For calibration, DiaSys TruCal U calibrator is recommended. The assigned values for total bilirubin have been made traceable to the NIST SRM 916 reference material. For internal quality control DiaSys TruLab N and P controls should be assayed. Each laboratory should establish corrective action in case of deviations in control recovery.

	Cat. No.	Kit size
TruCal U	5 9100 99 10 063	20 x 3 mL
	5 9100 99 10 064	6 x 3 mL
TruLab N	5 9000 99 10 062	20 x 5 mL
	5 9000 99 10 061	6 x 5 mL
TruLab P	5 9050 99 10 062	20 x 5 mL
	5 9050 99 10 061	6 x 5 mL

## Performance Characteristics

Measuring range up to 25 mg/dL (428 µmol/L) bilirubin (in case of higher concentrations re-measure samples after manual dilution with NaCl (9 g/L) or use rerun function)	
Limit of detection**	0.1 mg/dL (1.71 µmol/L) bilirubin
On-board stability	6 weeks
Calibration stability	7 days

Interfering substance	Interferences < 10%	Analyte concentration
Ascorbate	up to 30 mg/dL	0.771 mg/dL (13.2 µmol/L)
Hemoglobin	up to 125 mg/dL	0.254 mg/dL (4.35 µmol/L)
Hemoglobin	up to 500 mg/dL	0.776 mg/dL (13.3 µmol/L)
Lipemia (triglycerides)	up to 1000 mg/dL	0.355 mg/dL (6.07 µmol/L)

For further information on interfering substances refer to Young DS [5].

Precision (BX-3010)			
Within run (n=20)	Sample 1	Sample 2	Sample 3
Mean [mg/dL]	0.483	1.33	5.91
Mean [µmol/L]	8.26	22.8	101
Coefficient of variation [%]	1.50	1.33	0.628
Between run (n=20)	Sample 1	Sample 2	Sample 3
Mean [mg/dL]	0.509	1.36	6.50
Mean [µmol/L]	8.70	23.3	111
Coefficient of variation [%]	2.60	2.78	1.30

Method comparison (n=124)	
Test x	Bilirubin Auto Total FS (BioMajesty 6010C)
Test y	Bilirubin Auto Total FS (BX-3010)
Slope	1.04
Intercept	–0.010 mg/dL (0.176 µmol/L)
Coefficient of correlation	0.9998

\*\* lowest measurable concentration which can be distinguished from zero mean + 3 SD (n=20) of an analyte free specimen

### Conversion factor

Bilirubin [mg/dL] x 17.1 = Bilirubin [µmol/L]

## Reference Range [2]

		[mg/dL]	[µmol/L]
Neonates	24 h	< 8.8	< 150
	2 <sup>nd</sup> day	1.3 – 11.3	22 – 193
	3 <sup>rd</sup> day	0.7 – 12.7	12 – 217
	4 <sup>th</sup> – 6 <sup>th</sup> day	0.1 – 12.6	1.7 – 216
Children	> 1 month	0.2 – 1.0	3.4 – 17
Adults		0.1 – 1.2	1.7 – 21

Each laboratory should check if the reference ranges are transferable to its own patient population and determine own reference ranges if necessary.

## Literature

1. Guder WG, Zawta B et al. The Quality of Diagnostic Samples. 1<sup>st</sup> ed. Darmstadt: GIT Verlag; 2001; p. 18-9.
2. Thomas L ed. Clinical Laboratory Diagnostics. 1<sup>st</sup> ed. Frankfurt: TH-Books Verlagsgesellschaft, 1998: p. 192-202.
3. Tolman KG, Rej R. Liver function. In: Burtis CA, Ashwood ER, editors. Tietz Textbook of Clinical Chemistry. 3<sup>rd</sup> ed. Philadelphia: W.B Saunders Company; 1999. p. 1125-77.
4. Rand RN, di Pasqua A. A new diazo method for the determination of bilirubin. Clin Chem 1962; 6: 570-8.
5. Young DS. Effects of Drugs on Clinical Laboratory Tests. 5th ed. Volume 1 and 2. Washington, DC: The American Association for Clinical Chemistry Press 2000.
6. Bakker AJ, Mücke M. Gammopathy interference in clinical chemistry assays: mechanisms, detection and prevention. ClinChemLabMed 2007;45(9):1240-1243.

### Manufacturer



DiaSys Diagnostic Systems GmbH  
Alte Strasse 9 65558 Holzheim Germany

Chemistry Parameters 1				Sysmex BX-3010 Chemistry Analyzer Analytical Parameters			
Method No.	*	Method Name	TBIL	Reagent Name	Reagent (µL)	Water (µL)	
Print Name	Bilirubin total	MethodColor		R1	TBIL	100	
Sample Type	Serum			R2	TBIL	25	
Unit	mg/dL			Diluent	Disable		
Assay Type	End			Sample Ppt. Wash	Disable		
Measuring points		Start	End	Stirring Speed R1	Low	R2 Low	
		1	22 - 23				
		2	45 - 46				
Wave Length	Prim. 546	Sec. 660		Normal Range			
				No.	Normal Range Name	Min	Max
				1	Male-G1	*	*
				2	Male-G2	*	*
				3	Male-G3	*	*
				4	Female-G1	*	*
Normal	Sample Volume (µL)	Diluted Sample (µL)	Diluent (µL)	Technical Range	(Conc)	0.1	30.0
<input type="checkbox"/>	Low 0.0 < 2.5	Normal 2.5 < 0.0	High 0.0		(mAbs/10)	*	*
<input type="checkbox"/>	Diluent 0.0 < 2.5	< 0.0		Previous Result Comparison (%)	*	*	%
<input type="checkbox"/>	Diluent 0.0 < 2.5	< 0.0		Abnormal Range	(Conc)	*	*
<input type="checkbox"/>	Diluent 0.0 < 2.5	< 0.0		Panic Range	(Conc)	*	*
				Decimal Point	2	Profile SI	Disable

\*Entered by user

Chemistry Parameters 2				Sysmex BX-3010 Chemistry Analyzer Analytical Parameters		
Method No.	*	Method Name	TBIL	Sample	Serum	
Limit Checks				Blank measurement	Blank measurement:	Disable reagent blank and C1 blank
<input checked="" type="checkbox"/>	Duplicate Limit	50	mAbs/10	Measurement of Reagent Blank during Run:	None	
<input checked="" type="checkbox"/>	Sensitivity Limit	200	mAbs/10	Reagent blank measurement at calibration:	Reagent blank (No sample)	
<input checked="" type="checkbox"/>	Linearity Limit		%	The number of measurement:	Duplicate	
			(mAbs/10)/min	Reagent blank limit checks:	<input checked="" type="checkbox"/>	Duplicate Limit 20 mAbs/10
<input type="checkbox"/>	Prozone Limit	Higher	%	Instrument Factor	a	1.00
					b	0.00
	SL1-S		SL1-F			
	SL2-S		SL2-F			
	Sensitivity		mAbs/10			
<input checked="" type="checkbox"/>	Absorbance Limit					
	Abs. in reaction	Increase				
	Limit	25000	mAbs/10			

**Calibration Registration**

**Sysmex BX-3010 Chemistry Analyzer  
Analytical Parameters**

Method No.

Method Name

Sample Type

Replication

Check Interval

Test without calibration

Calibration Type

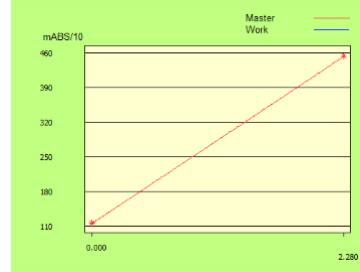
Reagent Lot

Calibrator Name

	Conc.	WORK	MASTER	Calibr. Lot No.	<input type="checkbox"/> All
C1	0	Automatic entry	Automatic entry	*	
C2	*	Automatic entry	Automatic entry	*	
C3	*				
C4	*				
C5	*				
C6	*				
C7	*				

K   C1 Blank  
 Reagent Blank for C1

Reagent Lot No.  
(R1)  Last   
(R2)



The calibration curve is lot dependent

Reagent blank  mAbs/10 Last

Blank  mAbs/10 Last

Calibration Curve  Conc.

Absorbance  mAbs/10

\*Entered by user

Chemistry Parameters		Sysmex BX-4000 Chemistry Analyzer Analytical Parameters																									
Method	* <input type="text"/>	Name	<input type="text" value="TBIL"/>		Reagent Name	Reagent (µL)	Water (µL)																				
Print Name	<input type="text" value="Bilirubin total"/>		R1	<input type="text" value="TBIL"/>	<input type="text" value="150"/>																						
Sample	<input type="text" value="Serum"/>		R2	<input checked="" type="checkbox"/> Enable	<input type="text" value="TBIL"/>	<input type="text" value="38"/>																					
Unit	<input type="text" value="mg/dL"/>																										
Assay Type	<input type="text" value="End"/>		Diluent	<input type="checkbox"/> Enable	<input type="text"/>	<input type="text"/>																					
Measuring points	Start	End	Decimal Points	<input type="text" value="2"/>																							
	1	<input type="text" value="33"/>	-	<input type="text" value="34"/>																							
<input type="checkbox"/> Enable	2	<input type="text" value="67"/>	-	<input type="text" value="68"/>																							
Wave Length	Prim.	<input type="text" value="546"/>	Sec	<input type="checkbox"/> Disable	<input type="text" value="660"/>																						
<table border="1" style="width:100%; border-collapse: collapse;"> <thead> <tr> <th>No.</th> <th>Normal Range Name</th> <th>Min</th> <th>Max</th> </tr> </thead> <tbody> <tr> <td>1</td> <td>Male-G1</td> <td>*</td> <td>*</td> </tr> <tr> <td>2</td> <td>Male-G2</td> <td>*</td> <td>*</td> </tr> <tr> <td>3</td> <td>Male-G3</td> <td>*</td> <td>*</td> </tr> <tr> <td>4</td> <td>Female-G1</td> <td>*</td> <td>*</td> </tr> </tbody> </table>								No.	Normal Range Name	Min	Max	1	Male-G1	*	*	2	Male-G2	*	*	3	Male-G3	*	*	4	Female-G1	*	*
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1	Male-G1	*	*																								
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<input type="checkbox"/> Dilution	<input type="text" value="3.8"/>	<input type="text"/>	<input type="text"/>	Technical Range	(Conc)	<input type="text" value="0.1"/>	-	<input type="text" value="30.0"/>																			
	Rerun (High/Prozone)				(mAbs/10)	<input type="text"/>	-	<input type="text"/>																			
<input type="checkbox"/> Dilution	<input type="text" value="3.8"/>	<input type="text"/>	<input type="text"/>																								
	Rerun (Low)																										
<input type="checkbox"/> Dilution	<input type="text" value="3.8"/>	<input type="text"/>	<input type="text"/>																								
				SPT Wash	<input type="checkbox"/> Enable	<input type="text"/>																					
				Stirring Speed	R1	<input type="text" value="Low"/>	R2	<input type="text" value="Low"/>																			

\*Entered by user

Chemistry Parameters		Sysmex BX-4000 Chemistry Analyzer Analytical Parameters					
Method No.	* <input type="text"/>	Name	<input type="text" value="TBIL"/>	Sample	<input type="text" value="Serum"/>		
<table border="0" style="width:100%;"> <tr> <td style="width:50%; vertical-align: top;"> <p>Limit Checks</p> <p><input checked="" type="checkbox"/> Duplicate Limit <input type="text" value="50"/> mAbs/10</p> <p><input checked="" type="checkbox"/> Sensitivity Limit <input type="text" value="200"/> mAbs/10</p> <p><input checked="" type="checkbox"/> Linearity Limit <input type="text"/> % <input type="text"/> (mAbs/10)/min</p> <p><input type="checkbox"/> Prozone Limit <input type="text"/> % <input type="text" value="Upper"/></p> <p>SL1-S <input type="text"/> - SL1-F <input type="text"/></p> <p>SL2-S <input type="text"/> - SL2-F <input type="text"/></p> <p>Sensitivity <input type="text"/> mAbs/10</p> <p><input checked="" type="checkbox"/> Absorbance Limit</p> <p>Reaction <input type="text" value="Increase"/></p> <p>Limit <input type="text" value="25000"/> mAbs/10</p> </td> <td style="width:50%; vertical-align: top;"> <p>Blank measurement</p> <p>Blank measurement: <input type="text" value="Disable reagent blank and S1 blank"/></p> <p>Measurement of Reagent Blank during Run: <input type="text" value="None"/></p> <p>Reagent blank measurement at calibration: <input type="text" value="Reagent blank (No sample)"/></p> <p>The number of measurement: <input type="text" value="Duplicate"/></p> <p>Reagent blank limit checks:</p> <p><input checked="" type="checkbox"/> Duplicate Limit <input type="text" value="20"/> mAbs/10</p> <hr/> <p>Instrument Factor</p> <p>a <input type="text" value="1.00"/> b <input type="text" value="0.00"/></p> </td> </tr> </table>						<p>Limit Checks</p> <p><input checked="" type="checkbox"/> Duplicate Limit <input type="text" value="50"/> mAbs/10</p> <p><input checked="" type="checkbox"/> Sensitivity Limit <input type="text" value="200"/> mAbs/10</p> <p><input checked="" type="checkbox"/> Linearity Limit <input type="text"/> % <input type="text"/> (mAbs/10)/min</p> <p><input type="checkbox"/> Prozone Limit <input type="text"/> % <input type="text" value="Upper"/></p> <p>SL1-S <input type="text"/> - SL1-F <input type="text"/></p> <p>SL2-S <input type="text"/> - SL2-F <input type="text"/></p> <p>Sensitivity <input type="text"/> mAbs/10</p> <p><input checked="" type="checkbox"/> Absorbance Limit</p> <p>Reaction <input type="text" value="Increase"/></p> <p>Limit <input type="text" value="25000"/> mAbs/10</p>	<p>Blank measurement</p> <p>Blank measurement: <input type="text" value="Disable reagent blank and S1 blank"/></p> <p>Measurement of Reagent Blank during Run: <input type="text" value="None"/></p> <p>Reagent blank measurement at calibration: <input type="text" value="Reagent blank (No sample)"/></p> <p>The number of measurement: <input type="text" value="Duplicate"/></p> <p>Reagent blank limit checks:</p> <p><input checked="" type="checkbox"/> Duplicate Limit <input type="text" value="20"/> mAbs/10</p> <hr/> <p>Instrument Factor</p> <p>a <input type="text" value="1.00"/> b <input type="text" value="0.00"/></p>
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<u>Registration Calibration</u>		<b>Sysmex BX-4000 Chemistry Analyzer Analytical Parameters</b>																																																	
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