BECKMAN COULTER CE

AU

Instructions For Use

TBili
TOTAL BILIRUBIN

REF OSR6112 4 x 15 mL R1 TBILC, 4 x 15 mL R1 TBILB
OSR6212 4 x 40 mL R1 TBILC, 4 x 40 mL R1 TBILB
OSR6612 4 x 173 mL R1 TBILC, 4 x 173 mL R1 TBILB

For *in vitro* diagnostic use only.

PRINCIPLE

INTENDED USE

Photometric colour test for the quantitative determination of total bilirubin in human serum and plasma on Beckman Coulter analysers.

OSR6612 for use on the AU5800, AU2700 and AU5400 systems only.

SUMMARY AND EXPLANATION

Reference[1,2]

80 – 85% of bilirubin produced daily originates from haemoglobin released by the breakdown of senescent erythrocytes, the remaining 15 – 20% results from the breakdown of haem-containing proteins such as myoglobin, cytochromes, catalases and from bone marrow as a result of ineffective erythropoiesis. A number of diseases affect one or more of the steps involved in the production, uptake, storage, metabolism and excretion of bilirubin. Depending on the disorder unconjugated or conjugated bilirubin or both are major contributors to the resulting hyperbilirubinemia. Hyperbilirubinemia can be classified as follows:

Prehepatic Jaundice: Diseases of prehepatic origin with predominantly unconjugated hyperbilirubinemia include corpuscular haemolytic anemias e.g. thalassemia and sickle cell anemia; extracorpuscular haemolytic anemia e.g. blood transfusion reaction due to ABO and Rh incompatibility; neonatal jaundice and haemolytic disease of the newborn.

Hepatic Jaundice: Diseases of hepatic origin with predominantly conjugated hyperbilirubinemia include acute and chronic viral hepatitis, liver cirrhosis and hepatocellular carcinoma.

Post hepatic Jaundice: Diseases of post-hepatic origin with predominantly conjugated hyperbilirubinemia include extrahepatic cholestasis and liver transplant rejection.

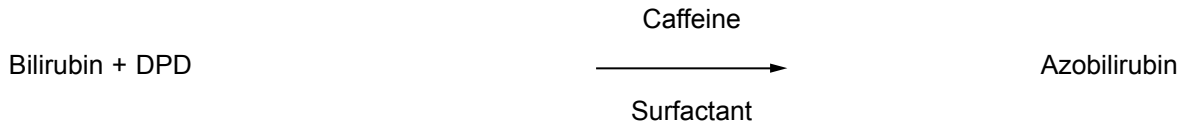
Chronic congenital hyperbilirubinemias include the unconjugated hyperbilirubinemias Crigler-Najjar syndrome and Gilbert's syndrome as well as the conjugated hyperbilirubinemias Dubin-Johnson syndrome and Rotor syndrome. The differentiation between chronic congenital hyperbilirubinemias and acquired types of bilirubinemia is accomplished via the measurement of bilirubin fractions and the detection of normal liver enzyme activities.

METHODOLOGY

Reference[3]

A stabilised diazonium salt, 3,5-dichlorophenyldiazonium tetrafluoroborate (DPD), reacts with conjugated bilirubin directly and with unconjugated bilirubin in the presence of an accelerator to form azobilirubin. The absorbance at 540 nm is proportional to the total bilirubin concentration. A separate sample blank is performed to reduce endogenous serum interference.

CHEMICAL REACTION SCHEME



SPECIMEN

TYPE OF SPECIMEN

Serum and EDTA or heparinised plasma. Protect samples from light.

Stable in serum and plasma for 7 days when stored at 2...8°C and 1 day when stored at 15...25°C.[4]

Haemolysed samples should be avoided.

REAGENTS

WARNING AND PRECAUTIONS

Exercise the normal precautions required for handling all laboratory reagents.

Dispose of all waste material in accordance with local guidelines.

REACTIVE INGREDIENTS

Final concentration of reactive ingredients:

Caffeine	2.1 mmol/L
3,5 Dichlorophenyl diazonium tetrafluoroborate	0.31 mmol/L
Surfactant	

The concentrations of the reactive components of the reagents shown on the kit label are the actual concentrations in the individual R1/R2 vials. The reagent composition which is shown in the Instructions For Use is the final concentration of these components in the reaction cuvette after addition of R1, Sample, and R2.

GHS HAZARD CLASSIFICATION

Total Bilirubin Blank R1	DANGER	
		
	H315	Causes skin irritation.
	H318	Causes serious eye damage.
	P280	Wear protective gloves, protective clothing and eye/face 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.
	P310	Immediately call a POISON CENTER or doctor/physician.
		Acetic Acid 0.1 - 1%
		Lithium dodecyl sulphate 2 - 5%
Total Bilirubin Color R1	DANGER	
		
	H315	Causes skin irritation.
	H318	Causes serious eye damage.
	P280	Wear protective gloves, protective clothing and eye/face 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.
	P310	Immediately call a POISON CENTER or doctor/physician.
		Acetic Acid 0.1 - 1%
		Lithium dodecyl sulphate 2 - 5%

SDS	Safety Data Sheet is available at beckmancoulter.com/techdocs

REAGENT PREPARATION

The reagents are ready for use and can be placed directly on board the instrument.

STORAGE AND STABILITY

The reagents are stable, unopened, up to the stated expiry date when stored at 2…8°C. Once open, reagents stored on board the instrument are stable for 90 days. Protect TBILC from light.

CALIBRATION

CALIBRATOR REQUIRED

System Calibrator Cat. No. 66300.

The calibrator total bilirubin value is traceable to the National Institute of Standards and Technology (NIST) Standard Reference Material (SRM) 916a.

Recalibrate the assay when the following occur:

Change in reagent lot or significant shift in control values;

Major preventative maintenance was performed on the analyser or a critical part was replaced.

QUALITY CONTROL

Controls Cat. No. ODC0003 and ODC0004 or other control materials with values determined by this Beckman Coulter system may be used.

Each laboratory should establish its own control frequency however good laboratory practice suggests that controls be tested each day patient samples are tested and each time calibration/blanking is performed.

The results obtained by any individual laboratory may vary from the given mean value. It is therefore recommended that each laboratory generates analyte specific control target values and intervals based on multiple runs according to their requirements. These target values should fall within the corresponding acceptable ranges given in the relevant product literature.

If any trends or sudden shifts in values are detected, review all operating parameters.

Each laboratory should establish guidelines for corrective action to be taken if controls do not recover within the specified limits.

TESTING PROCEDURE(S)

Refer to the appropriate Beckman Coulter AU analyser User Guide/Instructions For Use (IFU) for analyser-specific assay instructions for the sample type as listed in the Intended Use statement. The paediatric application is suitable for use with small volume serum/plasma samples.

CALCULATIONS

The Beckman Coulter analysers automatically compute the total bilirubin concentration of each sample.

REPORTING RESULTS

REFERENCE INTERVALS

Reference[5]	
Serum (Adults)	5 – 21 µmol/L (0.3 – 1.2 mg/dL)
Serum (Children)	
0 − 1 day	24 – 149 µmol/L (1.4 – 8.7 mg/dL)
1 − 2 days	58 – 197 µmol/L (3.4 – 11.5 mg/dL)
3 − 5 days	26 – 205 µmol/L (1.5 – 12.0 mg/dL)

Expected values may vary with age, sex, sample type, diet and geographical location. Each laboratory should verify the transferability of the expected values to its own population, and if necessary determine its own reference interval

according to good laboratory practice. For diagnostic purposes, results should always be assessed in conjunction with the patient's medical history, clinical examinations and other findings.

PROCEDURAL NOTES

INTERFERENCES

Results of studies conducted to evaluate the susceptibility of the method to interference were as follows:

Haemolysis:	Interference less than 10% up to 0.45 g/L haemoglobin
Lipemia:	Interference less than 10% up to 1,000 mg/dL Intralipid

In very rare cases gammopathy, especially monoclonal IgM (Waldenström's macroglobulinemia), may cause unreliable results.

N-acetyl-p-benzoquinone imine (metabolite of Paracetamol) will generate erroneously low results in samples for patients that have taken an overdose of Paracetamol.

Eltrombopag and its metabolites may interfere with this assay causing erroneously high patient results.

Refer to Young[6] for further information on interfering substances.

PERFORMANCE CHARACTERISTICS

PERFORMANCE CHARACTERISTICS

Data contained within this section is representative of performance on Beckman Coulter systems. Data obtained in your laboratory may differ from these values.

LINEARITY

The test is linear within a concentration range of 0 – 513 µmol/L (0 – 30 mg/dL).

SENSITIVITY

The lowest detectable level in serum on a DxC 700 AU analyser was estimated at 0.20 µmol/L.

The lowest detectable level represents the lowest measurable level of total bilirubin that can be distinguished from zero. It is calculated as the absolute mean plus three standard deviations of 20 replicates of an analyte free sample.

METHODS COMPARISON

Patient serum samples were used to compare this Total Bilirubin OSR6112 assay on the AU600 against another commercially available total bilirubin assay. Results of linear regression analysis were as follows:

y = 0.942x + 0.392	r = 0.998	n = 111	Sample range = 0.86 – 447.34 µmol/L

PRECISION

The following data was obtained on a DxC 700 AU using 3 serum pools analysed over 20 days.

n = 80	Within-run		Total	
Mean µmol/L	**SD**	**CV%**	**SD**	**CV%**
7.16	0.07	1.03	0.19	2.59
17.82	0.10	0.56	0.29	1.60
196.11	0.94	0.48	0.98	0.50

ADDITIONAL INFORMATION

DxC 700 AU requires that each reagent application has a standard format of abbreviated Closed Test Name. This Closed Test Name is required to allow automated loading of the calibrator information for each application as part of the DxC 700 AU Closed System. Refer to the table below for the Closed Test Name assigned to each application for this assay.

Test Name	Description
TBC1N	Total Bilirubin Colour (Serum)
TBB1N	Total Bilirubin Blank (Serum)
TBC1NP	Total Bilirubin Colour (Serum Paediatric)
TBB1NP	Total Bilirubin Blank (Serum Paediatric)

Setting Sheet Footnotes

‡ The above parameters must be entered twice using test names TBILC (colour) and TBILB (blank). Set the test as SAMPLE BLANK in the INTER RELATED TEST menu

User defined
† System Calibrator Cat. No.: 66300
* Values set for working in SI units (µmol/L). To work in mg/dL divide by 17.1

§ Set the factor range for the blank reagent at –99999 to 99999

ж Set the test as SAMPLE BLANK in the COMMON TEST PARAMETERS TEST NAME SAMPLE BLANK menu.

REVISION HISTORY

Added new languages

Preceding version revision history

Revised Interferences section.

REFERENCES

1. Thomas L. Bilirubin. In: Thomas L, ed. Clinical laboratory diagnostics. Use and assessment of clinical laboratory results. Frankfurt/Main: TH-Books Verlagsgesellschaft, 1998:192-202.

2. Balistreri WF, Shaw LM. Liver Function. In: Tietz NW, ed. Fundamentals of clinical chemistry. Philadelphia:WB Saunders Company, 1987:733-737.

3. Tolman KG, Rej R. Liver Function. In: Burtis CA, Ashwood ER, eds. Tietz textbook of clinical chemistry. Philadelphia: WB Saunders Company, 1999;1136-1137.

4. Ehret W, Heil W, Schmitt Y, Töpfer G, Wisser H, Zawta B, et al. Use of anticoagulants in diagnostic laboratory investigations and stability of blood, plasma and serum samples. WHO/DIL/LAB/99.1 Rev.2: 24pp.

5. Painter PC, Cope JY, Smith JL. Reference information for the clinical laboratory. In: Burtis CA, Ashwood ER, eds. Tietz textbook of clinical chemistry. Philadelphia:WB Saunders Company, 1999;1803pp.

6. Young DS. Effects of Drugs on Clinical Laboratory Tests, AACC, 5th ed. AACC Press, 2000.

EC REP Beckman Coulter Ireland Inc., Lismeehan, O'Callaghan's Mills, Co. Clare, Ireland +(353) (0) 65 683 1100

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