



**ELK Biotechnology**

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## **Total Bilirubin Assay Kit Instruction**

**(BC017 Total Bilirubin kit , TBIL      Chemical Oxidase Method)**

### **【Application】**

This kit was used for quantitative determination of total bilirubin in serum (blood plasma). Serum (blood plasma) total bilirubin is common in hepatitis, extrahepatic biliary tract obstruction, hemolytic disease and so on.

### **【Principle】**

Total bilirubin was oxidized by sodium nitrite in the presence of surfactant Triton X-100. The decrease in absorbance at 405 nm is proportional to its concentration.

### **【Composition: 96T】**

Reagent	Specifications	Composition	Concentration
Reagent 1	24ml×1 bottle	Tris- Hcl	50×10 <sup>-3</sup> mol/L
		Triton X- 100	10ml/L
Reagent 2	6ml×1 bottle	PBS	10×10 <sup>-3</sup> mol/L
		Nitrite	80×10 <sup>-3</sup> mol/L
Standard	1 bottle (Concentration reference label)		store in dark

### **【Storage Conditions and Validity period】**

The reagents can stable for 1 year at 2~8℃. Transport in summer is refrigerated, not frozen. It can be kept for 1 month at 2~8℃ after open.

### **【Sample Request】**

Serum or blood plasma. The blood should be separated in time to avoid hemolysis. And keep it avoid light



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**【 Inspection Methods 】**

**1 、 Operation**

	Blank	Standard	Assay
ddH <sub>2</sub> O (μl)	7		
Standard solution (μL)		7	
Sample (μl)			7
Reagent 1 (μl)	200	200	200
Mix thoroughly and incubate at 37℃ for 5min , Read OD values A1 at 450 nm.			
Reagent 2 (μl)	50	50	50
Mix thoroughly and incubate at 37℃ for 5min , Read OD values A2 at 450 nm. Δ A=A1-A2			

**Note :** Practice multi-channel pipettor operation.

**【 Formula 】**

$$T-BIL (\mu\text{mol/L}) = \frac{\Delta A_{\text{sample}} - \Delta A_{\text{Blank}}}{\Delta A_{\text{Standard}} - \Delta A_{\text{Blank}}} \times C_{\text{Standard}}$$

**【 Reference Range 】**

Healthy adults : 3.42-20.5μmol/L (0.2- 1.2mg/dL)

( It is recommended that each laboratory establish its own reference range. )

**【 Limitation 】**

- 1 、 The determination of TBIL is only one of the indicators of scientific research, and it is also necessary to make a comprehensive judgment based on the sample body's other experimental items and methods.
- 2 、 Interfering substance: NO interference when Hemoglobin ≤ 25g/ L, VC≤ 30mg/dl, TG≤ 2000mg/dl.



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**【 Performance Index 】**

Blank absorbance value:  $A_{450nm}$  (10mm)  $\leq 0.05$ ,

Linear range:  $3.4 \sim 684 \mu\text{mol/L}$  (a. Linear correlation coefficient  $R^2 \geq 0.995$ . b.  $3.4 -$

$100 \mu\text{mol/L}$ , Linear deviation  $\leq 10.0 \mu\text{mol/L}$ ,  $100 - 684 \mu\text{mol/L}$ , Linear deviation  $\leq 10.0\%$ ).

Accuracy: Relative deviation  $\leq 10.0\%$

Precision: Inter-batch CV  $< 4.0\%$ , Intra batch CV  $\leq 6.0\%$

Sensitivity : when the sample concentration is  $90 \mu\text{mol/L}$ , the OD value is not less than 0.04 ;

**【 Announcement 】**

- 1 、 The kit is for scientific research use only. If the reagent is accidentally splashed into the skin ,eyes and so on, you must wash with clean water. Accidental ingestion must be treated in the hospital。
- 2 、 The standard product is easy to decompose, so we don't provide it in the kit. You can calculate refer to our standard curve or calculation formula。
- 3 、 Sample and the reagents dosage can be adjusted according to you need ; Different batches of reagents must not be mixed.
- 4 、 Precautions should be taken when using and following all laboratory reagent operations. All wastes shall be disposed in accordance with local regulations。