Total Bile Acid (TBA) Assay Kit

(Cat/No.:BC064 Size:96T Microplate method)

1.Composition(The kit is valid for 6 months)

Reagents	Specifications	Remark
R1	18ml×1 bottle	Store at 2～8℃
（R2）	6ml×1 bottle	Store at 2–8℃ away from light.
Standard（50μmol/L）	0.2ml×1 tube	Store at 2–8℃ away from light.

2. Storage conditions and shelf life

The kit should be stored at 2–8°C, protected from light, for at least 6 months. During summer transport, refrigeration is necessary; freezing is not permitted.

3. Assay Significance

The kit is used for quantitative determination of total bile acid concentration in serum. Total bile acid (TBA) is mainly used for screening and follow-up of hepatobiliary diseases and as a market of liver parenchymal damage and cholestasis. In the case of hepatitis, Cirrhosis, alcoholic, alcoholic liver disease, drug induced liver damage and cholestasis, TBA are elevated.

4. Sample requirements

1、Isolate the serum within 2 hours of taking the blood. Its stable no more than 8 hours at 15～30℃,one week at 2～8℃and its stable 3 months at -20℃.

2、Interfering substance: When the conjugated bilirubin≤5mg/dl, the unconjugated bilirubin ≤ 20mg/dl, VC≤1mg/dl, Triglyceride≤9.25mmol/ L, and the hemoglobin≤100mg/dl, there is no effect on the result.

Assay Principle

$$Bile\ acid + \text{S-NAD} \xrightarrow{3\alpha\text{-}HSD} 3\text{-}ketosteroid + S\text{-}NADH$$

$$3\text{-}ketosteroid + S\text{-}NADH \xrightarrow{\text{Diaphorase}} NAD + Bile\ acid$$

The cyclic reaction is continuously completed, and the absorbance is measured at a wavelength of 405 nm. The change in absorbance is directly proportional to the concentration of bile acids.

Operation procedure: (ELISA reader)

	Blank tube	Standard tube	Assay tube
distilled water（μL）	3		
Standard products（μL）		3	
Test sample（μL）			3
R1（μL）	180	180	180
Gently shake to mix, incubate at 37°C for 5 minutes.			
R2（μL）	60	60	60
Gently shake to mix, incubate at 37°C for 1 min, and then read the absorbance A0 at 405 nm using an ELISA reader. After incubating at 37°C for 3 min, read the absorbance A1 again. Calculate ΔA = A1 - A0.			

5. Fomula

$$\text{TBA Content}\ (\mu mol/L) = \frac{\Delta A_{\text{Assay}}}{\Delta A_{\text{Standard}}} \times \underset{(\mu mol/L)}{S\ \text{tan}\ dard\ \ Concentration}$$

6. Reference range

0～10μmol/L(Human serum data are for reference only. It is recommended that each laboratory establish its own reference range.)

7. Product performance

1、Linear range： 0～180μmol/L（Decision basis：r^2≥0.990）；
2、Accuracy： Inaccuracy≤15.0%； Recovery rate within (100±20%).
3、precision： Within-lot deviation CV≤5.0%； relative range between batches≤10.0%；
4、Reagent blank absorbance： With a wavelength of 405 nm and an optical path of 1 cm, the measured absorbance value of the reagent was ≤0.7.
5、Standard accuracy： Relative deviation≤10.0%； Homogeneity:CV≤5.0%

8. Points to attention

1、 If the TBA concentration in the sample is greater than 150μmol/L, it is then diluted with normal saline and result is multiplied by the dilution factor.
2、 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.
3、 If the instrument does not have a filter with the wavelength required by this kit, choose a filter with a similar wavelength (but not too different).
4、 The ratio of sample to reagent can be adjusted according to the need.
5、 Mixing of different bathes of reagents is not recommended.
6、 If the ΔA value is low (≤0.003) during sample determination, the reaction time can be extended from 3 min to 5 min or 10 min.