

# N BIO - TOTAL BILE ACIDS

(Enzyme cycling method)

KIT NAME	KIT SIZE	CAT. NO
N BIO - Total Bile Acids	1 x 15 ml	DTBA01015M

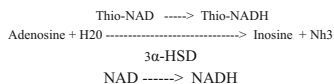


## INTRODUCTION

Total Bile Acids are steroid acids in the bile. In human, it includes taurocholic acid, lycocholic acid, and so on. Bile acids are synthesized in liver and released to bile and then get to help body digesting lipids and absorbing fat-soluble vitamins. Monitoring serum TBA levels is useful for diagnosing liver diseases, such as sometimes the elevated serum bile acids concentration indicates the development of colon cancer.

## METHOD PRINCIPLE

The Kit utilizes enzymatic and kinetic reactions to measure the amount of TBA ( $\mu\text{mol/L}$ ) in human serum or plasma.



3- $\alpha$ -hydroxysteroid dehydrogenase converts bile acids to 3-keto steroids and Thio-NADH in the presence of Thio-NAD. The reaction is reversible. With excess NADH, the cycling reaction enlarge the specific change of absorbance at 405 nm due to the formation of Thio-NADH. The rate of increase in absorbance at 405 nm is directly proportional to the TBA activity in the sample.

## KIT CONTENTS

Reagent Name	DTBA01015M
R1 TBA Reagent	1 x 11.25 ml
R2 TBA Reagent	1 x 3.75 ml
R3 Calibrator	1 vial

Please refer calibrator value mentioned in the vial. The reagents when stored at 2-8°C are stable up to expiry date printed on the package. Protect from light and avoid contamination.

## WORKING REAGENT PREPARATION AND STABILITY

Assay can be performed with use of separate R1-TBA and R2-TBA reagents.

## CONCENTRATIONS IN THE TEST

Tris buffer pH 3.0	30 mmol/L
Thio-NAD	2 mmol/L
NADH	2 mmol/L
3- $\alpha$ -HSD	$\leq 5 \text{ U/L}$

## SPECIMEN

Follow standard laboratory procedures to collect serum preventing hemolysis. It is recommended to perform test immediately after sample collection. If the test cannot be done immediately, specimens may be stored at 2-8°C for 14 days.

## PROCEDURE

These reagents may be used both for manual assay and in several automatic analyzers. Applications for them are available on request.

Wavelength	405 nm
Temperature	37°C
Cuvette	1 cm

## Pipette into the cuvettes:

Reagent	Standard (S)	Test (T)
R1 TBA Reagent	750 $\mu\text{l}$	750 $\mu\text{l}$
R3 TBA Reagent	250 $\mu\text{l}$	250 $\mu\text{l}$
Bring to assay temperature, then add		
Calibrator	20 $\mu\text{l}$	-
Sample	-	20 $\mu\text{l}$

Mix well and after exactly 60 sec read absorbance A1 of the test (T) and Calibrator (C) against air. After next 120 sec repeat absorbance reading (A2) and calculate  $\Delta A (A2 - A1)$  for the test and Calibrator.

## CALCULATION

TBA concentration =  $\Delta A(T) / \Delta A(C) \times$  calibrator concentration

## REFERENCE VALUES

upto 12  $\mu\text{mol/L}$

It is recommended for each laboratory to establish its own reference ranges for local population.

## QUALITY CONTROL

To ensure adequate quality control, each run should include assayed normal and abnormal controls. If commercial controls are not available it is recommended that known value samples be aliquoted, frozen and used as controls

## PERFORMANCE CHARACTERISTICS

Linearity	: up to 90 $\mu\text{mol/L}$ . Dilute the sample approximately and Multiply result with dilution factor.
Accuracy	: Bias proportion 90%~110%
Precision	: Within Run: CV $\leq$ 3%; Run-to-Run: Cvs $\leq$ 5%
Interference	: no interference detected for: ascorbic acid ( $\leq$ 40mg/dl), chylomicrons ( $\leq$ 2200NTU), bilirubin ( $\leq$ 20mg/dl), unconjugated bilirubin ( $\leq$ 20mg/dl) and hemoglobin ( $\leq$ 450mg/dl).

## WASTE MANAGEMENT

Please refer to local legal requirements.

## LITERATURE

1. Kirti Rani Sharma, Int J Pharm Biomed Sci 3(2):28-34(2012)
2. Shima T. et al., J Gastroenterol Hepatol. 15(3):294-9 (2000)
3. Brandon Bartling et al., Analyst 134:973-979 (2009)

## SYSTEM PARAMETERS

Method	End Point
Wavelength	405 nm
Zero Setting	Distilled Water
Temperature Setting	37° C
Incubation Temperature	37° C
Incubation Time	-----
Delay Time	60 secs
Read Time	120 secs
No. of Reading	2
Interval Time	60 secs
Sample Volume	0.02 ml (20 µl)
Reagent Volume	1.0 ml (1000 µl)
Standard Concentration	Refer Calibrator vial
Units	µmol/l
Factor	-----
Reaction Slope	Increasing
Linearity	90 µmol/l

**Reagent Blank Absorbance :** at 405nm wavelength and 10 mm optical diameter, O.D.  $\leq$  0.5



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