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(FOR RESEARCH USE ONLY. DO NOT USE IT IN CLINICAL DIAGNOSIS!)

# Direct Bilirubin Assay Kit

Catalog No.: BC00065

Size: 100T

If you have any questions or need further help during experiment, please don't hesitate to contact us through the following methods:


✉Email（Sale） order@enkilife.com

✉Email（Techsupport） techsupport@enkilife.com

Tel: 0086-27-87002838

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**Shelf life:** Please refer to the label on the outer package.

**Techsupport:** In order to provide you with better service, please inform us the lot number on the label of the outer package.

## Basic Information

| | |
|---|---|
| **Product Name** | Direct Bilirubin Assay Kit |
| **Detection Methods** | Colorimetric |
| **Sample type** | Serum, plasma |
| **Detection Type** | Quantitative |
| **Detection instrument and wavelength** | Microplate reader (450-470 nm , optimal detection wavelength is 450 nm) |
| **Range** | 12.5-100μM |
| **Sensitivity** | 1μM |

## Product Introduction

Direct Bilirubin (DBIL), also known as conjugated bilirubin, is the portion of total bilirubin that has been conjugated with glucuronyl transferase in the liver. The measurement of direct bilirubin is essential for assessing liver function and the health of the biliary system .

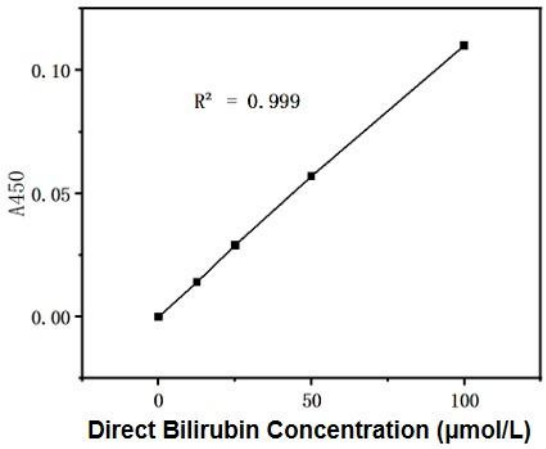
## Features

★ Easy to operate, the test can be completed within 5 minutes .

## Detection Principle

Direct bilirubin in serum In an acidic environment, serum-conjugated bilirubin can directly react with the diazo reagent to generate azobilirubin. The absorbance of this complex at 450 nm is proportional to the direct bilirubin concentration in the sample.

The figure below shows the standard curve of direct bilirubin determined by this kit. The following standard curve is for reference only:



## Product Composition

| Serial Number | Product Name | Packing Specifications | Storage |
|---|---|---|---|
| Reagent 1 | R1 | 30mL | Store at -20℃ away from light , store |
| Reagent 2 | R2 | 5mL | Store at -20℃ away from light , store |
| | 96-well ELISA plate | 1 plate | RT |
| | 96-well membrane | 2 pieces | RT |

## Storage Conditions

The unopened kit can be stored at -20°C for 6 months.

## Preparation before the experiment

### Sample processing

Serum and plasma should be separated promptly after blood collection to avoid hemolysis and light exposure. Samples should be measured as soon as possible.

### · Preparation of the kit

1. Before testing, the reagents in the kit were equilibrated to room temperature.
2. Dilution of different concentrations of standard: dilute reagent 3 with water in half to different concentrations such as 100, 50, 25, 12.5, 0 (blank well) μmol/L
3. Preparation of working solution: Mix reagent 1 and reagent 2 in a volume ratio of 1:1 to form working solution. Use immediately after preparation and store in a dark place. The best effect is achieved if the solution does not turn red significantly within 30 minutes.

## Operation Process

1. Standard wells: Take 10 μL of standard solutions of different concentrations and add them to the corresponding standard wells.
   Assay wells: Take 10 μL of sample and add it to the corresponding sample wells.
2. to the standard wells and assay wells in step “ 1 ” .
3. to the standard wells and sample wells in step “ 1 ” .
4. Oscillate on a microplate reader for 5 s and measure the OD value of each well at 450 nm.

**Operation table**

| | Standard wells | Assay wells |
|---|---|---|

| | | |
|---|---|---|
| Standard solutions of different | 10 | -- |
| Sample to be tested (μL) | -- | 10 |
| Reagent 1 (μL) | 250 | 250 |
| Working solution (μL) | 50 | 50 |
| Oscillate on the microplate reader for 5 s and measure the OD value of each well at 450 nm. | | |

## Result Calculation

Standard fitting curve: y = ax + b

Normal serum (plasma) sample, direct bilirubin concentration calculation formula:

Concentration (mmol/L) = (ΔA450 - b) ÷ a × f

annotation:

y: OD value of standard well - OD value of blank well (OD value when the concentration of standard is 0)

x: concentration corresponding to the absorbance

a: slope of the curve

b: intercept of the curve

ΔA450: Sample OD value - blank OD value (OD value when the standard concentration is 0)

f: dilution factor of the sample before adding it to the detection system

## Notes

1. The optimal detection wavelength of the ELISA reader is 450nm, and detection can be performed in the range of 450nm-470nm.
2. This product is limited to scientific research by professionals and shall not be used for clinical diagnosis or treatment, shall not be used as food or medicine, and shall not be stored in ordinary residences.