台塑生醫科技股份有限公司
FORMOSA BIOMEDICAL TECHNOLOGY CORP.

BILIRUBIN TOTAL TBIL

FOR BECKMAN CX AND LX SYSTEMS

INTENDED USE
For the quantitative determination of total Bilirubin in serum.

CLINICAL SIGNIFICANCE
Bilirubin, in bile, is derived primarily from the breakdown of hemoglobin when sensecent red blood cells are phagocytized. Normally, about 6 to 6.5 g of hemoglobin in aged red blood cells are broken down daily in an adult to form about 220 mg of Bilirubin; another 50 to 60 mg of Bilirubin originate from other sources. The exact mechanism to form Bilirubin is not well understood. But it is known that the heme group of hemoglobin converts to Bilirubin in reticuloendothelial system, which binds to albumin in plasma and esterified to Bilirubin diglucuromide (BDG), which is when secreted from liver as a waste product. It is present in serum in the free and conjugated forms. An increase in the formation or retention of Bilirubin in the body results in increased levels of serum Bilirubin and jaundice. This hyperbilirubinemia is classified as either pre-hepatic, hepatic or post-hepatic depending on the principal cause of condition. Therefore, determination of the total Bilirubin and its conjugated (direct) Bilirubin is important for the differential diagnosis of hyperbilirubinemia.

PRINCIPLE
Most chemical methods for the determination of Bilrubin are based on the reaction of Bilirubin with a diazolium salt to form an azobilirubin. Direct Bilirubin is water soluble and will react with diazolium salts in aqueous solution. However free Bilirubin will react only after the addition of a solubilizer. Malloy and Evelyn in 1937 developed the first useful quantitative method for Bilirubin determination. This method utilized methanol as solubilizer for free Bilirubin. Many chemicals had later been introduced as solubilizer for free Bilirubin. Among these, dimethyl sulfoxide discovered by Winston and Cehelrk and modified by Walters and Gerade had been recommended as a good solubilizer for free Bilirubin. The Total Bilirubin reagent presented here is a modification of Walters and Gerarde by using dimethyl sulfoxide as solubilizer which facilitates reaction of free Bilirubin with diazo reagent.
Sulfanilic acid reacts with sodium nitrite to give diazotized sulfanilic acid. In the presence of dimethyl sulfoxide, both direct and indirect bilirubin couples with diazotized sulfanilic acid to give a colored complex azobilirubin which is measured spectrophotometrically at 555 nm.

SPECIMEN COLLECTION AND PREPARATION
The sample be collected without hemolysis, since hemoglobin inhibits the diazo reaction.
Avoid direct light exposure to the specimen since Bilirubin values may decrease as much as 50 % in one hour (Valdze et.al). Serum specimen may be kept in dark in 2-8℃ for up to one week, and in freezer for 3 months without appreciable change in the Bilirubin levels.

REAGENT
Total Bilirubin reagent:
•••Each kit contains 2 cartridge of Total Bilirubin reagent (2 x 400 tests)
•••Ready to use

•Components :	Surlfanilic acid	27 mM
	Sodium nitrite	0.12 mM
	HCl	51 mM
	DMSO	45% (v/v)

Bilirubin calibrator: Total Bilirubin 51.3μmol/L (3 mg/dl); Direct Bilirubin 32 μmol/L(1.871 mg/dl).

PRECAUTIONS:
1. For in vitro diagnostic use only.
2. Since all specimens are potentially infectious, they should be handled with appropriate precautions and practices in accordance with Biosafety level 2 as recommended by USA NIH manual Biosafety in Microbiological and Biomedical Laboratories, and in accordance with National or local regulations related to the safety precautions of such materials.
3. Each laboratory has to perform the quality control test to assure the results being reliable before running the specimen tests.

PROCEDURE:
Use open channel and follows the attached parameters and procedures to perform the tests.

EXPECTED VALUES
Total Bilirubin: Adults < 20.5μmol/L(1.2 mg/dl)
Newborn < 205μmol/L(12mg/dl)

Serum Bilirubin levels are usually higher in the newborn, but fall rapidly after the first week and reach adult levels by the fourth week of life.
Since normal values are affected by age, sex, diet, geographical location, and other factors, each laboratory should establish its own 'normal' value based upon the specific situation in daily laboratory operation.

NOTE: It is generally recommended that each laboratory establish its own range of normal values for commonly performed tests.

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