Serum Bilirubin and Its Binding Parameters in Chinese Neonates

Kwang-Jen HSIAO[1,2], Yu-Lin WONG[3], Wen-Kuo TING[2]

Departments of Medical Research,[1] Laboratory[2] and Pediatrics[3] Veterans General Hospital Taipei, Taiwan, ROC

The incidence of neonatal jaundice in Chinese is much higher than that in Caucasian population. It was reported that the mean peak levels of total bilirubin (11-13 mg/dl) in Chinese neonates were twice of those in Caucasian neonates. Since the measurement of serum total bilirubin concentration alone can no longer be considered a sufficient means of assessing hazard to the infant with jaundice. In order to provide better laboratory aid to diagnosis and management of neonatal jaundice, we intended to study bilirubin binding parameters in addition to serum total bilirubin in Chinese neonates.

For establishing the reference background information, 300 neonates were randomly selected. For each of them, approximate 0.2-0.3 ml of blood were collected in capillary with brown vessel (Sarstedt No. 16,441) by skin-puncture technique on the 1st, 3rd and 5th day of age. Sera were separated by high-speed centrifugation and stored at -20°C if it was not assaied immediately. The total bilirubin concentration was determined spectrophotometrically with within-run and day-to-day precision of 0.5-2.0% and 1.0-1.9% (C.V.) in the working range, respectively. The unbound bilirubin was measured by the "peroxidase" method (Jacobsen & Wennberg, *Clin. Chem. 20,* 783-789, 1974). The within-run and day-to-day precision of this assay were determined to be 0.2-4.0% and 0.4-5.2% (C.V.), respectively. The reserve binding capacity may be estimated either by bilirubin titration with "peroxidase" method or the 2-(4'-hydroxyazobenzene) benzoic acid (HABA) binding method. Although the HABA method was shown with lower specificity and sensitivity, it required only 50μl of sample and was easier to perform than that of titration method. The dye-binding method with bromcresol green (BCG) was used to determine the concentration of albumin. The results were calculated by eliminating data out of 3SD range from the mean. Both the total bilirubin and unbound bilirubin were peaked at day three ($p < 0.001$). Unbound bilirubins were determined to be 0.44±0.50 nM (mean±SD; n=307), 0.73±0.64 nM (n=287), and 0.53±0.58 nM (n=236) for day one, three and five, respectively. The distribution of unbound bilirubin was not a normal distribution and based on the total data, the upper 97.5% limits for day one, three, and five were 1.65nM, 2.45nM and 2.70nM, respectively. The total bilirubin of day one, three and five were determined to be 5.9±1.9 mg/dl (n-303), 10.6±2.6 mg/dl (n=286) and 9.6±2.7 mg/dl (n=239), respectively. The reserve binding capacity determined by HABA method was decreased from 91±12% (n=157) of day one to 81±11% (n=166) of day three and day five. Albumin also decreased slightly from 4.1±0.3 g/dl of day one to 4.0±0.4 g/dl of day three and five. There were no statistical differences of reserve binding capacity and albumin for day three and five.

These data indicated that 30% of Chinese neonates would be considered pathological jaundice according to the criteria of Dr. T.A. Blumenfeld which were based on the serum total bilirubin. The correlation between those bilirubin binding

Proc. Post-APCCB Symposium on Recent Advances in Clinical Biochemistry; Taipei, pp. 32-34 (1982)

parameters and the bilirubin encephalopathy, and the guideline for the application of these bilirubin indexes to the diagnosis and management of neonatal jaundice in Chinese neonates remain to be established.

(This study was partially supported by a grant "NSC71-0412-B075-3" from the National Science Council of R.O.C.)