ABSTRACTS

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PIGMENTS–BILIRUBIN

THE SEPARATION OF DIRECT AND INDIRECT BILIRUBIN BY PAPER CHROMATOGRAPHY. *G. Gries, P. Gedigk and J. Georgi.* From the Medical Department and the Pathological Institute of the University of Marburg, Germany. Ztschr. Physiol. Chemie *298:* 132–139, 1954.

Protein-free, direct-reacting bilirubin was obtained from serum and urine of jaundiced patients. On paper chromatography it was found that with aqueous solvent systems, the Rf of this pigment varied little over the range from pH 2 to pH 12. On the other hand, crystalline bilirubin remained on the starting line from pH 2 to pH 7. Above pH 8 or 9, the Rf of crystalline bilirubin gradually increased with increasing pH, but never reached the Rf value of the direct bilirubin. In order to prevent oxidation of the pigments, the chromatograms were run in darkness under strictly anaerobic conditions.—*R.S.*

A CHROMATOGRAPHIC METHOD FOR THE DETERMINATION OF THE THREE BILE PIGMENTS IN SERUM. *Barbara H. Billing.* From the Department of Chemical Pathology, Postgraduate Medical School, London, England. J. Clin. Path. *8:* 126–129, 1955.

In an earlier publication it had been shown that in obstructive jaundice two water-soluble bile pigments can be demonstrated in the serum, both giving a direct van den Bergh reaction. The separation of these two polar pigments (pigments I and II) from the non-polar bilirubin is possible by reverse-phase chromatography. In comparing the amounts of pigment I and II present in the serum of twenty-seven jaundiced patients with the values obtained by the conventional Malloy and Evelyn method, a high degree of correlation was found between the values for direct reacting bilirubin and the total amount of pigments I and II. Thus the Malloy and Evelyn method for determining direct bilirubin in serum seems to give a reliable estimate of the total amount of polar pigments present. On the other hand, it was not possible to identify either of the directly reacting bile pigments with the promptly reacting "1 minute bilirubin."—*R.S.*

THE THREE SERUM BILE PIGMENTS IN OBSTRUCTIVE JAUNDICE AND HEPATITIS. *Barbara H. Billing.* From the Department of Chemical Pathology, Postgraduate Medical School, London, England. J. Clin. Path. *8:* 130–131, 1955.

In patients with obstructive jaundice and with hepatitis, quantitative determinations of bilirubin and of the two polar diazo-positive pigments present in the serum are reported. In all the sera, the proportion of pigment I was greater than that of pigment II or of bilirubin. On the other hand, in human bile more than 80 per cent of all the diazo-positive pigments were found to be pigment II. In the rat with bile-duct obstruction, pigment II was

shown to predominate in serum as well as in bile. The possibility is considered that pigment I is an intermediate compound between bilirubin and pigment II.—*R.S.*

THE QUESTION OF THE SO-CALLED DIRECT AND INDIRECT REACTION OF BILIRUBIN. *M. Jirsa.* From the 1st Medical Clinic, Charles University, Praha. Čas. lék. čes. *93:* 1380–1382, 1954.

The author prepared different colloidal solutions of pure bilirubin and of bilirubin stabilized with saponin as nonprotein stabilizer and with albumin. These solutions had the same properties as the indirect bilirubin in the serum. The author expresses the opinion that even the bilirubin in serum is present as a lyophobic colloid stabilized with albumin. The direct bilirubin is adsorbed on the surface of albumin micells, and albumin in this case behaves as an ion exchanger.—*M.N.*

DIAZO REACTION OF THE SERUM BILIRUBINS. *H. A. Kühn, R. Schneider and I. Spitzmüller.* From the Medical Clinic of the University of Freiburg, Germany. Ztschr. ges. exper. Med. *124:* 52–64, 1954.

In guinea pigs and rabbits, bilirubin was injected by intravenous route and its serum level was determined at several time intervals after the injection. In animals with healthy livers, the highest serum levels were reached immediately after the injection, and all the pigment gave the indirect diazo reaction. On the other hand, in animals with experimental liver necrosis, and in humans with cirrhosis, some of the injected bilirubin gave the "direct" diazo reaction. Direct-reacting bilirubin appeared in the serum only some time after the injection of the pigment, reaching a maximum after about one hour. In animals with severe liver damage up to 90 per cent of the total bilirubin present in the serum exhibited a "direct" diazo reaction. In vitro, serum of animals with liver cell necrosis was unable to convert added bilirubin into direct reacting material. The authors conclude that the hepatic cells are essential for the production of a direct diazo reaction. In liver cell necrosis, direct reacting bilirubin is regurgitated into the serum.—*R.S.*

THE NATURE OF DIRECT BILIRUBIN. *Barton Childs.* From the Harriet Lane Home, The Johns Hopkins Hospital, and the Department of Pediatrics, The Johns Hopkins University School of Medicine, Baltimore, Md. Bull. Johns Hopkins Hosp. *97:* 333–342, 1955.

Sera from jaundiced patients, containing both direct and indirect reacting bilirubin, were subjected to paper electrophoresis at different pH. At pH of 8.6, both bilirubin fractions migrated with the serum albumin fraction. At pH values of 6.5 and below, progressively more of the indirect pigment disassociated from the albumin, remaining at or near the starting point. This finding would indicate that direct bilirubin is more tightly bound to the serum protein than indirect bilirubin. It was further observed that incubation of serum containing mainly direct bilirubin with versene resulted in a relative increase in indirect reacting pigment. Similarly, dialysis of such serum against a 0.5 molar versene solution appeared to convert some of the direct into indirect reacting bilirubin. The author interprets these findings in support of the concept that direct reacting bilirubin is a bilirubin-metal-albumin complex.—*R.S.*

INCREASED PLASMA BILIRUBIN IN NEWBORN INFANTS IN RELATION TO BIRTH WEIGHT. *B. H. Billing, P. G. Cole and G. H. Lathe.* From the Bernhard Baron Memorial Research Laboratories, Queen Charlotte's Maternity Hospital, and the Institute of Obstetrics and Gynaecology, British Postgraduate Medical Foundation, University of London. Brit. M. J. *2:* 1263–1265, 1954.

A comparison has been made between birth weight of newborn infants and serum bilirubin levels. Babies with overt fetal hemolytic disease were excluded from the study. A high degree of correlation was found between decreasing birth weight and increase in physiologic jaundice. Very high serum bilirubin levels were found in premature infants with birth weights from 1 to 2 Kg. In contrast to erythroblastosis fetalis, the peak values for serum bilirubin levels were reached only around the 4th to the 6th day of life. Most of the bilirubin present was of the type giving the indirect van den Bergh reaction, and the

authors consider the greatly increased concentration of plasma bilirubin an important factor in the development of the central nervous system lesions which are frequently associated with prematurity.—*R.S.*

Is Neonatal Icterus Accompanied by Bilirubinuria? *V. Dietel and R. Schmöger.* From the Pediatric Clinic of the University of Leipzig, Germany. Arch. Kinderheilk. *148:* 235–240, 1954.

The neonatal jaundice in prematures is generally more outspoken and of longer duration than in full-term babies. In a group of 24 premature male infants the urinary bilirubin excretion was found to range from 0.11 to 3.61 mg./100 ml. Highest values occurred around the fifth day of life. Urinary bilirubin excretion seemed to depend largely on the plasma bilirubin level. It has long been a controversial matter whether or not in neonatal jaundice bilirubin is gaining access to the urine. At least for premature babies this question now seems to have been answered in the affirmative.—*R.S.*

Bilirubin Elimination from Blood during Acute Hypoxia. *E. Jalavisto, H. Lybeck, H. A. Salmi and I. Sundholm.* From the Institute of Physiology, University of Helsinki, Finland. Ann. Med. exper. et biol. Fenniae, *31:* 437–446, 1953.

The clearance by the liver of parenterally injected bilirubin was tested in normal dogs and in dogs subjected to acute hypoxia. Animals with and without anesthesia were exposed to oxygen-nitrogen mixtures containing from 6.7 to 9 per cent oxygen. No significant difference was found in the rate of disappearance of the injected bilirubin from the plasma of normal dogs and hypoxic dogs.—*R.S.*

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