Neonatal Hypothyroid Screening by Adaptation of Hybritech's Thyrotropin Tandem® Methodology, M. C. Haven, C. W. Ludvigsen, and M. M. McLochlin (Dept. of Pathol. and Microbiol., University of Nebr. Med. Center, 42nd and Dewey Ave., Omaha, NE 68105)

In this simple modification of the Hybritech immunoradiometric (immunoassay) technique for thyrotropin (TSH) we use a smaller sample, thus improving its suitability in screening programs for congenital hypothyroidism.

Serum was separated from capillary blood collected into a 700-µL Microtainer Tube (Becton Dickinson) from a neonate. To 50 µL of the specimen we added 150 µL of the zero standard, vortex-mixed, and followed the manufacturer's directions thereafter. The standard curve was fit by a hyperbolic function (I), and the concentration in the sample was determined from this curve.

Patient comparisons (35) to demonstrate the efficacy of diluting serum samples in the normal range gave the least-squares regression equation $y = 0.93x + 1.49$ ($r = 0.996$). At two concentrations the mean between-run precision was 7.2 (SD 0.82) and 30.16 (SD 3.07) micro-int. units/mL. For 80 normal neonates, postnatal ages one to 20 days, the mean TSH concentration with this modified technique was 4.08 (SD 2.95) micro-int. units/mL, the range 0.4 to 13.2. We selected an upper normal limit of 15 micro-int. units/mL. To negate dilution errors that may occur at low-normal values, values <6 are reported as "<6 micro-int. units/mL.”

This modification circumvents problems associated with sample collection on filter paper, fulfills sample volume requirements for a neonatal screening method, and significantly reduces costs, because the TSH can be measured in neonates with the same assay and in the same run as a routine TSH.

Reference


The "Beta Quik Stat" pregnancy kit (Pacific Biotech, Inc., San Diego, CA) detects specifically the intact molecule of chorionic gonadotropin (hCG). It is a solid-phase "sandwich"-type enzyme immunoassay procedure. A total of 65 serum samples which contained 5 to 185 int. units of hCG per liter (against 2nd International Standard) as quantified by the β-hCG RIA procedure were tested by Beta Quik Stat. The results indicate that Beta Quik Stat gives positive results for samples that contain hCG in concentrations >25 int. units/L and borderline-positive results for those containing 13 to 22 int. units/L. Similar results were obtained for urine samples containing known concentrations of hCG, prepared by adding standard intact hCG to a pooled specimen of urine from nonpregnant persons.

The Beta Quik Stat not only detects hCG in serum, plasma, and urine but also in whole blood. Either EDTA or heparin can be used as an anticoagulant. I compared more than 100 whole-blood pregnancy test results with those obtained from the corresponding plasma samples, with no discordant result. The use of whole blood for pregnancy testing is a convenient, time-saving procedure for dealing with urgent ("stat") orders. It requires less than 10 min to obtain the result after the blood is sampled. We have used this pregnancy test procedure to perform more than 300 whole blood tests since August 1985. Physicians are satisfied with the results which we have reported and the technologists are pleased with the simplicity of the assay procedure. Because the Beta Quik Stat detects only intact hCG and the production of free β-hCG is known to exist under certain pathological conditions, I recommend that the "Beta-Quik-V" kit (Pacific Biotech, Inc.) or β-hCG RIA procedure be used for tests on male patients and on those with cancer.

Pitfalls in the Assay of Acylcarnitine, Hideaki Yamamoto,¹ Makiko Okuyama,² and Shuichi Yamaguchi² (¹ Dept. of Radiology, ² Div. of Neurology (present address: Dept. of Pediatrics, Jikei Univ. Sch. Med., Tokyo, Japan), and ³ Div. of Endocrinol. and Metab., Saitama Children's Medical Center, 2100 Magome, Iwatsuki, Saitama, Japan-339 (address correspondence to S.Y.)

Recently the proportions of acylcarnitines are reported to be changed in several diseases involving mitochondrial dysfunctions such as Reye's syndrome and inborn error of metabolism of organic acids. Because various acylcarnitines present with several different acyl-chain lengths, it is important to evaluate the behavior of such mixtures for study of mitochondrial functions. Methods for the assay of non-acylated carnitine and acylcarnitine are based on a radioenzymatic procedure (I), later modified (2). Acylcarnitine is evaluated by subtracting the value for the non-acylated carnitine from that for total carnitine that was hydrolyzed with alkali before assay, because this method measures only non-acylated carnitine. The calibration is done with non-acylated carnitine as a standard. Unfortunately, however, the degree of hydrolysis of acylcarnitine depends on the acyl-chain length.

We synthesized L-acetyl-, L-propionyl-, and L-caproylcarnitine (3), and assayed non-acylated carnitines principally by the method of McGarry and Foster (2) with slight modifications. Acylcarnitines were dissolved in 0.1 M of 0.1 mol/L HEPES (pH 7.3) and 0.05 M of 0.4 mol/L KOH and allowed to stand for 1 h at 37 °C to hydrolyze acylcarnitines; the pH was then re-adjusted to 7.3 with HCl. Hydrolysis of the synthetic acylcarnitines exceeded 95%. At any concentration of acylcarnitines, the hydrolysis to non-acylated carnitine was greater for longer acyl-chains than for shorter ones (Figure 1). Individual identification of acylcarnitines in

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**Fig. 1.** Standard curves of free carnitine obtained from acetyl- (○), propionyl- (▲), and caproylcarnitine (■) after KOH hydrolysis.