



PKU and MSUD

Phenylketonuria (PKU) and Maple syrup urine disease (MSUD), branched-chain ketoaciduria are genetically transmitted metabolic diseases. PKU results in elevated Phenylalanine levels due to deficient activity of the enzyme Phenylalanine hydroxylase. In the MSUD state, elevated levels of leucine (Leu), isoleucine (Ile), valine (Val), and alloiso-leucine can be detected in blood. The increased concentration of the branched-chain amino acids is caused by an inherited deficient activity of the enzyme branched-chain keto-acid decarboxylase.

Failure to diagnose and treat these conditions in newborns can result in severe mental retardation (PKU) or even death (MSUD).

Dietary restriction of Val, Leu, and Ile for MSUD newborns must begin immediately once the diagnosis is made or suspected. MSUD patients must be monitored on a regular schedule in conjunction with the dietary therapy because even relatively small changes in the intake of the branched-chain amino acids may produce rather large fluctuations of their concentrations in the plasma. In this respect, MSUD is more difficult to control by dietary means than phenylketonuria (PKU), because each of the branched-chain amino acids has to be individually adjusted and monitored.

The Pickering Laboratories PKU/MSUD post-column system is a rapid, automated method for the quantitative analysis of Met, Leu + Ile, Tyr and Phe from whole blood samples. Between-injection time is 13 min while actual time is only 7 min. The Pickering lithium ion-exchange column has a matched set of eluant and regenerant to provide the best possible separation, sensitivity, and reproducibility. The post-column derivatizing reagent is TRIONE[®] Ninhydrin.

This chromatogram (right) is reproduced by
courtesy of Quynh Vantu and Wanda Andrews,
Virginia Newborn Screening Laboratory, Richmond, Virginia.

Screening for Phenylketonuria and Maple Syrup Urine Disease

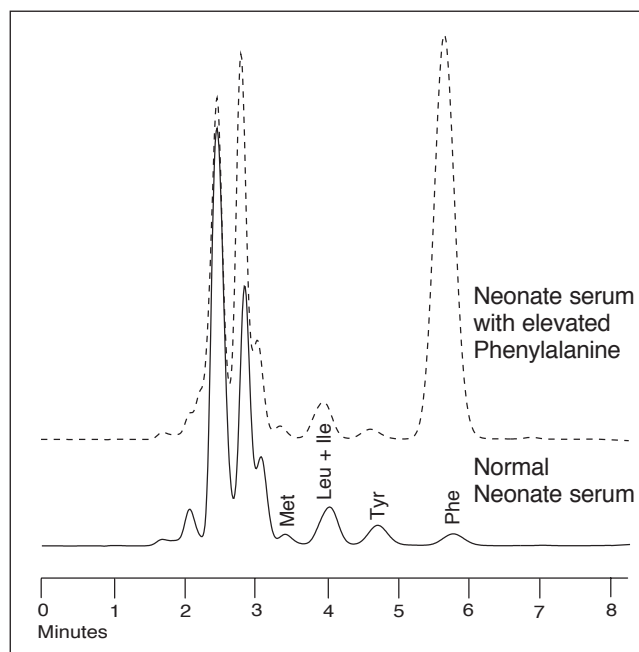
METHOD

Analytical Conditions

COLUMN:	PKU/MSUD column, 4.0 x 50 mm
	Catalog No. 0354050
TEMPERATURE:	65 °C Gradient
FLOW RATE:	0.4 mL/min
MOBILE PHASE:	Eluant (A): Li357, Regenerant (B): RG003
GRADIENT:	0–8.0 min 100% A
	8.1–10.0 min 100% B
	10.1–13.0 min 100% A

Post-column Conditions

POST-COLUMN SYSTEM:	PCX5200
REACTION DWELL:	0.5 mL
TEMPERATURE:	130°C
REAGENT:	TRIONE [®] Ninhydrin
FLOW RATE:	0.3 mL/min
DETECTION:	Visible @ 570 nm



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Pickering Laboratories also developed accelerated PKU-MSUD method for our high-efficiency Li cation-exchange column. This column is widely used for amino analysis of physiological fluids and since the accelerated method utilizes the same set of buffers used for full amino acid screen it can be easily incorporated into the sequence.

Analytical Conditions

COLUMN: High-efficiency Lithium cation-exchange column 4.0x100 mm, Catalog number 0354100A

LITHIUM GUARD COLUMN: 2.0x20 mm, Catalog number 0352020

TEMPERATURE: 38°C

FLOW RATE: 0.35 mL/min

Post-column Conditions

POST-COLUMN SYSTEM: Pinnacle PCX

REACTION DWELL: 0.5 mL

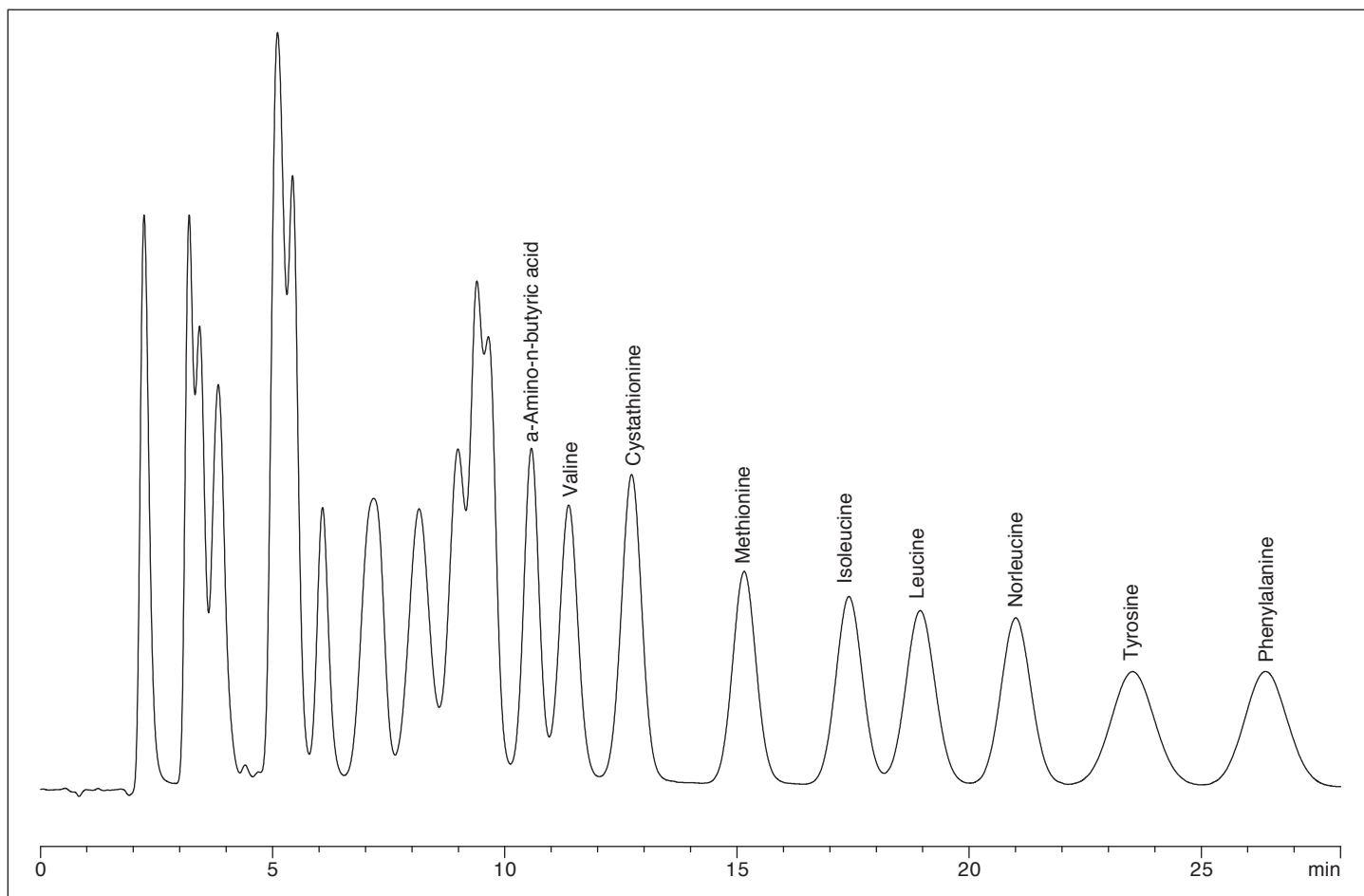
TEMPERATURE: 130°C

REAGENT: TRIONE® Ninhydrin

FLOW RATE: 0.3 mL/min

DETECTION: Visible @ 570 nm

Time [min]	Li275 [%]	Li750 [%]	RG003 [%]
0	86	14	0
25	73	27	0
25.1	0	0	100
30	0	0	100
30.1	86	14	0
42	86	14	0



TRIONE (U.S. Patent No. 4,274,833) is a registered trademark of Pickering Laboratories, Inc.