

Part One

## Approach to Diagnosis



## A

## Simple Tests in Urine and Blood

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Although many of the simple urine tests listed below are no longer being done in large metabolic centers, they are still of importance for small laboratories and in countries where new more sophisticated techniques are not available. Some are nonspecific, but a positive test may direct one to a more specific test indicating the need for further evaluations. To look at a urine sample and smell it should be a routine for a good metabolic laboratory.

**Table A.1.** Color (urine)

Color	Compound	Disorder – Source
Blue	Indican	Blue Diaper Syndrome, Hartnup disorder
Blue/brown	Homogentisic acid	Alcaptonuria
Brown	Met Hb	Myoglobinuria
Red-brown	Hb/Met-Hb	Hemoglobinuria
Red	Erythrocytes	Hematuria
Red	Porphyrins	Porphyria
Red	Pyrazolons	Drugs
Red	Phenolphthalein	Chemicals
Light red	Urates	Physiological, hyperuricosuria
Red	Beets	Nutritional
Yellow	Riboflavin	Vitamins

**Table A.2.** Odor (urine)

Odor	Compound	Disorder – Source
Musty, mousey	Phenylacetic acid	Classical PKU
Maggi – curry	2-Oxoisovaleric acid	Maple syrup urine disease (MSUD)
Maple syrup or burnt sugar	2-Oxoisocaproic acid, 2-Oxo-3-methylvaleric acid	MSUD
Sweaty feet	Isovaleric acid	Isovaleric acidemia 3-Hydroxy-3-methylglutaric aciduria MAD
Cat urine	3-Hydroxyisovaleric acid	3-Methylcrotonylglycinuria Multiple carboxylase deficiency
Cabbage-like	2-Hydroxybutyric acid	Methionine malabsorption Tyrosinemia 1
Rancid butter	2-Oxo-4-methiolbutyric acid	Tyrosinemia 1

**Table A.2** (continued)

Odor	Compound	Disorder – Source
Acid smell	Methylmalonic acid	Methylmalonic acidemia
Sulphurous	Hydrogen sulphide	Cystinuria
Fish market	Trimethylamine	Trimethylaminuria

**Table A.3.** Ferric chloride test (urine). This test is used for detection of oxo-acids, and is useful for detection of PKU. Besides phenylpyruvic acid many other compounds react with ferric chloride by forming different color complexes

Color	Compound	Disorder – Source
Blue-green	Phenylpyruvic acid	Classical PKU
	Imidazolepyruvic acid	Histidinemia
	Catecholamines	Pheochromocytoma
	Xanthurenic acid	Xanthurenic aciduria (B6 def.)
Transient blue-green	Homogentisic acid	Alkaptonuria
Greenish-gray	Branched chain oxoacids	MSUD
Green	p-Hydroxyphenylpyruvic acid	Tyrosinemia types 1 and 2
Gray-black	Melanin	Melanoma
Deep green	Bilirubins	Conjugated hyperbilirubinuria
Cherry red	Acetoacetic acid	Diabetic ketoacidosis, 3-oxothiolase def.
Purple red-brown	2-Oxobutyric acid	Methionine malabsorption
Purple	Ketones	3-Oxothiolase def.
	Salicylates	Drug treatment
Purple or green	Phenothiazines	Drug treatment

**Table A.4.** Reducing substances (urine). This test, also known as Clinitest (Ames Co.), detects various reducing substances in urine, by forming color complexes (green to orange)

Compound	Disorder – Source
Galactose	Galactosemia Galactokinase def. Fanconi-Bickel; Wilson disease
Fructose	Fructose intolerance Essential fructosuria
p-Hydroxyphenylpyruvic acid	Tyrosinemia types 1 and 2
Homogentisic acid	Alcaptonuria
Xylose	Pentosuria
Glucose	Diabetes mellitus Fanconi syndrome
Oxalic acid	Hyperoxaluria
Salicylates	Drug treatment
Uric acid	Hyperuricosuria
Hippuric acid	Treatment with sodium benzoate
Ascorbic acid	Excessive vitamin use

**Table A.5.** Dinitrophenylhydrazine (DNPH) and Acetest (urine). DNPH reacts with  $\alpha$ -oxo acids to form hydrazones which precipitate, and Acetest (Ames Co., Elkhart, IA) reacts with ketones

DNPH	Acetest	Positive compound	Disorder – Source
+	–	Phenylpyruvic acid	Classical PKU
+	–	2-Oxoisovaleric acid	MSUD
+	–	2-Oxoisocaproic acid	
+	–	2-Oxo-3-methylvaleric acid	
(+)	–	Imidazolepyruvic acid	Histidinemia
+	+	Acetone	3-Oxothiolase def.
–	+	2-Methylacetoacetate	Propionic acidemia, ketothiolase deficiency
–	+	Butanone	Methylmalonic acidemia
		Acetoacetate	SCOT-deficiency
+	–	p-Hydroxyphenylpyruvic acid	Liver disease, tyrosinemia types 1 and 2
+	–	2-Oxobutyric acid	Methionine malabsorption
+	+	Pyruvate	Lactic acidosis

**Table A.6.** Nitroprusside test (urine). This test reacts with sulfur containing acids to form pink to purple color complexes

Positive compound	Disorder – Source
Cystine	Cystinuria Hyperargininemia Generalized aminoaciduria Fanconi syndrome
3-Mercaptolactatecysteine-disulfide	3-Mercaptolactatecysteine-disulfiduria
Homocystine	Homocystinuria B12 def. and cobalamin C, D, E, G Methylene tetrahydrofolate reductase def. Cystathioninuria (bacterial)
Glutathione	Glutathionuria
Ketones + high creatinine	Dehydration

**Table A.7.** Routine chemistry in blood (serum or plasma)

Compound	Disorder – Source
Glucose ↓	Tyrosinemia type 1 MSUD 3-Methylcrotonylglycinuria 3-Methylglutaconic aciduria, other types 3-Hydroxy-3-methylglutanyl-CoA synthase def. 3-Hydroxy-3-methylglutanyl-CoA lyase def. HIHA-syndrome (hyperinsulinism + hyperammonemia) Methylmalonic acidemia 2-Ketoglutarate dehydrogenase complex def.

Table A.7 (continued)

Compound	Disorder – Source
Glucose ↓↓	Carnitine uptake defect Carnitine palmitoyltransferase 1 (CPT 1) Acylcarnitine translocase Carnitine palmitoyltransferase 2 (CPT 2) Very long-chain acyl-CoA dehydrogenase (VLCAD) Medium-chain acyl-CoA dehydrogenase (MCAD) Long-chain 3-hydroxyacyl-CoA dehydrogenase (LCHAD) Trifunctional protein Multiple acyl-CoA dehydrogenation (MAD) CDG-syndrome 1 A, 1 B Glycogen storage disease (GSD) types 1, 3, 6, 8, 9, 10 Galactosemia Hereditary fructose intolerance Fructose-1,6-diphosphatase def. Pyruvate carboxylase def. Glycerol kinase def. 3-Hydroxy-3-methylglutaryl-CoA synthase def.
Ammonia ↑↑	Isovaleric acidemia MSUD 3-Methylcrotonylglycinuria 3-Methylglutaconic aciduria, other types Biotinidase def. 3-Oxothiolase def. Methylmalonic acidemia Propionic acidemia L-2-Hydroxyglutaric aciduria Urea cycle disorders Hyperornithinemia – hyperammonemia – homocitrullinuria Hyperinsulinism-hyperammonemia (HIMA) (HHH) syndrome Dibasic aminoaciduria Carnitine uptake defect CPT 1 Acylcarnitine translocase def. MCAD Mitochondrial energy metabolism (MITEM) Severe liver disease, general $\Delta^1$ -pyrroline-5-carboxylate synthase def.
Blood gases (acidosis)	MSUD Isovaleric acidemia 3-Methylcrotonylglycinuria 3-Methylglutaconic aciduria, type 1 3-Methylglutaconic aciduria, other types 3-Hydroxy-3-methylglutaryl-CoA lyase def. Biotinidase def. Holocarboxylase synthetase def. Propionic acidemia 3-Oxothiolase def.

Table A.7 (continued)

Compound	Disorder – Source
Blood gases (acidosis)	3-Hydroxyisobutyric aciduria Methylmalonic acidemia Multiple carboxylase def. Methylmalonic acidemia, cobalamin C and D 2-Ketoglutarate dehydrogenase complex def. Malonyl-CoA decarboxylase def. Glutathione synthetase def. Pyroglutamic aciduria (5-Oxoprolinuria) Citrullinemia Argininosuccinic acidemia Carnitine uptake defect CPT 1 Acylcarnitine translocase def. CPT 2 VLCAD MCAD SCAD MAD Glycogen storage disease types 1 and 3 Galactosemia Hereditary fructose intolerance Fructose-1,6-diphosphatase def. D-Glyceric acidemia Glycerol kinase def. MITEM Pyruvate carboxylase def. Succinyl-CoA: 3-oxoacid-CoA transferase def.
Blood gases (alkalosis)	Urea cycle defects
Lactate ↑	Lipoid adrenal hyperplasia Biotinidase def. Holocarboxylase synthetase def. 3-Hydroxyisobutyric aciduria 2-Ketoglutarate dehydrogenase complex def. Fumarase def. Fructose-1,6-diphosphatase def. L-2-Hydroxyglutaric aciduria Hyperammonemia LCHAD, trifunctional protein GSD 1, 3, 6, 9, 0 Pyruvate carboxylase def. MITEM
Creatinine ↓	Creatine biosynthesis defects
Creatinine ↑	Lysosomal cystine transport def., infantile Lysosomal cystine transport def., adolescent Hyperoxaluria type 1
Urea ↑	Malonyl-CoA decarboxylase def. Lysosomal cystine transport def., infantile Lysosomal cystine transport def., adolescent

Table A.7 (continued)

Compound	Disorder – Source
Urea ↓	Hyperoxaluria type 1 Hyperammonemia Dibasic aminoaciduria Lysinuric protein intolerance
Uric acid ↑	MCAD Hereditary fructose intolerance GSD 1 GSD 7 Hypoxanthine phosphoribosyl transferase (HPRT) Phosphoribosylpyrophosphate synthetase (PRPS) Barth syndrome Fanconi-Bickel syndrome
Uric acid ↓	Molybdenum cofactor def. Purine nucleoside phosphorylase def. (PNP) Xanthine oxidase/dehydrogenase def. (XDH)
Aspartate amino- transferase, alanine aminotransferase (ASAT/ALAT) ↑	Tyrosinemia type 1 3-Methylglutaconic aciduria, other types Mevalonic aciduria 2-Ketoglutarate dehydrogenase complex def. Fumarase def. Malonyl-CoA decarboxylase def. Ornithine transcarbamylase def. Argininosuccinic aciduria Arginase def. Lysinuric protein intolerance Carnitine uptake defect Carnitine translocase def. CD 6 syndrome 1 B CPT 1 CPT 2 VLCAD MCAD SCAD LCHAD, trifunc. prot. Galactosemia Hereditary fructose intolerance Fructose-1,6-diphosphatase def. GSD 3–7 Cholesterol-7 $\alpha$ -hydroxylase def. 3 $\beta$ -Hydroxy- $\Delta^5$ -C <sub>27</sub> -steroid dehydrogenase (3 $\beta$ -HSDH) def. 3-Oxo- $\Delta^4$ -5 $\beta$ -reductase def. (5 $\beta$ -reductase def.) Wilson disease Zellweger syndrome Acid lipase def. (Wolman disease) $\alpha_1$ -Antitrypsin def. CDG-Ie 3-Hydroxy-3methylglutaryl-CoA synthase def.



Table A.7 (continued)

Compound	Disorder – Source
Creatine kinase (CK) ↑	3-Methylglutaconic aciduria type 1 3-Methylglutaconic aciduria, other types Mevalonic aciduria Dolicholphosphate-mannose synthase-1 def. CDG-Ic CPT 2 VLCAD LCHAD, trifunc. prot. GSD 2 GSD 3 GSD 5 Myoadenylate deaminase def.
Alkaline phosphatase (ALP) ↑	3 $\beta$ -Hydroxy- $\Delta^5$ -C <sub>27</sub> -steroid dehydrogenase (3 $\beta$ -HSDH) def. 3-Oxo- $\Delta^4$ -5 $\beta$ -reductase def. (5 $\beta$ -reductase def.)
Lactate dehydrogenase (LDH) ↑	Lysinuric protein intolerance Dibasic aminoaciduria GSD 5
Triglycerides ↓	Abetalipoproteinemia Hypobetalipoproteinemia
Triglycerides ↑	GSD 1 Glycerol kinase def. (pseudo-increase) Lipoprotein lipase def. Apolipoprotein C-11 def. Dysbetalipoproteinemia Hepatic lipase def. Lecithin cholesterol acyltransferase (LCAT) def.
Cholesterol ↓	3-Methylglutaconic aciduria, other types Mevalonic aciduria Abetalipoproteinemia Hypobetalipoproteinemia 3 $\beta$ -Hydroxy- $\Delta^5$ -C <sub>27</sub> -steroid dehydrogenase (3 $\beta$ -HSDH) def. 3-Oxo- $\Delta^4$ -5 $\beta$ -reductase def. (5 $\beta$ -reductase def.) Smith-Lemli-Opitz syndrome Barth syndrome Glucosyltransferase I def. CDG-Ic
Cholesterol ↑	Lipoprotein lipase def. Apolipoprotein C-11 def. Dysbetalipoproteinemia Hepatic lipase def. Hypercholesterolemia Defective apoB-100 LCAT def. Sterol 27-hydroxylase def. (cerebrotendinous xanthomatosis, CTX)
Myoglobin ↑	CPT 2 GSD 2 VLCAD LCHAD

Table A.7 (continued)

Compound	Disorder – Source
Ferritin ↑	Dibasic aminoaciduria Lysinuric protein intolerance
Hematological	Isovaleric acidemia
Neutropenia	3-Methylcrotonylglycinuria 3 Methylglutaconic aciduria type 2 GSD 1B Methylmalonic acidemia Propionic acidemia Carbamyl phosphate synthetase def. Lysinuric protein intolerance Hereditary orotic aciduria Wilson disease
Thrombocytopenia	Isovaleric acidemia Mevalonic aciduria 3-Methylcrotonyl-CoA carboxylase def. Methylmalonic acidemia Propionic acidemia Lysinuric protein intolerance 3-Phosphoglycerate dehydrogenase def.
Anemia (Hb) ↓	Tyrosinemia type 1 Methylmalonic acidemia Pyroglutamic aciduria (5-oxoprolinuria) γ-Glutamylcysteine synthetase def. Cobalamin C and D Lysinuric protein intolerance Hereditary orotic aciduria Pyrimidine-5-nucleotidase def. Familial LCAT def. Wilson disease Acid lipase def. (Wolman disease) 3-Phosphoglycerate dehydrogenase def. Methylentetrahydrofolate reductase def. Methioninsynthase def.
Vacuolated lymphocytes on peripheral smear	Nearly all lysosomal storage diseases
Reticulocytes ↑	γ-Glutamylcysteine synthetase def. γ-Glutamyl transpeptidase def. GSD 2