Current Practice

Set of Simple Side-room Urine Tests for Detection of Inborn Errors of Metabolism

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In 1962 the publication of a set of simple spot tests¹ for the detection of patients with metabolic disorders pioneered the search for further rapid and economical methods of detecting metabolic diseases. The success of the ferric chloride and more recently Guthrie's bacterial inhibition tests for the detection of phenylketonuria is well established, and wide-scale surveys based on these tests have yielded much information regarding the incidence of phenylketonuria in newborn infants and also in the apparently normal population. More recently the use of one-dimensional paper chromatography for amino-acids^{2,3} or other simple chemical tests⁴ in large-scale screening programmes has revealed the existence of many other previously unsuspected inborn errors of metabolism.^{5,6} It is not yet possible to treat all of these disorders. However, increased recognition of rare metabolic disease through such programmes has led to the development of successful treatment in several disorders provided that the disease is recognized in early life before the onset of mental retardation or other systemic manifestations.^{4,7}

The diseases which are known to be caused by inborn errors of metabolism are clinically so diverse⁸ that it may appear confusing to consider them collectively. However, the common factor that justifies grouping them together is that the correct diagnosis can always be established by appropriate biochemical tests even when no clinical evidence exists of the underlying metabolic disorder. The main difficulty in diagnosing these diseases lies in deciding which patients merit the detailed and often costly laboratory investigations necessary to identify the biochemical defect. It is therefore advantageous to develop a programme of screening tests which can be economically applied to large numbers of samples to detect abnormal metabolites, and in this way select the patients who should be further investigated.

This article describes a set of rapid, easy, and inexpensive urine tests which require a minimum of equipment. They can be performed in any surgery or hospital side-room and can provide information on the presence of many abnormal compounds and therefore on the existence of many potentially treatable underlying metabolic diseases.

TABLE I.—Diseases in Which a Specific Smell Has Been Described

Disease	Odour	Compound
Phenylketonuria ..	Mouse-like	Phenylacetic acid
Maple syrup urine disease	Maple syrup.* Sweetish	Branched chain. α -Keto-acids
Methionine malabsorption ⁹	Sour beery smell.† Reminiscent of a brewery	α -Ketobutyric acid
Cystinuria ..	Sulphurous	Hydrogen sulphide
Homocystinuria ..	"Sweaty feet. "Cheesy"	Isovaleric acid ¹²
Isovaleric acidemia ¹⁰ ..	Sweaty feet	Butyric acid. Hexanoic acid
Green acyl dehydrogenase deficiency ¹¹		

* Also has been likened to caramelized sugar, or white tobacco.¹²

† Also has been likened to burned sugar, dried curry, rancid butter, or "fishy."

Smell

Characteristic odours have been reported in several metabolic diseases, and a summary of these is shown in Table I. The odours are often most apparent in the urine, but the smell may be evanescent and difficult to describe and may be obscured by preservatives in the sample.

Reducing Substances

Reagent.—Clinitest (Ames).

Method.—The test is performed according to the manufacturer's instructions. Five drops of urine are added to 10 drops of water and the colour is noted after 15 seconds. A positive result is given by several other compounds besides glucose.

Results.—The compounds most often detected by this test are shown in Table II. The presence of glucose may be con-

TABLE II.—Reducing Substances Which May Produce a Positive Clinitest

Glucose—Diabetes mellitus, renal glycosuria, renal tubular dystrophies, Fanconi's syndrome, cystinosis, Lowe's syndrome, vitamin-D-resistant rickets (Dent type II)
Fructose—Fructosaemia (aldolase deficiency), essential fructosuria
Galactose—Galactosaemia and variants, galactokinase deficiency^{13,14}
Lactose—Lactase deficiency, congenital or acquired
Xylulose—Pentosuria
Homogentisic acid—Alkaptonuria
Phenols—Phenylketonuria, tyrosinosis, tyrosine transaminase deficiency
Drugs—Ascorbic acid, chloral hydrate, sulphonamides, P.A.S., tetracycline, chloramphenicol

firmed by the use of any commercially prepared dipstick containing glucose oxidase. Since these dipsticks depend on a glucose-specific enzyme they will not react with any other compound. Suitable preparations are Clinistix or Testape (Ames). Phenolic acids and sulphonamides will also give false-positive results in the ferric chloride test. More definitive identification of non-glucose-reducing compounds requires chromatographic techniques¹⁵ which are not readily adaptable as side-room procedures.

Ferric Chloride

Reagent.—10% aqueous ferric chloride. The reagent should be stored in a brown bottle in the refrigerator.

Method.—Add two drops of reagent to 1 ml. of urine. Mix and observe the colour immediately and two to three minutes later.

Results.—The colour reaction produced is not specific. A list of diseases and compounds which may give a positive result

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is shown in Table III. The test will detect classical phenylketonuria when the level of plasma phenylalanine exceeds 15 mg./100 ml., but will not differentiate classical phenylketonuria from the milder variants of hyperphenylalaninaemia which do not require rigid dietary therapy yet in which the plasma phenylalanine may reach 20 mg./100 ml. or more.^{13 14} The test will be negative in phenylalanine transaminase deficiency when no phenylpyruvic acid can be formed.¹⁸ In none of the diseases shown in Table III is the test invariably positive. A positive result will occur only when the metabolic disturbance is severe enough to cause excessive production of keto-acids from corresponding amino-acids. It is emphasized that while a positive ferric chloride test may suggest the existence of metabolic disease, confirmation of the diagnosis must be made by more precise analytical procedures. None the less, tests based on this reaction have been used successfully in preliminary mass screening programmes for phenylketonuria, in surveys of mental institutions, and even in such diseases as tuberculosis and in psychiatric clinics to assess whether patients are taking their medication.

TABLE III.—Diseases and Compounds Giving a Reaction with Ferric Chloride and/or Phenistix

	Ferric Chloride	Phenistix
Phenylketonuria	+ Blue-green	+
Tyrosine transaminase deficiency, tyrosinosis, tyrosyluria (p-hydroxyphenylpyruvic acid)	+ Transient blue-green	+
Histidinaemia	+/- Grey/green	+/-
Hyperglycinaemia	+/- Green	+
Maple syrup urine disease (α-ketoisovaleric acid)	+ Blue	+
(α-ketoisocaproic acid)	Yellow	+
(α-ketomethyl valeric acid)	Blue-green	+
Methionine malabsorption (α-ketobutyric acid)	Purple fading to red/brown	+
Lactic acidosis ¹⁶	+/- Grey	+
Pyruvic acidemia ¹⁷	Yellow	Yellow
Pyridoxine disorders, kynureninase deficiency (3-OH-anthranilic acid)	Immediate brown	Yellow
(xanthurenic acid)	Deep green→brown	—
Homogentisic acid	Transient blue-green	—
Acetoacetic acid	Brown/red	—
Conjugated bilirubin	Bluish green	+
Melanin	Grey precipitate→black	+
Salicylates	Purple	Purple
P.A.S.	Purple-brown	“
Isonicotinic acid hydrazide	Grey	“
Phenothiazines	Purple-brown	Purple
Phenols	“	“
Vanillic acid	Red/brown-mauve	“

* Not tested.

Dinitrophenylhydrazine (For Keto-acids)

Reagent.—100 mg. of 2,4-dinitrophenylhydrazine in 100 ml. of 2 N hydrochloric acid. The reagent should be stored in a dark bottle in the refrigerator.

Method.—The reagents should be used at room temperature. Add 10 drops of reagent to 1 ml. of clear urine.

Results.—A yellow or chalky white precipitate forming within 10 minutes indicates a positive reaction. The test indicates the presence of keto-acids in the urine, and a positive result is therefore likely in phenylketonuria (phenylpyruvic acid); maple syrup urine disease (branched chain keto-acids); tyrosinosis and tyrosyluria (p-hydroxyphenylpyruvic acid); histidinaemia (imidazole pyruvic acid); methionine malabsorption (or Oasthouse syndrome) (α-ketobutyric acid). A positive reaction is also given by acetone, which may be anticipated in hyperglycinaemia, isovaleric-acidaemia, and glycogen storage disease types 1, 3, 5, and 6. Definitive identification of dinitrophenylhydrazones can be achieved by chromatographic techniques, which are not easily performed outside a laboratory.

Cyanide-nitroprusside (For Sulphur-containing Amino-acids)

Reagents.—(A) 5% aqueous sodium cyanide. (B) 5% aqueous sodium nitroferricyanide (sodium nitroprusside). (C)

2 N sodium hydroxide. Reagents A and B should be stored in a brown bottle in the refrigerator. Reagent A should be made freshly each month. Both solutions are dangerous and require caution in handling and storage.

Method.—With pH paper alkalize 2.5 ml. of urine with reagent C to pH 6–8. Add 1 ml. of reagent A to the urine. After mixing and standing for 20 minutes add one drop of reagent B.

Results.—A pink-red or beet colour within five minutes indicates an abnormal urine. A false-negative result will occur if the urine is too acidic. The colour reaction is given by compounds which possess a free sulphhydryl group or disulphide bond. Cystine and homocystine are therefore detected by the test, but cystathionine, methionine, and taurine do not react. The test is sensitive enough for even heterozygotes of cystinuria types 2 and 3 to give a positive result.¹⁹

Cetyltrimethylammonium Bromide (For Mucopolysaccharides)

Reagents.—(A) 50 g. of cetyltrimethylammonium bromide is dissolved in 1,000 ml. of 1 M sodium citrate buffer pH 5.75. Warming may be necessary to facilitate solution. (B) 1 M sodium citrate buffer: Dissolve 210 g. of citric acid monohydrate in 800 ml. of water. Add 150 ml. of 20 N sodium hydroxide. Adjust the final pH of the solution to 5.75 at room temperature by the further addition of 20 N sodium hydroxide. Finally the buffer is taken to 1,000 ml. by the addition of water. Both reagents are stable at room temperature.

Method.—Add six drops of the cetyltrimethylammonium bromide reagent to 1 ml. of clear urine at room temperature and mix.

Results.—The appearance of a cloudy or flocculent precipitate within 20 to 30 minutes indicates a positive test. Though all the diseases shown in Table IV are usually associated with

TABLE IV.—Disorders of Mucopolysaccharide Metabolism

Disease	Clinical Features	Abnormal Compound
<i>Probable Primary Disorders</i>		
Hurler's syndrome (autosomal recessive)	Severe classical gargoylism	Chondroitin sulphate B and heparitin sulphate
Hunter's syndrome (sex-linked recessive)	Gargoylism less severe. Cornea is often clear	
San Filippo's syndrome	Much milder clinical manifestations. I.Q. more variable, may be very low	Heparitin sulphate
Scheie's syndrome	Occurs in adults. Stiff joints. Aortic regurgitation. Cloudy cornea. I.Q. may be normal. Coarse facies	Chondroitin sulphate B
Maroteaux-Lamy syndrome	Severe bony changes. Intellect intact. Cloudy cornea	“
Morquio-Brailsford disease	Severe bony changes. Intellect normal. Cloudy cornea	Kerato sulphate. Chondroitin sulphate B
Morquio-Ullrich syndrome	Less severe bone disease. Mental retardation. Cloudy cornea	Kerato sulphate
<i>Other Diseases in which Mucopolysacchariduria has been Found</i>		
Diaphysal aclasia	Multiple exostoses	Chondroitin sulphate A
Arthro-onychodysplasia	“Nail patella syndrome”	Hyaluronic acid
Marfan's syndrome	Arachnodactyly	?
Mastocytosis	“Urticaria pigmentosa”	Chondroitin sulphate B
Rheumatoid arthritis	“	?
Cretinism	“	?
Carcinomatosis	“	?

excess mucopolysaccharides in the urine, only Hurler's, Hunter's, San Filippo's, and Scheie's syndromes consistently show a positive result with the tests.²⁰ The clinical and biochemical abnormalities of the inherited disorders shown in Table IV result from excess tissue storage of mucopolysaccharides. The test is therefore not usually helpful until after early infancy, before which time false-positive and false-negative results are often obtained.

Toluidine Blue Stain (Alternative Test for Mucopolysaccharides)

Reagents.—(A) Toluidine blue 0.1 g., glacial acetic acid 5 ml., 95% ethanol 10 ml., water to 500 ml. (B) 95% ethanol.

Method.—Place two drops of urine on a piece of thick filter paper (Whatman No. 3 or equivalent). Allow to dry, then soak in reagent A for one minute. Dip the paper in reagent B to decolourize the background. Allow the paper to dry and observe for colour.

Results.—When excess mucopolysaccharides are present the dried urine stains a reddish-purple colour. The background colour remains pale blue. Not infrequently a thin purple ring will be seen around the edge of a drop of normal urine. This test is at least as sensitive as the cetyltrimethylammonium bromide test.

Nitrosonaphthol Test (For Tyrosine Metabolites)

Reagents.—(A) 2.63 N nitric acid (1 vol. of concentrated nitric acid in five volumes of water). (B) 2.5 g. of sodium nitrite dissolved in 100 ml. of water. (C) 100 mg. of 1-nitroso-2-naphthol dissolved in 100 ml. of 95% ethanol. All three reagents should be stored in the refrigerator.

Method.—To 1 ml. of reagent A add 1 drop of reagent B and 10 drops of reagent C. Mix. Then add three drops of urine and mix again.

Results.—An orange-red colour within three to five minutes indicates a positive result. The colour is given only by tyrosine and its metabolites, and a positive result may therefore be anticipated in tyrosine transaminase deficiency, tyrosinosis (tyrosinaemia), transient tyrosinosis of the newborn, and the tyrosyluria of scurvy.

Isatin Test (For Proline)

Reagents.—(A) 20 mg. of isatin, 96 ml. of acetone, 4 ml. of glacial acetic acid. The reagent will be stable for three months if kept in the refrigerator. (B) 1 N hydrochloric acid.

Method.—Dip some filter paper (Whatman No. 3 or equivalent) into reagent A. Allow to dry at room temperature. Add one drop of urine and dry at 100° C. for 10 minutes. Dip in reagent B and wash with water.

Results.—A deep blue colour of the urine spot suggests that proline is present in excess. Confirmation of this must be made by chromatographic methods.⁵

Ninhydrin Test (For Excess Amino-acids)

Reagent.—1 g. of ninhydrin dissolved in 500 ml. of 95% ethanol. The reagent should be kept in a dark bottle in the refrigerator.

Method.—Add three drops of urine to 1 ml. of reagent in a clean test-tube and mix. Examine for colour after three to four minutes.

Results.—A blue or purple colour is taken to indicate the presence of excessive amino-aciduria, but a positive result may be caused by an artifact such as concentrated urine or a high ammonia content of the sample. Conversely, elevation of single amino-acids may not be detected by this method if the overall quantity of urinary amino-acids is not increased. For these reasons the test has been discontinued in this laboratory owing to the development of rapid paper chromatographic methods for screening for amino-aciduria.^{5 21} This latter is not a technique easily adapted as a side-room procedure.

Ehrlich's Aldehyde (For Porphobilinogen)

Reagent.—2% *p*-dimethylaminobenzaldehyde in 2 N hydrochloric acid.

Method.—Add 1 ml. of reagent to 5 ml. of fresh urine. Examine for colour after 10 minutes.

Results.—A pink colour is given by urobilinogen and porphobilinogen. These compounds may be distinguished by adding 5 ml. of chloroform to the tube and shaking. The pink colour produced by porphobilinogen is not extracted into the chloroform layer. Indoles in the urine may also give a positive result.

Sulphuric Acid (For Hydrindicuria)

Reagent.—Concentrated sulphuric acid.

Method.—Add 0.5 ml. of reagent to 5 ml. of urine. Observe the colour after 10 minutes.

Results.—A strong red or violet colour is regarded as a positive result for hydrindic acid. Certain drugs, notably phenothiazines, will also cause a positive reaction. This recently described test²² has not been widely employed, and it is not yet known whether the frequency of hydrindicuria is such as to warrant the routine inclusion of this test in a screening programme of this kind.

O-Toluidine Test (For Copper)

Reagent.—O-toluidine 0.1 g., ammonium thiocyanate 0.5 g., acetone 5 ml. *Mixed just before use.*

Method.—Put two drops of urine on thick filter paper (Whatman No. 3 or equivalent). Allow to dry, then add one drop of the freshly made reagent.

Results.—Appearance of a blue colour within 30 seconds suggests that copper is present in excess and that the diagnosis of Kinnier Wilson's disease should be considered. Definitive assay for ceruloplasmin should then be made.

Urine Microscopy

Reagent.—2% aqueous methylene blue.

Method.—The test should be performed on fresh urine samples, otherwise the renal cells may be destroyed. Centrifuge

TABLE V.—*Microscopical Findings Suggestive of the Presence of Metabolic Disease*

Disease	Stain	Results	Disease	Compound	Appearance
<i>Abnormal Cellular Constituents</i>			<i>Abnormal Crystalline Constituents</i>		
Metachromatic leucodystrophy (sulphatide lipoidosis)	2% Aqueous methylene blue	Blue, intracellular* granules within at least 10% of cells	Cystinuria. Cystinosis	Cystine	Crystals have 120° angles and are usually six-sided. Nitroprusside test is positive
The mucopolysaccharidoses	1% Toluidine blue in 1 N acetic acid	Small purplish intracellular granules in at least 10% of cases*	Fanconi's syndrome. liver atrophy	Acute Leucin.	Crystals are circular*. May be confused with acetyl sulphamide but dissolves on warming
Familial nephritis with or without hyperprolinaemia	No stain necessary	Increased number of pus cells and tubular epithelial cells which may contain lipid	Gout, sex-linked neuropathic hyperuricaemia	Uric acid	Various forms*
Fabry's disease	Dark-ground illumination	Refractile lipid deposits in renal epithelial cells	Tyrosinosis and liver necrosis	Tyrosine	Small rosettes*
			Orotic aciduria	Orotic acid	Fine spindles*
			Xanthinuria	Xanthine	Lemon-shaped*

* See Fig.

the urine and decant the supernatant fluid. Add two drops of stain to the sediment. Examine by high-power microscopy after 5 to 10 minutes.

Results.—Metabolic diseases which may be associated with microscopical abnormalities are shown in Table V. The appearance of the pathological materials is shown in the Fig.

Microscopical appearance of urinary constituents which may be associated with metabolic diseases. A, Cystine crystals. B, Leucine crystals. C, Uric acid crystals. D, Tyrosine crystals. E, Orotic acid crystals. F, Xanthine crystals. G, Metachromatic granules in sulphatide lipoidosis. H, Metachromatic granules in Hurler's syndrome.

Cellular Pathology.—With the methylene blue stain, metachromatic granules show blue within the renal epithelial cells. In older urine the cells will have ruptured and the granules may be seen extracellularly. A positive result requires more than 10% of cells to be stained and indicates the diagnosis of sulphatide lipoidosis (metachromatic leucodystrophy). A modification of this stain has been proposed by Lake.²³ A similar test with 1% toluidine blue in 1 N acetic acid is reported to be positive in certain mucopolysaccharidoses. In this case the granules stain purplish and are smaller and finer than those in sulphatide lipoidosis (see Fig.) These granules should be differentiated from toxic granulation of the leucocytes. The presence of other cells should be noted as suggesting the existence of underlying renal disease. An inborn error of proline metabolism has been described in hereditary nephritis which may be associated with deafness or other manifestations of Alport's disease.²⁴

Crystals.—The identification of crystals in urine may be difficult, since several compounds may assume different shapes under varying conditions. A more detailed description of urinary sediments is given by several authors.^{25 26} Uric acid crystals dissolve in an alkaline pH and phosphates dissolve on the addition of a dilute acid. Uric acid crystals occur in normal urine, but large amounts of this compound suggest the advisability of checking the plasma uric acid levels. We have seen one case of familial hyperuricaemic neuropathy diagnosed in infancy by this means.

All of the tests except urine microscopy for cellular abnormalities and Ehrlich's aldehyde for porphobilinogen can be performed on stored urine. Samples may be kept in the refrigerator for a week, but if the tests are to be delayed longer the specimen should be frozen. It is our policy to perform the tests initially on random urine samples. When an abnormal result is found a further sample is obtained for repeat testing. When the second test is abnormal the results are confirmed by more refined techniques on accurately timed urine samples.

Discussion

Over the past few years wide-scale screening programmes^{3 4 27-32} of patients with mental retardation or diseases of unknown origin have demonstrated many new inborn errors of metabolism.^{5 8} Most of these diseases are inherited in a recessive manner. They are usually caused by the absence or inactivity of a specific enzyme required for normal metabolic activity. The biochemical disturbance produced by the missing enzyme may be severe enough to cause serious symptoms in the newborn period or be so minor that no clinical symptoms can be attributed to it. It is now apparent, moreover, that many of these diseases have widely varying clinical manifestations and may occur in patients who appear quite normal at the time of examination. Patients suffering from homocystinuria may vary from the grossly abnormal phenotype of a mentally retarded patient with the picture of Marfan's syndrome to an adult of normal appearance and intelligence. None the less, so far as can be judged by present knowledge both these patients seem to be equally at risk in terms of developing severe visual disturbance through dislocation of the lenses and early death through abnormal clotting mechanisms or a dissecting aneurysm of the aorta.

It has recently been shown that very high plasma levels of phenylalanine may occur in early life and yet produce no clinical signs of phenylketonuria. It seems likely that several different types of phenylketonuria may exist. For the most part it is not known why apparently identical biochemical disturbances should have such widely varying clinical manifestations.

The clinical and biochemical abnormalities of these groups of diseases are not usually detectable until after birth, presumably because, prenatally, accumulation of any abnormal metabolites is cleared through the mother's circulation. In view of the fact that many of these diseases cause severe damage it is important to detect affected patients in early life in order to develop further effective remedies and to study the effect of such therapy on the subsequent course of the disease. It is equally important that methods should be available to detect heterozygotes or asymptomatic homozygotes in order that the natural history of such metabolic diseases can be elucidated and appropriate therapeutic and genetic counselling provided for such patients.

At present it is not practical that all potential patients can be subjected to the detailed biochemical tests necessary to establish a precise diagnosis. This is particularly true since there is no knowledge of the true incidence of many metabolic diseases in the normal population. None the less, by the selection of patients whose symptoms suggest the possibility of underlying metabolic disease, and the use of a set of simple chemical tests, it is often possible to detect patients who require full biochemical evaluation and whose symptoms may be cured by appropriate therapy.

A list of the conditions which appear to be most commonly associated with metabolic abnormalities is shown in Table VI. Currently all patients who exhibit any of the disorders shown in this Table are screened in the laboratory. It is not always

TABLE VI.—Some Conditions in Which a Metabolic Error Should be Suspected

Mental retardation	Many possible causes
Psychiatric illness	" " "
Failure to thrive and dwarfing ..	" " "
Unknown diagnosis	" " "
Unexplained deaths at any age ..	" " "
Relatives of patients with known metabolic disease	" " "
Food intolerance and distaste for protein foods	Fructosaemia, Disorders of the urea cycle. Disaccharidase deficiencies. Isovaleric acidemia
Renal calculi	Cystinuria, Xanthinuria. Disorders of uric acid metabolism
Cataracts and dislocated lenses ..	Diabetes. Homocystinuria. Galactosaemia
Bone diseases of uncertain origin ..	Mucopolysaccharidoses. Fanconi's syndrome
Liver disease of uncertain origin ..	Galactosaemia. "Tyrosinosis." Mucopolysaccharidoses. Glycogen storage diseases
Marfan's syndrome	Homocystinuria
Speech disorders	Histidinaemia

necessary to use all the screening tests to establish a diagnosis provided that the clinical picture of the suspected condition is recognized. For example, in patients with renal calculi or dislocated lenses the nitroprusside-cyanide test alone will provide conclusive proof of the existence of excess cystine or homocystine in the urine.

Apart from the tests for porphobilinogen and for abnormal cells, the samples can be preserved for many days in a refrigerator and will keep for months if frozen. It is therefore possible to perform the tests on samples collected over any period of time, and this leads to a more rapid and efficient system. The reagents used in these tests can be made by any hospital pharmacist and the tests can be run in bulk for about five shillings per sample.

In this area of rapidly expanding knowledge and increasing awareness of the incidence of rare metabolic diseases there are great benefits to be gained through the detection of new patients suffering from known diseases and also the likelihood of discovery of further inborn errors of metabolism.

The tests described above do not pretend to detect all types of metabolic diseases, but they can be relied upon to detect many of the diseases which cause mental retardation or progressive systemic conditions. As the number of treatable diseases increases it is certain that further screening tests will be developed, and it is likely that just as preventive screening for tuberculosis, diabetes, phenylketonuria, and many other diseases is now feasible and rewarding, so will screening for the prophylactic diagnosis of metabolic disease become commonplace. Many other tests can be suitably adopted for screening programmes. Tests based on the principle of the bacterial inhibition test for phenylketonuria have been developed by Guthrie⁷ for 10 other diseases of amino-acid metabolism. The simple one-dimensional descending paper chromatography used in this laboratory^{2, 3, 5, 27} will detect nearly all the known amino-acidopathies. Several different tests are available for the detection of galactosaemia.^{7, 33-36} A set of screening tests for the detection of red cell enzyme defects have been described by Beutler.^{35, 36} Aisen *et al.*³⁷ have described a test for the detection of Kinnier Wilson's disease, but in our hands the test has not been very reliable. The use of dipsticks has been extended to the detection of bacteriuria* and relatively easy chromatographic techniques have been developed for porphyrins,^{38, 39} indoles,⁵ phenolic acids,⁴⁰ and nucleic acid derivatives.⁴¹ The use of the computer⁴² in this field seems to set the seal on the value of this approach to preventive medicine.

Summary

This article describes a group of simple inexpensive tests which can be used to detect the presence of a group of metabolic diseases. The tests can be performed in any hospital side-room, clinic, or surgery with little or no previous experience. Currently, methods for detecting these diseases are not easily available, and therefore, though each disease is rare, collectively they cause an unknown amount of morbidity and mortality.

The use of these tests combined with judicious clinical selection of high-risk patients can provide conclusive proof of the existence of several diseases and can act as a screening method to select patients who merit more precise biochemical investigation in many other instances. Ideally, such tests should be used as part of the routine management of all healthy infants

so that early diagnosis and treatment can be provided before clinical manifestations occur.

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