GENETIC FACTORS IN RELATION TO DRUGS[1,2]

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In this review, recent developments in the area of pharmacogenetics will be emphasized that, according to the author's view, are especially important. References prior to 1964, the time covered by the previous review for this series by Kalow (79), have been excluded except where it seemed essential for clarity. Since the last general review appeared, numerous authors (38, 39, 80–82, 88, 89, 110, 112, 127, 128) have either surveyed the general field of pharmacogenetics or considered certain aspects of the area. Each has emphasized certain disciplines. In the following, consideration will be limited to those areas wherein there is a clear link between genetic factors and drug effects in man. Animal studies have been excluded except when they provide better understanding of studies in man. Also omitted from consideration here is the large area of resistance of microorganisms to the action of drugs, studies of which have provided so much basic information on the mechanisms of antibacterial chemotherapy. Similarly, the resistance of insects to insecticides has also been excluded.

Drug-Induced Hemolysis

The history of hemolysis of erythrocytes by drugs began more than a century ago, and the number of hemolytic agents known today approximately parallels the development of pharmaceutical chemistry. Dausset & Contu (25) listed more than 50 drugs reported to have induced hemolysis of red blood cells under certain circumstances. These conditions include hemolysis in normal erythrocytes, in red blood cells deficient in certain enzymes or cofactors, in the newborn, and in individuals with various hemoglobinopathies.

Although normal individuals may show hemolysis of red blood cells when exposed to a sufficiently high concentration of the hemolytic drugs, we are concerned here with those subjects in whom hemolysis occurs following a dose that ordinarily would have no effect in normal individuals. In sensitive individuals, the response, in numerous instances, results from some hereditary deficiency or abnormality in metabolism of the red blood cell or in hemoglobin structure.

The normal duration of active function and survival of the mature red cell is remarkable since it has no nucleus, mitochondria, endoplastic reticulum,

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Golgi apparatus, or ribosomes (13), and synthesizes no protein or lipid (108). The energy of the cell is derived from ATP production via anerobic glycolysis (Embden-Meyerhof pathway); normally, 8 to 10 per cent of the glucose-6-phosphate (G-6-P) formed by the action of hexokinase on glucose enters the aerobic pentose phosphate pathway, but ultimately the product of this shunt, ribulose-5-phosphate, is converted to members of the glycolytic pathway, glyceraldehyde-3-phosphate and fructose-6-phosphate (Fig. 1).

Several authors (8, 33) have suggested that drugs and chemicals cause acute hemolysis simply by accelerating the normal red cell aging process. If this is true, then a consideration of the factors concerned in aging and death of the red cell would seem important for understanding drug-induced hemolysis. Most known changes leading to death of the erythrocyte reflect both increased oxidative processes (or loss of reducing potential) and energy loss (33). The maintenance of reducing potential has been emphasized in Figure 1, by showing the enzymatic reactions that provide for the continued formation of reduced nicotinamide adenine dinucleotide ($NADH_2$) and reduced nicotinamide adenine dinucleotide phosphate ($NADPH_2$).

Whereas the activities of most of the glycolytic enzymes of red cells decrease linearly with aging, glyceraldehyde-3-phosphate dehydrogenase and glucose-6-phosphate dehydrogenase (G-6-PD) have been reported to decrease exponentially (95). The first reaction generates $NADH_2$ and the second, $NADPH_2$; thus, it would seem that a lowering of the total reducing potential may be especially important in the death of the red cell. NADPH, is required for the maintenance of glutathione in the reduced state, and this tripeptide is apparently essential for maintenance of activity of hexokinase (130, 135), which controls the entry of glucose into the glycolytic cycle. The importance of hexokinase is stressed by the finding that a type of hereditary hemolytic anemia associated with hexokinase deficiency has been reported (155). Similarly, deficiencies in the glycolytic enzymes, pyruvate kinase (16, 153) and triosephosphate isomerase (139, 154), are associated with variable, often severe, hemolysis. Other hemolytic anemias reportedly are associated with adenosine triphosphatase deficiency (62), or with certain glycolytic mutases (14, 97), or with an abnormal lipid composition of the erythrocyte membrane (72). In addition to hexokinase, glyceraldehyde-3-phosphate dehydrogenase also requires reduced glutathione for activity. Such interdependence of reactions makes it extremely difficult to define the primary event in aging. Decreasing glycolysis also implies less available ATP for control of Na^+ and K^+ exchange, with concomitant increased osmotic fragility. ATP levels have been correlated with red cell survival during *in vitro* storage (114). Furthermore, a diminution of $NADH_2$ from lowered glyceraldehyde-3-phosphate dehydrogenase activity would be expected to cause less activity of the $NADH_2$-dependent reduction of methemoglobin by the diaphorase enzyme (71). Some evidence has been presented that the content of methemoglobin is greater and its rate of reduction less in older red

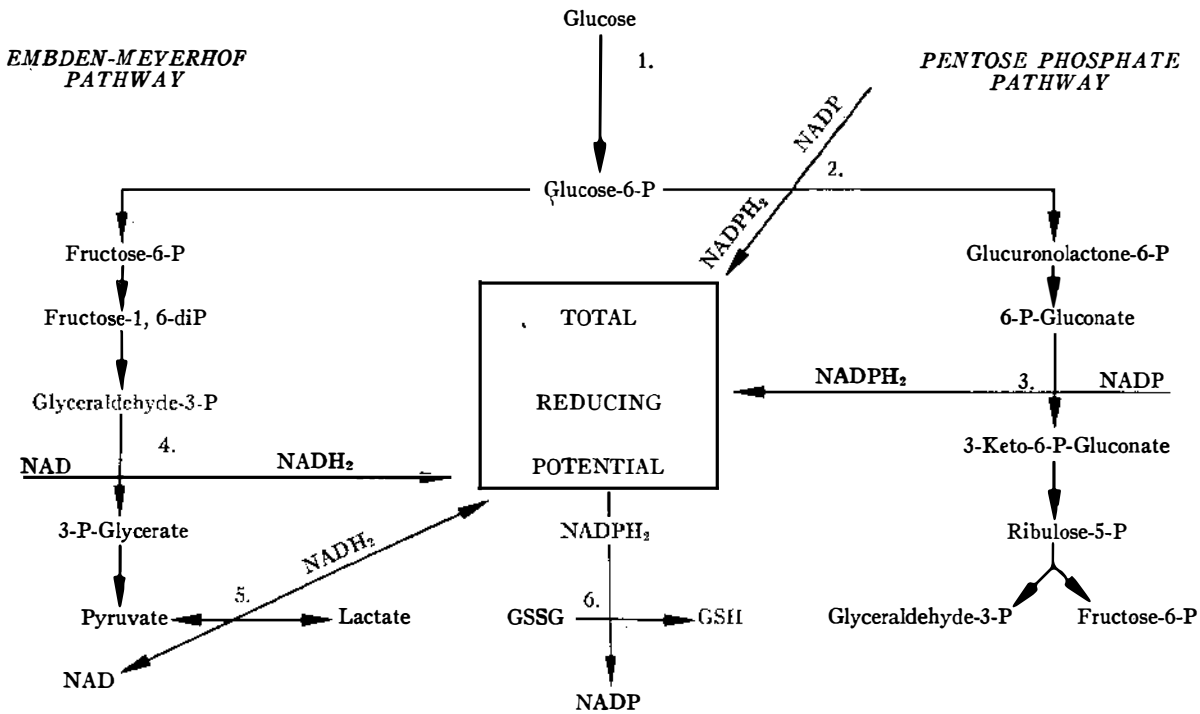

FIG. 1. Broad outline of metabolic pathways in the red cell.

cells (73). The further oxidation of methemoglobin or hemoglobin itself to a number of products, viz., sulfhemoglobin, cholehemoglobin, verdohemoglobin, and their subsequent polymerization may result in the formation of Heinz bodies. That such insoluble erythrocytic inclusions are products of the oxidative breakdown of hemoglobin, that hemoglobin thiol groups are involved in these oxidations, and that reduced glutathione protects hemoglobin from oxidation have been emphasized earlier (3, 74). These authors, as well as Barnes (8), have stressed the parallel between the appearance of Heinz bodies in drug-induced anemias and the normal red cell aging process.

Thus, it seems apparent that the delicate balance of interdependent reactions in the red cell may be easily disturbed by deficiencies of controlling reactions for survival. Such deficiencies have been shown in numerous instances to predispose individuals to acute hemolytic episodes following exposure to certain agents. Recent work in various deficient or abnormal states is discussed below.

G-6-PD deficiency.—It has become apparent that a very extensive polymorphism exists for this enzyme in red cells. A particular enzyme variant may be described in terms of (*a*) quantitation of its enzymatic activity; (*b*) its electrophoretic mobility; (*c*) its Michaelis constants for NADP and G-6-P; (*d*) its relative affinities for alternative substrates such as NAD, 2-deoxyglucose-6-phosphate or galactose-6-phosphate; and (*e*) its relative thermostability.

G-6-PD deficiency is probably the most prevalent and heterogeneous hereditary enzymatic defect of clinical significance. The trait is estimated to be present in more than a hundred million people and its incidence is extremely variable among different population groups, ranging from zero in some to a maximum of 70 per cent of Kurdish Jews in the village of Maoz Zion in the Jerusalem mountains (84, 105, 150). The extreme heterogeneity in different populations is manifest as variations in the severity of the decrease in enzyme activity in red cells, white cells, and other tissues of affected subjects, as differences in the properties of the enzyme and as differences in the susceptibility of affected individuals to hemolysis after exposure to drugs or other agents. G-6-PD deficiencies are apparently not the result of the same mutation in different, affected populations.

The mode of inheritance of G-6-PD deficiency is due to a gene of intermediate dominance, which is linked to the X-chromosome (103, 115). Deficiency is transferred from mother to children and is found most often among males. Females are genetic mosaics in regard to G-6-PD activities. This is believed to result from a permanent, but random, inactivation of one or the other X-chromosome very early in embryonic life (115). The deficiency is manifest in variations in the severity of the decrease in enzyme activity as well as other factors mentioned above. It is not yet known whether such heterogeneity results from different mutations at a single locus on the X-chromosome or mutations at different loci which determine the formation, regulation, or function of the dehydrogenase enzyme.

Normal G-6-PD.—Yoshida (169) reported the isolation and purification

of normal human erythrocyte G-6-PD. The molecular weight was estimated as 240,000, and removal of NADP caused dissociation into inactive subunits. Complete dissociation in 4 *N* guanidine hydrochloride into subunits of molecular weight 43,000 indicated that the enzyme consisted of six similar units. It is highly specific for G-6-P and NADP; of numerous other carbohydrate phosphates examined, only galactose-6-phosphate and 2-deoxyglucose-6-phosphate were substrates. However, these compounds were oxidized at rates only 2 to 3 per cent of that using the natural substrate. NAD was a poor substitute for NADP.

Variants of G-6-PD.—A number of known variants are distinguishable from the normal enzyme, designated B^+, by their electrophoretic mobility (84). However, these variants–faster moving ones called A^+ and Barbieri, and other slower moving ones designated C–are associated with approximately normal or slightly reduced levels of activity of G-6-PD in red cells. Recently, a variant was reported to be electrophoretically identical to normal phenotype B^+ but to have four times the normal enzymatic activity (31). No obvious clinical or hematologic manifestations have been associated with these types of G-6-PD.

Approximately 20 per cent of American Negro males possess an enzyme of high electrophoretic mobility, form A^+, but are not deficient. The cells of another 10 to 15 per cent have a G-6-PD variant, designated A^-, which is electrophoretically indistinguishable from A^+, but have only 10 to 15 per cent of normal activity. Approximately 20 per cent of Nigerian Negro males have this same deficient red cell G-6-PD. This deficiency seems to be expressed principally as a predisposition to drug-induced anemia (primaquine sensitivity) (101). The levels of enzyme in nucleated tissues, such as leukocytes and liver of A^- Negro subjects, were normal or only slightly deficient, and no consistent differences could be found between A^+ and A^- enzymes in enzymatic and immunologic properties. But differences between A^+ and A^- enzymes were reported in regard to heat stability and elution patterns from diethylaminoethyl-Sephadex columns. Further studies (170) using carboxymethyl cellulose-Sephadex columns, also yielded good physical separation of the A^+ and A^- enzymes. These authors demonstrated that the two enzymes have about the same serological characteristics and specific enzymatic activities as does the normal B^+ enzyme. They concluded that the low enzymatic activity of A^- subjects resulted from a reduction in the number of active A^- enzyme molecules in older red cells. Young cells had normal activities. The mechanism of loss of enzyme in the older cells is not clear, but the A^- enzyme was irreversibly inactivated by dialysis in the absence of NADP. Similar treatment of the A^+ enzyme yielded reversible inactivation. The enzyme deficiency in A^- red cells seems to be caused by an increased rate of degradation in older cells rather than by a decreased rate of synthesis by young cells.

A variant (Seattle) very similar to, but different from, the A^- form has been reported in Caucasians (87). It migrates more slowly than the normal B^+ form but has other properties similar to the A^- form.

Another faster-moving electrophoretic variant (Canton) has been found

in G-6-PD-deficient Chinese. This enzyme migrates faster than normal but not quite as fast as the A^- or A^+ enzymes found in Negro populations. Only limited population studies are available, but one report gives an incidence of 5.5 per cent in adult males from Kwangtung Province (21) and deaths resulting from kernicterus following neonatal exposure to moth balls (naphthalene) have been reported (113). Deficient cells have from 4 to 24 per cent of normal activity and exhibit slightly reduced thermostability and Michaelis constants lower than normal for G-6-P and NADP (107). This deficiency confers greater risk than in the case of the A^- enzyme, since affected subjects may exhibit severe neonatal jaundice and are susceptible to hemolytic episodes precipitated by common hemolytic drugs or by exposure to fava beans.

In contrast, individuals with deficient variants that migrate in starch gel electrophoresis, as does the normal B^+ type, seem to be sensitive to a wider range of drugs than do G-6-PD-deficient Negroes (105). This type, found in varying percentages in Jews, Sardinians, and Greeks, has been designated the Mediterranean type. Affected subjects are susceptible to severe hemolytic reactions from exposure to the fava bean. Some G-6-PD-deficient Sardinian and Greek infants develop neonatal hyperbilirubinemia without apparent exposure to any hemolytic agents. G-6-PD activity is very low toward G-6-P but surprisingly high toward 2-deoxyglucose-6-phosphate. The red cell enzyme shows altered pH optima and is labile; its concentration is decreased not only in erythrocytes but also in other cells such as liver, saliva, and skin (86).

Apparently, the most severe expression of G-6-PD deficiency is found in congenital nonspherocytic hemolytic disease (85). Spontaneous disintegration of red cells may occur and the hemolysis may be accelerated by hemolytic drugs. However, this disease state is not uniformly coincident with a single type of G-6-PD-deficient variant. In fact, variants range from one said to be normal, except for an extreme thermolability, to variants showing less than 2 per cent of normal activity. These include the Chicago, Oklahoma, and Eyssen variants (84).

The continually expanding list of apparently distinct variants of G-6-PD with varying properties is indeed puzzling. Single variants are not separable into isozymes. That they are distinct mutant proteins is suggested by the simultaneous starch gel electrophoresis of the Mediterranean, Seattle, and Canton variants. Different migrations were revealed by using both G-6-P and 2-deoxyglucose-6-P as substrates. Each variant was separated but the two activities of each migrated together (84).

Previously used screening tests for detecting G-6-PD-deficiency have been reviewed and a simple one-stage visual test has been devised by Jacob & Jandl (69).

6-Phosphogluconic acid dehydrogenase (6-PGD) deficiency.—In the conversion of 6-phosphogluconate to ribulose-5-phosphate, NADP is an essential cofactor in the oxidation of the substrate to the ketoform before decarboxy-

lation to the pentose phosphate. (See Fig. 1).—Thus, deficiency in 6-PGD may also be expected to result in lowered amounts of $NADPH_2$ in the red cell. Eight phenotypes, including the normal form, are known but only four exhibit activities of 50 per cent or less than normal. Inheritance is autosomal. As yet, no studies have reported any pathological significance of these deficient states (15, 17, 54, 122), and primaquine administration to an individual with half the normal 6-PGD levels did not produce a clinically significant hemolytic response (32). Sensitivity to other potential hemolytic agents is unknown.

Glutathione reductase deficiency.—The importance of this enzyme in maintaining the integrity of the erythrocyte and in protecting it from drug-induced hemolysis may be suggested, indirectly by the observation of primaquine sensitivity in a subject whose G-6-PD activity was normal but who exhibited a deficiency of $NADPH_2$-dependent glutathione reductase (19). Nonspherocytic anemia is associated with this deficiency and severe hemolysis has been precipitated by drugs (19, 45, 96, 158). Löhr & Waller (97) have observed ten homozygotes and five heterozygotes in a family distribution consistent with autosomal recessive inheritance. There appears to be no association between this deficiency and G-6-PD variants except in regard to the final pathologic manifestations and drug sensitivity.

Glutathione deficiency.—That glutathione levels are important in the integrity of the red cell can also be deduced from the biochemical defect of glutathione deficiency resulting from impairment of its synthesis (129). The defect is also associated with a clinical picture of nonspherocytic hemolytic anemia. Affected subjects are sensitive to fava beans, primaquine, and other drugs (98, 118, 157). Deficient red cells have less than 10 per cent of the normal amounts of reduced glutathione, but G-6-PD and glutathione reductase activities of the affected subjects were normal. Using deficient cells, Prins et al. (129) could not demonstrate incorporation of the precursors, glycine-C^{14} or glutamine-C^{14}, into glutathione. Apparently, the defect is due to a low rate of glutathione synthesis in the abnormal cells rather than an increased rate of turnover. Erythrocyte life span was shortened to approximately 30 days instead of the 100 to 120 days of normal red cells. Family studies were consistent with an autosomal recessive pattern of inheritance.

A severe depletion of intracellular glutathione is apparently necessary to have dire consequences on red cell survival. Jacob & Jandl (67) found that glutathione levels of normal red cells could be reduced by 90 per cent without changing metabolism *in vitro* or survival time *in vivo*. However, very small amounts of *p*-mercuribenzoate markedly decreased the viability of the erythrocytes, despite the maintenance of normal intracellular level of reduced glutathione. This reagent can react with sulfhydryl groups, but does not enter the cell. Apparently, sulfhydryl groups of the cell membrane are much more important than intracellular glutathione for maintenance of cellular integrity.

Other defects of erythrocytes.—An increase in the proportion of methemoglobin in the red cells is also of pharmacologic interest; such an increase may be either inherited or induced by a normally harmless drug. Liddell & Lehmann (94) differentiated three types of hereditary methemoglobinemia: the NAD-dependent methemoglobin reductase deficiency (20); the hemoglobin M variants, both of which are characterized by a clinical picture of persistent cyanosis; and a number of other hemoglobin variants characterized by mild hemolytic anemias that are particularly sensitive to drugs. This last class includes a number of hemoglobin variants whose primary structural variations in the globin molecule from the normal are known. Examples are hemoglobin Zurich and hemoglobin Seattle, wherein substitutions for normal amino acids in the β-chains of globin close to the point of attachment of the heme group result in hemoglobins more prone to oxidative denaturation by hemolytic agents.

An entirely different group of hemoglobin variants which can result in drug-induced methemoglobinemia are those in which the protein component of the normal adult hemoglobin molecule, composed of two each of the α- and β-polypeptide chains, is diminished or replaced by tetramers of α- or β-chains. These hereditary disorders are usually referred to as the thalassemias; the α-form is due to diminished or absent synthesis of α-chains, and β-thalassemia is due to decreased synthesis of β-chains. These are the most prevalent and most studied, although others, corresponding to the presence of tetramers of the other known existing types of globin peptide chains (γ, δ, ϵ), could theoretically exist (44, 111, 160). Thalassemia major, the homozygous state, is lethal before birth or shortly thereafter. The clinical presentation of thalassemia minor, the heterozygous state, is a variable, usually mild, symptomatic state characterized by hypochromia and microcytosis of red cells, reduced hemoglobin concentration in the peripheral blood, slightly shortened red cell survival, and, occasionally in severe forms of the disorder, hyperbilirubinemia and splenomegaly. α-Thalassemia patients with hemoglobin H, i.e. tetramers of four β-chains, have a chronic anemia with reticulocytosis and a shortened red cell survival time of about 45 days. The amount of hemoglobin H in this condition varies from a few per cent to as much as 40 per cent of the total hemoglobin. Gabuzda (47) emphasized that hemoglobin H is a complete failure as an oxygen-transporting protein because of its extremely high affinity for oxygen—so high that the gas is never released to the tissues at physiological oxygen tensions. Furthermore, hemoglobin H is much more readily oxidized than the normal hemoglobin. Motulsky (111) and Marks (104) have emphasized the heterogeneity of the thalassemias, and attempts to define the defect at the molecular level have been pursued by Bank & Marks (7).

Role of other factors besides drugs.—It is evident that all the factors that combine to determine blood concentrations for a hemolytic drug may influence its actual effect in a susceptible subject. Some of these factors are the

dosage administered and the absorption, metabolism, and rate of excretion of the drug. Several hemolytic anemias caused by drugs administered in therapeutic doses have been observed in subjects with normal red blood cells but with renal impairment that resulted in abnormally high levels of the drug in the blood (30). Furthermore, a recent report (18) points out a relatively unemphasized aspect of G-6-PD-deficiency anemia—the frequency with which illness in itself acts to precipitate hemolysis. Hemolytic episodes, of the acute type associated with G-6-PD-deficiency and attributable to no other known factor, occurred more frequently in association with illness alone than with drug therapy alone. Most cases were seen during the course of bacterial or viral infection. Of particular note was the occurrence of eight episodes of hemolysis, not attributable to drug ingestion or concurrent infection, in association with diabetic keto-acidosis. Infection-induced anemias have been reported by others (108) to occur three to four times more frequently in G-6-PD-deficient subjects than in nondeficient subjects. In addition, the hemoglobinopathies, including the thalassemias, are particularly prone to the induction of severe crises by the stress of infections or minor illnesses (166).

Dausset & Contu (25) and Huguley, Lea & Butts (65) have recently reviewed the occurrence of hemolytic anemia of allergic origin. Why only certain individuals are susceptible is unknown, but such effects may result from a combination of several factors, one of which may have a genetic basis. Germane to this possibility is the demonstrated genetic control of the anaphylactoid reaction in rats (164).

Mechanism of drug-induced hemolysis.—The exact reaction sequence by which hemolysis is induced by individual drugs in sensitive subjects is not known. However, by analogy with other basic studies, mechanisms can be suggested. Thus, Kiese (83) has investigated in detail the process of methemoglobin formation by aromatic amines in dogs, rats, and other animals. Nitroso derivatives are formed from aniline or substituted aniline derivatives by enzymes present in the liver and other tissues but absent from erythrocytes. These nitroso compounds enter the circulation, penetrate the red cell, and are reduced to hydroxylamino compounds by the $NADPH_2$ derived from the pentose oxidation pathway.

In a "coupled oxidation," the hydroxylamino compound and hemoglobin are oxidized to a nitroso metabolite and methemoglobin, respectively. The nitroso derivative can again be reduced to the hydroxylamino compound via $NADPH_2$, thus producing more compound for reaction in the coupled oxidation above. The initial nitroso derivative then has a catalytic effect (as long as $NADPH_2$ is available), and only small amounts of the drug metabolite are needed to convert considerable amounts of hemoglobin to methemoglobin and cause depletion of available $NADPH_2$. Recently, it has been suggested that this mechanism might explain hemolysis induced by the anti-leprotic drug, diaminodiphenylsulfone (63). The hemolytic effects of this drug in both

normal and G-6-PD-deficient subjects have recently been reinvestigated (29). Whether this mechanism can explain hemolysis by other agents containing aryl amino groups is as yet untested.

In a similar manner, when G-6-PD-deficient cells are exposed to acetylphenylhydrazine, there is a rapid drop in glutathione level and in catalase activity, a transitory increase in oxidized glutathione, oxidation of hemoglobin to methemoglobin, and finally, an appearance of a mixture of denatured products of hemoglobin, which precipitates in the form of Heinz bodies (25). The appearance of these oxidative derivatives of hemoglobin in insoluble form within the red blood cells is a phenomenon common to all drug-induced hemolysis.

Many simultaneously operating reactions are involved in hemolysis, however. As pointed out by Jaffe (71), there are mechanisms for the prevention of oxidation of hemoglobin to methemoglobin, as well as for the reduction of methemoglobin. $NADPH_2$, $NADH_2$, or glutathione can directly protect hemoglobin from oxidative denaturation. In addition, generation of hydrogen peroxide plays a role in drug-induced hemolysis (23) by the oxidation of hemoglobin to methemoglobin. In contrast to the normal red cell, G-6-PD-deficient cells are low in catalase activity; therefore, one would expect a greater susceptibility to oxidation by hydrogen peroxide. Hydrogen peroxide is also decomposed by the enzyme glutathione peroxidase, which catalyzes the reaction of peroxide and glutathione to form water and oxidized glutathione. Glutathione peroxidase activity is dependent upon the generation of adequate amounts of glutathione through the glutathione reductase reaction which requires $NADPH_2$ (68, 71).

Scott, Duncan & Ekstrand (140) compared the rates of reduction of purified methemoglobin by different reducing systems found in normal human erythrocytes. The relative rates were: $NADH_2$-dependent diaphorase 73, ascorbic acid 12, glutathione 9, and $NADPH_2$-dependent diaphorase (methemoglobin reductase) 6. The point of major importance may be the overall decrease in the reducing potential ($NADH_2$ and $NADPH_2$) in deficient cells, coupled with an increased rate of utilization of the limited amounts of $NADPH_2$ in the metabolism of the hemolytic agents themselves. Under the circumstances that produce a severe depletion of reducing potential, vital enzymes or constituents within the red cell and of its membrane may become susceptible to oxidative denaturation. A summary of factors influencing the interconversion of hemoglobin and methemoglobin has been presented in Figure 2.

Drugs and Porphyrias

Undoubtedly, the most serious manifestation of an interaction of a genetic defect and a drug effect is the exacerbation of certain types of porphyria by analgesics, sedatives, and other drugs. From a description of patients by Dean (27), it is apparent that the symptoms of the disease (abdominal pain,

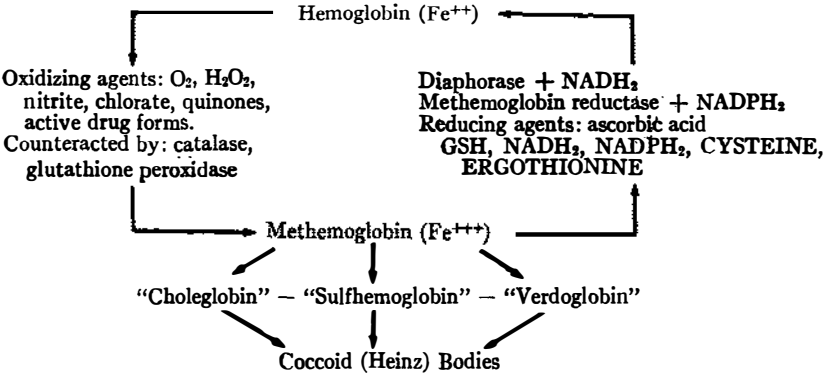

FIG. 2. Factors influencing interconversion of hemoglobin and methemoglobin.

neural disturbances) suggest the use of palliative drug therapy; however, sulfonamides and barbiturates, in many cases, have caused paralysis and death of porphyric individuals.

Numerous reviews are available covering both the types of porphyria known to be determined by hereditary factors and the types of drugs which can cause acute attacks and death (22, 57, 132, 136, 137, 152, 159). Any attempt to completely review this extremely complex field by the author would be inadequate. However, certain points appear important enough to be emphasized here.

In contrast to other metabolic errors, porphyrias are characterized, not by a lack or deficiency or an enzyme, but by increased synthesis of the enzyme, δ-aminolevulinic acid synthetase. This mitochondrial enzyme is the first, and the rate-controlling, step of the porphyrin biosynthetic chain. Increased activity results in excessive formation of δ-aminolevulinic acid, porphobilinogen (the monopyrrole precursor of porphyrins), and the porphyrins, formed as metabolic by-products during the biosynthesis of heme. Normally, the excretion of these materials is very small in relation to the over-all rate of heme synthesis, but in hereditary porphyria excessive amounts of these compounds and their derivatives are found in the excreta. Both erythropoietic and hepatic porphyrias are known, in parallel with the known high activity of the liver and erythroid elements of the bone marrow in heme synthesis.

A common characteristic of the types of drugs known to cause acute attacks in otherwise latent porphyria is the induction of increased synthesis of the δ-aminolevulinic acid synthetase. Numerous animal experiments have suggested this common mechanism. Perhaps the most convincing parallel between animal experiments and the induction of porphyria in man is the epidemic of toxic porphyria in Turkey caused by the consumption of bread

made from wheat treated with the fungicide, hexachlorobenzene. The wheat seed had been provided for planting only. It has been estimated that over 5,000 young children were affected (27). Hexachlorobenzene was strongly implicated as the porphyrinogenic toxin; it was subsequently demonstrated that this compound readily produces toxic porphyria in rats. The frequency of occurrence in the Turkish population contradicted the possibility that the children were genetically predisposed to the toxic effect. Therefore, this occurrence is generally considered as a purely acquired, rather than a genetically determined, form of hepatic porphyria. It appears to represent the human counterpart to the experimental forms of porphyria inducible in animals by various agents. These include, besides hexachlorobenzene, unrelated compounds of three other groups or types: (*a*) dialkyl-substituted acetamides, acetylcarbamides, and barbiturates, when one substituent is an allyl group; (*b*) a collidine derivative, 3,5-diethoxycarbonyl-1, 4-dihydro-2,4,6-trimethylpyridine; and (*c*) the antibiotic, griseofulvin. The work of Granick (57) primarily has shown that these diverse unrelated agents have the common property of inducing the synthesis of increased amounts of δ-aminolevulinic acid synthetase in livers of experimental animals. All except the collidine compound have been implicated in the precipitation of acute attacks in porphyric human subjects. Other drugs reported capable of causing exacerbation of symptoms in various types of human porphyria include sulfonamides (132), chloroquine (132), anticoagulants (12), tranquilizers (24), stilbestrol (92), and oral contraceptives (28).

Granick (56) has devised a simple method for determining whether a given chemical is likely to be porphyrogenic. Good correlation of the results of this method with clinical experience and with the production of porphyria in experimental animals testifies to its validity thus far. Hopefully, widespread use of this test, especially for new analgesics and sedatives, will prevent future iatrogenic attacks of acute porphyria in susceptible subjects.

Pseudocholinesterase Variants

Choline esters are hydrolyzed rapidly in the body by enzymes present in numerous tissues. On the basis of substrate specificity, the enzymes are divided into two classes: true cholinesterase, which hydrolyzes acetylcholine rapidly and other choline esters more slowly; and pseudocholinesterase, which is more active against choline esters containing longer aliphatic or aromatic acid moieties such as butyryl, propionyl, or benzoyl.

Interest in pseudocholinesterase of serum was stimulated in the past by the finding that certain individuals were particularly sensitive to the short-acting muscle relaxant, succinylcholine chloride, a dicholine ester of succinic acid. In the usual patient or subject receiving 30 to 100 mg of this agent for general anesthesia or as an adjunct to electroconvulsive therapy, the resulting muscle paralysis and apnea last for only about two minutes. In sensitive individuals, prolonged effects of the drug are associated with low activities of serum pseudocholinesterase that are a result of genetically controlled

variants of the enzyme. Numerous reviews of this subject have appeared (81, 90, 109, 133, 143), since Kalow (79) last summarized progress in this area.

Three variants from normal serum pseudocholinesterase that result in increased sensitivity to succinylcholine chloride have been clearly defined; the atypical, the fluoride-resistant, and the "silent" types. Of these, the atypical form was the first known and has been most extensively studied. This form is a result of a deficiency of an autosomal gene, and sensitivity is expressed clinically when the subject is homozygous for the abnormal gene. The gene frequency has been determined in several populations, using the per cent inhibition by dibucaine to ascertain the heterozygote. Gene frequencies ranging from 1.0 per cent in Australian aborigines to 8.5 per cent in Czechoslovakians have been found, with other populations exhibiting frequencies within this range (142). Neither the high nor the low frequencies were judged significantly different from those in other populations. These observations have resulted in the view that there is a remarkable similarity in the frequencies of the usual and atypical alleles among different populations. The only divergent opinion from this view has been that of Omoto & Goedde (117), who failed to detect any instance of this atypical type (either heterozygous or homozygous) in over 200 Japanese subjects. In populations in which this type is found, the homozygous state occurs with an approximate frequency of one per 2500 individuals. Tests of the hydrolysis of concentrations of suxamethonium expected *in vivo* during muscle relaxation induced by this drug showed that the usual enzyme was highly active but that the atypical variant exhibited virtually no hydrolytic effect (138).

The fluoride-resistant variant is characterized by high sensitivity to the inhibitor dibucaine and low sensitivity to fluoride, in contrast to the atypical variant which exhibits low sensitivity to dibucaine and high sensitivity to fluoride. Normal pseudocholinesterase is highly sensitive to both dibucaine and fluoride. Although the fluoride-resistant variant has not been studied as extensively, the frequency of occurrence of the heterozygous state resulting from the combination of the normal and fluoride-resistant genes is similar to that of the atypical heterozygote in several populations, including the Japanese (117). Individuals homozygous for the fluoride-resistant variant are rare, but families have been found (58, 91, 165) wherein the homozygote has been detected. These individuals are highly sensitive to suxamethonium. The combination of atypical and fluoride-resistant genes yields a phenotype that is also sensitive to suxamethonium.

An allele of the normal and atypical genes, which determines the complete or near complete absence of pseudocholinesterase activity, has also been discovered. The homozygous state is extremely rare, but highly sensitive to suxamethonium. Heterozygotes combining this "silent" gene with the atypical or fluoride-resistant gene would be expected to be highly sensitive. In studies using starch-gel electrophoresis, Hodgkin et al. (64) suggested the complete absence of the enzyme in serum of homozygotes. Im-

munologic investigations yielded no evidence of a mutant inactive enzyme, nor did serum apparently lacking the enzyme inhibit normal pseudocholinesterase activity. Other workers (75) obtained essentially identical results while studying a Bantu schoolgirl, who was homozygous for the silent gene. In addition, transfusion of this subject with plasma from donors possessing the usual type of pseudocholinesterase resulted in a half-life estimation of the enzyme of approximately 10 days. This value closely paralleled the regeneration rate of normal plasma pseudocholinesterase after the administration of isofluorophate (116). However, in contrast to these studies that suggest an absence of any pseudocholinesterase enzyme, Goedde, Gehring & Hofmann (50, 51), using a micromanometric assay procedure, presented evidence that the activity of the silent-gene sera was 2 to 3 per cent of that of normal serum. In addition, results of immunologic tests suggested that the weak activity is due to a qualitative difference between the silent gene protein and the normal pseudocholinesterase protein.

It is especially interesting that all persons homozygous for this silent variant are normal healthy individuals. Either the presence of an active pseudocholinesterase is not essential for health or, in these subjects, other enzymes serve the normal (unknown) function of the pseudocholinesterase enzyme.

Some assessment of the relative importance of the different variants to increased sensitivity to suxamethonium may be obtained from the data of Simpson & Kalow (143). They examined sera from 104 surgical patients who had exhibited prolonged apnea (>30 minutes) during exposure to suxamethonium. Fifty of these were found to contain only the atypical enzyme, which could have resulted from the homozygous atypical or heterozygous atypical-silent gene combinations; 11 were of the intermediate phenotype resulting from the combination of the usual and atypical genes; two were heterozygous for the atypical and fluoride-resistant characteristics; one was due to the homozygous silent state, and one due to the phenotype from the combination of the usual and fluoride-resistant genes. A surprisingly large number of this succinylcholine chloride-sensitive group had the usual type of enzyme. These 39 sera exhibited a mean of pseudocholinesterase activity characteristic of larger normal random groups, and the distribution was Gaussian. In a similar, more detailed study, Thompson & Whittaker (151) examined the pseudocholinesterase activities in 78 patients sensitive to suxamethonium. Thirty were homozygous for the atypical enzyme; 10 were heterozygous for the combination of the usual: atypical forms; seven and six were heterozygous, combining the atypical and fluoride-resistant mutants and the usual:fluoride-resistant combinations, respectively; and 25 were homozygous for the normal enzyme. The cause of the suxamethonium-sensitivity in subjects possessing the usual enzyme is unknown in this study and that of Simpson & Kalow. However, as pointed out by Lehmann & Liddell in their recent review (90) and by Thompson & Whittaker (151), numerous pathologic conditions can cause a lowering of serum pseudocholin-

esterase activities. Furthermore, others (66) have suggested the occurrence of atypical pseudocholinesterase enzymes exhibiting qualitative characteristics that differ from known inhibition patterns using dibucaine and fluoride.

Although not associated with succinylcholine chloride sensitivity, variants that can be recognized by starch-gel electrophoresis have also been described. Usually, this procedure yields only four distinct pseudocholinesterase bands–the atypical and normal enzymes give this same pattern. In a few sera, a fifth, slower-moving band was found by Robson & Harris (134) and by Ashton & Simpson (6). This component is designated the C_5 fraction. Those subjects exhibiting the C_5 component had mean pseudocholinesterase activities 30 per cent higher than normal. Inheritance was apparently autosomal, but the gene for the C_5 component is at another locus from the pseudocholinesterase genes recognized by dibucaine or fluoride inhibition characteristics (60, 141). Neitlich (116) has also described the occurrence of a slower-moving band by disc electrophoresis, using acrylamide gel. The study of these families showed that this component was probably inherited as an autosomal dominant. The possession of this variant, however, is associated with three times more activity than normal and an increased resistance to the muscle-relaxant action of succinylcholine chloride. Finally, further slow variants, detected by electrophoresis, that are slightly different from the C_5 component of Harris or the slower-moving component of Neitlich, have been reported in African populations (156). These new variants have not been detected in Caucasians; but others (6) have seen variants migrating near C_5 in some members of a Brazilian population. Sensitivity to suxamethonium was not reported.

Polymorphic Acetylation of Drugs

Another well-authenticated instance of genetic polymorphism known to account for the occurrence of drug toxicity in man is related to the inactivation of isoniazid (78). Shortly after the introduction of this compound for the treatment of tuberculosis, it was found that there existed unusually large person-to-person differences in the conversion of the parent drug to inactive metabolites. These differences were stable, reproducible, individual characteristics that were due to metabolic, rather than physiologic, parameters. It was first concluded, on the basis of studies in Caucasians, that rapid inactivation of isoniazid is an autosomal dominant character (43, 78), yielding a bimodal distribution of subjects classifiable as rapid or slow inactivators. However, using slightly different criteria in studies of Caucasian, Negro, and Japanese populations, Dufour, Knight & Harris (35) obtained a trimodal distribution of subjects and confirmed the earlier conclusion (149) that rapid and slow inactivators are homozygous, that intermediate inactivators are heterozygous, and that neither gene is dominant. These two hypotheses have not yet been reconciled, but since much of the work reviewed herein has been performed in Caucasians or tissues derived from this group, the follow-

ing considerations are from the view that two groups of inactivators, rapid and slow, are involved. In these terms, Caucasian and Negro populations contain about equal numbers of rapid and slow inactivators, and Mongoloid populations are predominantly rapid inactivators. Inactivator status seems to make little difference for antituberculous therapy, but without supplemental pyridoxine, slow inactivators more frequently exhibit toxic side effects (peripheral neuropathies) to isoniazid treatment (34, 40).

This area has been extensively reviewed in recent years by Kalow (79), Evans (39), La Du (88), and Porter (128). Reports since Kalow's review (79) are primarily concerned with more detailed studies on the establishment of acetylation as the reaction responsible for the polymorphism and studies of other compounds that may be acetylated polymorphically. Shortly after Evans & White (41), using biopsy specimens, showed that the difference between rapid and slow inactivators was attributable to the activity of an acetyltransferase in the liver, Jenne (76) reported essentially the same results using liver obtained at autopsy from subjects whose inactivation capacities were determined before expiration. The similarity of a number of properties (Michaelis constants, heat inactivation, substrate specificities) of the semipurified enzymes obtained from rapid or slow acetylators suggested that amount of enzyme, rather than specific activity of the enzyme, accounted for the difference between the two types of acetylators (76). Concurrently, another approach was used by Peters, Miller & Brown (126), who studied the excretion of isoniazid and its derivatives in a group of normal subjects following the administration of isoniazid and its metabolites, acetylisoniazid and isonicotinic acid. These authors concluded that acetylation was the primary reaction for the determination of inactivator status, that differences in amounts of isonicotinic acid derivatives excreted were a result of the availability of differing amounts of acetylisoniazid for degradation, that isonicotinic acid and isonicotinyl glycine were the major end products of isoniazid and acetylisoniazid metabolism in man, and that no relation existed between the capacity of the individual to acetylate and to form isonicotinyl glycine from isonicotinic acid. Since derivatives of CoA are involved in both acetylation and conjugation with glycine, it was suggested, but not proved, that availability of CoA was not a factor in different acetylating capacities since rapid or slow acetylators did not show corresponding abilities to conjugate isonicotinic acid with glycine.

Other drugs found to be acetylated polymorphically by the liver acetyltransferase are sulfamethazine and hydralazine (41). In contrast, *p*-aminobenzoic acid (41), sulfanilamide (41), *p*-nitroaniline (76), and *p*-aminosalicylic acid (76) were not acetylated by the liver homogenates. These results *in vitro* could be correlated with other findings that subjects receiving sulfamethazine excreted amounts of free and acetylated sulfamethazine that reflected rapid or slow acetylation (41, 124). However, urinary excretion of sulfanilamide (124) or *p*-aminosalicylic acid (76) and their acetylated derivatives did not provide any distinction between subjects known to be rapid or

slow acetylators of isoniazid. In another study (148), intact subjects of known isoniazid acetylator status exhibited plasma levels of sulfisoxazole after administration, indicating that this sulfonamide was also inactivated (acetylated?) polymorphically; but no correlation could be discerned between inactivation of isoniazid and inactivation of *p*-aminobenzoic acid and *p*-aminosalicylic acid. It is apparent that both polymorphic and monomorphic acetylation can occur in human populations.

Furthermore, in a study of the influence of acetylator phenotype on the effects of treating depression with phenelzine (*β*-phenylethylhydrazine), Evans, Davidson & Pratt (42) found that severe adverse effects after phenelzine were more common among slow acetylators of isoniazid than among rapid acetylators on the same treatment schedule. As in the case of isoniazid studies, only drug toxicity could be correlated to acetylator phenotype; therapeutic response to phenelzine showed no significant difference between slow or rapid acetylators.

Paresthesia observed in about 10 per cent of patients receiving pharmacologic doses of hydralazine may also be based on poor acetylation by susceptible subjects. As in the case of isoniazid-induced neuropathy, this side effect of hydralazine treatment can be eliminated by pyridoxine supplementation (131).

The properties of the human transacetylase enzyme isolated from liver of a rapid acetylator were recently examined in more detail by Weber (161). He found that the purified enzyme had rather broad specificity. Michaelis constants for different substrates (relative to isoniazid as 1.0) were: *p*-aminosalicylic aicd, 6.3; sulfamethazine, 1.5; hydralazine, 0.50; *p*-phenetidine, 0.15; and aniline, 0.041 (162). Jenne (76) had reported earlier that the purified enzyme exhibited tenfold greater affinity for isoniazid than for *p*-aminosalicylic acid, that the lower enzymatic activity in a slow acetylator was not due to presence of an inhibitor, and that, using acetylisoniazid as the substrate, the enzyme did not exhibit deacetylase activity. Transacetylation studies as carried out previously, using semi-purified rat or pigeon liver preparations (11, 70), have not yet been reported employing the human enzyme. Evidence had been obtained previously that a transacetylation reaction can occur between acetylisoniazid and *p*-aminosalicylic acid *in vitro*, using pigeon liver extracts (93), and *in vivo* in the rat (123).

The knowledge that human subjects excrete acetylated metabolites of sulfanilamide, *p*-aminobenzoic and *p*-aminosalicylic acids, even though human liver homogenates *in vitro* did not acetylate these compounds, suggests extrahepatic sites of acetylation. Evans & White (41) and Jenne (76) have presented preliminary evidence that human intestinal mucosa is capable of acetylating isoniazid and other amines, although the work thus far has not defined completely whether polymorphic or monomorphic distributions are predominant. Studies based on use of mammalian tissues have shown that kidney, stomach, duodenum, ileum (61), and reticuloendothelial cells (55) of certain species are capable of acetylating sulfanilamide.

Attempts to find animal species that acetylate isoniazid or other compounds polymorphically have been partly successful. Frymoyer & Jacox (46) observed a clear-cut polymorphism for sulfadiazine acetylation in rabbits. Pedigree studies led to the conclusion that acetylation capacity was inherited autosomally, with rapid acetylation dominant to slow. *p*-Aminosalicylic acid was acetylated to the same extent, i.e. monomorphically, regardless of differing abilities to acetylate sulfadiazine. A bimodal distribution of isoniazid inactivation was not clearly demonstrated however, although a relationship between the relative rates of inactivation of isoniazid and sulfadiazine was demonstrated. Apparently the complexity of the metabolism of isoniazid made it more difficult to discern a bimodality of isoniazid inactivation.

Detailed studies of the rabbit liver supernatant enzyme capable of acetylating isoniazid, sulfadiazine, sulfamerazine, sulfamethazine and sulfanilamide have been reported by Weber & Cohen (163). The characteristics and kinetic properties of the purified preparation of the liver enzyme were reported, and comparison with the enzyme obtained from intestine (jejunum) suggest that, using either sulfamethazine or isoniazid as substrates, liver was 20 to 25 times more active than intestine.

Subhuman primates have also been tested as possible models of man for polymorphic acetylation of isoniazid. Goedde, Schoepf & Fleischmann (53) studied a group of 37 African green monkeys (*Cercopithecus aethiops*). They reported a possible polymorphism of isoniazid inactivation and showed that two animals with extremely high and low levels of the drug in the plasma under the test conditions exhibited activities of liver enzymes that were correspondingly high and low in acetylating isoniazid. In later, more detailed studies (52) using rhesus monkeys (*Macaca mulatta*), a clear inverse relationship between level of isoniazid in the plasma and specific activity of acetylation by liver homogenates was found. However, the distribution of activities in the 49 animals studied was more consistent with a continuous spectrum, rather than with a bi- or trimodality. Peters, Gordon & Brown (125) also found that 17 rhesus monkeys exhibited varying capacities to acetylate isoniazid, as measured by urinary excretion studies. In contrast to observations in human subjects (124), acetylation capacities for isoniazid and sulfamethazine were poorly related in the rhesus monkeys. These authors (125) also investigated isoniazid acetylation in 12 cynomolgus monkeys (*M. cynomolgus*) and 12 mangabey monkeys (*Cercocebus fulliginosus*). The former animals exhibited no clear modality although the latter presented a distribution of acetylation capacities suggesting a bimodality. More extensive studies are obviously required on both the human and animal transacetylase enzymes. In any event, a finding of unimodal distribution of enzyme activities does not preclude the possibility of a genetic basis for enzymatic activity, since Harris (59) found that an over-all unimodal distribution of red cell acid phosphatase activities in man was actually a summation of a series of

separate but overlapping distributions corresponding to each of five qualitatively different phenotypes.

Resistance to Coumarin Anticoagulants

The exact mode of action of the coumarin anticoagulants is not known, but available information (1) suggests interaction of these drugs with enzyme complexes in the liver responsible for the synthesis of the circulatory clotting factors II, VII, IX, and X. The ability of vitamin K to neutralize the effects of these anticoagulant drugs demonstrates the essential role of this agent. None of the metabolites of these drugs thus far identified has been found to have anticoagulant activity in man; therefore, it is assumed that only the parent drugs are active. Consequently, factors that influence inactivation of the drug would be expected to influence the biologic effect since a high degree of correlation has been observed, in the case of warfarin, between the plasma level and the prothrombin response in a group of 24 normal subjects (1).

Genetic influences may play a role in the metabolism of the coumarin anticoagulants, but in the earliest study Motulsky (110) found little evidence to support this concept. Other studies (144) with 29 subjects did not generate any evidence that inactivation capacities were under the influence of hereditary factors in man. In a preliminary communication, Solomon & Schrogie (145) reported that, in rabbits, both the susceptibility to anticoagulant effects of bishydroxycoumarin and the capacity to inactivate the drug are under genetic control.

Nevertheless, O'Reilly et al. discovered a family in some members of which the dose of warfarin required to increase the prothrombin time to a satisfactory therapeutic level was far outside the range required by normal subjects. The result of the study of this family suggested that the abnormal resistance to the drug was under the control of a single autosomal dominant gene or of an X-linked dominant one (119, 121). The propositus showed equal resistance to bishydroxycoumarin, but no aspect of the physiologic disposition of warfarin was different from normal. The rate of disappearance of the clotting factors dependent on vitamin K from the plasma following coumarin anticoagulants was also normal, but the propositus was extremely sensitive to the antidotal action of vitamin K. The probable explanation of the extreme resistance to the coumarin anticoagulants is that the affected members of the family have a genetically controlled abnormal enzyme or receptor site that has either a decreased affinity for the anticoagulants or an increased affinity for vitamin K (120). The resistance is apparently related to both quantitative and qualitative factors since anticoagulant effects can be obtained by sufficiently high doses of the drug in resistant subjects. In addition, the potentiation of anticoagulant action of warfarin by phenylbutazone occurred in a resistant subject as well as in normal subjects (2).

Soulier & Blatrix (146) have also described three patients in whom re-

sistance to coumarin anticoagulants by mechanisms similar to those described above have been found. In addition, reports were made on two patients whose resistance also seemed to be related to increased rate of inactivation of the drugs. No family studies were reported.

Taste Sensitivities for Drugs

One of the earliest known instances of the influence of heredity on drug response is the sensitivity to the bitter, unpleasant taste of phenylthiocarbamide (phenylthiourea, PTC) (78). Early work showed that the essential chemical grouping for differentiation into so-called tasters and non-tasters was the thiourea moiety. Thus, thiouracil and thiopental yield the same taste threshold polymorphism as phenythiocarbamide does. Metabolic degradation is apparently unrelated to taste sensitivity since tasters and non-tasters did not differ in their capacities to degrade methylthiouracil and thiopental (37). Sensitivity has generally been considered to be a dominant autosomal trait, yielding bimodal distributions; the frequency of tasters in different populations is about 50 per cent in Australoids and 90 to 95 per cent in Negroes and Chinese, with frequencies of other populations falling within this range.

However, the recent studies of Lugg (99, 100) in Australian aborigines, Koreans, and Japanese, wherein previously undetected, extremely high acuities of taste for PTC have been found, suggest the existence of a multimodal pattern of distribution of taste thresholds. Such distributions depart from the classical pattern of inheritance usually considered to explain taster :non-taster distributions.

Confirming and extending earlier studies of others, Eaton & Gavan (36) have emphasized that the threshold for tasting PTC is not a neutral trait, since sensitivities may be enhanced by increasing the amount of tyrosine in the saliva. This observation, coupled with the fact that compounds containing the thiourea moiety are goitrogenic (i.e. they block the synthesis of thyroxine in the thyroid gland), suggests an intimate relationship between thyroid function and taster status. Widström & Henschen (167) reported that nontasters of PTC exhibit subnormal protein-bound-iodine values, whereas tasters have high values. In addition, the ability to taste this compound is associated with an increased rate of skeletal maturation (77). It is difficult at this time to separate cause from effect. Nevertheless, many studies in the past have suggested that taster status is important in determining susceptibility to certain types of thyroid disease (78, 128). On the other hand, sensitivity to PTC is also known to be altered by disease since patients with cystic fibrosis showed an abnormal distribution of sensitivities, although their parents and siblings did not (102).

Recently, Goedde & Ohligmacher (48, 49) reported a taste polymorphism for anetholtrithione, 5-(4′-anisyl)-1,2-dithiol-3-thione. This compound does not contain the thiourea group characteristic of PTC and related compounds.

The trimodal distribution found in unrelated subjects suggested that those showing intermediate thresholds were heterozygotes. On the basis of family and twin studies, autosomal inheritance was suggested. Poor correlation between sensitivities to the thione compound and PTC implied an independence of the two polymorphisms. Dawson, West & Kalmus (26) confirmed the existence of the polymorphism for taste sensitivities to the thione derivative, but concluded that it was identical to the PTC polymorphism, as both showed a bimodal distribution in 97 unrelated subjects. Also, there was individual concordance in sensitivities to the two compounds. These authors suggested that the use of 30 per cent ethanol as solvent for the thione compound by Goedde & Ohligmacher may have influenced taste sensitivities. This suggestion may be valid in regard to studies of unrelated subjects, but it is difficult to accept it as an explanation for the discordance between the two taste sensitivities in the twin studies reported (49).

Miscellaneous Observations

Two instances of inherited anatomic features that influence drug effects are open-angle glaucoma and muscular subaortic stenosis. In the first case, corticosteroids and corticosteroid derivatives such as dexamethasone or betamethasone were found to cause abnormal increases of intraocular pressure in persons genetically predisposed to simple chronic (open-angle) glaucoma (5). Becker & Hahn (10), on the basis of two levels of hypertensive response among normal eyes, postulated that clinical open-angle glaucoma was a heritable trait representing the homozygous recessive state, but Armaly (4) detected three levels of response and suggested several alternate genetic models. It is not yet clear which model is correct, but the recent finding (9) of a trimodal distribution of response to betamethasone treatment in siblings of patients with primary, open-angle glaucoma suggests other models than that of a homozygous recessive trait for this type of glaucoma. The importance of these studies is that such drug tests detect hereditary predisposition to glaucoma and thus allow early institution of treatment and, possibly, the prevention of blindness in later years.

In muscular subaortic stenosis (168), there is abnormal hypertrophy of the basal portion of the ventricular septum and adjacent anterior wall of the left ventricle. This feature is apparently inherited as an autosomal dominant characteristic. If digitalis glycosides are administered to such individuals because of a mild degree of cardiac failure, unexpected death may occur, since this drug increases the severity of the muscular obstruction.

Comment

It is apparent that in every case cited here, the genetic defect may be considered as an inborn error in which some adverse manifestation to drugs has been demonstrated. In many of the situations described here, however,

the normal function of the controlling system (e.g. taste sensitivity, plasma pseudocholinesterase, liver transacetylase) has not been clearly defined. The diverse nature and frequency of inborn errors of metabolism (147) suggest that as new drugs are developed, individuals showing unusual sensitivities will be found. Hopefully, a better understanding and wider recognition of past experiences wherein genetic factors have predisposed individuals to the adverse effects of drugs will result in fewer such occurrences with new drugs in the future.

LITERATURE CITED

1. Aggeler, P. M., O'Reilly, R. A., *Thromb. Diath. Haemorrhag.*, Suppl. **21**, 227–56 (1966)
2. Aggeler, P. M., O'Reilly, R. A., Leong, L., Kowitz, P. E., *New Engl. J. Med.*, **276**, 496–501 (1967)
3. Allen, D. W., Jandl, J. H., *J. Clin. Invest.*, **40**, 454–75 (1961)
4. Armaly, M. F., in *Drug Mechanisms in Glaucoma*, 191–229 (Paterson, G., Miller, S. J. H., Paterson, G. D., Eds., J. & A. Churchill, London, 320 pp., 1966)
5. Armaly, M. F., Becker, B., *Federation Proc.*, **24**, 1274–78 (1965)
6. Ashton, G. C., Simpson, N. E., *Am. J. Human Genet.*, **18**, 438–47 (1966)
7. Bank, A., Marks, P. A., *Nature*, **212**, 1198 (1966)
8. Barnes, A., *J. Am. Med. Assoc.*, **198**, 151–56 (1966)
9. Becker, B., Chevrette, L., *Arch. Ophthalmol.*, **76**, 484–87 (1966)
10. Becker, B., Hahn, K. A., *Am. J. Ophthalmol.*, **57**, 543–51 (1964)
11. Bessman, S. P., Lipmann, F., *Arch. Biochem. Biophys.*, **46**, 252–54 (1953)
12. Birchfield, R. I., Cowger, M. L., *Am. J. Diseases Children*, **112**, 561–65 (1966)
13. Bishop, C., in *The Red Blood Cell*, **147**–88 (Bishop, C., Surgenor, D. M., Eds., Academic Press, New York, 566 pp., 1964)
14. Bowdler, A. J., Prankerd, T. A. J., *Acta Haematol.*, **31**, 65–78 (1964)
15. Bowman, J. E., Carson, P. E., Frischer, H., de Garay, A. L., *Nature*, **210**, 811–13 (1966)
16. Bowman, H. S., McKusick, V. A., Dronamraju, K. R., *Am. J. Human Genet.*, **17**, 1–8 (1965)
17. Brewer, G. J., Dern, R. J., *Am. J. Human Genet.*, **16**, 472–76 (1964)
18. Burka, E. R., Weaver, Z., Marks, P. A., *Ann. Internal Med.*, **64**, 817–25 (1966)
19. Carson, P. E., Brewer, G. J., Ickes, C., *J. Lab. Clin. Med.*, **58**, 804 (1961)
20. Cawein, M., Behlen, C. H., Lappat, E. J., Cohn, J. E., *Arch. Internal Med.*, **113**, 578–85 (1964)
21. Chan, T. K., Todd, D., Wong, C. C., *Brit. Med. J.*, **2**, 102 (1964)
22. Chu, T. C., Chu, E. J.-H., *Clin. Chem.*, **13**, 371–87 (1967)
23. Cohen, G., Hochstein, P., *Abstr. Intern. Congr. Biochem., 6th, New York, 1967*, p. 227 (1964)
24. Cowger, M. L., Labbe, R. F., *Lancet*, **1**, 88–89 (1965)
25. Dausset, J., Contu, L., *Ann. Rev. Med.*, **18**, 55–70 (1967)
26. Dawson, W., West, G. B., Kalmus, H., *Ann. Human Genet.*, **30**, 273–76 (1967)
27. Dean, G., *The Porphyrias* (Lippincott, Philadelphia, 118 pp., 1963)
28. Dean, G., *S. African Med. J.*, **39**, 278–80 (1965)
29. Degowin, R. L., Eppes, R. B., Powell, R. D., Carson, P. E., *Bull. World Health Organ.*, **35**, 165–79 (1966)
30. De Leeuw, N. K. M., Shapiro, L., Lowenstein, L., *Ann. Internal Med.*, **58**, 592–607 (1963)
31. Dern, R. J., *J. Lab. Clin. Med.*, **68**, 560–65 (1966)
32. Dern, R. J., Brewer, G. J., Tashian, R. E., Shows, T. B., *J. Lab. Clin. Med.*, **67**, 255–64 (1966)
33. Desforges, J. F., *New Engl. J. Med.*, **273**, 1310–21 (1965)
34. Devadatta, S., Gangadharam, P. R. J., Andrews, R. H., Fox, W., Ramakrishnan, C. V., Selkon, J. B., Velu, S., *Bull. World Health Organ.*, **23**, 587–98 (1960)
35. Dufour, A. P., Knight, R. A., Harris, H. W., *Science*, **145**, 391 (1964)
36. Eaton, J. W., Gavan, J. A., *Am. J. Phys. Anthropol.*, **23**, 381–88 (1965)
37. Evans, D. A. P., *Am. J. Med.*, **34**, 639–62 (1963)
38. Evans, D. A. P., *J. Chronic Diseases*, **18**, 59–76 (1965)
39. Evans, D. A. P., *Ann. N. Y. Acad. Sci.*, **123**, 178–87 (1965)
40. Evans, D. A. P., Clarke, C. A., *Brit. Med. Bull.*, **17**, 234–40 (1961)
41. Evans, D. A. P., White, T. A., *J. Lab. Clin. Med.*, **63**, 394–403 (1964)
42. Evans, D. A. P., Davidson, K., Pratt, R. T. C., *Clin. Pharmacol. Therap.*, **6**, 430–35 (1965)
43. Evans, D. A. P., Manley, K. A., McKusick, V. A., *Brit. Med. J.*, **2**, 485–91 (1960)
44. Fink, H., *Ann. N. Y. Acad. Sci.*, **119**, 369–850 (1964)
45. Fornaini, G., Bianchini, E., Leoncini, G., Fantoni, A., *Brit. J. Haematol.*, **10**, 23–35 (1964)
46. Frymoyer, J. W., Jacox, R. F., *J. Lab. Clin. Med.*, **62**, 891–904, 905–09 (1963)
47. Gabuzda, T., *Blood*, **27**, 568–79 (1966)
48. Goedde, H. W., Ohligmacher, H., *Humangenetik*, **1**, 423–36 (1965)

49. Goedde, H. W., Ohligmacher, H., *Acta Genet. Statist. Med.*, **16**, 350–54 (1966)
50. Goedde, H. W., Gehring, D., Hofmann, R. A., *Biochim. Biophys. Acta*, **107**, 391–93 (1965)
51. Goedde, H. W., Gehring, D., Hofmann, R. A., *Humangenetik*, **1**, 607–20 (1965)
52. Goedde, H. W., Schloot, W., Valesky, A. *Arzneimittel-Forsch.*, **16**, 1030–34 (1966)
53. Goedde, H. W., Schoepf, E., Fleischmann, D., *Biochem. Pharmacol.*, **13**, 603–08 (1964)
54. Gordon, H., Keraan, M. M., Vooijis, M., *Nature*, **214**, 466 (1967)
55. Govier, W. C., *J. Pharmacol. Exptl. Therap.*, **150**, 305–08 (1965)
56. Granick, S., *J. Am. Med. Assoc.*, **190**, 475 (1964)
57. Granick, S., *Ann. N. Y. Acad. Sci.*, **123**, 188–97 (1965)
58. Griffiths, P. D., Davies, D., Lehmann, H., *Brit. Med. J.*, **2**, 215–16 (1966)
59. Harris, H., *Cancer Res.*, **26**, 2054–63 (1966)
60. Harris, H., Robson, E. B., Glen-Bott, A. M., Thornton, J. A., *Nature*, **200**, 1185–87 (1963)
61. Hartiala, K. J. W., Terho, T., *Nature*, **205**, 809–10 (1965)
62. Harvald, B., Hanel, K. H., Squires, R., Trap-Hensen, J., *Lancet*, **2**, 18 (1964)
63. Hjelm, M., de Verdier, C.-H., *Biochem. Pharmacol.*, **14**, 1119–28 (1965)
64. Hodgkin, W. E., Giblett, E. R., Levine, H., Bauer, W., Motulsky, A. G., *J. Clin. Invest.*, 44, 486–93. (1965)
65. Huguley, C. M., Jr., Lea, J. W., Jr., Butts, J. A., *Progr. Hematol.*, **5**, 105–36 (1966)
66. Irwin, R. L., Hein, M. M., *Biochem. Pharmacol.*, **15**, 145–54 (1966)
67. Jacob, H. S., Jandl, J. H., *J. Clin. Invest.*, **41**, 779–92, 1514–23 (1962)
68. Jacob, H. S., Jandl, J. H., *J. Biol. Chem.*, **241**, 4243–50 (1966)
69. Jacob, H. S., Jandl, J. H., *New Engl. J. Med.*, **274**, 1162–67 (1966)
70. Jacobson, K. B., *J. Biol. Chem.*, **236**, 343–48 (1961)
71. Jaffe, E. R., in *The Red Blood Cell*, 397–422 (Bishop, C., Surgenor, D. M., Eds., Academic Press, New York, 566 pp., 1964)
72. Jaffe, E. R., Gottfried, E. L., Bradley, T. B., Jr., *J. Clin. Invest.*, **45**, 1027 (1966)
73. Jalavisto, E., Solantera, L., *Acta Physiol. Scand.*, **46**, 273–83 (1959)
74. Jandl, J. H., Engle, L. K., Allen, D. W., *J. Clin. Invest.*, **39**, 1818–36 (1960)
75. Jenkins, T., Balinsky, D., Patient, D. W., *Science*, **156**, 1748–50 (1967)
76. Jenne, J. W., *J. Clin. Invest.*, **44**, 1992–2002 (1965)
77. Johnston, F. E., Hertzog, K. P., Malina, R. M., *Am. J. Phys. Anthropol.*, **24**, 253–55 (1966)
78. Kalow, W., *Pharmacogenetics: Heredity and Response to Drugs*, 93–104 (Saunders, Philadelphia, 231 pp., 1962)
79. Kalow, W., *Ann. Rev. Pharmacol.*, **5**, 9–26 (1965)
80. Kalow, W., in *Drugs and Enzymes*, 245–55 (Brodie, B. B., Gillette, J. R., Capek, R., Eds., Macmillan, New York, 504 pp., 1965)
81. Kalow, W., *Federation Proc.*, **24**, 1259–65 (1965)
82. Kalow, W., *Appl. Therap.*, **8**, 44–47 (1966)
83. Kiese, M., *Ann. N. Y. Acad. Sci.*, **123**, 141–55 (1965)
84. Kirkman, H. N., McCurdy, P. R., Naiman, J. L., *Cold Spring Harbor Symp. Quant. Biol.*, **29**, 391–98 (1964)
85. Kirkman, H. N., Rosenthal, I. M., Simon, E. R., Carson, P. E., Brinson, A. G., *J. Lab. Clin. Med.*, **63**, 715–25 (1964)
86. Kirkman, H. N., Schettini, F., Pickard, B. M., *J. Lab. Clin. Med.*, **63**, 726–35 (1964)
87. Kirkman, H. N., Simon, E. R., Pickard, B. M., *J. Lab. Clin. Med.*, **66**, 834–40 (1965)
88. La Du, B. N., *Toxicol. Appl. Pharmacol.*, **7**, 27–38 (1965)
89. La Du, B. N., *Federation Proc.*, **24**, 1287–92 (1965)
90. Lehmann, H., Liddell, J., in *The Metabolic Basis of Inherited Disease*, 2nd Ed., 1356–69 (Stanbury, J. B., Wyngaarden, J. B., and Fredrickson, D. S., Eds., McGraw-Hill, New York, 1434 pp., 1966)
91. Lehmann, H., Liddell, J., Blackwell, B., O'Connor, D. C., Daws, A. V., *Brit. Med. J.*, **1**, 1116–18 (1963)
92. Levere, R. D., *Blood*, **28**, 569–72 (1966)
93. Levy, D. A., Spicer, W. S., Bachur, N. R., Bessman, S. P., *Am. Rev. Respirat. Diseases*, **78**, 839 (1959)
94. Liddell, J., Lehmann, H., *Ann. N. Y. Acad. Sci.*, **123**, 207–11 (1965)

95. Löhr, G. W., Waller, H. D., *Klin. Wochschr.*, **37**, 833–43 (1959)
96. Löhr, G. W., Waller, H. D., *Med. Klin. (Munich)*, **57**, 1521–25 (1962)
97. Löhr, G. W., Waller, H. D., *Folia Haematol.*, **8**, 377–97 (1963)
98. Löhr, G. W., Baum, P., Kamm, G., *Med. Klin. (Munich)*, **58**, 2111–20 (1963)
99. Lugg, J. W. H., *Ann. Human Genet.*, **29**, 217–30 (1966)
100. Lugg, J. W. H., *Nature*, **212**, 841–42 (1966)
101. Luzzatto, L., Allan, N. E., *Biochem. Biophys. Res. Commun.*, **21**, 547–54 (1965)
102. Manlapas, F. C., Stein, A. A., Pagliara, A. S., Apicelli, A. A., Porter, I. H., Patterson, P. R., *J. Pediat.*, **66**, 8–11 (1965)
103. Marks, P. A., in *The Red Blood Cell*, 211–41 (Bishop, C., Surgenor, D. M., Eds., Academic Press, New York, 566 pp., 1964)
104. Marks, P. A., *New Engl. J. Med.*, **275**, 1363–69 (1966)
105. Marks, P. A., Banks, J., *Ann. N. Y. Acad. Sci.*, **123**, 198–206 (1965)
106. Marks, P. A., Gellhorn, A., Kidson, C., *J. Biol. Chem.*, **235**, 2579–83 (1960)
107. McCurdy, P. R., Kirkman, H. N., Naiman, J. L., Jim, R. T. S., Pickard, B. M., *J. Lab. Clin. Med.*, **67**, 374–85 (1966)
108. Mengel, C. E., Metz, E., Yancey, W. S., *Arch. Internal Med.*, **119**, 287–90 (1967)
109. Mone, J. G., Mathie, W. E., *Anaesthesia*, **22**, 55–68 (1967)
110. Motulsky, A. G., *Progr. Med. Genet.*, **3**, 49–74 (1964)
111. Motulsky, A. G., *Cold Spring Harbor Symp. Quant. Biol.*, **29**, 399–413 (1964)
112. Motulsky, A. G., *Ann. N. Y. Acad. Sci.*, **123**, 167–77 (1965)
113. Naiman, J. L., Kosoy, M. H., *Can. Med. Assoc. J.*, **91**, 1243–49 (1964)
114. Nakao, K., Wada, T., Kămiyama, T., Nakao, M., Nagano, K., *Nature*, **194**, 877–78 (1962)
115. Nance, W. E., *Cold Spring Harbor Symp. Quant. Biol.*, **29**, 415–25 (1964)
116. Neitlich, H. W., *J. Clin. Invest.*, **45**, 380–87 (1966)
117. Omoto, K., Goedde, H. W., *Nature*, **205**, 726 (1965)
118. Oort, M., Loos, J. A., Prins, H. K., *Vox Sanguinis*, **6**, 370–73 (1961)
119. O'Reilly, R. A., *New Engl. J. Med.*, **272**, 108 (1965)
120. O'Reilly, R. A., Aggeler, P. M., *Federation Proc.*, **24**, 1266–73 (1965)
121. O'Reilly, R. A., Aggeler, P. M., Hoag, M. S., Leong, L. S., Kropatkin, M. L., *New Engl. J. Med.*, **271**, 809–15 (1964)
122. Parr, C. W., *Nature*, **210**, 487–89 (1966)
123. Peters, J. H., Gill, M., Hayes, V. E., *Arch. Intern. Pharmacodyn.*, **159**, 340–52 (1966)
124. Peters, J. H., Gordon, G. R., Brown, P., *Life Sci.*, **4**, 99–107 (1965)
125. Peters, J. H., Gordon, G. R., Brown, P., *Proc. Soc. Exptl. Biol. Med.*, **120**, 575–79 (1965)
126. Peters, J. H., Miller, K. S., Brown, P., *J. Pharmacol. Exptl. Therap.*, **150**, 298–304 (1965)
127. Porter, I. H., *Toxicol. Appl. Pharmacol.*, **6**, 499–511 (1964)
128. Porter, I. H., *Diseases Nervous System*, **27**, 25–36 (1966)
129. Prins, H. K., Oort, M., Loos, J. A., Zurcher, C., Beckers, T., *Blood*, **27**, 145–66 (1966)
130. Rapoport, S., Scheuch, D., in *Drugs and Enzymes*, 393–98 (Brodie, B. B., Gillette, J. R., Capek, R., Eds., Macmillan Co., New York, 504 pp., 1965)
131. Raskin, N. H., Fishman, R. A., *New Engl. J. Med.*, **273**, 1182–85 (1965)
132. Rimington, C., *Proc. Roy. Soc. Med.*, **57**, 511–14 (1964)
133. Robertson, G. S., *Brit. J. Anaesthesia*, **38**, 355–60 (1966)
134. Robson, E. B., Harris, H., *Ann. Human Genet.*, **29**, 403–8 (1966)
135. Scheuch, D., Kahrig, C., Ockel, E., Wagenknecht, C., Rapoport, S. M., *Nature*, **190**, 631–32 (1961)
136. Schmid, R., in *Drugs and Enzymes*, 405–18 (Brodie, B. B., Gillette, J. R., Capek, R., Eds., Macmillan, New York, 504 pp., 1965)
137. Schmid, R., in *The Metabolic Basis of Inherited Disease*, 2nd Ed., 813–70 (Stanbury, J. B., Wyngaarden, J. B., Fredrickson, D. S., Eds., McGraw-Hill, New York, 1434 pp., 1966)
138. Schmidinger, S., Held, K. R., Goedde, H. W., *Humangenetik*, **2**, 221–24 (1966)
139. Schneider, A. S., Valentine, W. N., Hattori, M., Heins, H. L., Jr., *New Engl. J. Med.*, **272**, 229–35 (1965)
140. Scott, E. M., Duncan, I. W., Ek-

strand, V., *Federation Proc.*, **22**, 467 (1963)
141. Simpson, N. E., *Am. J. Human Genet.*, **18**, 243–52 (1966)
142. Simpson, N. E., Kalow, W., *Am. J. Human Genet.*, **17**, 156–62 (1965)
143. Simpson, N. E., Kalow, W., *Ann. N. Y. Acad. Sci.*, **134**, 864–72 (1966)
144. Solomon, H. M., Schrogie, J. J., *Clin. Pharmacol. Therap.*, **8**, 65–69 (1967)
145. Solomon, H. M., Schrogie, J. J., *The Pharmacologist*, 9, 78 (1967)
146. Soulier, J. P., Blatrix, Ch., *Thromb. Diath. Haemorrhag.*, Suppl. 21, 257–67 (1966)
147. Stanbury, J. B., Wyngaarden, J. B., Fredrickson, D. S., Eds., *The Metabolic Basis of Inherited Disease*, 2nd Ed. (McGraw-Hill, New York, 1434 pp., 1966)
148. Sunahara, S., *Proc. Intern. Tuberculosis Conf., 16th, Toronto, 1961*, **2**, 513–40 (Excerpta Medica Foundation, New York, 1961)
149. Sunahara, S., Urano, M., Ogawa, M., *Science*, **134**, 1530 (1961)
150. Szeinberg, A., in *The Genetics of Migrant and Isolate Populations*, 69–72 (Goldschmidt, E., Ed., Williams & Wilkins, Baltimore, 369 pp., 1963)
151. Thompson, J. C., Whittaker, M., *Acta Genet. Statist. Med.*, **16**, 209–22 (1966)
152. Tschudy, D. P., *J. Am. Med. Assoc.*, **191**, 718–30 (1965)
153. Valentine, W. N., Tanaka, K. R., in *The Metabolic Basis of Inherited Disease*, 2nd Ed., 1051–59 (Stanbury, J. B., Wyngaarden, J. B., Fredrickson, D. S., Eds. McGraw-Hill, New York, 1434 pp., 1966)
154. Valentine, W. N., Schneider, A. S., Baughan, M. A., Paglia, D. E., Heins, H. L., Jr., *Am. J. Med.*, **41**, 27–41 (1966)
155. Valentine, W. N., Oski, F. A., Paglia, D. E., Baughan, M. A., Schneider, A. S., Naiman, J. L., *New Engl. J. Med.*, **276**, 1–11 (1967)
156. Van Ros, G., Druet, R., *Nature*, **212**, 543–44 (1966)
157. Waller, H. D., Gerok, W., *Klin. Wochschr.*, **42**, 948–54 (1964)
158. Waller, H. D., Löhr, G. W., Zysno, E., Gerok, W., Voss, D., Strauss, G., *Klin. Wochschr.*, **43**, 413–26 (1965)
159. Watson, C. J., *J. Am. Med. Assoc.*, **197**, 1074–80 (1966)
160. Weatherall, D. J., *The Thalassaemia Syndromes* (Blackwell, Oxford, 272 pp., 1965)
161. Weber, W. W., *Federation Proc.*, **26**, 683 (1967)
162. Weber, W. W. (Personal communication, 1967)
163. Weber, W. W., Cohen, S. N., *Mol. Pharmacol.*, **3**, 266–73 (1967)
164. West, G. B., Harris, J. M., *Ann. N. Y. Acad. Sci.*, **118**, 439–52 (1964)
165. Whittaker, M., *Acta Genet. Statist. Med.*, **14**, 281–85 (1964)
166. *World Health Organ., Tech. Rept. Ser.*, **338**, 40 pp. (1966)
167. Widström, G., Henschen, A., *Scand. J. Clin. Lab. Invest.*, **15**, Suppl. 69, 257–61 (1963)
168. Wigle, E. D., *Federation Proc.*, **24**, 1279–86 (1965)
169. Yoshida, A., *J. Biol. Chem.*, **241**, 4966–76 (1966)
170. Yoshida, A., Stamatoyannopoulos, G., Motulsky, A. G., *Science*, **155**, 97–99 (1967)

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