Inherited Metabolic Diseases of the Nervous System

The pathogenesis and strategies for the control of heritable neurological disorders are examined.

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Reasonably good evidence exists that inborn enzymatic defects are the underlying causes of nearly 150 metabolic disorders. Central and peripheral nervous system damage occurs in many of these situations. That still other heritable disorders will be discovered can be predicted with reasonable confidence; some of these possibilities are cited in this article. The known inherited neurological conditions, enzymatic defects, diagnostic procedures, detection of heterozygotes, or monitoring of pregnancies at risk have been summarized and tabulated in reviews (*1*) and textbooks (*2*). In this article I try to develop systematic approaches to understanding the underlying pathological physiology and chemistry by analyzing representative examples of the various types of disorders. This approach seems useful since we now appear to be between the era of rapid discoveries of metabolic defects and the development of effective therapy for them. Elucidation of subtle biochemical relationships should be helpful for devising rational bases for therapeutic attempts. The complexities and difficulties of such an undertaking have recently been discussed (*3*).

Disorders of Amino Acid Metabolism

It seems reasonable to begin by considering abnormalities of amino acid metabolism since one of these, phenylketonuria (PKU), is the commonest heritable metabolic disorder in which the central nervous system (CNS) is susceptible to damage. The incidence of the condition is approximately one in 11,000 births (*4*). Some of the original, correct conceptualizations of metabolic abnormalities were made in the study of PKU (*5*), which is the first disorder of amino acid metabolism where the specific enzymatic defect was established (Fig. 1) (*6*), and which is one of a small group of such conditions that is benefited by regulation of the diet (*7*).

Many theories have been advanced and numerous experiments have been conducted to provide insight into the basis of the cerebral disorder in PKU (*8*). The alternative pathways of phenylalanine metabolism, which are all greatly enhanced in PKU, permit numerous possibilities for pathogenetic mechanisms in this disorder. Chief among these are the potential toxic effects of metabolites such as phenylpyruvic acid, phenyllactic acid, and phenylacetic acid, whose formation and excretion are elevated manyfold in PKU.

Conceivably, a compound such as phenylpyruvic acid could interfere with pyruvate carboxylase or pyruvate decarboxylase, thereby impairing carbohydrate metabolism and interfering with the large energy requirement of the brain. Phenyllactic acid may impede the reversible reduction of pyruvate to lactate, an important reaction in the CNS. If phenylacetic acid can be activated to the corresponding coenzyme A (CoA) derivative, a multiplicity of metabolic abnormalities could ensue. For example, the citric acid cycle may be impaired by competition of such a derivative with acetyl CoA for condensation with oxalacetate. Fatty acid synthesis could be impaired by blocking the carboxylation of acetyl CoA to malonyl CoA or by direct competition for the acetyl CoA site on fatty acid synthetase. One would expect abnormalities of cholesterol, phospholipid, and sphingolipid anabolism to occur in patients with PKU.

A number of years ago, Menkes postulated reduced synthesis of proteolipid, a major myelin component, and diminished cholesterol and cerebrosides in PKU (*9*). However, the pitfalls of extrapolations of this type were demonstrated by Silberberg, who examined the effects of phenylalanine metabolites on myelination in explants of cerebellar culture (*10*). Much higher concentrations of phenylacetic acid and phenylpyruvic acid as compared to those present in the blood of PKU patients were required in vitro before defects in myelination were observed. One of the principal difficulties in interpreting experiments with such compounds is the uncertainty concerning the levels of these substances in the tissues of patients. The situation is further complicated by the observation that indole derivatives were comparatively much more toxic than phenylpyruvic or phenylacetic acids. An attempt was made to explain this effect by citing the observation that phenylalanine competes for the absorption of tryptophan from the intestine, resulting in microbial catabolism of the latter to indole, which is then absorbed in toxic quantities by PKU patients (*11*).

These findings are in contrast with results obtained in a similar study with α-ketoisocaproic acid. Excessive quantities of this metabolite are found in the urines of patients with maple syrup urine disease. This substance was highly myelinotoxic in vitro (*12*). Thus, although I favor this kind of investigative approach for establishing the pathogenesis of brain damage in PKU, correct answers may not be forthcoming with such simple direct protocols. I think a fair analogy in support of this remark may be derived from the extensive investigations on the elucidation of the mechanism of action of clofibrate (Fig. 1), a compound used in humans to lower serum cholesterol. It is unlikely that direct inhibition of cholesterol or fatty acid synthesis by this compound adequately explains its physiological effect (*13*). Because of the structural similarities of this substance and the phenylalanine derivatives, a similar multiplicity of factors might be involved in the pathogenesis of PKU.

p-Chlorophenylalanine (Fig. 1) has been used in conjunction with high doses of phenylalanine to produce behavioral changes in a rat model of PKU (*14*). Some structural similarities to the human disorder are evident; however, the lack of a myelin defect in the experimental animals that is frequently seen in young patients with PKU is still unexplained.

The altered behavior in PKU patients may be a consequence of abnormalities of catecholamine metabolism. Several factors may contribute to the decrease of catecholamine concentrations in various areas of the brains of these patients (*15*). High levels of phenylalanine could interfere with the hydroxylation of tyrosine derived from the diet and thereby impair

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whatever catecholamine formation might occur from this source. Conceivably the orthohydroxylated derivatives of phenylalanine found in increased quantities in PKU patients (*16*) (Fig. 1, from left-hand arrow) also interfere with the multiplicity of reactions required for the conversion of catecholamines to physiologically active derivatives (*17*).

Still other considerations have been advanced to account for some of the mental and neurological disturbances in PKU, including the possible biosynthesis of suboptimal or even abnormal proteins because of the altered ratio of phenylalanine to other amino acids. An experiment carried out by Copenhaver and associates showed that ribosomes in the brains of fetal rats were disaggregated when the pregnant females were injected with *p*-chlorophenylalanine and large quantities of phenylalanine (*18*). If protein synthesis is required for memory consolidation (*19*), such a lesion might result in the learning defect reported in PKU patients and animals treated with *p*-chlorophenylalanine (*14*).

The biochemistry of PKU illustrates some of the attempts at elucidating the pathogenesis of the observed derangements in this disorder and the animal models that, in a number of respects, mimic the human situation. When definitive answers are found, such information may be applicable to other disorders of amino acid metabolism. Obviously, special situations are encountered, depending on the amino acid involved.

1) For example, increased quantities of glycine are found in the blood, cerebrospinal fluid, and urine in various metabolic disorders such as methylmalonic acidemia, isovaline acidemia, carbamyl phosphate synthetase deficiency, and disorders of propionic acid carboxylation. Glycine levels are also greatly increased in a primary disorder of glycine catabolism (*20*). The condition is associated with severe mental retardation, listlessness, decreased spontaneous movements, respiratory difficulties, and seizures. There is good evidence that glycine may be a natural transmitter substance for postsynaptic inhibition in the spinal cord and brainstem, a contention supported by the isolation of specific glycine-accumulating synaptosomes from these structures (*21*). It seems logical that the characteristic lethargy in these patients is a consequence of the excessive quantities of glycine; however, the correlation of this finding with the seizures some of these patients experience is in a less satisfactory state.

2) In the metabolism of glutathione (GSH), two possible derangements warrant discussion. The first of these is a deficiency of the enzyme required for the reduction of glutathione disulfide to two molecules of GSH (*22*). Such individuals have neurological involvement and a hemolytic anemia that may suddenly appear after the administration of drugs such as primaquine, chloroquine, aspirin, and phenylbutazone. Most patients have some spasticity and show high-frequency electroencephalographic waves. The second is that abnormally low amounts of GSH are defects in the γ-glutamyl cycle involved in GSH formation (*23*). One patient had multiple neurological difficulties while a second, who was younger, had normal mental development at the time of examination. The metabolism of glutathione is particularly interesting from a neurochemist's viewpoint since large quantities of the reduced form of this material are present in the brain along with high activity of the enzymes involved in GSH metabolism.

It seems reasonable to offer some thoughts concerning the possible role of GSH in the nervous system and how disturbances in its formation and maintenance of a proper proportion in the reduced state may alter brain function. A pertinent observation was made several years ago when it was discovered that impulse conduction in isolated squid giant axons was particularly susceptible to block by chemicals such as quinones with a redox potential greater than 0.5 volt (*24*). If the agent is applied gradually, the first observable effect is a slight decrease in the resting potential of the

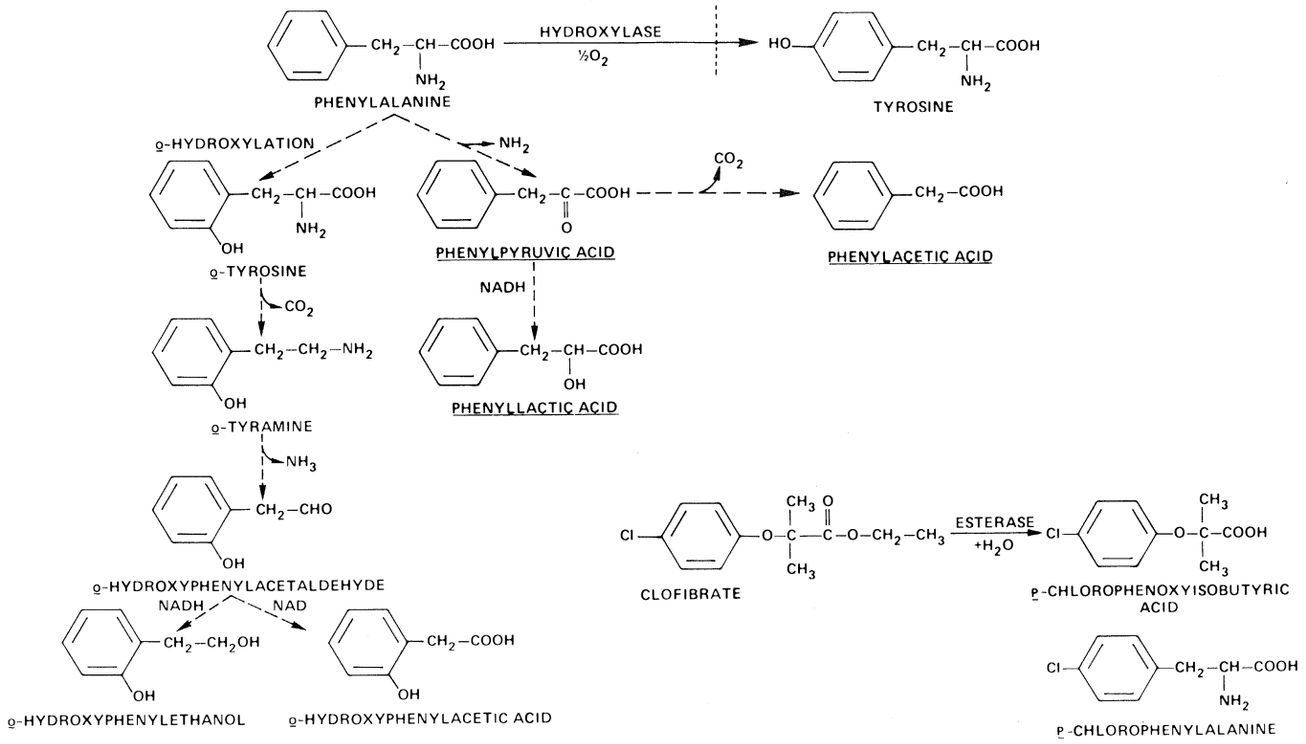

Fig. 1. (Left) Pathways of phenylalanine metabolism in patients with phenylketonuria. (Lower right) Comparison of the structures of *p*-chlorophenylalanine with clofibrate and its derivative from which the ester group was removed.

nerve (Fig. 2). When the amount of the substance is increased, there is spontaneous electrical discharge of the nerve. The critical concentration of oxidizing agents such as quinone, iodine, bromine, and hypochlorite appears to be about 0.2 m*M*. A similar effect is produced by hydrogen peroxide although a higher concentration is required. If the amount of oxidizing substance is increased still further, irreversible depolarization of the nerve occurs. These observations suggest that an electrophysiologically active component of the axonal membrane must be kept in a reduced state for physiological activity. Although the substance has not been identified, it is conceivable that GSH is the agent that participates in the maintenance of the reduced state, and therefore alterations of GSH metabolism result in neurological abnormalities.

Still another of the amino acid disorders affecting mental function is the condition known as aspartylglycosaminuria. Abnormally large quantities of aspartamido-*N*-acetylglucosamine are found in the urines of these patients because of a lack of *N*-aspartyl-β-glucosaminidase, the enzyme that catalyzes the cleavage of the *N*-glycosidic bond between aspartic acid and *N*-acetylglucosamine (*25*). Most patients with this metabolic disorder are mentally retarded, have facial dysmorphism, and other structural abnormalities (*26*). It seems likely that the glycosylated amino acid is a result of the faulty catabolism of glycoproteins since this type of linkage is common in these substances (*27*). Of particular interest is the possibility that there are patients with another type of glycosaminoaciduria who excrete abnormally large quantities of *N*-acetylgalactosaminylthreonine and possibly *N*-acetylgalactosaminylserine in the urine. *N*-Acetylgalactosamine is linked to these amino acids in glycoproteins by *O*-glycosidic bonds that have an α-anomeric configuration. Thus patients deficient in this α-hexosaminidase presumably may be identified because human placental tissue has high activity of this enzyme (*28*). It is also present in other tissues, and, in fact, the enzyme has been partially purified from human and pig liver (*29*). One would suspect that the incidence of patients with deficient α-hexosaminidase activity might be nearly as frequent as those with Tay-Sachs disease who lack β-hexosaminidase activity (*30*). Certainly investigators should be aware of this possibility when confronted with a mentally retarded patient whose signs and symptoms are incompatible with a diagnosis of known metabolic disorders.

Disorders of Carbohydrate Metabolism

There are a number of disorders where carbohydrate metabolism is abnormal and where there is malfunction of the nervous system. For example, excessive quantities of glycogen are found in nerve cells in the brain, spinal cord, and sympathetic ganglia in Pompe's disease (*31*). Mental retardation is said to occur often in this condition (*32*). Here the metabolic defect is a deficiency of α-glucosidase activity (*33*). Particular interest has centered about this disorder since several therapeutic trials were carried out in these patients with impure enzyme preparations obtained from the fungus *Aspergillus niger* (*34*), and although there was evidence of decreased hepatic glycogen in the liver after the intravenous administration of the enzyme preparation, severe pyrogenic reactions occurred in the recipients. There was no indication of a decrease in the quantity of glycogen in the brain or muscle and no clinical improvement was observed. Similar results were obtained with a highly enriched α-glucosidase preparation obtained from human placental tissue (*35*). It seems reasonable to conclude that there was an insufficient uptake of the exogenous enzyme in muscles and brain to exert a beneficial effect. Consequently, work along this line has been in abeyance for several years. However, recent developments lend encouragement for further experimentation. Engel and co-workers have developed a technique for the propagation of human muscle cells in tissue culture (*36*), and these investigators have shown that cells obtained from patients with Pompe's disease manifest the metabolic defect in vitro (*37*). This technique lends itself to the investigation of the effect of exogenous α-glucosidase on the glycogen accumulation in these cells. Since previous workers were unable to demonstrate an effect of α-glucosidase on muscle glycogen in vivo, it might be anticipated that it would be unlikely that the cultured muscle cells would take up the enzyme, particularly if it is isolated from tissues other than muscle. Accordingly, the feasibility of administering α-glucosidase encapsulated in liposomes, which are microspherules made from lipids such as lecithin, cholesterol, and phosphatidic acid (*38*), should be examined in vitro. It seems likely that the lipid components of the liposomes may be varied so that uptake by muscle cells may be enhanced, and increased specificity for such a target tissue might be provided in vivo.

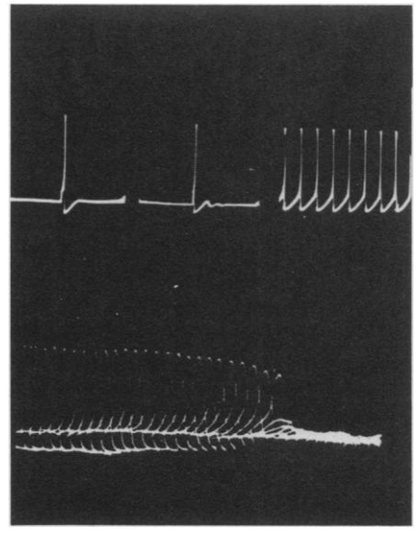

Fig. 2. Effect of quinone on the action potential of the squid giant axon. (Top) Left, control; center, $5 \times 10^{-5}M$ quinone; right, $2 \times 10^{-4}M$ quinone. (Bottom) $1 \times 10^{-3}M$ quinone.

Disorders of Complex Carbohydrates

Another group of heritable disorders is known as the mucopolysaccharidoses. Most patients with these diseases manifest diffuse nervous system dysfunction in addition to visceral and bone abnormalities. There are increased quantities of polymeric carbohydrates such as dermatan sulfate and heparan sulfate (Fig. 3) in the urine, spinal fluid, and various tissues including the brain in these patients. The metabolic disturbances have been identified in at least seven of these disorders as deficiencies of various sulfatases and glycosidases required for the stepwise catabolism of these glycosaminoglycans (*39*). The principal aspect that warrants consideration concerns the pathogenesis of the CNS injury. It is generally agreed that these acidic mucopolysaccharides arise from proteoglycans whose turnover is rapid in the developing nervous system (*40*). One of the developmental processes that probably contributes to this phenomenon seems likely to involve modifications of the neural plasma membranes that occur in synaptogenesis (Fig. 4). A significant portion of the glycoproteins on the surfaces of cells that participate in synapse formation is probably catabolized in the region of the synapse in order to permit functional approximation of these cells to form excitable junctions. Synapse formation may therefore be conceived of as being initiated by the action of one or more specific extracellular or surface-bound glycosidases secreted by one or both of the cells involved. The catabolism of specific glycoproteins on

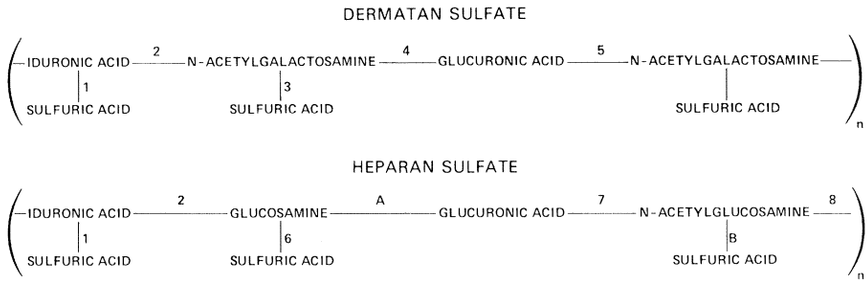

Fig. 3. Structures of dermatan sulfate and heparan sulfate which accumulate in patients with mucopolysaccharidoses. Specific enzymatic defects have been identified at the numbered sites.

the surfaces of the involved cells is thereby initiated. This concept is reminiscent of the lock and key hypothesis involving surface glycosyltransferases (*41*); however, in the present instance, catabolic enzymes and specialized glycoproteins are viewed as the primary determinants of synaptogenesis. A particularly important phenomenon that appears relevant to this process is that the formation of morphologically correct synapses occurs even though the target cells might have abnormal locations in the brain and the timing of the cell-cell contact has been significantly altered (*42*). Furthermore, this phenomenon has been reproduced in vitro. Thus, the development of sensitive histochemical assays should provide much needed information regarding the particular enzymes and reactants involved in synaptogenesis.

In order to explain the intraneuronal accumulation in patients afflicted with mucopolysaccharidoses (*43*), it is presumed that, once the catabolism of surface glycoproteins has been initiated, most of the reactions involved in the degradation of these materials occur after endocytosis of the partially modified components. This concept is strengthened by the demonstration that the carbohydrate composition of the oligosaccharide portion of glycoproteins critically determines whether such proteins will be pinocytized (*44*). One way to examine the validity of this speculation would be by comparing the intracellularly accumulating mucopolysaccharides with those that are elevated in the cerebrospinal fluid in these patients (*45*).

Disorders of Lipid Catabolism

There are ten heritable disorders of lipid metabolism for which the abnormal enzymology is well established. Many of these diseases are named for the clinicians who first described patients with specific signs, symptoms, and pathological features. Among the most frequent of these disorders are Gaucher's disease, Niemann-Pick disease, Fabry's disease, and Tay-Sachs disease. Excessive quantities of the class of lipids known as sphingolipids accumulate in various tissues of patients with these disorders. These lipids have a portion of their structure in common called ceramide. Ceramide is a fatty *N*-acylamide of sphingosine [CH_3-$(CH_2)_{12}$-CH=CH-CH(OH)-CH(NHCOR)-CH_2OH]. Various oligosaccharides or phosphorylcholine are attached to carbon atom 1 of ceramide. For example, the lipid that accumulates in patients with Gaucher's disease is glucocerebroside (ceramideglucose) and that in Niemann-Pick disease is sphingomyelin (ceramidephosphocholine). The rate of synthesis of the accumulating materials is normal, and they arise from the normal turnover of cells in various tissues and the blood. The activity of an enzyme required for the hydrolytic cleavage of the accumulating lipid is diminished in each of the disorders. For example, in Gaucher's disease, the β-glucosidase that cleaves glucose from glucocerebroside is lacking in the tissues of these patients (*46*). When the nature of the enzymatic abnormalities became known, tests were devised for the diagnosis of the affected individuals (*47*) and the detection of heterozygous carriers (*48*). Now accurate prenatal diagnosis is readily available for each of these diseases (*49*).

Some concepts that seem pertinent to the therapy of these disorders merit discussion. The administration of purified enzymes obtained from normal human tissue such as the placenta has been shown to have a measurable effect on the accumulated lipids. When purified placental ceramidetrihexosidase was injected intravenously into patients with Fabry's disease (in which this enzyme is lacking), there was a significant decrease in the level of plasma ceramidetrihexoside (ceramide-glucose-galactose-galactose) that is considerably elevated in patients with this disorder (*50*). Observations made in the course of this study indicated that the exogenous enzyme appeared to catabolize the accumulating lipid after it had been taken up by tissues such as the liver, rather than acting directly on the material in the bloodstream. Thus, the decreased ceramidetrihexoside in the circulation was a consequence of the lowered level in the tissues and the subsequent redistribution of the lipid due to the equilibrium that exists between this material in the tissues and in blood. There are several important consequences of this observation. The first is that lowering the blood ceramidetrihexoside by enzyme replacement would be expected to reduce the rate of accumulation of this lipid in various organs and tissues and thereby prevent or even reverse the renal damage that is a result of this disease. Second, there could also be a salutary effect on the neuralgia these patients experience due to a reduction of ceramidetrihexoside in peripheral nerves. Third, it is possible that the lipid accumulation in the walls of the blood vessels of the heart and brain that often results in myocardial infarctions and strokes in these individuals might also be reversed by the exogenous enzyme. If the quantity of lipid in the blood vessels proves to be susceptible to such reduction, an important principle will be established that in turn may serve as a model for the control of atherosclerosis through the use of still unidentified enzymes.

Investigations of enzyme replacement in Gaucher's disease indicate that intravenously administered glucocerebrosidase can reduce the quantity of lipid that has accumulated in the livers of afflicted individuals (*51*). There was also a long-term decrease in the quantity of circulating glucocerebroside in these patients (*52*). Most patients with Gaucher's disease have the "adult" form, in which there is lipid accumulation in the parenchymal organs, but no signs of the nervous system involvement that occurs in the "infantile" and "juvenile" forms. In contrast to many lipid storage diseases that have cerebral involvement, such as Tay-Sachs where there is overt accumulation of lipid in the nerve cells, glucocerebroside in the brains of those Gaucher patients with the infantile and juvenile forms is found in perivascular cells in the Virchow-Robin spaces and there is only modest storage in the neurons of the brainstem and basal ganglia. If a glucocerebrosidase were injected both intravenously and intraventricularly, the quantity of lipid in the perivascular cells might be reduced, since the cells would be exposed to the enzyme in both blood and cerebrospinal fluid (*53*). However, prior to such an undertaking in humans,

investigators would prefer to carry out a comprehensive investigation with a suitable animal model. It appears that a condition somewhat analogous to Gaucher's disease has been found in Sydney silky dogs (*54*). However, there are two notable differences between the human and the canine forms. The first is the absence of lipid-laden "Gaucher cells" in the spleen of the dog whereas this organ may be filled with sheets of these cells in affected humans. Second, there was much more extensive neuronal involvement in the brain of the Gaucher dog than in humans with the infantile or juvenile forms of Gaucher's disease. Thus, although enzyme replacement studies with such a model might provide much valuable information, extrapolation of the findings to the situation in humans may not be completely feasible.

A potentially important development that should be mentioned here concerns enzyme replacement in disorders such as Tay-Sachs disease where the CNS is primarily involved. The effect of intravenous administration of hexosaminidase A was examined several years ago in a patient with the O-variant form of Tay-Sachs disease (Sandhoff-Jatzkewitz disease) (*55*). These patients have a drastic deficiency of all hexosaminidase activity in their tissues (*30*). When the exogenous enzyme was injected, a normal level of hexosaminidase A was quickly produced in the blood. The enzyme was very rapidly cleared from the circulation, with a half-time of 8 minutes. None of the injected enzyme appeared to reach the brain, probably because it could not cross the blood-brain barrier. There was no evidence of improvement of the patient's clinical condition. However, a 55 percent decrease in the amount of globoside (ceramide-glucose-galactose-galactose-*N*-acetyl-galactosamine) was observed in the blood 4 hours after injection of the enzyme, again indicative of a physiological effect after the enzyme had been taken up by tissues. When the amount of hexosaminidase activity was compared in a liver biopsy specimen taken before infusion with that in a similar sample obtained 45 minutes after injection of the enzyme, it was found that there was at least 2.4 times more total hexosaminidase A activity in the liver that had actually been infused (*56*). This effect was similar to an augmentation of α-galactosidase activity that was observed in liver biopsy specimens obtained from a patient with Fabry's disease when ceramidetrihexosidase, an α-galactosidase, was injected (*50*). These observations (*50, 56*) prompted the hypothesis that the active exogenous enzymes had

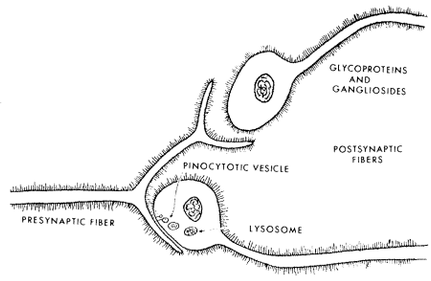

Fig. 4. Hypothetical reduction of glycoproteins on the surfaces of cells during synapse formation (lower right).

caused a change in the configuration of the patients' mutated catalytically inactive enzymes and therefore conferred activity to the aberrant enzymes (*56*). Support for this hypothesis is derived from the following observations.

1) It is well known that catalytically inactive *Escherichia coli* β-galactosidase can be activated in many instances when it is mixed with an antibody prepared against the active, wild-type enzyme (*57*).

2) Bakay and Nyhan observed that coelectrophoresis of hypoxanthine-guanine phosphoribosyltransferase (HGPRT), the enzyme lacking in patients with the Lesch-Nyhan syndrome (*58*), with a red blood cell hemolyzate from a patient with this disorder resulted in the recovery of more catalytic activity than had been applied (*59*).

3) Srivastava and Beutler have reported that patients with Sandhoff-Jatzkewitz disease have a protein in their tissues that cross reacts with antibodies developed against human placental hexosaminidase A indicating the presence of the inactive enzyme (*60*). These findings provide considerable support for the hypothesized activation of the mutant enzyme by a subunit of the enzyme derived from normal tissues.

The finding that intravenously injected hexosaminidase A did not reach the brain is discouraging for the treatment of Tay-Sachs and related diseases of the CNS by enzyme replacement therapy. Much innovative conceptualization and methodology is required in order to effectively reduce the pathological quantities of stored materials in such disorders. Thoughts along this line include modification of the natural active enzyme to increase its hydrophobicity, so that such an altered molecule will traverse the blood-brain barrier (*61*). So far, all attempts to effect such an alteration in hexosaminidase A have been accompanied by a loss of catalytic activity.

Alternatively, much consideration is being given to the possibility of temporarily opening the blood-brain barrier by intraarterial injection of hypertonic solutions (*62*). The rationality of this approach seems in flux at present. It was suggested that an experiment be carried out with cultured neuronal cells to determine whether the exogenous enzyme would actually be taken up by such cells (*56*). An experiment was recently carried out along this line in my laboratory. There was no indication that cultured N-18 mouse neuroblastoma cells took up placental hexosaminidase A (*63*).

These negative results seem to make enzyme replacement therapy for metabolic disorders involving the CNS even more difficult. However, some recent information compels concerned scientists to maintain an open mind toward this formidable task. Feder has obtained good evidence of the exchange of β-glucuronidase between cells in the brain and other tissues in tetraparental mice derived from high and low β-glucuronidase strains (*64*). This important observation again raises the hope for enzyme replacement for heritable nervous system disorders. Several possibilities now seem apparent. The first is to attempt to devise a suitable procedure for obtaining the requisite enzyme from human brain. Since the logistic problems of such an undertaking appear enormous, recourse to other strategies to obtain an effective enzyme seem necessary. One approach might be to examine the effect of modifying the carbohydrate portion of the molecule since hexosaminidase A has been shown to be a glycoprotein (*65*) and it is known that the uptake of enzymes can be influenced by the oligosaccharide portion of the molecule (*44*). This alteration might be accomplished through enzymatic addition or removal of selected hexoses, hexosamines, or sialic acid residues. If an appropriate modification is discovered, it may be possible to screen enzymes from various sources for the proper carbohydrate composition with the use of lectins of known carbohydrate affinities (*66*). Once an enzyme with the requisite properties is obtained, it might then be useful to consider procedures to deliver the enzyme to the brain, such as incorporation of the missing enzyme into the patient's own polymorphonuclear leukocytes (*56*). This type of incorporation has been elegantly demonstrated by Cohen and coworkers (*67*).

Disorders of Lipid Anabolism

The scientific community should be aware of an entirely new type of hereditary abnormality of the nervous system that is caused by deficiencies of synthet-

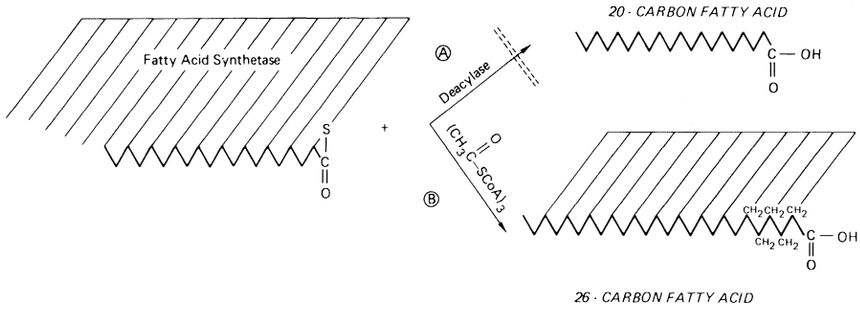

Fig. 5. Postulated alteration in the synthesis of long-chain fatty acids in sudanophilic leukodystrophy. (Top right) Pathway in normal brain and adrenals; (lower right) pathway in patients with the disorder.

ic enzymes. The mucopolysaccharidoses, sphingolipidoses, and most glycogen storage diseases are due to deficiencies of hydrolytic catabolic enzymes. For years it was considered likely that mutations involving synthetic enzymes would be lethal in utero. However, the prototype of anabolic disorders has now been identified. The propositus was a male infant with respiratory difficulties and convulsions a few days after birth. Physical and motor development were poor, the skin was thickened and coarse, the liver and spleen were enlarged, and bilateral inguinal hernias were present (*68*). The child died at 3½ months of age. The brain showed spongy degeneration and a severe lack of myelin in several major areas (*69*). The principal biochemical abnormality was a dramatic alteration of the composition of sphingolipids known as gangliosides (*70*). Most gangliosides in normal human brain are comprised of ceramide linked to an oligosaccharide chain consisting of glucose, galactose, *N*-acetylgalactosamine, galactose along with one to four molecules of sialic acid joined to the internal and terminal molecules of galactose. The only gangliosides in the brain of the patient consisted of ceramide-glucose-galactose and one or two molecules of sialic acid linked to the galactose. These gangliosides are called G_{M3} and G_{D3}, respectively. The enzymatic defect was shown to be a dramatic reduction in the activity of the enzyme that catalyzes the addition of the molecule of *N*-acetylgalactosamine to G_{M3} to form the trihexosylganglioside G_{M2} (*71*). [This metabolic defect is exactly the opposite of that in Tay-Sachs disease, where there is a deficiency of the hexosaminidase that catalyzes the cleavage of *N*-acetylgalactosamine from G_{M2} to form G_{M3} (*72*)]. Since ganglioside synthesis occurs in a stepwise fashion by the addition of molecules of hexoses, hexosamine, and sialic acid to the oligosaccharide moiety (*41*), the block in the conversion of G_{M3} to G_{M2} prevented the formation of the tetrahexosyl gangliosides normally present in human brain. It seems likely that this metabolic defect was inherited as an X-chromosomal recessive characteristic since a maternal uncle of the child had exactly the same outward appearance and died at 2 months of age 30 years earlier. Recently a brother of the propositus was born with features identical to those of the patient. A similar metabolic block has been observed in cells transformed with tumorigenic viruses (*73*). The pathophysiological consequences of the lack of higher ganglioside homologs in the brain is the subject of another article (*74*).

The etiology of another disease involving the CNS that might be thought of as a "mixed-function" disorder of anabolism deserves mention. Patients with the hereditary form of sudanophilic leukodystrophy (Schilder's disease) have widespread demyelination, dementia, and neurological deficits along with atrophy of the adrenal cortex (*75*). A potentially important observation concerning the abnormal biochemistry in this condition was reported by Igarashi and coworkers (*76*). These investigators found that cholesterol esters in the brain and adrenal glands in these patients had an unusually high proportion of fatty acids with a chain length of 24 to as many as 30 carbon atoms. The fatty acids in cholesterol esters from control tissue samples were mostly 20 carbon atoms in length. Fatty acid synthesis in the brain results in the formation of free fatty acids that have been cleaved from a sulfhydryl group on the synthetic enzyme (*77*). The liberation of free acids is probably catalyzed through the action of a deacylase that is present in high activity in brain and other tissues (Fig. 5). Attenuation of this function might promote further addition of two-carbon fragments to the growing fatty acid chain, resulting in the formation of the very long fatty acids. Experiments to determine whether there is diminished activity of this thiolase can be readily devised. However, several other perhaps equally plausible hypotheses can be advanced. These theories include a mutation of the amino acid composition of the fatty acid synthetase in the hydrophobic portion of the molecule so that the aliphatic portion of the fatty acid is bound more tightly to the enzyme than normal and the formation of longer hydrocarbon chains is favored. Another possibility involves substitution of amino acids in the esteratic site of the enzyme which decreases the way in which water can approach this portion of the enzyme. An impairment of fatty acid oxidation could result in the occurrence of very long chain fatty acids that occur in patients with this condition. Whatever the cause, it is likely that the exceptionally long chain cholesterol esters interfere with the function of the myelin-specific cholesterol ester hydrolase (*78*). Impairment of myelinogenesis may well be a consequence of such an impediment since it is well known that the proportion of cholesterol esters to free cholesterol decreases with normal brain development. One might imagine that such a metabolic impairment would also be involved in the abnormalities of steroidogenesis in the adrenal cortex of these patients.

Summary

This overview was designed primarily to provide examples of hereditary metabolic disorders that result in nervous system dysfunction. Some of the more frequently encountered pathological conditions were selected in order to illustrate the mechanisms and the consequences of the metabolic derangements. Therapeutic approaches for the correction of such disorders are discussed where it appears appropriate. In time the precise etiology for those eponymous genetic conditions with stereotyped pathologic and clinical manifestations such as Huntington's chorea (*79*) and Friedreich's ataxia (*80*) will be disclosed. It is possible that some forms of epilepsy (*81*) and perhaps certain psychiatric disturbances (*82*) will be shown to be inherited metabolic disorders. As our knowledge and skill increase, this logic may eventually be extended to biochemical explanations of variation in individual skills and talents. Certainly innovative extrapolation and novel research directions will be necessary to provide an understanding of these differences. However, it is axiomatic in research that each useful contribution serves largely as a point of departure for further accomplishments.

References and Notes

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