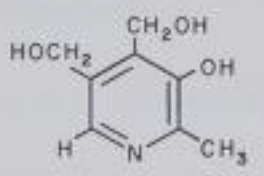
51: Pyridoxine

György[1] first recognized vitamin B_6 as a distinct entity in 1934 and less than 5 years later, another worker succeeded in crystallizing the material.[2] Shortly thereafter, the term, pyridoxine, was introduced by György and Eckhardt[3] to designate the isolated B_6 derivative. There are three interconvertible forms of vitamin B_6; namely, pyridoxine, pyridoxal and pyridoxamine (Fig 51-1). Pyridoxal phosphate and pyridoxamine phosphate are the derivatives that serve as coenzymes, whereas pyridoxic acid is the principal B_6 metabolite that is excreted in the urine.

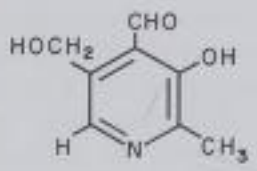
BIOCHEMICAL RELATIONSHIPS

Absorption

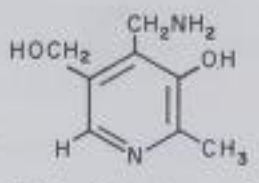
Members of the B_6 group of vitamins are readily taken up by the intestine. They are also synthesized by many normal microbial inhabitants of the gut, but not to an extent sufficient to meet the demands of the host.



PYRIDOXINE



PYRIDOXAL



PYRIDOXAMINE

Fig. 51-1. Structures of the biologically active forms of vitamin B_6.

Transport

The principal forms of the vitamin in the blood and tissues are the phosphorylated derivatives of pyridoxal and pyridoxamine.

Function

The B_6 group of vitamins are essential for growth in all animal species. Pyridoxal phosphate serves as an important coenzyme in reactions involving fats, carbohydrates, amino acids and porphyrins (Table 51-1). Vitamin B_6 metabolism is influenced by hormone balance.[4] Glucocorticoid administration, for example, enhances the require-

Table 51-1. Coenzymic actions of pyridoxal phosphate

Type of Metabolism	Prosthetic Group Involvement
Amino acid	1. Decarboxylation of tyrosine, arginine, glutamic acid and certain other amino acids
	2. Deamination of serine and threonine
	3. Desulfhydration of cystine
	4. Transamination of all naturally occurring amino acids
	5. Racemization of amino acids
	6. Conversion of tryptophan to quinolinic acid and niacin
	7. Decarboxylation of 5-hydroxytryptophan to form serotonin
	8. Conversion of serine to cystathionine
Lipid	1. Conversion of protein to fat
	2. Fat storage
	3. Unsaturation of linoleic acid to form arachidonic acid
Carbohydrate	1. Lactic acid dehydrogenase
	2. Aldolase
	3. Catalase
	4. Phosphorylase
Porphyrin	1. Incorporation of glycine and succinate into heme

ment of this vitamin by accelerating carbohydrate, lipid and protein metabolism.

Pyridoxine plays an important role in the immune process by catalyzing the biosynthesis of nucleic acids required for cellular proliferation and the formation of specific antibodies.[5] Therefore, lack of the vitamin is not only associated with an impairment in the production of circulating antibodies, but also a delay in hypersensitivity responses.

Pyridoxine depletion causes an interesting increase in the myristic and palmitoleic acid content of serum triglycerides and cholesterol esters.[6] Available evidence suggests that γ-aminobutyric acid, a substance with unique occurrence in vertebrate central nervous tissue and vitamin B_6 are involved in the neuronal excitability mechanism.[7] It is assumed that vitamin B_6 deficiency may decrease the stability of the neuronal membranes, thereby contributing to increased excitability and consequent convulsive seizures.

Excretion

Huff and Perlzweig[8] are credited with first isolating pyridoxic acid from human urine. Others showed that this compound was the major metabolite of vitamin B_6 excreted in the urine by man.[9] It is important to recognize, however, that early estimates of the excretion of pyridoxic acid were spuriously high due to a lack of specificity of existing analytical techniques.[10]

LABORATORY EVALUATION

Fluorometric Techniques

The pyridoxic acid formed by oxidizing pyridoxine or pyridoxal can be converted into a highly fluorescent lactone by heating in an acid medium. Pyridoxine is first transformed into pyridoxamine prior to formation of the lactone. Methods based on this fluorometric response have been used to quantitate members of the B_6 group in both blood and urine. The technique has been adapted to microquantities of the specimen and is considered much more sensitive and specific than spectrophotometric or colorimetric methods.[11]

The 4-formyl group of pyridoxal and pyridoxal phosphate takes part in the cyanohydrin reaction.[12] This has served as a basis for quantitating pyridoxal phosphate and pyridoxal, but not as yet in blood or tissues.

Enzymatic Techniques

Pyridoxal phosphate serves as a coenzyme for the conversion of tryptophan to indole, pyruvic acid and ammonia. This member of the B_6 group may be measured in terms of the indole liberated in a system containing the test material, substrate and the apoenzyme, apotryptophanase.[13] The procedure has been applied to serum, leukocytes, as well as tissue.[14]

Chromatographic Techniques

Gas liquid chromatography provides a rapid method for the analysis of individual analogues in the B_6 group.[15] Acetyl derivatives of pyridoxine, pyridoxal and pyridoxamine can be separated in this manner using either hydrogen flame ionization or β-ionization detectors.

Manometric Techniques

A pyridoxal-requiring reaction involving the decarboxylation of tyrosine with consequent liberation of carbon dioxide has been used for quantitation purposes.[16] This procedure, with slight modification, has been widely used to measure the vitamin B_6 content of the blood and tissues.[17] It is not capable of accurately quantitating levels of the vitamin below 10 mμg/ml.[17]

NORMAL VITAMIN B_6 LEVELS

The normal young adult is considered to require a minimum of 1.5 to 1.75 mg of vitamin B_6 daily.[18] This need varies with the protein intake and, if more than 100 gm of protein is ingested, the daily requirement of the vitamin may be slightly increased. Possible influences of physical stress, pregnancy, sex and age, as well as other factors on B_6 requirement have not yet been adequately explored.

Examination of pyridoxine levels in maternal and cord blood has shown that the latter contains less B_6 than the former.[19] There is a striking decrease in circulating pyridoxal phosphate concentration with age[20] (Table 51-2). Comparisons of transaminase values in serum with and without added pyridoxal phosphate disclose that addition of the coenzyme causes a pronounced increase in measured enzyme activity. The alteration in pyridoxine metabolism that occurs with aging is prob-

Table 51-2. The plasma pyridoxal phosphate content in various age groups[20]

Age (yr)	Number of Subjects	Mean Pyridoxal Phosphate Level (mμg/ml)
0 to 1	14	16.3
20 to 29	13	11.3
30 to 59	11	7.1
60	21	3.4

ably related to a defect in absorption or phosphorylation of the vitamin.

ABNORMAL VITAMIN B_6 LEVELS

Alterations in vitamin B_6, both of a nutritional and metabolic nature, seem associated with a number of disorders. In some diseases, it is unclear whether or not there is an actual deficiency due to a nutritional lack or some secondary impairment in the normal utilization of the vitamin, leading to an increased requirement. One of the most interesting disturbances is that which seems to involve a genetic defect in pyridoxine metabolism.

Pyridoxine-responsive Anemia

A number of cases of spontaneously occurring anemia alleviated by pyridoxine have recently been recognized.[21] Although the hematologic findings are quite variable, there are always characteristic abnormalities in iron metabolism. Most patients with pyridoxine-responsive anemia have high serum iron levels accompanied by a decreased serum iron-binding capacity. There is a high incidence of hemosiderosis of the liver and other clinical evidence of an iron overload. Ferrokinetic data indicate that the failure in hematopoiesis is responsible for the iron excess.

Renal Lithiasis

The deposition of oxalate in the urinary tract is a serious complication of several disorders in man. Induced deficiencies of pyridoxine provoke oxaluria in man as well as animals.[22] This suggests a possible relationship between a lack of B_6, the endogenous production of oxalic acid and, perhaps, the etiology of urinary stone formation.

Celiac Disease

Urine levels of various tryptophan metabolites are abnormally high in patients with celiac disease.[23] This is an expression of a failure to metabolize tryptophan in a normal manner and is attributable to a lack of vitamin B_6.[24] Hence, such patients require supplemental vitamin B_6.

Drug-induced Deficiency

Penicillamine, a powerful chelating agent, is useful for treating Wilson's disease and other cases of heavy metal intoxication. This drug also forms a complex with pyridoxal phosphate, thereby interfering with the participation of the coenzyme in tryptophan metabolism. In man, this results in an increase in the 24-hour output of xanthurenic acid and kynurenine, which are tryptophan metabolites.[25] Therefore, pyridoxine supplementation should always be included with long-term penicillamine therapy.

Rheumatoid Arthritis

Patients treated with isoniazid for tuberculosis often develop rheumatic symptoms.[26] The drug promotes the excretion of pyridoxine and disturbs the tryptophan metabolite pattern of the urine. Persons with active rheumatoid arthritis excrete abnormally low levels of the B_6 vitamins and related metabolites in urine.[27] Supplements of the vitamin may be therapeutically beneficial in arthritic individuals.

Miscellaneous Diseases

The excretion of abnormally large amounts of xanthurenic acid in a patient with acute disseminated lupus erythematosus has been alleviated by pyridoxine therapy.[28] Subjects with scleroderma eliminate excessive amounts of tryptophan metabolites after receiving a loading dose of the amino acid.[29] Abnormalities in tryptophan metabolism are evident in various forms of cancer and Hodgkin's disease.[28] Pyridoxine-responsive xanthurenuria has been noted in patients with schizophrenia,[28] acute alcoholism[30] and epilepsy.[31]

Nutritional Deficiency

A variety of animals and also man, when depleted of vitamin B_6, show characteristic deficiency signs (Table 51-3). For many years, it was considered highly unlikely that a lack of the vitamin sufficient to cause clinical symptoms could occur naturally in man. However, the condition has been observed in two mentally retarded infants who received a pyridoxine-restricted diet.[32,33]

A typical clinical picture observed in a group of infants including abdominal distress, increased irritability, as well as seizures and unconsciousness was ultimately traced to the ingestion of a commercially prepared formula that contained minimal amounts of the vitamin.[34] A subsequent national survey of infants receiving this formula disclosed that about 300 cases with overt symptoms were responsive to pyridoxine therapy.[35] Sterilization and autoclaving of the milk used in the formula were responsible for the reduction in vitamin B_6.

Pyridoxine Dependency

A number of cases of vitamin B_6-preventable seizures in newborn infants has been described that are not related to an actual deficiency of the vitamin.[36] Extremely large doses of the vitamin are required to avoid further seizures. Moreover, electroencephalograms, which are abnormal dur-

Table 51-3. Symptomatology of vitamin B_6 deficiency in experimental animals and man

Type of Mammal	Major Deficiency Symptoms
Rat	Severe dermatitis, occasionally hemolytic anemia, reduction in total body fat
Syrian hamster	Muscular weakness, atrophy of lymph glands and adipose tissue, increased urinary excretion of xanthurenic acid
Rabbit	Desquamative dermatitis of the ears, mild anemia, convulsions, hypercoagulability, creatinuria, paralytic collapse, death
Dog	Seropurulent conjunctivitis, blepharitis, dermatitis with loss of hair around eyes, arteriosclerotic complications
Rhesus monkey	Invariable arteriosclerosis, dental caries, fatty degeneration or cirrhosis of the liver, sclerosis of the pancreas, disturbance of the central nervous system
Man	Seborrheic and desquamative dermatitis of skin and mucosa, various neurologic and mental disorders

ing such convulsive episodes, revert to normal during pyridoxine therapy.

Available evidence, although admittedly incomplete, strongly favors the postulate that pyridoxine dependency is familial in nature. However, the underlying metabolic defect involving vitamin B_6 is obscure. It appears likely that repeated stillbirth sometimes results from pyridoxine dependency *in utero*.[37] Prophylactic pyridoxine is indicated in any pregnant female with a previous history of giving birth to a child with the vitamin dependency.

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