BIOCHEMICAL PATHOLOGY IN PORPHYRIN METABOLISM

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The normal biochemical pathway of porphyrin synthesis has only recently been elucidated.[1] This has led to a better understanding of various clinical disturbances related to porphyrin metabolism, but both clinicians and biochemists are still far from understanding all the problems related to the porphyrias. Certain aspects of the porphyrin synthesis pathway, such as the production of isomers belonging to type III series, rather than type I, under normal conditions; the role of certain control processes regulating the rate at which the porphyrin nucleus is formed, such as the place of feedback mechanism and non-porphyrin pathway utilization of δ -amino levulinic acid, are not clearly understood. However, present knowledge of the various steps of the normal synthetic pathway is specific enough to permit the biochemist to make certain assumptions and speculations. The chemical findings with regard to aberrations of porphyrin metabolism in the porphyrias will be discussed in this paper.

Porphyrin Synthesis

The normal biochemical pathway is schematized in figure 1. A few aspects of the synthesis of the porphyrin nucleus have been thoroughly investigated; the synthesis of δ -amino levulinic acid, and the formation of the precursor pyrrole, porphobilinogen, from this compound are clearly understood. However not much is known about the condensation of the four pyrroles which unite to give an assymetrical porphyrin, uroporphyrinogen III. This is indicated in figure 1. Not enough is known about the synthesis of abnormal isomers, such as uroporphyrinogen I and II, that occurs in excess under certain pathologic conditions; nor is any normal physiologic function of these abnormal molecules known if, in fact, any function exists. Also, although the pathway leading from uroporphyrinogen III to protoporphyrin IX is clear, not much is known about the enzyme systems that catalyze this conversion nor about the inclusion of iron in the protoporphyrin IX nucleus which finally produces the heme molecule. The heme molecule in turn is used as a building block for various heme proteins in any living cell. Additionally, not enough is

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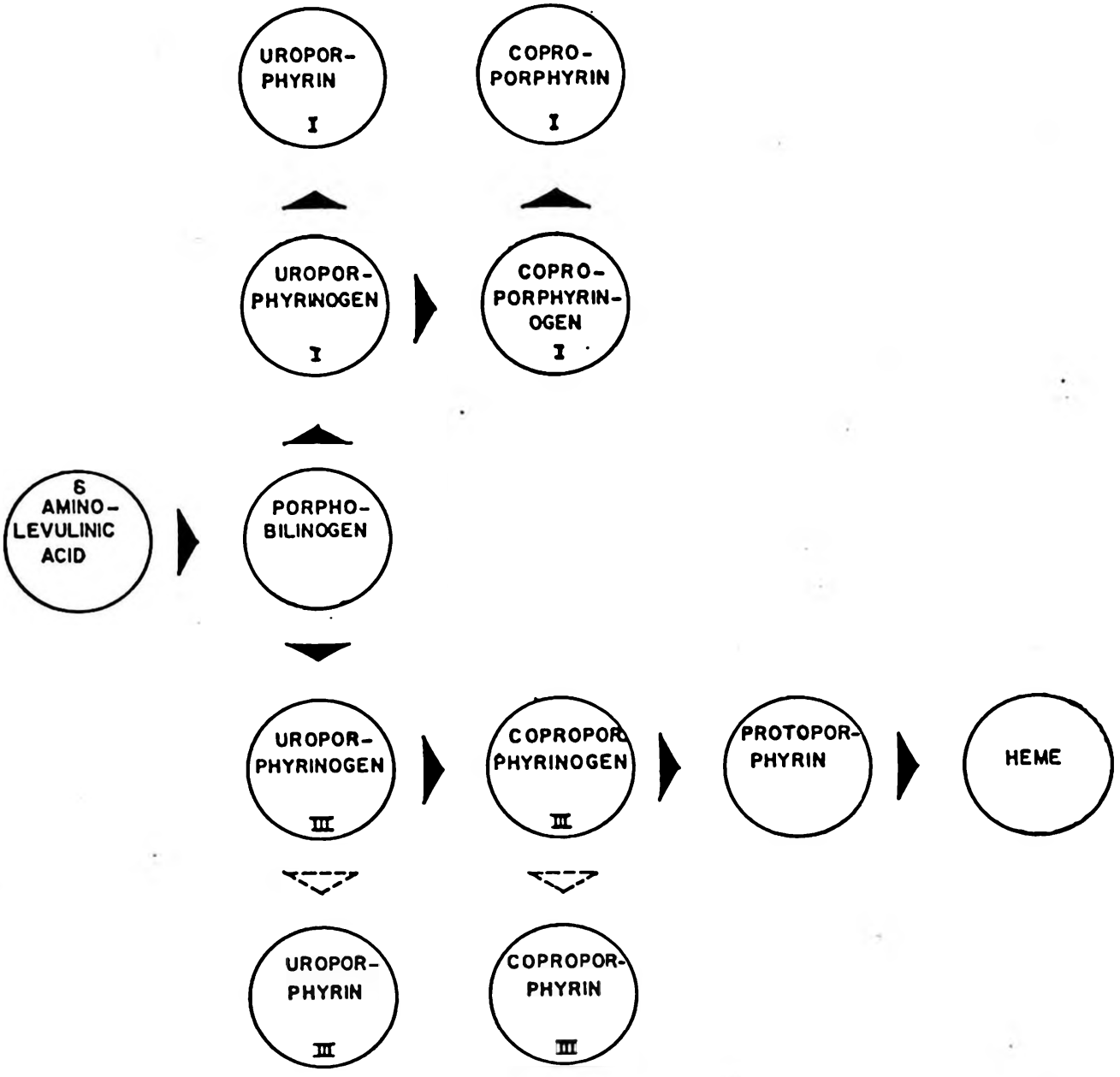


Fig. 1. An abbreviated scheme for the synthesis of heme. Dotted arrow indicates the formation of porphyrins of normal isomeric series.

known about the mechanisms that control this formation; there is therefore no clear knowledge about the various metabolic errors either occurring congenitally or arising from intoxication by certain drugs. We are far from explaining such errors with reasonable chemical certainty.

Metabolic Turnover of Porphyrins

We know that heme, once synthesized, becomes part of the structure of many proteins. One of the best worked-out examples of this process is the formation of hemoglobin from heme and globin. Heme is synthesized in the red blood corpuscles and, judging from the decay time of the average erythrocyte in man, approximately 300 milligrams of heme must be synthesized per day, in adult bone marrow, to produce the required amount of hemoglobin.[2] The heme nucleus also participates in the formation of heme proteins other than hemoglobin, such as the cytochromes. Cytochromes exist in almost all the cells which carry on aerobic oxidation of glucose. Not much information is available about their turnover rate. Drabkin[3, 4] reported a 12.7 per cent turnover per day in the cytochrome *c* of liver tissue in animals that had undergone partial hepatic lobectomy, corresponding to the rate of appearance of new cytochrome *c* during liver regeneration. This represents an appreciably greater daily turnover than that of hemoglobin. The half life of liver catalase, which is another heme protein, is reported to be several days. Such a high rate of turnover requires that a considerable amount of porphyrin nucleus be synthesized.[5] The metabolic fate of these heme proteins will not be described in this paper as it is not germane to the discussion.

From these facts it is apparent that the rate of heme, and hence protoporphyrin, synthesis must be very large compared to the amount of porphyrins and their precursors excreted in urine and feces, which is very small. These compounds, and their precursors, must be efficiently retained in the cells forming them since, under ordinary circumstances, only a small amount is encountered in urine and feces.

Delta-amino levulinic acid is excreted mainly in the urine, the excretion of this compound in bile is negligible.[6] About 2 milligrams of this compound are excreted daily.[7, 8] If this chemical, labeled at 1 and 4 positions with C^{11}, is injected into a human volunteer, labeled CO_2 will appear in the breath but only a very small portion of the labeled compound will be found in heme, indicating that little is used for heme synthesis.[6, 9, 10] The reason for this is the rapid excretion

of this compound in urine; in fact most of the injected δ -amino levulinic acid is excreted in the urine unchanged.[6] Also the penetration of this compound into the immature red cells seems to be a rate limiting factor. However, the formation of even a small amount of labeled heme from intravenously injected labeled δ -amino levulinic acid indicates that some of this compound does indeed penetrate the immature red cells and is used to synthesize porphobilinogen and protoporphyrin. Indeed both these latter compounds are found in the urine and bile of persons who have received labeled δ -amino levulinic acid injections.

As much as 1.0 to 1.5 milligrams of porphobilinogen is excreted per day.[7, 8] Porphobilinogen determination is very difficult and with routinely used methods one cannot accurately measure the amount excreted in urine.[11] Porphobilinogen cannot pass the cell membrane, hence injected material is largely recovered in the urine and fecal excretion; the synthesis of porphyrins from porphobilinogen is insignificant.[12]

The other intermediary products of the porphyrin pathway are also found in urine and feces. The compounds belonging to the porphyrinogen series are readily oxidized to porphyrins by exposure to light and air. Uroporphyrin is excreted mainly in the urine. Most of the excreted uroporphyrin in normal subjects is type I; however small amounts of type III isomer can also be detected.[13, 14] The intermediary porphyrins which contain 7, 6, 5 carboxylic acid side chains are also encountered in the urine.[15, 16] Coproporphyrin is excreted in both bile and urine. In fact the bulk of urinary porphyrin under normal conditions is coproporphyrin, which is excreted at the rate of about 100 to 300 milligrams per day.[14, 17] Both types I and III isomers are seen in normal subjects with varying ratios.[15, 18] Coproporphyrin is however chiefly excreted through the bile at a daily rate of 400 to 1000 milligrams as demonstrated by Schwartz. The preferential excretion route for this compound is the biliary route unless simultaneous liver damage is present.[19] Protoporphyrin is also excreted in the bile and is a normal constituent of bile and feces, however it is not encountered in the urine under normal conditions.[20]

The Porphyrias

The term porphyria is used for a group of diseases in which disturbance of porphyrin metabolism is the predominant finding. Until recent years the porphyrias were considered, except for porphyria

erythropoietica, to be diseases of adulthood. In the summer of 1955, however, a new type of porphyria appeared in southeastern Turkey in epidemic form. The epidemic was touched off by the consumption of hexachlorobenzene treated wheat and most of those afflicted were children. Thus the subject of porphyria has now entered the domain of pediatrics.

Porphyrias can be classified as follows :

I. Porphyria Erythropoietica
   1. Congenital type
   2. Protoporphyric type

II. Porphyria Hepatica
   1. Type chiefly affecting children and adolescents (porphyria turcica)
   2. Types chiefly affecting adults
      a. Swedish
      b. South African (whites)
      c. Cutaneous
      d. Multiform

III. Non-Hereditary Toxic Porphyrias
   (lead poisoning)

In this classification the terms are intended to reflect genetic patterns rather than geographic factors. For example, in the porphyria hepatica group, some Swedish patients have been observed to exhibit symptomatology and excretory patterns typical of South African porphyria. Conversely, South African patients have occasionally showed findings typical of the Swedish type.[21] In Holland two porphyric families have been observed by Dean et al.[21] in one of which the disease resembled the South African type and, in the other, the Swedish type. Thus it needs to be emphasized that in classifying cases of porphyria hepatica the country in which the patient resides does not necessarily determine the type of disease from which he suffers. This is determined by the clinical and chemical findings.

Beyond this there remain cases of porphyria which do not fit comfortably into any of these categories. Until a satisfactory classification can be found, these cases may be grouped under the heading *porphyria multiforme.*

The third form includes the cases where an environmental factor regularly leads to disturbance of porphyrin metabolism irrespective of hereditary predisposition. A notable example of this form of porphyrin metabolism disturbance is that attributable to lead poisoning.

As biochemical pathology is helpful in diagnosing cases of porphyria and at times is the unique means of establishing the type of porphyria involved; blood, urine and stool findings typical of various forms of the disease will be presented in this paper together with a discussion of the metabolic derangement leading to the biochemical pathology.

Chemical Findings in Erythropoietic Porphyrias

In these porphyrias the disturbance of porphyrin metabolism occurs only in the erythropoietic system. There are two subdivisions of this group; one of which (congenital erythropoietic porphyria) has long been known and the other (protoporphyria erythropoietica) has only recently been described.

1. Congenital Erythropoietic Porphyria. In this form of the disease there is increased excretion of porphyrins in the urine. As much as 500 milligrams of uroporphyrin daily can be found in the urine of such patients.[22] The uroporphyrin is mostly of type I although a small amount of type III isomer is also present,[23] and some porphyrins with less than eight carboxyl groups can be demonstrated. A large amount of coproporphyrin is found in the urine but it is less than the amount of uroporphyrin. The main isomer of the coproporphyrin present is type I, type III being present but in lesser quantities.[24] Porphobilinogen cannot be detected with conventional techniques in the urine of such patients although with more advanced techniques it has been found to be present in normal amounts.[25, 26] In this disease increased amounts of coproporphyrin are excreted in stool, and here again the isomer is mainly type I with a small amount of type III; only a very small amount of uroporphyrin is excreted in the feces, and protoporphyrin excretion by the fecal route is usually not increased.[24, 27] Both uroporphyrin and coproporphyrin are demonstrable in plasma. [26, 28] A large amount of uroporphyrin and a somewhat smaller amount of coproporphyrin are present in circulating erythrocytes; the highest concentrations of these porphyrins are seen in bone marrow and spleen. The isomers present seem to be type I. In this type of porphyria there is an excessive production of porphyrins, mainly type I, and the defect seems to be confined to the hematopoietic system.

The enzyme systems responsible for the formation of the two isomeric types of porphyrins are not well worked out; however there is good indirect evidence for the presence of more than one system.[20, 30] Under normal conditions, the enzyme system that synthesizes type III isomer is operative and only a very small amount of the compounds belonging to type I series are synthesized. In erythropoietic porphyria the reverse holds true. Schmid has demonstrated by the fluorescence technique that not all the bone marrow cells belonging to the erythroid series fluoresce; hence he had concluded that there are two lines of erythropoietic cells, one being normal and the other one manifesting the defect.[35] A similar finding is reported for bovine erythropoietic porphyria.[27] This would suggest that some of the erythroid cells of the bone marrow in porphyria erythropoietica patients undergo a mutation which leads to the preferential formation of type I isomers. These hypotheses, however, and the very nature of the defect itself must be established by direct experimental evidence.

It has been shown that the erythrocytes of patients with erythropoietic porphyria can form both type I and type III porphyrinogens from δ -amino levulinic acid and porphobilinogen.[31] The larger part of porphyrins in bone marrow are present in the developing erythroid cells and, in fact, absorption spectra of developing bone marrow cells indicate that both porphyrin and hemoglobin synthesis proceed very actively in maturing red cells.[1, 32] It seems likely that these cells do produce a large amount of uroporphyrin I isomer via the uroporphyrinogen I formed. This conversion may be increased by light and in fact some patients with acute porphyria do excrete more uroporphyrin I during the summer months when they are exposed to more sunlight than during the winter.

The uroporphyrinogen I molecules which escape the oxidation that may be light-induced, are decarboxylated to coproporphyrinogen I molecules. During this process some compounds with less than eight, more than four, carboxyl groups are formed which are in turn oxidized to their respective porphyrins and then excreted in urine. There is not much information about the mode of release of these abnormal porphyrins from the immature red blood cell elements. The hemolysis seen in this disease is responsible for only a small fraction of the excreted porphyrins;[34] the erythroid hyperplasia resulting from hemolysis seems to contribute to this excretion.

Coproporphyrinogen I and its oxidized form, coproporphyrin I, are also excreted as no metabolic way exists for this isomer to proceed

beyond this step, i.e. to be decarboxylated and then oxidized to the corresponding protoporphyrin. However, in certain other types of congenital porphyria, such as the disease seen in congenitally affected cattle, the erythrocytes contain excessive amounts of protoporphyrin. This is in contrast to the human disorder where protoporphyrin values in the erythrocytes are found to be normal.[33] In congenitally affected cattle the erythrocytes seem to contain the type I isomer of uroporphyrin and a protoporphyrin of undetermined isomer type. Though it cannot be stated conclusively it is altogether possible that this latter compound may belong to an abnormal type I series, and that cattle may have enzyme systems which can decarboxylate coproporphyrinogen I to a corresponding protoporphyrin.[27]

2. Protoporphyria Erythropoietica. Seven cases belonging to this category have been published so far and, as can be seen from the case cited by Magnus et al.,[36] the excreted amounts of urinary coproporphyrin, porphobilinogen and δ -amino levulinic acid were all within normal limits but the excretion of fecal coproporphyrin and protoporphyrin were consistently and grossly raised. The erythrocytes contained greatly increased amounts of coproporphyrinogen and protoporphyrin although uroporphyrin was absent except for a trace amount found on one occasion. During a period when the patient was highly photosensitive, bone marrow biopsy was performed. Under ultraviolet light the biopsy specimen was seen to contain numerous red-fluorescent normoblasts.

In congenital erythropoietic porphyria cases, porphyrin isomers belonging to the abnormal type I are synthetized in large quantities. The defect is thought to become operative at the stage of the condensation of precursor porphobilinogen molecules to uroporphyrinogen which, in this disease, leads to the formation of the abnormal uroporphyrinogen I isomer. This compound, once formed, is either decarboxylated at the acetyl side chains to give coproporphyrinogen I or is oxidized to uroporphyrin I. As no enzymes exist that would decarboxylate coproporphyrinogen I to its corresponding protoporphyrin, both uroporphyrin and the oxidized end product of coproporphyrinogen, coproporphyrin, are excreted.

In Magnus' case, however, the coproporphyrin isolated belonged to the normal type III series and a significantly increased formation of protoporphyrin (the normal isomer protoporphyrin IX) was detected. The possibility of lead poisoning was excluded by determination of lead levels in the urine and plasma. In view of these facts and since

this form of porphyria is manifested largely in the erythropoietic system, it is assigned to the hitherto undescribed category of «protoporphyria erythropoietica.»

The biochemical defect leading to the excessive production of coproporphyrin and protoporphyrin belonging to the normal isomeric series is not known. However the biochemical picture is entirely different from that in porphyria erythropoietica, and, in fact, these two diseases have nothing in common except the site of the disturbance which is the erythropoietic system in both cases.

Chemical Findings in Hepatic Forms of Porphyria

Several different types of porphyrin disease are included under this heading but all are characterized by the presence of large amounts of porphyrin in the liver and impairment of hepatic function. The classification offered in this paper is, of necessity, a provisional one. Further division of this group must wait until the underlying metabolic defects, in the various forms of the disease which have been reported, are more clearly understood.

1. Porphyria Turcica. This is a new type of porphyria which appeared in southeastern Turkey in 1955. Almost 5000 individuals were reported to have been afflicted in the epidemic which was found to have been related to the accidental ingestion of hexachlorobenzene treated wheat by the victims. The reader is referred to the work of Doğramacı, et al.[37, 38, 39, 40, 41] for extensive discussion of the disease.

In the cases of this type of porphyria so far described, increased fecal excretion of protoporphyrin, coproporphyrin and uroporphyrin was manifest, in contrast to the situation in the Swedish type of porphyria which will be discussed next. Porphyria turcica cases resemble more closely the cases described by Dean and Barnes[21] of adult hepatic porphyria observed among the South African white population. However in contrast to both the Swedish and South African varieties, the urinary excretion of δ -amino levulinic acid and porphobilinogen was within normal limits in the Turkish cases even during the acute phase of the disease.

In porphyria turcica cases the porphyrin isomers that were isolated from stool belonged to the normal isomer type III with some type I isomer also present. In the Turkish type of porphyria the liver seems to be the main site of metabolic derangement. Unlike both

TABLE 1

BIOCHEMICAL FINDINGS IN PORPHYRIA TURCICA

		Patients					
		C. T.	S. A.	V. C.	S. C.	H S.	S. G.
Fecal porphyrin microgram/ 100 Gm. wet weight	Copro	168	3917	2916	1937	2600	3917
	Proto	27	408	511	116	291	150
	Uro	73	2093	1375	1185	1000	1156
Urine porphyrin microgram/ day	Copro	229	473	913	130	195	1596
	Uro	1321	6134	8248	1560	4021	19950
	P.B.G.	0	0	0	0	0	0
	ALA	192	150	294	102	204	231
Packed cells microgram/ 100 cc.	Copro	1.2	1.7	1.7	2.5	9.6	2.9
	Proto	25	144	14	27	16	83
	Uro	1.4	1.4	1.5	1.7	2	2.1
Serum microgram/ 100 cc.	Copro	2.0	4.5	22	4.8	5.6	18.1
	Uro	3.8	8.6	12.4	4.5	1.4	24.8

Copro : Coproporphyrin
Uro : Uroporphyrin
Proto : Protoporphyrin
P.B.G. : Porphobilinogen
ALA : δ amino levulinic acid.

Swedish and South African types of hepatic porphyria, porphyrin precursors are not excreted in abnormal amounts in the urine and feces in the Turkish type. Although there is excessive deposition of porphyrins in the liver no evidence could be obtained for their presence in the erythropoietic cells of the bone marrow. The laboratory findings discussed in these cases correlated well with the urinary and fecal porphyrin excretion data obtained for porphyric rabbits which had been treated with hexachlorobenzene.[42, 43] However in rats, the excretion pattern and perhaps the lesions produced by experimentally administered hexachlorobenzene differ from those observed in clinical cases among human beings.[44] Increased excretion of δ-amino levulinic acid and porphobilinogen in urine was observed in hexachlorobenzene poisoned rats, whereas this could not be demonstrated in porphyria

turcica cases. Whether this is due to a species difference, or whether hexachlorobenzene produces a different type of lesion in rats is something to be clarified.

The exact nature of the metabolic defect is not known, but it is altogether possible that a negative feedback mechanism may be operative in this type of porphyria together with other so-far uninvestigated factors related to the control mechanism of the porphyrin synthesis. For example, Lascelles found that when *rhodopseudomonas spheroides* cells were grown in the light but under iron deficient conditions, they accumulated large quantities of porphyrin in the medium; addition of iron resulted in a decreased production of porphyrin. Hemin has been observed to repress the formation of two enzyme systems required for porphyrin synthesis, namely δ -amino levulinic acid synthetase and δ -amino levulinic dehydrase.[45] The inhibitory action of hemin has been demonstrated in crude extracts of this organism and Burnham has even demonstrated it on a 20-fold purified δ -amino levulinic acid synthetase.[46] This negative feedback mechanism may represent a very important regulatory mechanism in heme synthesis, particularly if it can be proved to occur in other tissues and with sufficiently pure preparations of the enzyme. Such feedback mechanisms have been shown to be operative in other metabolic pathways and inborn errors of metabolism resulting from imperfectly functioning feedback mechanisms have been reported. A well known example in man is orotic aciduria.[47] Negative feedback mechanisms in various microorganisms have been shown to control the formation of purine precursors,[48] arginine[49] and isoleucine.[50] A better understanding of this feedback control may give clues to the excessive porphyrin formation observed in hepatic forms of porphyria.

Factors such as reduced reductive capacity of the cell may also have a part in producing the clinical picture. In regard to this possibility the experiments conducted by Wolf et al.[51] are pertinent. These authors could not find any change in the reduced glutathione content of the liver in rats that had become porphyric due to hexachlorobenzene intoxication. This conflicts with the view that the main disturbance in hepatic forms of porphyria derives from a defect in the cellular antioxidant systems which prevent the oxidation of porphyrinogens to porphyrins. Williams has shown that halogenated benzene compounds are conjugated and excreted as mercaptide derivatives. This supposedly led to the transient reduction of reduced glutathione in the liver. However hexachlorobenzene intoxication was not found

by Wolf et al. to produce a similar decrease in hepatic glutathione content. Similar investigations need to be carried out, in other species, to determine whether such a correlation between cellular reductive capacity and porphyria does indeed exist.[52]

In conjuction with hexachlorobenzene intoxication the role of certain other drugs with a reputation for inducing porphyria in susceptible subjects, or experimentally in animals, will be discussed although no direct correlation exists between this group of drug induced porphyrias and porphyria turcica. In fact patients with this latter disease showed no untoward results after use of these drugs.[53] However the biochemical pathway underlying this group of diseases is as mysterious as that of porphyria turcica. Better understanding of the action of hexachlorobenzene, certain barbiturates and sedormid may contribute to the solution of various biochemical problems related to the synthesis of porphyrins and its control mechanism.

Other Drug-Induced Porphyrias. The oldest drugs with a reputation for causing porphyrinuria are sulfonal and trional. Garrod reported patients who developed porphyria after the use of such drugs and referred to other cases previously reported. At that time the inadequacy of the techniques used, and the lack of knowledge about the various porphyrins, prevented identification of the porphyrins involved. It is possible that sulfonal and trional were merely the agents which precipitated a mild or severe porphyria in constitutionally liable subjects. Waldenström observed that administration of trional to apparently healthy near-relatives of patients with congenital porphyria was followed by the appearance of uroporphyrin in urine.[54] It has been very hard to reproduce this type of porphyria in experimental animals. Administration of near lethal amounts of these drugs to rabbits failed in most cases to produce manifest disease symptoms[55] and in the mild cases that occurred occasionally the workers could not obtain enough porphyrin to examine its character.

Much more striking changes in porphyrin metabolism have been attributed to sedormid (allylisopropylacetylcarbamide).[56] It was found that upon feeding sedormid to rabbits for several consecutive days the daily urinary excretion of ether-insoluble porphyrins increased from 0.02 to 60 milligrams. This was paralleled by an increase in porphobilinogen excretion. In studying the organs of sedormid-poisoned rabbits, a marked increase in porphyrin and porphobilinogen was found in the liver.[57] In contrast to the liver findings, no porphobilinogen and no increase in porphyrin concentration was detected

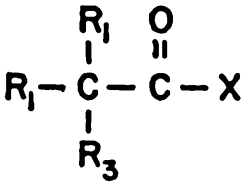
in the erythropoietic system. The urinary pattern of excretion was characterized by increased excretion of porphobilinogen and uroporphyrin; increased coproporphyrin and protoporphyrin were noted in bile and feces. Schmid and his group made the very interesting observation that the administration of sedormid produced a rapid and progressive fall in liver catalase activity.[57] This is not due to inhibition of catalase activity, as occurs with another porphyria-causing drug: 3-amino, -1, 2, 4- triazole, which inhibits catalatic activity noncompetitively.[58] Sedormid probably interfered with the formation of the enzyme, or increased the breakdown of catalase, as indicated by the marked reduction of inclusion of labeled glycine into catalase heme in sedormid treated rats.[57] Sedormid apparently had little effect on the catalase formed in red blood cells, because administration of the drug for as long as 42 days did not produce an appreciable decrease in the catalase activity of bone marrow and red blood cells. The exact mechanism leading to decreased catalase activity is not known, however, such a blockage exists and no other effect of this drug on other heme enzymes can be demonstrated.[57] Schwartz et al. also described the accumulation of «green pigments» in the liver parallel to the accumulation of porphyrins.[59]

Pharmacological studies have been carried out with drugs related to sedormid which led to the discovery of a basic structure that gave rise to the development of experimental porphyria in animals. This structure has been confirmed by at least two groups of workers, [60, 61] and is described by the basic formula presented in figure 2.

As previously mentioned, certain barbiturates can also precipitate or produce attacks of porphyria. Stich et al.[60] have found that allylethyl barbituric acid, allylisopropyl barbituric acid, allylbutyl barbituric acid, allylisobutyl barbituric acid, and diallyl barbituric acid were particularly effective in this regard. The formula of these drugs is shown in figure 2.

The effect of these compounds is comparable to that of sedormid. Another drug which can also lead to porphyria and which can depress liver catalase activity[62] is 3-amino-1, 2, 4- triazole. Margoliash et al. reported the non-competitive inhibition of catalase by this compound.[58]

2. Swedish Type of Porphyria. This form of porphyria is extremely rare in childhood and generally makes its appearance after puberty. The biochemical defect responsible for this condition is far



R_1 = CH_2= CH − CH_2−

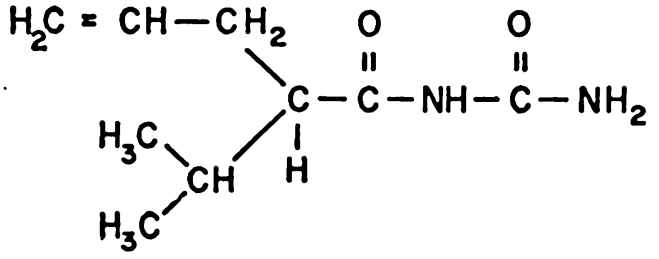
R_2= Alkyl −

R_3= H− (or Alkyl−)

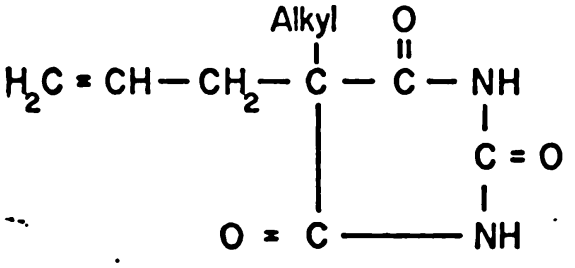
X = − NH_2 or

R_3+ X = − CO − NH − CO − NH

chemical structure of porphyrogenic compounds.



chemical structure of sedormid.



chemical structure of porphyrogenic barbiturates.

Fig. 2. The chemical structure of certain porphyrogenic compounds, barbiturates and sedormid.

from being clearly understood although it has been established that here again the liver is the main site of metabolic disturbance.

The most important chemical finding is the increase in urinary porphobilinogen excretion noted during the acute manifestations of the disease; values of 50 - 170 milligrams of excreted urinary porphobilinogen have been reported during acute attacks.[54, 8, 63] These values are reported to be lower during periods of latency. Together with porphobilinogen, δ-amino levulinic acid is excreted in large amounts in the urine; as much as 180 milligrams per day of this compound is excreted during attacks, much less during the remission phase. Other porphyrins, including porphomethenes and porphodimethenes, seem to be present in the urine of these patients.[64, 65, 66] However these porphyrins were rather poorly defined in the investigations reported and controversial results were obtained. There is little doubt that most of the porphyrins are actually formed after the urine is passed, from porphyrin precursors or even from porphobilinogen. This may account in part for the change of color noticed in such urine upon standing and also for the seemingly contradictory results obtained by various workers.[67, 68, 69] It also seems that both type I and type III porphyrin isomers are present in the urine.

Not much fecal coproporphyrin or protoporphyrin is found in this condition.[63] A somewhat increased amount of porphyrin precursors, uroporphyrin and coproporphyrin, is noted in the liver,[34, 70] together with large amounts of porphobilinogen. However similarly increased amounts of porphobilinogen are not encountered in the spleen, bone marrow or muscle.

In this type of porphyria the liver is the main site of metabolic disturbance and, in contrast to the acute erythropoietic type, porphyrin precursors are present in large amounts in the liver and are excreted to a great extent by the kidneys. The exact nature of the metabolic defect is not known but it has been proposed that either the formation of δ -amino levulinic acid is increased or its normal utilization, its metabolism in the non-porphyrin pathway, is diminished, causing porphyrins and their precursors to be overproduced. This might well be true if a block in the non-porphyrin pathway of δ -amino levulinic acid could be demonstrated. In sedormid-induced porphyria, the increased formation of porphyrins is associated with decreased purine synthesis;[71] this was amply demonstrated in experiments with chick embryos where it was found that exposure to sedormid led to decreased formation of purines in the eggs and to a con-

comitant rise in porphyrins.[72] The percentage of δ -amino levulinic acid converted to urinary porphobilinogen is several times greater in patients with porphyria than in normal subjects.[6] The inhibition of liver catalase formation has been demonstrated in animals with experimental sedormid-induced porphyria,[73] but the relationship of this defect to the overproduction of porphyrins has not been well worked out. Furthermore determinations of liver catalase activity in human porphyria have failed to show any significant decrease in the enzyme activity.[1] Additionally no increase in the amount of coproporphyrin and protoporphyrin formation has been noted in this type of hepatic porphyria although such an increase has been observed in patients with the South African type of porphyria. Had there been a block in heme-protein synthesis, as seen in sedormid porphyria in the formation of liver catalase, one would have expected to see a significantly increased concentration of coproporphyrin and protoporphyrin as well.

It is possible that the feedback mechanism proposed earlier may well play a part in the pathogenesis of this disease. At the step of δ -amino levulinic acid synthesis the cell may possess a regulatory, or negative feedback mechanism by which porphyrin and heme-biosynthesis is controlled. If the synthesis of heme is blocked this may result in increased synthesis of δ -amino levulinic acid and porphobilinogen, as was indicated by Burnham's demonstration of the inhibition of δ -amino levulinic acid synthetase enzyme by protohemin IX. The situation encountered here may well be analogous to that in orotic aciduria. Proof of these speculations must of course wait for future work.

It was noticed that, during latency periods, patients with Swedish porphyria continued to excrete more than the usual amounts of δ -amino levulinic acid and porphobilinogen.[77, 63] This suggests that, even during remission periods, the metabolic disturbance is still active but the reasons for the resulting clinical fluctuations are not yet clearly understood. Further work is needed on this aspect of the disease and also on its relation to gonadal activity; as the onset comes, in most cases, after puberty.[7]

3. South African Type of Porphyria. This disease is common among European settlers in South Africa and its origin can be traced to a pair of immigrants from Holland who married at the Cape of Good Hope in 1688.[74, 21]

This type differs chemically from the Swedish type in that it involves a greatly increased excretion of coproporphyrin and protoporphyrin in the feces.[63, 75] However, during acute attacks, increased urinary excretion of porphobilinogen and δ -amino levulinic acid is noticed, as in the Swedish type. Whether the biochemical defect is the same in both types of the disease is not clear.

4. Cutaneous Porphyria. In most cases that are classified as cutaneous porphyria, skin lesions are the predominant findings. Chemically, increased urinary excretion of uroporphyrin and coproporphyrin is noted, whereas fecal porphyrin is not significantly augmented. Increased porphobilinogen excretion is consistently absent in the Italian cases reported by Radaeli;[76] however, increased excretion of porphobilinogen and δ -amino levulinic acid is occasionally observed in the Bantus.[77]

5. Multiform Porphyria. Patients in this group exhibit a very mixed symptomatology and no uniform pattern of pyrrole-porphyrin excretion can be found. It should be admitted that most of the cases are grouped under the multiform heading simply because there is no other choice at the present stage of understanding of the problems associated with porphyrin synthesis.

Summary

The normal biosynthetic pathway of porphyrins and their metabolic turnover are reviewed. The chemical findings in various forms of porphyria are presented and the biochemical defects and mechanism that may lead to the observed biochemical pathology are outlined.

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