STUDIES OF PANTOTHENIC ACID METABOLISM

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Introduction

My first experience with the problem of differing pharmacologic effects produced by chemical compounds with closely related molecular structure was reported in a study of the vasodilating capacity of nicotinic acid and related pyridine chemicals.[1] Several of the compounds we tested were mildly toxic. Woolley[2a] demonstrated the harmful effects of 3-amino pyridine in experimental animals. The concept of molecular antagonists is that kindred chemical compounds compete for space on the surface of protein enzymes. If active compounds got priority, enzyme activity progressed naturally. If the available places are all taken by compounds which preempt surface space but have no active portion free enzyme action halts. An early application to clinical problems was Woods[2b] observation of the antagonism by paraaminobenzoic acid of sulfanilamide in inhibiting bacterial growth. Reasoning by analogy, I tried to find compounds which, by slowing or stopping the action of B-complex vitamins might give a clue to early and single vitamin deficiencies. Pyridine-3-sulfonic acid, which bears the same relation to nicotinic acid that sulfonilamide bears to PABA, was given to a number of patients with pellagra. No harmful effects occurred but the studies were not adequate to show whether the principle was wrong, the material inert or not given in adequate amounts or time. Later the soundness of the idea was demonstrated by the development of folic acid antagonists and the use of pyridoxine antagonists to produce human pyridoxine deficiency.[2c]

In spite of the fact that pantothenic acid was established as essential for growth and health of experimental animals and certain lower organisms, nothing was known of its possible role in human nutrition and metabolism when we undertook our studies. The increasing importance of coenzyme A in many important metabolic and enzymatic actions was being unfolded.

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After a period of two years of investigation using pantoyl-taurine which proved to be inert, we began a series of observations using omega methyl-pantothenic acid and a purified ration devoid of pantothenic acid.

In previous investigations of pantothenic acid metabolism in human subjects, Bean, Hodges, Daum, and Thornton[1, 2, 3] described metabolic abnormalities and clinical signs which occurred in normal young men given the pantothenic acid antagonist omega-methylpantothenic and a diet devoid of pantothenic acid. The signs were those of an illness characterized by torpor, apathy and depression; cardiovascular instability especially in the erect position; a neuromotor disorder with paresthesias, burning sensations and muscle weakness; abdominal pains and disturbance of alimentary function; and frequent infections. Biochemical alterations included an inconstant reduction in the percentage of PABA excreted in the urine in the acetylated form; irregularities in glucose tolerance and increased sensitivity to insulin; a failure of ACTH to induce eosinopenia; an irregular reduction in 17-ketosteroid excretion; and the development of a histamine refractory achlorhydria without any disturbance in gastric motility. The illness induced in the subjects caused us to abandon the planned recovery period, and employ cortisone, a rich diet and added vitamins. Because the abnormalities had not been anticipated, because of the hazard and the inherent difficulties in such studies in human subjects, the experimental design was not without limitations very obvious in retrospect. For these reasons it was desirable to repeat and extend the observations.

Experimental Design

We planned the present study with more elaborate control periods (Figure 1). Period I was designed to establish baselines and the test routines while

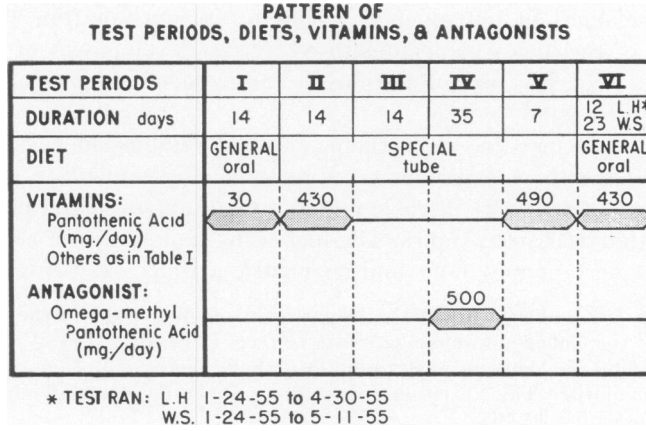

FIG. 1. The experimental design

employing a general hospital diet. Period II was to test the adequacy of the formula with mineral and vitamin supplements and the effects of intubation since the formula was intolerable by mouth. Period III was designed to see whether the deficient diet, without the antagonist, might induce changes in two weeks. Each of these periods in plan and in fact was two weeks long. Period IV with the deficient diet plus 500 mg. of omega methylpantothenic acid, was planned for four weeks. At the end of this time we extended it for one week because the clinical state of the subjects suggested that they had not reached a danger period. Their symptoms of illness were not as striking as those in the earlier study after four weeks of deficiency. Because 10 weeks is about the limit of endurance in taking the tubes, the planned recovery Period V with the formula plus pantothenic acid but without the antagonist was reduced from two weeks to a single week. During this week the general condition of one subject improved whereas the other had a progressing emotional disorder. Only when the chemical data were assembled did we find that some of the changes had not returned to normal till Period VI, with a normal diet.

Procedures

The formula, vitamin and mineral supplements are indicated ie Table I. The pattern is similar to that reported previously.[1-3] The calorie intake in

TABLE I

Composition of Formula		Vitamin Supplement		Composition of Salt Mixture	
			mg.		*mg.*
Granulated sugar...	290 gm.	Thiamine hydrochloride.....	1.2	Calcium biphosphate.....	67.9
Cornstarch.....	75 gm.	Riboflavin.....	1.8	Calcium lactate..	163.5
Water.....	750 ml.	Pyridoxine.....	2.0	Ferric citrate....	14.9
Vitamin-free casein*.....	125 gm.	Ascorbic acid....	50.0	Magnesium sulfate.....	68.5
Corn oil.....	90 gm.	Niacin.....	12.0	Dibasic potas- **	
Cystine.....	750 mg.			sium phosphate	119.9
Vitamin A.....	5330 U.S.P. units			Sodium biphosphate.....	43.6
Vitamin D.....	1070 U.S.P. units			Sodium chloride.	21.8
$NaHCO_2$.....	10 gm.				
Calories.....	3000				

* Contains 20 mcg. Vitamin B_{12} by assay.

** Coffee and soft drinks, allowed ad libitum but measured, brought the average intake of potassium to 1.2 grams a day.

We were enabled to do the study with prisoners as volunteers through the thoughtful cooperation of Mr. Roy Purcell, warden of the Iowa State Reformatory at Anamosa, and with the authorization of the State Board of Control, Mr. Henry W. Burma, Chairman.

Periods I and VI was the same as during the time the formula was used. W.S. was the only normal subject on whom we collected complete data. In another young man the experiment was stopped after two weeks of Period IV because of a breach of control. The other subject, a 31 year old white woman had obesity and the adrenogenital syndrome. We wanted to see whether the antagonist and deficient diet might reduce her adrenal cortical overactivity. We used a 1000 calorie reducing diet throughout the whole study. She got only a third of the quantity of formula but the same supplement of vitamins and minerals.

In our present studies we concentrated on fluid and electrolyte problems while repeating many of the studies we had done previously.

The original plan was somewhat different for the patient L.H. Her control period on a general diet lasted 8 weeks from January 12, 1955, to March 7, 1955. On March 7 she started the pantothenic acid deficient tube formula and 500 mg. of omega methyl-pantothenic acid. She then had a 5-week period exactly like that of Period IV of W.S. Period V likewise was identical with that of subject W.S. as was Period VI with the exception that in the case of L.H. it lasted only 12 days for W.S. it was for 23 days.

Results, Clinical

H.T. had no symptoms until the beginning of the second week of Period III. Then he first noted increasing fatigue, slight weakness, and transient unsteadiness upon standing upright. These symptoms, although non-progressive, were unrelenting. No personality change was noted. He remained pleasant and cooperative. Four days after the start of Period IV, he first had flexor spasms of the right forearm and hand. Three days later, paresthesias of the upper and lower extremities began. Repeated neurological examination failed to reveal any abnormalities until 8 days after the start of Period IV when a slight but definite decrease occurred in the tendon reflexes on the right side. This progressed for three days and then remained stationary. No other symptoms or abnormal signs appeared. Subject W.S. was asymptomatic until the beginning of the second week of Period III. Then he noticed the onset of increasing fatigability, a generally tired out feeling, mild weakness, and unsteadiness of gait on arising in the morning. Three days after the beginning of Period IV, he had cramps of the right anterior thigh muscles and severe pain in the right Achilles tendon. At the end of the first week of Period IV paresthesias of the upper and lower extremities began. By the next day he was extremely torpid; and for the first time the tendon reflexes on both sides had decreased. Otherwise the neurological examination and physical examination were normal. Two weeks after the start of Period IV W.S. was troubled by severe muscular spasms of his hands, forearms and legs. Although the paresthesias persisted there was

no muscle tenderness. He felt that his over-all strength was improving. A week later he had less muscular cramping. The paresthesias persisted. He was much more lethargic and somnolent. He spent most of the time in bed. It was only with difficulty that he could be aroused in the morning. After four weeks of Period IV, for the first time a positive Trousseau sign was demonstrated and the tendon reflexes became still more inactive. Two days before the end of Period IV he had repeated bouts of nausea, increasing somnolence and moderately severe muscle cramping and paresthesias. One day after the start of Period V nausea became troublesome. It was only with difficulty that he could retain the tube feeding. Only 24 hours after Period VI was begun there was complete cessation of the paresthesias in subject W.S. and he became less somnolent. A week later the muscular cramping was gone and the Trousseau sign was negative. He felt wonderfully well. All lethargy, somnolence, and weakness were gone. Diminution of tendon reflexes remained.

L.H. remained asymptomatic until 11 days after the start of Period IV when she had muscle pains in the legs, calf muscle tenderness and slight unsteadiness of gait. No definite reflex changes were demonstrable. Seventeen days after beginning Period IV she had intermittent spontaneous carpopedal spasm in addition to severe "menstrual cramps" without bleeding. Thigh and calf muscles were extremely tender. Paresthesias had developed in the lower extremities. The remainder of the neurological examination was negative. After 23 days of Period IV she noted some lessening of the muscle cramps but the paresthesias persisted along with a definite increase in lethargy and somnolence. During the last week of Period IV she was nauseated and refused her tube feeding on 4 occasions. After four days of Period V her clinical status was unchanged except that some disorientation and hallucination occurred. She kept saying, "People are taking pictures of me." During the last few days of Period V she became extremely depressed, cried frequently, and often repeated, "I want to kill myself." After five days of Period VI she was completely asymptomatic. She felt better than she had for many months. No abnormalities of the tendon reflexes could be demonstrated.

No significant lability of pulse rate or change in blood pressure or pulse pressure occurred in any subject. Electrocardiograms failed to reveal any abnormalities in subject H.T. until the beginning of Period IV when the U-waves in the precordial leads became prominent. In W.S., repeated electrocardiograms were normal until the beginning of the second week of Period IV at which time T-wave and U-wave changes in the precordial leads were those seen in hypokalemia (Figure 2). In L.H. control electrocardiograms revealed a prolonged QTC interval and abnormal T-waves. As the experiment progressed she also developed additional abnormalities.

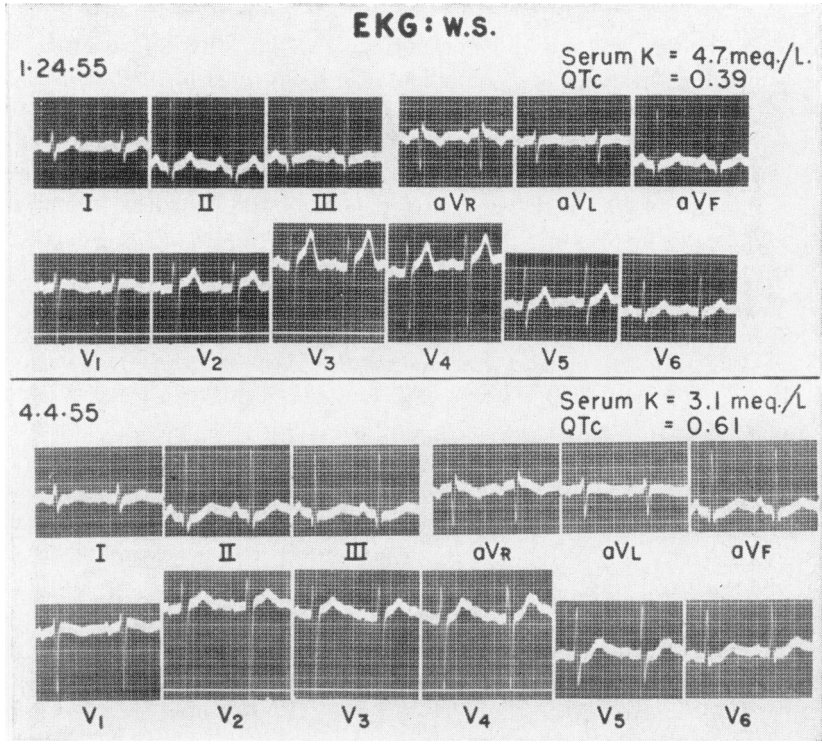

FIG. 2. The electrocardiograms before and during hypokalemia

Ballistocardiograms of H.T. were normal until the first week of Period IV when definite abnormalities appeared. In patient L.H. control ballistocardiograms of H.T. were normal until the first week of Period IV when definite abnormalities appeared. In patient L.H. control ballistocardiograms revealed bizarre complexes. In W.S., no abnormalities developed. Chest films failed to reveal any alterations. Gastrointestinal changes were similar to those reported previously.[3] During Period IV none of the subjects had upper respiratory infections or other manifestations of decreased bacterial resistance. W.S. and L.H. had a progressive increase in the sedimentation rate after the beginning of Period IV (Figure 3). It promptly returned to normal early in Period VI. No alteration in the sedimentation rate of subject H.T. occurred during the control period but it increased after one week of Period IV. During the experiment there was no significant change in the weight of subjects W.S. and H.T. L.H. lost weight on the 1000 calorie diet at the same rate in all periods. No significant alterations were noted in the hemoglobin, red blood count, white blood count or

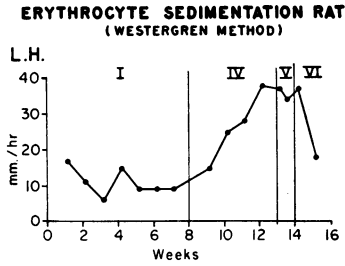

FIG. 3. The increase in sedimentation rate

urinalysis. Weekly eosinophil counts failed to reveal any significant variation.

Results, Metabolic

Eosinopenic response to ACTH

In one subject H.T. no significant alteration was noted. In both L.H. and W.S. there was a definite decrease in the eosinopenic response to ACTH beginning in Period IV and continuing through Periods V and VI (Figure 4).

Kepler-Power-Robinson Test

Subject H.T. had one abnormal test during Period II but all other tests were normal. Subject W.S. had an abnormal test during Period I and II

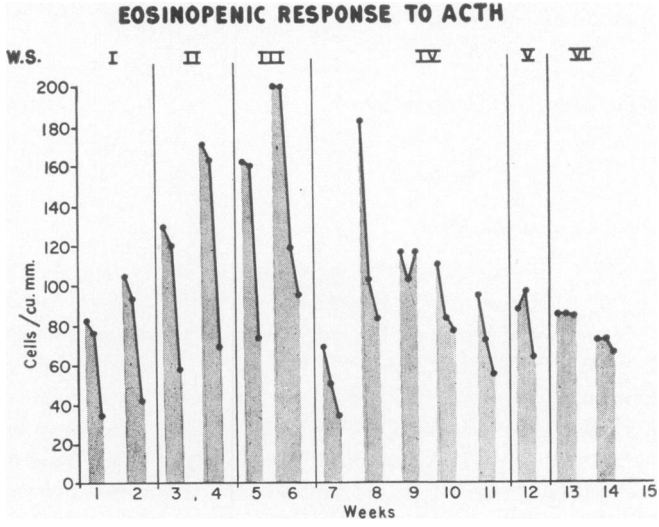

FIG. 4. The eosinopenic response to ACTH

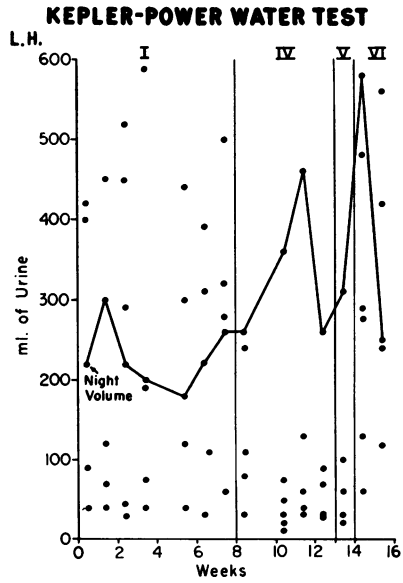

FIG. 5. Water excretion test

but subsequently it became and remained normal. A striking aberration occurred in L.H. Numerous tests during the control period were all well within normal limits. During Period IV she developed an extremely abnormal response as manifested by an inability to excrete the water load. The test remained abnormal until after the start of Period VI (See Figure 5).

Para-aminobenzoic Acid Acetylation

Acetylation of a standard dose of PABA did not change significantly in any subject throughout the study.

17-Ketosteroid Creatinine Ratio

In subject H.T. no significant change occurred. In subject W.S. a definite decrease in the excretion of 17-ketosteroids occurred toward the end of the Period IV. It persisted through Period V and the early part of Period VI. Before the study L.W. had a greatly increased 17-ketosteroid excretion. At the beginning of the control period para-aminobenzoic acid was given to test acetylation. As soon as PABA was given there was a prompt fall in the urinary excretion of 17-ketosteroids. (See Figure 6). It again became markedly elevated when para-aminobenzoic acid administration was stopped. While again excreting large amounts of 17-ketosteroids in the

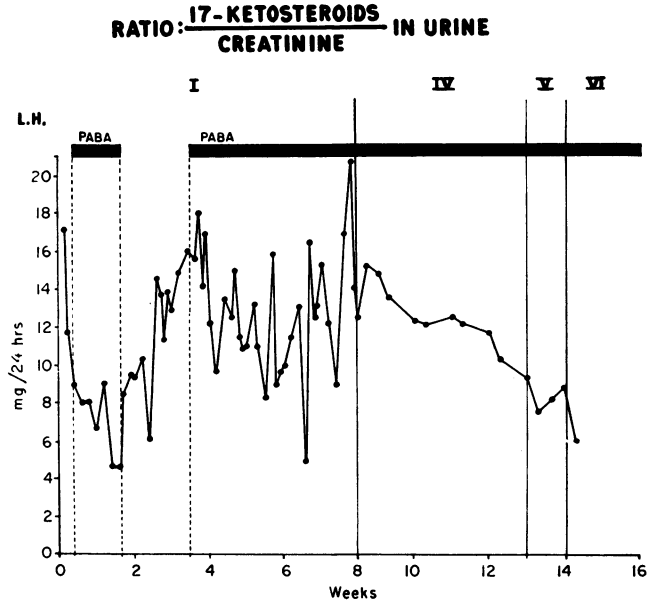

FIG. 6. Acetylation of PABA

urine, paraminobenzoic acid was again started. This was followed by a slight but definite decrease in the amount of excreted 17-ketosteroids. Excretion of 17-ketosteroids, however, was still above normal. A definite decrease in urinary excretion of 17-ketosteroids from this level was noted in Periods IV and V as well as Period VI in this subject.

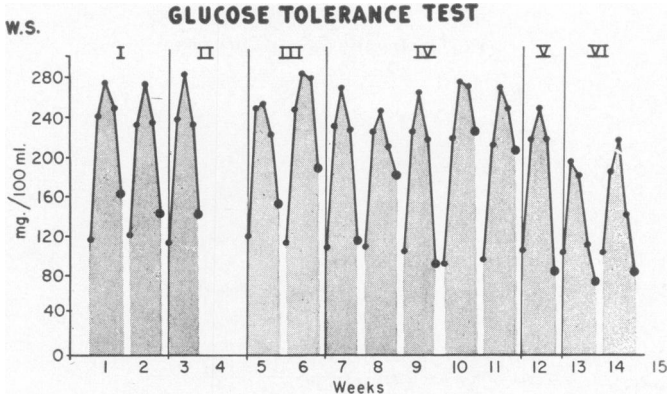

FIG. 7. Glucose tolerance test

Glucose Tolerance Test

In W.S., marked alterations occurred. Although he had an abnormality of his glucose tolerance test throughout the entire study, during the latter part of Period III and throughout Period IV, a new abnormality occurred (Figure 7). This was characterized by a persistent elevation in the 2-hour blood sugar determination. This alteration was lost promptly during Period V and did not recur. Likewise, in L.H. a similar alteration occurred. During Periods IV, V and the early part of Period VI an elevation of the 2-hour blood sugar occurred. The curve returned to the control form after the first week of Period VI.

Insulin Tolerance Test

In subject H.T. an increase in sensitivity to insulin was noted during Period III and IV with the 20-minute blood sugar much lower than in the control tests. In subject W.S. a marked increase in sensitivity to insulin

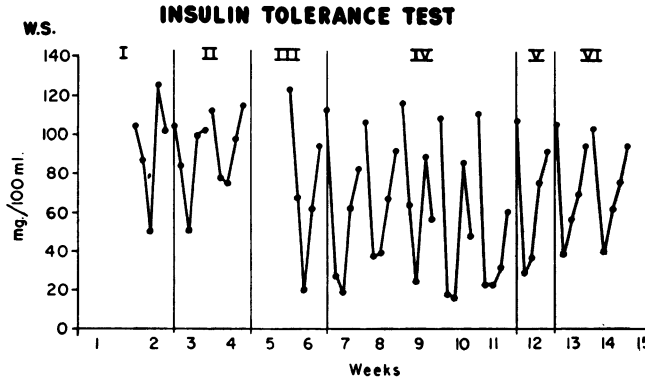

FIG. 8. Insulin sensitivity test

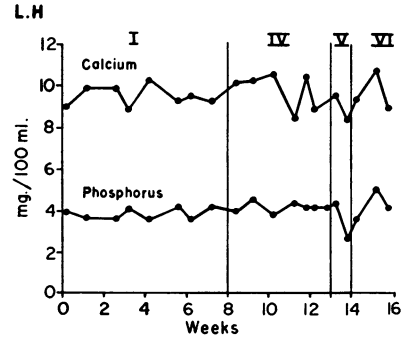

FIG. 9. Calcium and phosphorous

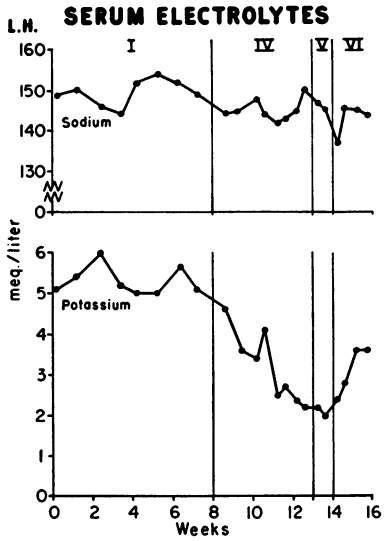

FIG. 10. Serum sodium and potassium

began in Period III and persisted through Periods IV and V (See Figure 8). L.H. likewise had a great increase in insulin sensitivity.

Serial determinations of serum calcium and phosphorus failed to reveal any alteration (Figure 9). W.S. had a slight elevation of alkaline phosphatase during the latter half of Period IV, which persisted during V and VI. Serial blood urea nitrogen determinations revealed no alterations from the control values nor did repeated sodium determinations. Alterations in the serum potassium serum chloride and CO_2 combining power were of striking magnitude. In subject H.T. a trend toward the development of hypochloremic alkalosis with hypokalemia began during the first week of Period IV. In subjects W.S. and L.H. extreme degrees of hypochloremic alkalosis and hypokalemia developed during Periods III, IV, and V with a rapid return to normal during Period VI (Figures 10 and 11).

Serum Protein Studies

In all subjects the albumin and total proteins remained normal. There were definite alterations in the electrophoretic pattern with an increase in the alpha I and alpha II globulins during the deficient periods in all three subjects with a return towards control levels during Period VI. In L.H. there was also a definite decrease in the beta globulin during Period IV. No significant alterations in total protein, or albumin or globulin occurred (Figure 12).

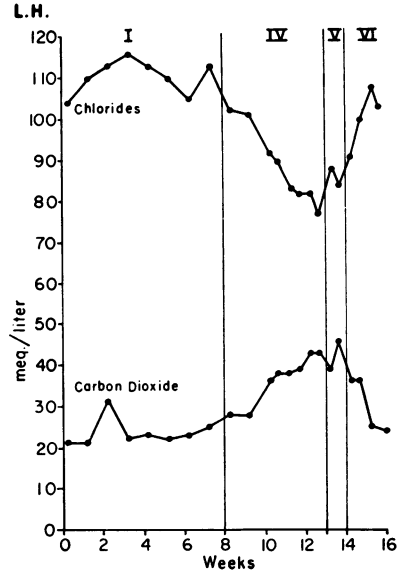

FIG. 11. Serum chlorides and carbon dioxide

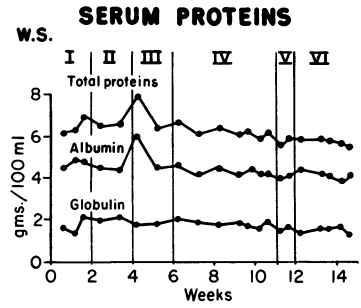

FIG. 12. Plasma globulin fraction

Liver Function Tests

Liver function studies including the bromsulphalein test, thymol turbidity, cephalin flocculation, zinc sulfate turbidity and serum bilirubin determinations were performed every two weeks on subject W.S. No alterations were noted.

Cholesterol and Cholesterol Esters

Figure 13 gives a representative curve for the changes in cholesterol and esters. There was a sharp fall in both components while the tube feedings were used, with a return to normal when the normal diet was restored. Total serum fat and phospholipids showed no significant changes.

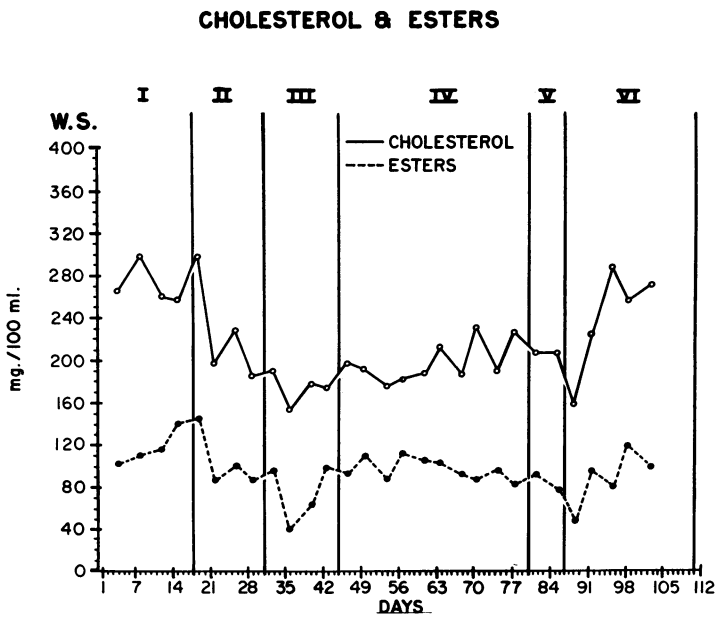

FIG. 13. Cholesterol and esters

Prothrombin Activity

A ratio of the control prothrombin time to the patients' prothrombin time times 10 was arbitrarily selected as a measure of prothrombin activity. Using this ratio there was a definite decrease in prothrombin activity during the deficient periods in subject W.S. with a rather prompt return to control levels during Period VI. A similar but somewhat less marked decrease in prothrombin activity occurred in subject L.H. (Figure 14).

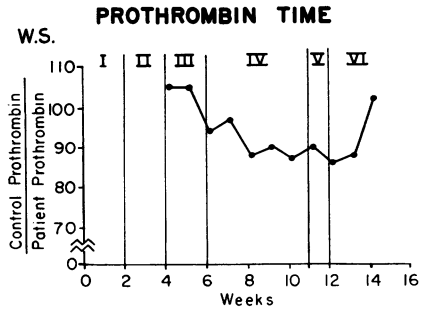

FIG. 14. Prothrombin time

Discussion

In our previous studies, we were impressed by the clinical illness which occurred when the deficient diet and omega methylpantothenic acid were used together. In the present study there was a similar weakness, fatigue and decrease in spontaneous activity. Mood changes, though they did develop, were not as impressive as those observed in previous tests. Dizziness and unsteadiness were severe. Sometimes changes in position were awkward because of the temporary instability. At least some of the fatigue, weakness, and awkwardness seemed to be caused by the neuromuscular disorder. It is impossible to say whether mental disturbances were responsible for one of the subjects going A W O L but such behavior is hardly surprising under the circumstances of the test. One subject developed a psychosis with paranoid features which continued into Period V but rapidly vanished during Period VI. The torpor and somnolence began to diminish very soon after the antagonist was stopped and pantothenic acid was given in Period V, suggesting strongly that the induced state of pantothenic acid deficiency was responsible for such changes. The neuromuscular abnormalities consisting of paresthesias, weakness, cramps, tenderness and alteration in tendon reflexes, muscle tenderness and the positive Trousseau sign did not all clear up till Period VI but there was a very definite improvement in Period V when pantothenic acid was substituted for the antagonist.

The subjects in the present test did not exhibit much vascular instability, postural hypotension, labile pulse, or easily provoked tachycardia which had all been so prominent in earlier studies. The ballistocardiograms became abnormal in one subject. Having been abnormal in the control period of another subject, they changed towards normal. Such observations do nothing to clarify the nature of the circulatory status of the subjects or the vagaries of the ballistocardiogram.

There was no tendency for the subjects to have infection though the conditions of the metabolic ward and of the entire experiment were unchanged from previous tests when infections prevailed during the period of deficiency. There was, however, a striking increase in sedimentation rate which returned to normal in Period VI. The reason for the increased speed of sedimentation is not known.

Several metabolic changes were studied in detail. In subject W.S., the eosinopenic response to ACTH did not decline until Period IV when the antagonist was given. By the third week of the deficient period, the response had diminished conspicuously. In contrast to our previous observation, this did not come back to normal during the recovery period. The failure of restoration to normal when pantothenic acid was given and when the diet was normal may mean that the mechanism responsible was disturbed more seriously than in the earlier studies when the deficiency-and-antagonist period was 4 rather than 5 weeks in duration. Another possi-

bility is that cortisone, which was used at the termination of the experimental period in the earlier test, had some other effect on the adrenals. The capacity of one subject to excrete a normal amount of urine in response to an ingestion of a large quantity of water was seriously impaired during the deficiency period but came back to normal during Period VI.

The excretion of 17-ketosteroids is recorded in Figure 6. In one subject the initial levels were higher than normal and fell very promptly while PABA was given. They then returned to the previous level when it was discontinued. Subsequent administration was followed by a diminution of the ratio and then a tendency to level off at the previous control level. During the period of induced deficiency, there was a slow but rather steady decline which was still continuing at the time the last test was done during the recovery period. The significance of these findings is uncertain.

The changes in glucose tolerance were characterized by prolonged elevation of the blood sugar which even after two hours tended to stay up around 200 or higher particularly during the latter part of Period IV. This returned to normal during Period V and remained normal. Study of the insulin tolerance test revealed that with the deficient diet alone a sharp increase of the insulin sensitivity occurred; and this persisted throughout Period IV. It was slowly being restored towards normal in Periods V and VI.

Figure 10 shows the very significant fall in serum potassium and the relative stability of the level of serum sodium throughout the experiment. These changes in serum potassium occurred during a period when the intake of potassium was constant. Figure 11 shows the striking changes in blood chlorides and carbon dioxide. Unfortunately we do not have data from balance studies so cannot say whether there was a potassium diuresis, an absorption defect or whether potassium was stored in the body. The electrocardiographic changes indicate that there was indeed a cellular depletion of potassium. Since the absolute requirement of potassium is not known, the possibility of an inadequate supply exists.

Consistent, but not very extensive declines in the prothrombin occurred during the deficient period but there was no sign of any liver malfunction nor was clinical bleeding at any time a problem.

Speculation

This report deals with work in progress. Slowly we are improving the experimental design which we hope eventually will enable us to understand the nature of the changes we have produced. So far we cannot be sure that I) we have produced a defect in pantothenic acid metabolism by employing a metabolic antagonist which interferes with the diverse functions of coenzyme A, II) whether omega methylpantothenic acid is a more powerful toxic agent working as a general protoplasmic poison, or III) whether some unrecognized deficiency exists in the experimental diet.

As far as I is concerned, many of the changes induced are similar to those induced in animals by pantothenic acid deficiency. Perhaps too much emphasis should not be put on the fact that glucose tolerance returned to control levels in Period V without change in diet but when pantothenic acid replaced the antagonist (see Figure 7). Many other abnormalities did not disappear until Period VI. In retrospect Period V was too short. We must emphasize the well known fact that correcting a specific deficiency in diet does not necessarily correct a lesion induced by the deficiency. Too little, too brief or too late may explain failures in correcting what has been called the humpty-dumpty situation.[4] Final proof will depend on our ability to titrate the human deficiency syndrome, if such it is, to the stage where it is still quickly reversible by merely replacing antagonist with vitamin. The interpretation of any vitamin antagonist's action must be made with knowledge that many subtle metabolic bypasses may enable the cellular and humoral economy to make remarkable adjustments.

Not knowing, except by inference and extrapolation what coenzyme A and pantothenic acid do in human metabolism, interpretation of our data, at the present merely tentative stage of work in progress, is impossible. If one is willing to compare our observations with a miscellany of observations in a variety of animals, and some of man, it may be that potassium deficiency alone is adequate to explain many of the clinical and biochemical changes.[5-7] Our next tests will inquire into that possibility. We propose to study the effects of large doses of potassium at a time when it is low. Likewise, administration of Coenzyme A may tell us whether that substance can correct the metabolic errors. The possibility that our metabolic machine is related to stimulation of aldosterone production and reduced production cortisone-like compounds is a stimulating speculation.

If we have an unrecognized deficiency, a longer Period V should provide the clue.

Conclusions

Work on a syndrome induced in normal subjects, and one with evidence of adrenal cortical overactivity, confirms and extends our observations. A clinical state of lethargy, weakness, burning paresthesias and cramps with signs of tetany was observed. Low serum potassium might account for many of the clinical findings. Likewise hypokalemia, hypochloremia, hypochlorhydria metabolic alkalosis and a defect in carbohydrate metabolism were observed. Further studies are in progress to elucidate the mechanisms whose disordered function leads to these disorders.

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DISCUSSION

Dr. Henry St. George Tucker, Jr. (Richmond); I was not entirely certain about one point. Were these evidences of adrenal insufficiency produced equally by the diet and by the pentothenic-acid antagonist? Of course, the obvious question is, If they were produced by the antagonist, have you yet given it to a patient with overactive adrenals, a patient with Cushing's syndrome?

Dr. Joseph L. Lilienthal, Jr (Baltimore): I want to ask Dr. Bean, if I may, whether—in view of the changes in the serum carbon dioxide and potassium, which suggest the alkalosis which occurs in potassium depletion—you had a chance to secure the evidence of loss in potassium in the urine during this period or, by any chance, muscle biopsy, to see whether this affected muscles.

Dr. Irving S. Wright (New York): Inasmuch as Dr. Bean went to such extreme measures to produce this deficiency, I wonder if he has encountered any indications which he considers justifies the use of pantothenic acid therapeutically.

Dr. Julian M. Ruffin (Durham): I would like to ask Dr. Bean if the picture which he has just described ever occurs spontaneously in man in this part of the world? In other words, is a pantothenic acid deficiency of clinical importance?

Dr. William B. Bean (Closing): I did not have time to go into a lot of things. Others, undoubtedly, I did not make clear.

We have tried giving the antagonist in large doses to people on a normal diet. So far, over a period up to five weeks, we have not been able to induce a single one of the changes we have demonstrated in subjects on a deficient diet. We have not gone longer than six weeks on merely the deficient diet without the antagonist. Only after we have concluded the study we have in progress now may we be able to tell you whether a deficiency can be induced by depletion alone.

We have tried giving omega methylpantothenic acid to patients with advanced or terminal carcinoma, in the hope that we might be able to induce a medical and correctible adrenocortical deficiency; and we have given it to one person with adreno-

genital syndrome. In cancer we induced no clinical improvement; nor did we have laboratory evidence that we had done anything important metabolically.

As for Dr. Lilienthal's question, when we began to pick up the spilled and broken pieces and analyzed the data when the experiment was over, we realized we had left undone those things we ought to have done and had done those things which we ought not to have done; and we were sort of worried about it. We are now doing a balance study. This all happened on a relatively fixed and we hope normal intake of potassium and other electrolytes. We have studies going now on the urine and feces as well as what comes in. You might explain those data by saying (1) potassium was not absorbed, (2) that it was lost in excessive amounts in the urine. We have no reason to suspect that they lost any extra amounts in the stool.

It is obvious to those who are studying the latest medical reports in LIFE, TIME and other medical journals which keep us up to date (laughter) that, if I were an aldosterone man, I might say that these subjects might have been manufacturing extra aldosterone; in other words, we may have been stimulating one part of the adrenal cortex and diminishing another. At least, one can explain these things on the basis that there is too much aldosterone, or that there is simply a wastage of potassium from somewhere in the system. But we have not studied either urine or muscles.

The implication is that pantothenic acid is useful; but our terrible difficulty in trying to find a deficiency in anything which resembled food should be emphasized. We measured anywhere from two to three, four, or five times the actual pantothenic acid in the diet that the tables indicate is present in the food.

A warning, I think, should be voiced here. The work of Becker and Friedenwald in alloxan-induced diabetes in rats has demonstrated that those animals, when given a pantothenic-acid deficient diet, are spared some of the unhappy consequences of alloxan diabetes; when given pantothenic acid, the vascular changes occur either accelerated in time or worse in degree, suggesting that an imbalance may stir up the adrenal cortex under those peculiar circumstances.

Thus, because pantothenic acid is present in a liberal quantity in any conceivable diet—it is probably a waste of money to put in more pantothenic acid. We do not know enough about it. I trust that the vitamin magnates and manufacturers do not seize upon our data as reason to needle their already curiously needled vitamin packets with even more vitamins.

As far as Dr. Ruffin's comment is concerned, there has been in nutritional literature—I should say "malnutritional literature," if I may call it that (laughter)—over the last hundred years, a description of burning feet. Our subjects developed paresthesias which were precisely similar, as far as you can judge somebody else's burning feet, to what had been described in medical literature.

A man by the name of Gopalan, in 1946, described the burning-foot syndrome among Indians in India. These people were literally starving, and ate a most peculiar diet. The Indian diet, as you know, varies in all kinds of different directions. He gave them nicotinic acid, thiamine—I do not think he had any B-6 at the time—riboflavin singly and in combination. There were dozens of subjects; so he had a good experimental group. Nothing happened to the burning feet. Then he gave calcium pantothenic, and the burning feet no longer burned. I suspect that there may have been a spontaneous pantothenic-acid deficiency in these people.

The evidence, however, looked at from this distance, is merely a clinical inference. We have tried it on many people with various neuropathies, and have not seen that it has done any good.