mechanism in which liver cells play an important rôle, the effects of the bacterial invasion may be far-reaching and necessitate adjustments which tax the reserve power of the organism beyond the limit of its capacity.

The question is: are the effects shown by the polarograph the result of the infection or the manifestation of the chemical changes involved in the readjustment of the system to meet the new conditions imposed by the bacterial invasion. Obviously this question can not be answered fully with the available evidence. We do not yet know the full significance of the changes in protein as indicated by the current-voltage curves. From the resemblance of these with the curves obtained with cancer sera and also with the sera from a limited number of patients suffering from different inflammatory conditions it would seem that the effects we get are not entirely specific for pneumonia. Of course, further study may reveal some phase of the effect which is definitely related to the peculiar conditions of pneumonia and lead to a better understanding of the disease. We can not tell from the evidence if the changes in the proteins are of the same type and follow the same course. It is possible that the cleavage of the proteins follows a definite pattern laid down by the enzymatic tools furnished by the specific bacteria. From our results it appears that not only do the sera from the pneumonia dogs give progressively higher peptone values when denatured but also show increasing amounts of such degradation products in the whole sera with the progress of the disease.

Preliminary results indicate that the treatment of the experimental pneumonia with sulfanilamide and sulfapyridine did not affect either the height or the shape of the characteristic current-voltage curves. This was also true of normal dogs given these drugs. Their curves were similar to those previously obtained when no drug had been administered. Whatever the function of these drugs in curing the disease, there is no evidence that they influence the course of the disease as indicated by the protein changes manifested by the polarographic results. This may mean that the essential function of the drug is to aid in destroying the bacterial infection and thus bring about a check of the disease. The blood changes measured by the polarograph may be part of the result of the infection and may run a regular course which is not effected by the chemotherapy. Further information should throw more light on this phase of the problem.

Besides, the cystine values of the whole sera of the pneumonia dogs are lower than those for the normal animals. The cystine drops progressively with the advance of the pneumonia until it reaches a minimum at the height of the disease. Then, if the animal recovers, it begins to rise and continues upward until it reaches the normal level, when the dog is well. This runs parallel with the rise and fall in the typical peptone wave. It also seems from the preliminary data that with rapid onset and severity of the pneumonia there is a correspondingly rapid decline in the cystine values. The drop in cystine corresponds to the decrease in total nitrogen of the serum and is additional evidence that the sera of the pneumonia dogs contain less proteins than those of the normal animals.

These polarographic results are consistent with the evidence obtained from the x-ray photographs showing the progress of the pneumonia. With the increase in the congestion of the lungs there is a rise in the peptone wave or increase in protein degradation products. Correspondingly there is observed a fall in the cystine, this tending to reach its minimum value when the pictures show the congestion at its height. Similarly, when the x-ray pictures show the dog to be improving, that is a progressive resolution of the involved areas, the polarograms also show the animal to be returning to normal. In general the polarographic results seem to give a more accurate picture of the animal's condition, showing a steady progress toward normalcy when the x-ray pictures fail to show definite evidence of the disease. The animal appears well before the polarograms indicate this condition.

While these results are interesting and appear to be significant they are not complete enough to justify a conclusion as to the specific chemical changes in pneumonia and relate cause and effect in connection with the manifestation of the symptoms of the disease. The investigation continues and it is hoped that in time sufficient data will be secured to allow of a definite correlation of the chemical changes and the symptoms of the disease. Such results would help to provide a rational basis for chemotherapy and improve the efficiency in predicting what chemical agents will have the greatest chance of success in combating infectious diseases.

THE IMPORTANCE OF MICROORGANISMS IN VITAMIN RESEARCH

By Professor ROGER J. WILLIAMS
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THE beneficial relationships of bacteria and microorganisms to mankind are almost as well recognized as their harmful effects. In very recent years, however, these tiny distant cousins have become useful to man in an entirely new way.

The history of their usefulness in vitamin research goes back to 1901—ten years before the term "vitamin" was coined—when in Ide's laboratory in Louvain, Belgium, it was discovered that yeast plants require for their proper nutrition something which was then called "bios" but is now known to be essentially a mixture of several of the water soluble "B vitamins."

A relationship of "bios" to what we call vitamins was first definitely established when it was found in the author's laboratory about ten years ago (1930) that pure vitamin B_1 had, under appropriate conditions, a tremendously stimulating effect on yeast growth and hence was a constituent of the hypothetical "bios" discovered about thirty years before.

A second link between "bios" and vitamins has become apparent since pantothenic acid has been demonstrated to be a vitamin required by all animals which have been tested. This substance was discovered and isolated in the writer's laboratory, because of its stimulating effect on yeast growth and was undoubtedly a highly important constituent of the original "bios." It appears to be a universal constituent of all types of living matter and to be part of a metabolic mechanism common to all living things.

Three more links between "bios" and vitamins have been discovered within the past two years—two of them within a few months. Pure vitamin B_6, now called pyridoxin, was found to be effective as a stimulator of yeast growth, and hence it may be deduced that it was also a contributing factor toward the original "bios" effect.

One of the most recent findings relating "bios" to vitamins has to do with the very interesting substance "biotin" discovered and isolated by Kögl in Utrecht. This substance stimulates yeast growth under conditions quite different from those used in Ide's laboratory in 1901, but was nevertheless one of the several effective substances present in the original "bios" mixture. It, too, is a vitamin as has been demonstrated very recently by György and du Vigneaud, and the name "vitamin H" will now drop out of the literature and be replaced by the name *biotin*, which is that of a single definite substance.

The latest connecting link between the old "bios" and vitamins came with the discovery by Woolley that inositol is a vitamin required by at least one animal—the white mouse. Inositol as a chemical substance has been known for decades. It was discovered in the late Lash Miller's laboratory in Toronto to be a "bios" constituent over a decade ago, but it has been only within a few months that it has been found to be a necessary constituent of the food of an animal.

It is a highly significant fact that every one of the five pure substances known possess "bios" properties, namely, thiamin (vitamin B_1), pantothenic acid, pyridoxin (vitamin B_6), biotin (vitamin H), and inositol is, without exception, a vitamin required by animals. Conversely, most of the known members of what was earlier called the "vitamin B complex" have under proper conditions at least some "bios" activity. If vitamin chemists had years ago set out seriously to determine what "bios" is, they would have discovered most of the members of the "vitamin B complex" which are now known. Because testing with yeast, though not without pitfalls, is incomparably more rapid than testing with animals, it is reasonable to assume that progress would have been much more rapid using the "yeast route" than it has been using animals as test organisms.

But yeasts are not the only microorganisms which have yielded results which are valuable from the standpoint of vitamins and if the hypothetical vitamin chemists mentioned above had broadened the scope of their investigations and had investigated the nutrition not only of yeasts but of other non-pathogenic microorganisms (and excluded animals from consideration), they would have found *all* the members of the "vitamin B complex" which are now known. The vitamin-like properties of nicotinic acid (which as a chemical compound had been known for many years) were in fact first discovered by Knight in England, who found it to be essential for the growth of *Staphylococcus aureus.* This finding was rapidly followed by the discovery of Elvehjem and coworkers that nicotinic acid (or its amide) is the anti-pellagra vitamin and will cure blacktongue in dogs.

Riboflavin (for a time called vitamin B_2) is one of the "B vitamins" which under ordinary conditions has not been observed to affect yeast growth in any striking way. It, like pantothenic acid and several other "B vitamins," is required by lactic acid bacteria (among others) and it could have more readily been discovered using these organisms than by using animals.

It is hardly profitable, of course, to discuss at length what might have happened if vitamin research had taken a different course. It is interesting, however, to note that microorganisms have played a leading role in the discovery of four of the seven members of the "vitamin B complex" which have been isolated or identified as possessing vitamin properties, *i.e.*, pantothenic acid, nicotinic acid, biotin and inositol.[1] This is in spite of the relative lack of emphasis on microbiological nutrition.

It is not only in the discovery of vitamins that microorganisms have been and will be useful. They may be expected to play an important part in the discovery of how vitamins function in the various enzyme systems.

A most important requirement necessary for the

[1] Since the above was written, another member of the "B family" of vitamins, *p* aminobenzoic acid, has been identified by Ansbacher. It represents an additional case in which studies with microorganisms have led to the identification of a vitamin.

study of how and where vitamins function is that methods be available for their quantitative determination. Though each individual substance presents its peculiar problems it is safe to say that microbiological methods are destined to play a highly important role in the future determination of "B vitamins." The microbiological methods can be rapid and fairly accurate, and at the same time they can be applied to extremely small amounts of material.

Workers now associated with the writer have described acceptable assay methods for three of the "B vitamins," namely, riboflavin, pantothenic acid and biotin. Methods for others will be forthcoming shortly from our laboratory at the University of Texas. E. E. Snell, in the writer's laboratory, has been a pioneer in this field. The usefulness of these methods is particularly outstanding when the available material to be tested is far too little to be used in animal tests and even too small for most chemical tests, in case such are available.

Investigations with microorganisms have made it seem apparent that the "B vitamins" (or at least some of them) are more fundamental and of more far-reaching importance than the other vitamins. This is true in the sense that they function in the mechanisms of diverse forms of life. I believe there is no reason to think that vitamins A, D, E, K, or even C, function in the life of yeast cells, and for other microorganisms also they may be unimportant. The recent observation that cockroaches can be raised to maturity without vitamin A and that an assay of their bodies indicates its absence, is further evidence regarding non-universality at least of this one fat-soluble vitamin. On the other hand, so far as thiamin, pantothenic acid, nicotinic acid, biotin and riboflavin are concerned, the evidence indicates that they may be present and function in all forms of life from the highest to the lowest. These five have not been investigated with equal thoroughness. The two other "B vitamins," pyridoxin and inositol, have been investigated even less thoroughly but appear to be very widespread.

Summary: In spite of a general lack of emphasis on this field, yeasts and other microorganisms have been used in the discovery (and/or isolation) of four out of seven of the recognized members of the "vitamin B complex."[1] Their use in the study of how vitamins function shows much promise, particularly in that methods have been worked out or are in process of development for determining quantitatively on a micro-scale practically all the members of the B family of vitamins. Indications are cited that the "B vitamins" are unusually important, *i.e.*, for all forms of life, in contrast to the other vitamins which appear not to be universally distributed.

OBITUARY

HERBERT FREUNDLICH
1880–1941

HERBERT FREUNDLICH was born in Charlottenburg, Germany, on January 28, 1880, and died suddenly in Minneapolis, Minnesota, on March 30, 1941, of a coronary thrombosis.

Professor Freundlich was the son of Phillip and Ellen (Finlayson) Freundlich. He graduated from the gymnasium in Wiesbaden in 1898. He studied general science for one year at the University of Munich and then specialized in chemistry at the University of Leipzig under the distinguished leadership of Professor Wilhelm Ostwald. Here in 1903 he took his Ph.D. degree with a dissertation dealing with the coagulation of colloidal sols by electrolytes.

For the following eight years Professor Freundlich remained at the University of Leipzig, teaching analytical and physical chemistry, attaining the rank of Privatdocent in October, 1906, on the basis of his studies on adsorption from solution.

He was called to the professorship of physical chemistry and inorganic technology at the Technische Hochschule, Braunschweig, in the autumn of 1911. Here he remained until February, 1916, when at the invitation of Fritz Haber he joined the staff of the Kaiser Wilhelm Institut fur Physikalische und Electrochemie at Berlin-Dahlem to conduct and direct research work on the adsorption of war gases and to study charcoals and other adsorbents for use in gas mask canisters. In these studies he achieved notable success.

In January, 1919, he resigned his professorship at Braunschweig to remain permanently at the Kaiser Wilhelm Institut as chief of the Division of Colloid Chemistry and Applied Physical Chemistry. Still later he was appointed associate director of the Institut.

In 1923 he was made honorary professor of chemistry at the University of Berlin, and he received the same honor in 1930 from the Technische Hochschule of Berlin. In 1925 he accepted the invitation of the University of Minnesota and the Colloid Committee of the National Research Council to be guest scholar at the second National Colloid Symposium held at the University of Minnesota, and he remained in residence at Minnesota, giving a series of lectures on colloid chemistry in the following summer session. Here he captivated his American colleagues by his charming personality and established many enduring friendships. The invitation to be foreign guest scholar at the fourteenth annual National Colloid Symposium at the Uni-