Physician's bulletin commemorating the 25th anniversary of the discovery of insulin.

COMMEMORATING THE 25th ANNIVERSARY OF THE DISCOVERY OF INSULIN

Physician's Bulletin

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Foreword

This Memorial Edition of the PHYSICIAN'S BULLETIN *has been prepared in response to many requests and suggestions from those who attended the International Diabetes Clinic at the Indiana University Medical Center, Indianapolis, September 23, 1946, to celebrate the twenty-fifth anniversary of the discovery of Insulin. It seemed more appropriate to the occasion to publish only the main addresses, rather than a transcript of the entire proceedings, in a format that could be permanently treasured. To this end, only the size and collation of the* PHYSICIAN'S BULLETIN *have been retained, in order to facilitate production and distribution of the volume and to make the articles available for bibliographic purposes. Otherwise the issue is completely individual.*

The speakers and distinguished guests whose co-operation made the program possible are among those who have written the living history of modern treatment of the disease diabetes. From the four corners of the earth they were gathered together the previous week at the University of Toronto, with the members of the American Diabetes Association, in solemn convocation to commemorate the epochal discovery of Banting and Best. Except for officers of the American Diabetes Association, representatives of official medical organizations, and the foreign guests, the meeting at Indianapolis was local. It was attended by some 900 of the medical

student body of Indiana University, members of the Lilly organization, and the medical profession of Indianapolis and vicinity.

The first experimental evidence that Banting and Best had isolated a potent antidiabetic principle was obtained on July 27, 1921, at the University of Toronto, when they observed a definite lowering of the blood-sugar level of a depancreatized dog following injection of 5 cc. of a crude extract of pancreas. The participation of the Indianapolis group in the problem came later, in the spring of 1922, and by the end of the year the production of Insulin had been fairly well worked out as a result of close co-operation between the two groups. This volume, therefore, marks the silver anniversary of the participation of the Indianapolis group in the developmental aspect of large-scale Insulin production.

In arranging the program, and again in this volume, the presentations have been chronological insofar as possible. The omission of a number of exceedingly interesting extemporaneous discussions was necessitated by limitation of space, including the remarks of Dr. Moses Barron, whose original publication germinated the birth of Banting's original idea, and the minutes of practically the entire evening session. To J. O. Ritchey, Professor of Medicine, to Lady Banting, and to all those who participated and whose names are too numerous to mention individually—our supreme thanks.

FRANKLIN B. PECK, M.D., *Editor*
ASSOCIATE DIRECTOR
DIVISION OF MEDICINE
LILLY RESEARCH LABORATORIES

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*For Professors Carl and G. T. Cori, Washington University, St. Louis, Missouri.

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Makers of Fine Pharmaceuticals and Biologicals

COMMEMORATING

THE 25TH ANNIVERSARY

OF THE DISCOVERY

OF INSULIN

Courtesy of Dr. Fred Hipwell

FREDERICK GRANT BANTING, K.B.E.

1891–1941

Surgeon and physician, an illustrious scientist,
a noble gentleman, and a brave soldier.
By his discovery of Insulin with Charles H. Best,
he made possible the control of a disease that had been
a scourge of mankind for a thousand years.
"... through the years since 1941 the memorials to Banting
have grown and multiplied. The most important of these
are not the institutions and lectures and buildings and
foundations and ships and other tangible memorials which
bear his name, but the miracles that happen
every day throughout the world—the miracles
of human beings brought back to health and
*hope by Insulin."**

*SEALE HARRIS, *Banting's Miracle*

John S. Steele

CHARLES HERBERT BEST, C.B.E.

1899–

Codiscoverer of Insulin with Banting; eminent physiologist and coauthor of several texts on physiology; soldier for Canada in World War I; Surgeon Captain and Director, Royal Canadian Navy Medical Research Division, World War II; Director, Banting and Best Department of Medical Research, and Professor of Physiology, University of Toronto; D.Sc., 1928; F.R.S. (London), 1938; Commander of the Order of the British Empire, 1944; Baly Medalist, Royal College of Physicians, England, for distinguished service in physiology; the only person to receive both the Ellen and Charles Mickle Fellowships, University of Toronto; initiator of investigations on numerous aspects of the Insulin problem, and other studies leading to discovery of the enzyme histaminase, the dietary factor choline, and the use of heparin in prevention of thrombosis.

A Note on Sir Frederick Banting

JOHN R. WILLIAMS, M.D.

Down through the pages of history there emerges from obscurity now and then an individual who, without the benefit of long training and elaborate institutional facilities, projects an idea which in time influences the destiny of man. For example, in the sixteenth century a young English clergyman, William Lee, distressed by the long arduous toil of the housewife in knitting the family stockings, designed a machine to replace the fingers of woman in this laborious task. This was the first attempt to relieve the drudgery of hand labor by machinery. Lee laid the cornerstone of the modern industrial age.

More familiar is Sir Isaac Newton, born of humble parentage in the seventeenth century. At the age of twenty-three, Newton wrote an original treatise on mathematics known today as differential and integral calculus and laid the foundation for the modern sciences of chemistry and physics with their vast and unpredictable implications.

Today we honor the memory of Frederick Grant Banting. By his discovery of Insulin he made possible the control of a disease that had been a scourge of mankind for a thousand years. But he did more. To medical

research throughout the world he gave an impetus that has proved most productive. Banting's biography is now well known. On this occasion, let us consider his fine qualities of mind and heart. Foremost of these were loyalty and curiosity.

Loyalty is that sacred quality of character which binds one devotedly to a worthy cause, be it friendship, patriotism, or the search for truth. Curiosity is that eager concern which prompts one to explore the darkness of the unknown in the quest of light. That fortunate individual who is blessed with these attributes seeks little other reward than the infinite satisfaction of achievement. Loyalty and curiosity are foundation stones of success, be it in business or in scientific research. They were strikingly evident in Banting's character. He was loyal to the friends of his humbler days. These he never forgot even when his fame became world-wide. He was loyal to his Alma Mater, to whom he gave the valuable patent rights of his discovery for the promotion of research.

This loyalty is further illustrated by an episode which I had the privilege of witnessing. In May, 1922, when Banting was but little known and his discovery of Insulin not yet wholly accepted or appreciated, he visited Rochester. He shared with me his anxieties and difficulties, and they were many. Debt and lack of means for the research which he hoped to continue hung heavily over him. We were at a luncheon as guests of the late George Eastman, founder of the Kodak Company and benefactor of many cultural and humanitarian institutions. Mr. Eastman, in a roundabout way, had heard of Banting's distressing problems and tactfully offered him a sum of money adequate to equip and operate a research laboratory provided he would affiliate with the new University of Rochester Medical School, in which Mr. Eastman was much interested. Banting promptly declined with thanks, stating that his duty was to his own country and that it was his greatest desire to see Canada take its place in the field of scientific research. Mr. Eastman was deeply moved by this reaction and, turning to me, said, "If ever one deserved a halo, this young man does."

He was also loyal to his country in the throes of war and to his comrades-in-arms, as evidenced at the battle of Cambrai in 1918; although seriously injured himself, he remained on duty at grave risk to his own life, tending the wounded and dying all day and far into the night. Finally, and most important was the fine loyalty to his associates, with whom he ever insisted on sharing full credit for the accomplishments of his laboratory.

Curiosity was another of his praiseworthy characteristics. This was insatiable. He rarely was satisfied with usually accepted explanations as, for example, those concerning the problem of cancer and the cause of death in

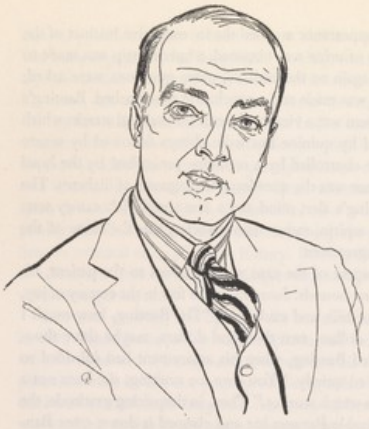
drowning, sudden death in heart disease, and blackout of aviators. Indeed, it was while engaged in this last piece of investigation in the service of his country that he lost his life. These are but a few and in the field of medicine. However, his curiosity was really universal in scope. He was intrigued by the legends and habits of the American Indian and by the optical effects of sunlight on the snows of the frozen North which he delighted to reproduce in oils. Far afield from these, the principles involved in the study and detection of crime interested him greatly. Once when in New York together, we spent several exciting hours in the office of a world-renowned expert exploring this fascinating subject and examining the protocols of some of the famous criminal cases of recent history. Banting had the mental equipment of a Sherlock Holmes. To a high degree he combined logical deduction with scientific experiment.

Late one evening, shortly after the announcement of the discovery of Insulin, Banting received an urgent phone call from a Toronto hotel. An anxious voice with a foreign accent demanded his services immediately. Explanations that he did not practice medicine failed to satisfy or assuage the distressed patient. It so happened that none of Banting's clinical colleagues was available at that hour; piqued by curiosity as to the identity and needs of the alarmed stranger, he volunteered to give emergency aid. The patient proved to be an opulent banana exporter from a Central American country. He had a long and fantastic history of bouts of illness which had been diagnosed as diabetes. The doctors of his native land, the specialists of Harley Street, and the savants of Paris had studied, pondered, and disagreed over his case. The patient continued to suffer. He had returned home and was filled with despair. Then one day there was flashed across the world the news of the wonderful discovery of Insulin. Here at last was hope. Urgent cablegrams and energetic New York agents soon had an ampoule of the precious medicine at his bedside. Since the anxious doctor felt that speedy and heroic treatment was imperative, he injected the entire contents of the ampoule at one dose. The result in a few minutes was both startling and violent—a severe Insulin reaction.

In most hotels there will be found an all-wise mind who has all the answers. In this hotel, the home of the patient, the genius of infallibility was the porter. When it became evident that the victim would survive, the porter ventured that he knew the famous Dr. Banting, the discoverer of the miraculous remedy, and that he had worked as an orderly in the Toronto General Hospital. So with all the speed of travel, the thoroughly alarmed sick man hastened to Toronto and lost no time in contacting the great doctor.

Something about his appearance aroused the investigative instinct of the young scientist. A sample of urine was obtained; a hurried trip was made to the laboratory and back again to the bedside; more questions were asked; then a dramatic diagnosis was made and a conclusion was reached. Banting's inquiry revealed that the man was a victim of recurrent malarial attacks which were temporarily relieved by quinine but were always followed by severe muscle pains. These were controlled by a medicine prescribed by the hotel drug clerk; next in sequence was the questionable diagnosis of diabetes. The combination of Dr. Banting's alert mind and a few simple laboratory tests suggested that too much aspirin, rather than diabetes, was the cause of the international medical disagreement.

When this puzzling aspect of the case was explained to the patient, his relief and gratitude knew no bounds. Jumping to his feet in the ecstasy of joy, he pulled out a big roll of bills and exclaimed, "Dr. Banting, how much I owe you—one thousand dollars, two thousand dollars, maybe three thousand dollars?" The startled Banting, when his amazement had subsided so that he could speak, replied quietly, "You owe me nothing, sir. I am not a practicing doctor, just a research worker." Then, in despairing gratitude, the patient picked up his valuable Panama hat and clapped it down over Banting's ears, saying, "Well, if you won't take money, keep this to remember me by." This amusing story reveals the active, alert, reasoning mind of the scientist and the kind, sympathetic nature of the man. In 1923 King Gustave of Sweden presented Banting with the highest award in medicine, the Nobel prize, and in 1934 King George V of England conferred knighthood upon him. But long before, the King of all Kings had set the seal of true nobility upon his brow. Fred Banting was indeed one of Nature's noblemen.



The Discovery of Insulin

CHARLES H. BEST, M.D.

WHEN I saw the first draft of the program, Dr. Russell Wilder was to give a ten-minute discussion on "Diabetes in the United States." I thought that since Insulin was only one part of the treatment of diabetes, I would have lots of time inasmuch as I had been asked to limit myself to the early work. I still think that in the few minutes allotted, I will have the opportunity to review with you early happenings.

As Dr. Banting and I always tried to point out, we felt most sincerely our intense obligation to our predecessors in this field of medical research. Of course, there had been hundreds of articles, some of them of relatively little use but a few of very great importance; we based our findings and our working hypotheses, as formulated by Banting, on these papers. A hypothesis which is as productive as the one which Banting originated (at least it was independently originated by him) has served the purpose if the goal or any interesting goal is attained. That certainly was true of the idea which initiated the work on Insulin in Toronto.

I have paid my tribute many times to Banting's perseverance, his intense energy, the drive which compelled him to give up his medical practice. He gave up the possibility of becoming a surgeon, and he was a grand surgeon. Experimentally he has done a full amount of clinical surgery and undoubtedly would have had success if he had stayed in that field.

We had many advantages over our predecessors, but I think the greatest single advantage undoubtedly was the method of doing blood sugars quickly and accurately. On reading over the papers of previous workers, one can see how they might have been guided along the right path if their blood-sugar methods had been accurate or if they had had procedures available to do good estimations on very small amounts of blood.

Our partnership began on the seventeenth of May, 1921. Banting had come to Toronto, as most of you know. He felt himself completely able to handle the surgical approach to the problem, but he needed a collaborator on some of the scientific problems. Many stories have been written about this phase of the work, but the simple truth is that I think that I was the only student in the Department of Physiology who had actually registered with Dr. Macleod to stay there another year, and it was quite natural that he should ask me if I cared to undertake this work with Banting. Of course, we also had the great advantage of being left completely uninterrupted during four or five months. Many people have said that they have seen the early work on Insulin, but as I remember there was no one in the laboratories except the two of us, the caretaker, and one or two intimate friends. A little later in the autumn when the results had developed slightly, four of our medical friends and colleagues dropped in to see how we were getting along. I think we have to attribute the results that came quickly to good fortune.

It is true that when the opening wedge had been driven and we saw the fall in blood sugar in our completely diabetic dogs, we did fight the problem as hard as we could. We made great lists of things we hoped to get done even while we were working on the perfectly obvious development of the initial discovery. Some of these experiments have never been carried out, in spite of the 20,000 papers that have been written on the subject since 1921. As a matter of fact, Banting and I had planned just before his death to do some work on the interference with hepatic function caused by ligation of the bile duct and on the various effects arising out of that. I should like to pay tribute also to the work of Professor Macleod and Dr. Collip in the development of Insulin. As I said the other day in Toronto, I owe a very great debt to Dr. Macleod because of the training he gave us as students. In my fifth year in the University of Toronto, which was completed just before I began working

with Banting, Professor Macleod had given us an intensive course in carbohydrate metabolism three to four hours a week while he was writing articles and books on sugar metabolism. Eight or ten of us who were senior students had a very great advantage in delving into this research and in hearing the opinion of a very great expert. It was Dr. Macleod who guided the development of Insulin in the autumn of 1921 when he returned to the laboratory and found a great deal of complete evidence that Insulin had been isolated. I also owe a very great debt to Dr. Collip for his work on the purification of Insulin. His work, with others, on the testing and various aspects of the physiology of Insulin was outstanding.

There was a tense atmosphere in Toronto in the autumn of 1921 and in 1922, and every bit of knowledge that came along was eagerly applied. We were working up to the point where we could give the first dose of Insulin to each other and then make it available for clinical trial.

Also, I should like very sincerely to pay my tribute to Eli Lilly and Company, to Dr. Clowes, and to Mr. Lilly particularly, but perhaps I have seen more of these men during the past twenty-five years than most of you. I think I came to Indianapolis nine or ten times in 1922, to try to work out with the experts here procedures for the large-scale preparation of Insulin, which was, of course, in great demand. I would hesitate to say how many times Dr. Clowes came to Toronto in 1922, but his visits were much more frequent than mine here. So, I should like to pay my tribute to this great organization and, in addition, to thank them for making this meeting possible today.

Twenty-Five Years of Insulin

JOSEPH H. BARACH, M.D.

TWENTY-FIVE years of Insulin. What has it wrought and what has it brought? We who have lived through the past twenty-five years are fortunate indeed to be celebrating the twenty-fifth year since the discovery of Insulin. That being so, this is the proper occasion to summarize some of our experiences and at the same time to clarify in our minds what we have attained and how much better off we are for having had Insulin for a quarter of a century. Perhaps, in giving over to our successors, we might do well to give the coming generation the fruits of our experience as well as the priceless remedy itself.

The fleeting moments of today will permit mention of only a few achievements by the doctor and only some of the benefits which have come to the patients because of Insulin. First of all, I should say that the need for intelligent use of Insulin is forcing us to a better understanding of what constitutes a physiological diet for man. Even though final words have not yet been said on this complex subject, the problem as a whole is becoming clearer from year to year.

The clinician is coming to understand how much carbohydrate is desirable for a day's sustenance; he is nearer to knowing how much protein a day will meet nutritional requirements; and we are at the threshold of a better understanding of how fat is metabolized and how much fat is physiologically desirable in a day's rations. We know that we pool carbohydrate, protein, and fat to supply the needs of metabolism. We should like to know more about each of these substances in turn, by what processes and functionings the final fuel substance is produced, and when and where insulin enters and takes its part in the maintenance of a normal metabolism. Because of all this that we do not know as yet, it is just as well to continue to be circumspect and to avoid finality and hasty conclusions at this time. Great gains have been achieved these twenty-five years, but these acquisitions have only increased our needs and desires for facts.

In 1922, Insulin was handed to the practicing physician on a platter of gold, so to speak. Insulin had been isolated, it was purified, it was perfected, and it was given to us for clinical use with such suddenness that we hardly knew what to do with it. When I was younger than I am today, I used to enjoy reading one of the well-known philosophers of that day who said, "Good happy truth seldom lives over ten years." Of course, he was commenting on the rapidity of change and the progress of those days. Insulin, however, is a good happy truth and has already lived twenty-five years; and as far as we can see today, Insulin will live many times twenty-five years and will continue to live on until a better Insulin or an endogenous or exogenous substitute for Insulin is discovered. With alloxan we destroy the beta cells selectively. Who can say that we will never build or construct selectively?

Regular Insulin, crystalline Insulin, Protamine Zinc Insulin, and many modifications, one after another, have changed the life and outlook of the diabetic from a sickly and a difficult one to one of health and frequently to one of luster. Last week Rabinowitch, of Montreal, reported that with an average diet of carbohydrate, 250 Gm., protein, 100 Gm., and fat, 45 Gm., he finds acetonuria in only three-fourths of 1 percent of his patients.

In 1921, before Insulin was available, our records show that on a high-fat diet 43 percent of our diabetics at one time or another during the course of a year showed ketonuria. By 1932, on a low-fat diet without Insulin, only 3 percent of our diabetics showed ketonuria. Of those who were on a low-fat diet and taking Insulin, 2 percent showed ketones. Our diets provided 90 Gm. of fat, whereas the Rabinowitch diet calls for only 45 Gm. of fat per day. Therefore, it is evident that lowering the fat in the diet lowers the incidence of ketonuria. Another interesting point made by Rabinowitch is that only

one out of four of his diabetic patients requires Insulin. With approximately 90 Gm. of fat and 200 Gm. of carbohydrate, we find it necessary to give Insulin to approximately 50 percent of our patients. Although these clinical observations do not have the exactness of laboratory experiments, it is clear that patients do well on diets relatively low in fat. Now and then we do see patients who seem to have done well on high-fat diets, but they are the exceptions rather than the rule. So much for diet.

Herbert Marks, of the Metropolitan Life Insurance Company, told us last week that the death rate among diabetics of all ages up to fifty-five is less than one-half of what it was before Insulin. In 1900, of the young diabetics, or those under twenty years of age, 753 out of every thousand died per year. In the present period the death rate has been reduced 98 percent in the diabetics under twenty years of age. At ages of forty to sixty, today the death rate is 68 percent lower than it was formerly. So much for those that die.

Now for those among the living and those to be born. According to various studies in 1909, 55 percent of mothers survived. In 1935, 96 percent of mothers survived. Today, 100 percent of mothers are expected to survive. Today, with Insulin and hormonal therapy, according to Priscilla White, 100 percent of mothers and 97 percent of infants survive. This is what Insulin has wrought, and this is what Insulin has brought.

General surgery in the diabetic has lost its horror because of Insulin and all the knowledge that has come since the advent of Insulin. Diabetic gangrene—whether it be slight or extensive, whether it involve the toe or the foot or a leg—is dealt with in comparative safety and ease because our patients will not sink and drown while operations are being performed. The mortality rate in amputations for patients with diabetic gangrene in one group of New York hospitals at one time ranged from 70 to 85 percent, while in another it was reported to be 45 percent. Beverly Smith, of New York, who reported on these results, believes that this high death rate was due largely to thigh amputations where leg amputations or operations might better have been performed. He also warns that in less than 10 percent of elderly amputees will a thigh-cross be achieved successfully. Low operations minimize shock and yield a low mortality rate of 8.6 percent, especially if prophylactic treatment against infection is instituted before surgery is undertaken.

Diabetic coma cases before Insulin ended in death. Joslin, Root, and their colleagues last year reported 123 consecutive cases of coma with a mortality of 1.25 percent. In the last forty-six consecutive cases that I have treated, there was one death, a mortality of 2.4 percent. And yet, alongside these experiences, when neglected cases are sent to hospitals that are unprepared—

hospitals in which the staff is unprepared mentally and physically for prompt and adequate treatment—cases of diabetic coma still have a mortality rate of 50 percent. That is not one bit better than it was before Insulin was available. These contrasts are striking and the figures are true, but there are extenuating circumstances. One must of necessity be sure that he is considering comparable types of cases. Old age and youth, physically fit and physically deteriorated individuals, and, above all, patients with and without infections give contrasting mortality rates. My own impression is that, first of all, we should segregate those who have infections from the noninfected group before making comparisons. With increasing experience and knowledge it is possible today to remove most of the drama and all the tragedy of diabetes. If we put into use what is already known, the coming generation will have a grand opportunity and also a great responsibility insofar as taking care of the diabetic population is concerned.

There is indeed much more to be said, more than can even be mentioned today. It was my duty to speak only of the past twenty-five years; others will project the future possibilities. It is twenty-five years since we have had Insulin, a product of pure science at its highest level. Insulin as today manufactured is of the highest refinement and utmost purity. During the past twenty-five years many thousands of my patients have used it once, twice, or thrice daily by injection. Only once have I seen an abscess result at the site of injection, and that was in one of the most careless patients that I have ever had in a large, free dispensary service.

It is the function of the American Diabetes Association, a group for which I am honored to speak on this occasion, to study products and methods, to stimulate investigation, to standardize the treatment of the disease diabetes, and to bring to the diabetic patient all the good that comes with an advancing science. As a member of the American Diabetes Association, for our group today and in the memory of Frederick Banting, let me express thanks to Charles Best and to every one of his colleagues, and thanks to the men and women in the laboratories who make and prepare Insulin with such refinement and exactitude. Insulin indeed is a sovereign remedy. Insulin has wrought and Insulin has brought much to our generation of doctors and patients.

Etiology of Diabetes

BERNARDO A. HOUSSAY, M.D.

THE CAUSE of diabetes is as yet unknown. In all cases of permanent diabetes there is insulin insufficiency, since the pancreas is unable to secrete the amount necessary to maintain a normal glycemia. This insulin insufficiency may be due to a primary alteration in the pancreas or to metabolic disturbances in the tissues outside the pancreas, which require and provoke an increase in insulin secretion with subsequent damage to the islets.

Diabetes can be produced experimentally in the following ways:

(a) By *total or subtotal pancreatectomy.*

(b) By *injection of anterohypophyseal extract*, which first produces extrapancreatic metabolic disturbances and then secondary lesions in the islet cells.

(c) By the *administration of thyroid*, which produces diabetes only when the islets have been previously damaged or diminished in number by partial pancreatectomy.

(d) By *injection of alloxan*, which selectively destroys the beta cells.

(e) By *injection of some of the corticoadrenal hormones*, which can produce a transitory diabetes in the rat.

A diet rich in sugar or other carbohydrates favors the diabetogenic effect of the hypophysis, the thyroid, and subtotal pancreatectomy but does not

produce diabetes if there is an intact, normal pancreas. In the rat, a diet rich in fat favors the onset and increases the severity of diabetes caused by alloxan or subtotal pancreatectomy.

In 41 to 75 percent of diabetic cases, autopsy reveals definite lesions in the Langerhans islets, but in others there are no alterations. Lesions in the pancreatic islets are found in 15 percent of nondiabetic subjects.

Diabetes is inherited in the Mendelian recessive proportion. Some investigators suppose there is inheritance of an inadequate capacity of the pancreas to secrete insulin—a diminished insular reserve. It is also possible that what is inherited is a metabolic abnormality, in which the tissues utilize or inactivate insulin in amounts greater than normal; thus the beta cells would have to secrete it in excess and would be damaged by fatigue and subsequent exhaustion.

Obesity and overeating frequently precede the onset of diabetes. Hypophyseal and thyroid diabetes are more easily produced when the diet is rich in sugar; they can be cured by the early administration of hypoglycemic agents. In the subtotally pancreatectomized rat, the onset of diabetes is accelerated and its severity is increased by a diet rich in fat or an abundant mixed diet. On the other hand, its frequency can be decreased by reducing the amount of food ingested and by dividing the normal daily ration into several meals.

Sometimes extrapancreatic factors have an evident effect. For instance, (a) the anterohypophyseal extract is intensely diabetogenic in animals deprived of the pancreas and the hypophysis; (b) in hypophyseal diabetes the blood sugar rises before the capacity to secrete insulin diminishes; (c) the anterohypophyseal extract produces growth in puppies, and diabetes appears only when growth ceases; (d) some cases of diabetes have been cured by extirpation of corticoadrenal tumors.

Probably there is first an increase in insulin secretion and then a secondary alteration in the islet cells in the following cases: (a) in overfeeding; (b) during growth; and (c) during the administration of thyroid, anterohypophyseal, and corticoadrenal hormones, which increase resistance to insulin in the tissues.

In future research it will be necessary (a) to intensify the study of metabolic disturbances in prediabetic and diabetic conditions; (b) to measure insulin secreted, circulating and inactivated, so as to know the variation in its production and consumption; (c) to estimate quantitatively the endocrine factors which regulate carbohydrate metabolism and which are unbalanced in diabetes; and (d) to perfect the administration of Insulin so as to obtain a continuous action. Finally, it is necessary to find ways of preventing damage to the islets and of obtaining the regeneration of beta cells.

The discovery of Insulin is one of the greatest of scientific achievements; it was obtained by research in animals, and the correct application of this knowledge to patients was also thus acquired.

ON THE 30TH OCTOBER, 1920, FREDERICK GRANT BANTING ORIGINATED THE HYPOTHESIS THAT THE FAILURE THERETOFORE TO ISOLATE THE INTERNAL SECRETION OF THE PANCREAS HAD BEEN DUE TO ITS DESTRUCTION BY THE FERMENTS LIBERATED DURING THE PROCESS OF EXTRACTION.

HE DEVISED AN EXPERIMENTAL METHOD BY WHICH THIS DESTRUCTION COULD BE AVOIDED AND THE INTERNAL SECRETION (NOW KNOWN AS INSULIN) OBTAINED.

IN MAY, 1921, BANTING AND CHARLES HERBERT BEST, BOTH GRADUATES OF THE UNIVERSITY OF TORONTO, CONDUCTED IN THIS ROOM THE EXPERIMENTS WHICH CULMINATED IN THE ISOLATION OF INSULIN.

This bronze plaque marks the small laboratory in the Physiology Department at the University of Toronto, where the original experiments were conducted.

Modifications of Insulin

HANS C. HAGEDORN, M.D.

THE EFFECTS of different hormones vary widely as to duration and intensity. It is no wonder, then, that the first substance of this kind successfully given by mouth was the thyroid hormone, which has a long-lasting effect in the body. We cannot exclude the possibility of the existence of hormones not yet known on account of short duration of their effect.

The duration of the effect of some therapeutically useful hormones is so short that methods of continuous injection have been developed for some patients by the help of a clockwork operating a syringe, but such gadgets only show how far we are from Nature's wonderful methods.

Let us consider the case of Insulin. It is easily soluble in the body fluids; and when it is injected into the subcutaneous tissues or elsewhere, we are not able to alter the rate of its absorption—that is, the quantity absorbed during the unit of time—without altering the total dose. No matter whether we inject the same quantity of a more concentrated solution or a greater quantity of a solution of unaltered concentration, the increased dose will mean a greater rate of absorption. This is the reason that Insulin has to be injected at short intervals.

When the good news of the discovery of Insulin came to us, Professor August Krogh visited Toronto and at an early date obtained information from Dr. Banting, Dr. Best, Professor Macleod, and Dr. Collip, who allowed us to start preparation of Insulin in Denmark. Later on we had great help from publications from Shaffer. The first preparations, of course, were not purified. It was noticed then that some preparations which gave local trouble had a considerably prolonged action, which appeared on the blood-sugar curve and was also noticed by the patient. Since then we have been very much interested in products giving a delayed action without any local irritation, and various attempts have been made by many other investigators to make Insulin preparations which will allow the injection of a greater dose without an increase in the rate of absorption.

In general, three different lines have been followed. (1) The injection, together with the Insulin, of some substance influencing the circulation at the site of the injection. So, among others, adrenaline has been used (Figure 1); on a very superficial investigation it may appear that the effect is prolonged

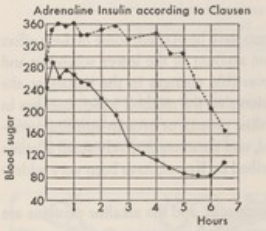


Figure 1

Ordinary Insulin and adrenaline Insulin compared in diabetic patients

(Whole line: Insulin. Broken line: adrenaline Insulin. Injection immediately after first blood sample)

(From Clausen: Kliniske Undersgelser over Insulinresorptiones Paavirkelighed af Adrenalin, Pituitrin og Ephetonin [with an English Summary], Diss. Copenhagen, 1934, p. 30, figure 15.)

because the maximum effect is observed later after the injection than when the same dose of Insulin without adrenaline is given. Closer examination, however, shows that there is a period during which no, or only very little, Insulin is absorbed; and when the spasm of the vessels is relaxed, a very high rate of absorption sets in. (2) The injection of dry Insulin in an oil suspension. This method has been used in some very mild cases of diabetes with some success. Some of the best results as far as I know have been obtained by the

late Dr. Leyton, of London, but on the whole it has not been found of very much practical value. (3) The third method consists in giving the Insulin together with a substance with which it forms a combination relatively insoluble in the tissue fluids (Figures 2 and 3).

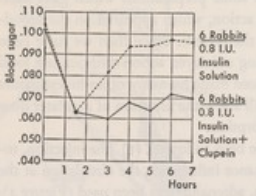


Figure 2

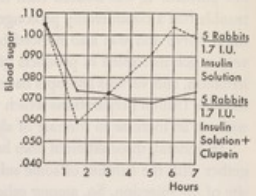


Figure 3

Unpublished experiments on rabbits in Nordisk Insulinlaboratorium, by Dr. Marie Weitze (Injections immediately after first blood sample)

Different protein-precipitating agents have been tried; some of them effect a considerable delay, partly perhaps on account of the lower solubility and partly on account of more or less reversible changes in the tissues at the injection site. It is the general impression that the method which consists in using Insulin in combination with alkaline protein or protein-splitting products has so far been the most successful, and the comparatively small alkaline molecules of the protamines, first described by Kossel, have been very widely used.

Insulin can be considered a very weak acid, and the alkaline proteins are weak bases; by bringing those substances together at proper reaction, something formally analogous to the formation of a salt takes place. We can also say that the two colloids precipitate under mutual discharge; therefore, products are easily obtained which are relatively insoluble at the reaction obtaining in the subcutaneous tissue. By injecting such precipitate products of protamine and Insulin as a suspension or as a solution brought about by surplus of protamine, a considerable delay in the duration of absorption is observed (Figures 4 and 5).

Scott and Fisher have made the very important discovery that this delay can be augmented considerably by adding zinc in small quantities, thereby

at the same time obtaining better stability of some preparations. Although Protamine Zinc Insulin has been most widely used in America, we have continued the use of Protamine Insulin without zinc in Denmark.

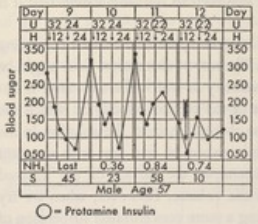


Figure 4

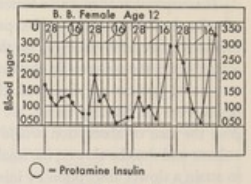


Figure 5

Ordinary Insulin and Protamine Insulin compared in diabetic patients at the Steno Memorial Hospital

(U = International Insulin units. Encircled figures represent injections of Protamine Insulin. H = hour. The large vertical arrow signifies a slight hypoglycemic reaction.)

The whole problem of absorption of Protamine Insulin from the tissues can be studied more closely now that it is possible to use suspension of crystals. Abel succeeded in crystallizing Insulin, and later Scott demonstrated

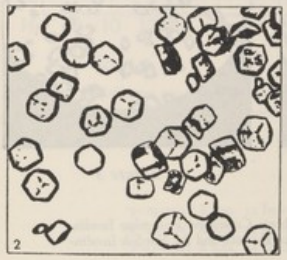


Figure 6

Zinc-Insulin crystals, formed at *p*H 6 (From Scott: Endocrinology, *25*: 440, 1939, figure 2.)

that the presence of a metal, preferably zinc, was necessary to obtain crystals (Figure 6). Krayenbühl and Rosenberg, working in our laboratories, have succeeded in crystallizing Protamine Insulin with zinc (Figures 7 and 8). The crystals appear to be useful in the treatment of diabetes, since free Insulin added to the crystal suspension is not adsorbed to the same extent that it is when the precipitate is amorphous.

If a suspension of an active material is injected, we have a depot consisting of a dissolved and an undissolved part. The rate of absorption then might be expected to depend either (1) on the velocity at which the substance is absorbed from the solution in the depot or (2) on the velocity at which the suspended particles are dissolved. If the absorption from the solution in the depot were the limiting factor, we might expect to find that with constant dose the rate of absorption would depend on the surface of the injected volume and thus be slower, the smaller the volume. However, when we try to attain a slower absorption by injecting a more concentrated suspension of

Figure 7

Figure 8

Protamine Insulin crystals
(*See* Krayenbühl and Rosenberg: Crystalline Protamine Insulin. Reports of the Steno Memorial Hospital and the Nordisk Insulinlaboratorium. In press.)

Protamine Insulin, we find the proportion between dose and rate of absorption unaltered or only altered very little (Figure 9). A little of the expected effect is observed, perhaps by mere coincidence; at any rate, it is much smaller than should be expected if the theory under test held good.

The other possibility was that the limiting factor is the rate of dissolution of the solid particles. No easy way to attack this problem was found until crystals of Protamine Insulin were available. Mr. Krayenbühl was kind enough to prepare for my purpose two suspensions of crystals with the same content of Protamine Insulin, but with one containing larger crystals so that the main surfaces of all the small crystals were about two times as great as those of the

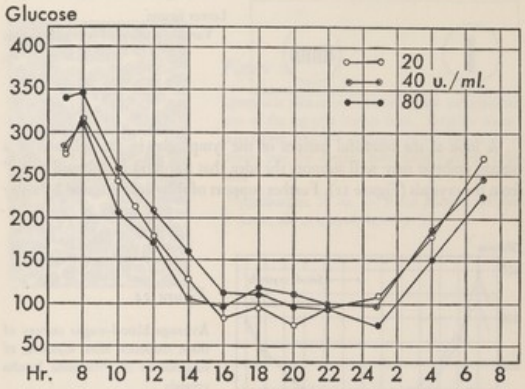


Figure 9

Blood-sugar curves of a diabetic patient (case record No. 2,138) after injection of suspensions of Protamine Insulin

9–19–1941 8 a.m. 24 International units 40 units per ml.
9–22–1941 8 a.m. 24 International units 80 units per ml.
9–25–1941 8 a.m. 24 International units 20 units per ml.

other (Figure 10). The rate of absorption, however, was nearly the same (Figure 11). It may be then that the presumption from which we started is erroneous, that no depot of suspension exists, that the fluid is absorbed rapidly, and that the crystals remain "dry" in the tissue.

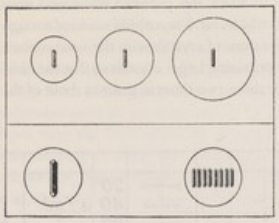


Figure 10

Upper figure:
Varying surface of injected volume

Lower figure:
Varying surface of injected crystals

A look at the beautiful pattern of the lymphatics in the subcutis of a human embryo may well support the idea that the fluid is "filtered away" from the crystals (Figure 12). Further support of this view is found by x-ray

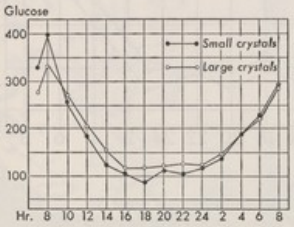


Figure 11

Average blood-sugar curves of three diabetics after injection of suspensions of Protamine Insulin crystals

Case record No. 2,134 2–14–1942 8 a.m. 28 International units, large crystals
Case record No. 2,134 2–17–1942 8 a.m. 28 International units, small crystals
Case record No. 2,248 2–15–1942 8 a.m. 20 International units, large crystals
Case record No. 2,248 2–17–1942 8 a.m. 20 International units, small crystals
Case record No. 2,262 3– 4–1942 8 a.m. 24 International units, large crystals
Case record No. 2,262 3– 2–1942 8 a.m. 24 International units, small crystals

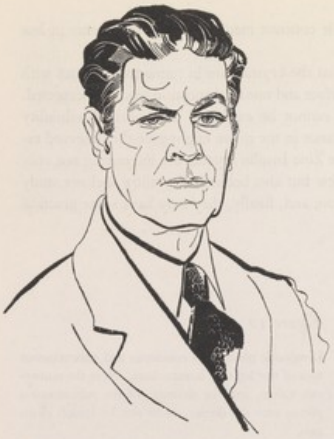
examination of injected soluble contrast medium, which disappears in less than ten minutes.

But even if we presume that the crystals are in immediate contact with the cells, a relation between surface and rate of absorption would be expected. The observations, therefore, cannot be explained as results of solubility alone. A splitting of the substance in the tissue might give the observed result. Therefore, the Protamine Zinc Insulin crystals are interesting not only from a theoretical point of view but also because they allow a closer study of the mechanism of absorption; and, finally, they may have some practical value in therapy.

Figure 12

Lymphatic plexus in a cutaneous and subcutaneous area of the leg of a human fetus. Note the numerous valves, and the drainage of the subcutaneous plexus into the deeper, more slender lymph channels.

(Magnification: × ca. 30. From Kampmeier: Am. J. Anat., *40*: 442, 1928, figure 31.)



Hemochromatosis and Complications of Diabetes

R. D. LAWRENCE, M.D.

As a diabetic myself, I owe a great personal debt to Lilly Insulin, for which I should now like to give public thanks. I have had Insulin a long time. True, I am not one of the many people in many countries of the world who profess to have had Insulin first, but I ran that fairly close. When I started Insulin early in 1923, nearly twenty-four years ago, it had only begun to come into commercial production in England. You cannot blame us for that; we are always a bit behind the first. At any rate, I was very glad to get what I could. About April, 1923, wasted, thin, fit for nothing, in a cautious way I began using Insulin which was produced by a commercial firm in England. I understood that there was enough for me in my hospital to carry me on for about a month, then the grace of God would have to supply me with a new set-up.

But something developed. A girl (a doctor's daughter) in diabetic coma was admitted. To bring her out, she got the Insulin on which I had hoped to

live the next fortnight. Well, I was in (I don't know what you call it here) a jam, an awful fix. Of course, God looks after His own children. Somebody somehow brought me a bottle of Lilly Insulin the next day. I looked at it with suspicion, but I was glad to have that really—anything with an Insulin label on the bottle. As the next fortnight wore on (I mean we used as little Insulin as we could; it kept me for about a fortnight) I looked upon it with less suspicion, and by the end of the fortnight with complete confidence, which I have never had any reason to change in supervening years. And so you see, standing here and talking to you, I am giving thanks to this great firm which so quickly produced Insulin in large amounts—not merely lip service on my part. Far from it.

Now, ladies and gentlemen, particularly those interested in more scientific aspects, I am to talk here on "Hemochromatosis and Complications of Diabetes." This, I think, may seem curious and require a little explanation, but I assure you I am not drawing any red herring across the sense of this meeting. There is no doubt at all that with Insulin properly used, or even somewhat improperly used, full health and activity may be expected. At our meetings of the American Diabetes Association in Toronto, the main background of clinical interest was the evidence presented that diabetic complications develop, in spite of treatment, as the years go on. These changes, so well known, are mostly vascular, giving rise to the typical diabetic hemorrhages in the eyes, changes in the medium-sized blood vessels, and, coupled with high blood pressure, renal complications.

For about ten years I thought, and I think most of us thought, that these changes were associated with the mild, middle-aged diabetic in the fifties and sixties. They were not very obvious in the children or the young adult diabetic. The diabetes did not have to be severe, but the history had to be long, and the disease was a matter of living without control. Because I had mild diabetes for so many years, I thought that I would exhibit these symptoms. Since we have had Insulin for over twenty years now and have applied it to many patients, we have been able to keep the young alive long enough to study. It is an upsetting thing to all of us, and Dr. Joslin has emphasized it, that so many of our children and young adults with prolonged diabetes but otherwise in good health are beginning to develop these complications.

This does not apply to all of them, and it has been my impression that it is mostly in the wildly uncontrolled diabetic person who lives for weeks or months loaded with sugar that these conditions develop. I think, to go back to the personal note, that I have had my diabetes for some twenty-six years; and, at least when I left London a fortnight ago, I could swear that I had no

retinal hemorrhages and that my blood pressure was normal. I do not think I would like any of the doctors to take it this afternoon, for I think I might legitimately have a little hypertension.

Both of these conditions are intimately related to the clinical problem of treatment of diabetes at the moment, and that is why I turn to hemochromatosis ("bronzed" diabetes), which may help in elucidating this problem. An explanation is required as to what this disease is. Hemochromatosis, or bronzed diabetes as it is called, is a very rare form of diabetes. Some doctors might go through their whole careers without seeing a case, or at any rate without recognizing it. We know to what it is due. It is due to an imbalance of iron metabolism by which, throughout the patient's whole life, some organic ore, possibly some inorganic, and probably some other forms are deposited in the tissues throughout the body, under the skin, in the liver, and in the pancreas —practically everywhere except in the muscle tissue. This causes no ill effects for about fourteen to fifteen years; but when the processes have gone on so long, it brings degeneration and some enlargement of these organs. The typical practical four symptoms achieved by these people are this brown pigmentation (a slaty, bronze color) of the skin caused by deposited iron; an enlarged cirrhotic liver; sexual atrophy; and, what interests me today, quite severe diabetes, which can come on quite suddenly and which we know quite definitely (we do not know all that about the other forms of diabetes) is due to a direct attack by the fibrous tissue on the islets of Langerhans of the pancreas and its insulin productive capacity.

This disease is very rare, but I have seen perhaps some forty cases. Not much. However, I have been looking for them, and in these forty I have never seen any of the vascular diabetic complications in the eyes, feet, kidneys, and so on. Although forty cases are nothing, I thought I would bring up the subject at this meeting. Here is a rare condition; no one has many cases. It is a matter of co-operation. If, throughout this country, perhaps under the auspices of the American Diabetes Association, those who had cases, numerous or otherwise, of hemochromatosis would make a survey of them and send them into a center, we could get at some facts. I think this is important, because if these people are found not to develop the diabetic complications and if their diabetes is severe (they use a lot of Insulin; they are just as uncontrolled in their sugar as anybody else), then we have to look for some other factor than merely the high blood sugar, cholesterol, or the ketosis, or whatever it is. We have to look for a factor that produces not only the diabetes but the complications.

You might ask me what it might be. I don't know. My answer would be pure speculation. If, on the other hand, they do get these complications, it means, I think, that this disease is directly due to probable infection of the islets of Langerhans; for if it is true that the control of the diabetes or the uncontrol of it over these years is bringing these hateful complications, then we must set about to control the uncontrolled, reviewing our treatment and keeping the sugar down as well as we can, and perhaps better subsequently than we do today. Of course, we all know we cannot control severe diabetes by any present forms of Insulin treatment that are available. The only way, I think, that we can control the diabetic almost physiologically (like a normal human individual) would be to give a little Insulin after every meal and a dose at night. Now, nobody is going to stand for that.

Until we get a more physiologically acting Insulin or indeed a cure, which I think is shortly coming, before our next meeting here in another twenty-five years we have to get farther along and closer to the light and the truth by collecting more data on hemochromatosis and by developing a treatment that will safeguard our diabetics, not merely in health but in longevity and security from the complications of the future. That is the problem that I have no chance to share with any other people but which I think is a fundamental one, perhaps of interest to your progressive American Diabetes Association.

The Conquest of Diabetic Coma

ELLIOTT P. JOSLIN, M.D., AND JOSIAH K. LILLY

YESTERDAY at the Indianapolis City Hospital I talked with the coma patients Dr. Peck had gathered together for me to present today. There were two recovered cases from each of the eight decades of life. Not every one of them is here today—we did not need them all to play their parts. In fact—

"All the world's a stage,
And all the men and women merely players.
They have their exits and their entrances;
And one man in his time plays many parts,
His acts being seven ages.

Case 1. C.A. (*coma at fourteen months*)

"At first the infant,
Mewling and puking in the nurse's arms.

Case 2. C.M. (*coma at fifteen years*)

"And then the whining school-boy, with his satchel
And shining morning face, creeping like snail
Unwillingly to school.

Case 3. R.M. (*coma at twenty-two years*)

"And then the lover,
Sighing like furnace, with a woful ballad
Made to his mistress' eyebrow.

Case 4. E.S. (*coma at thirty-six years*)

"Then a soldier,
Full of strange oaths, and bearded like the pard;
Jealous in honour, sudden and quick in quarrel,
Seeking the bubble reputation
Even in the cannon's mouth.

Case 5. A.Y. (*coma at forty-nine years*)

"And then the justice,
In fair round belly with good capon lined,
With eyes severe and beard of formal cut,
Full of wise saws and modern instances;
And so he plays his part.

Case 6. E.M. (*coma at fifty-five years*)

Case 7. A.W. (*coma at sixty-eight years*)

"The sixth age shifts
Into the lean and slipper'd pantaloon,
With spectacles on nose and pouch on side;
His youthful hose, well saved, a world too wide
For his shrunk shank; and his big manly voice,
Turning again toward childish treble, pipes
And whistles in his sound.

Case 8. A.K. (*coma at seventy-four years*)

"Last scene of all,
That ends this strange eventful history,
Is second childishness, and mere oblivion,
Sans teeth, sans eyes, sans taste, sans everything."*

**Shakespeare: As You Like It*

First of all, these cases all represent diabetic coma at the different ages that I have tried to describe briefly. We do not have the swearing soldier "bearded like the pard," so we miss him, but we have the others. Dr. Best emphasized the value of blood sugars in the discovery of Insulin. I want to emphasize the value of blood sugar in the treatment of diabetic coma. In two of our cases, the blood sugar was 1,300 to 1,600 mg. and the Insulin 1,224 units. Thereafter you see the blood sugar steadily decline. That, I think, is confirmation again of the value of that single test in the treatment of diabetic coma.

In the treatment of diabetic coma today, we hear much about comparisons

between one hospital, one city, one clinic, and another. That must cease. *That must cease.* Remember, Mr. Lilly, prior to the time you gave Insulin for all these patients who have recovered from diabetic coma, prior to that, I told my patients for twenty-five years that coma represented the end of the disease —that there was nothing else that could be done. I absolved myself. But today, gentlemen, there is something different in the treatment of coma. Pay no attention to what the other man has done, but look up your own cases. One cannot put all the blame for deaths from coma in one's own clinic entirely on complications and say, "That case was inevitable; that was the Lord." Do not put that unkind thought on Him. That must go.

A week ago, I think it was, Dr. White told me Mrs. B left the hospital. She was sixty-nine. She entered in coma. She had a bacteremia with temperature upward of 104°. Penicillin, streptomycin, sulfadiazine—and she finally got better; then she had a thrombosis and the vessels of her leg closed up, but even then we did not put the blame on kind heaven. We took off her leg; when she went home, it is true she went without her leg, but she did go out with her life. I think we must stop assuming and concluding that cases of coma are desperate and are not going to recover!

There was one happy instance in the analyses of these coma cases I saw yesterday. I asked sincerely (and I had the records of them all), I especially asked about the amount of soda that was used in their recovery. They all recovered, *they all recovered;* but the soda, it so happens, was given to only one patient, and this patient received 5 Gm. of soda. I think that is the end of the soda era in the treatment of diabetic coma.

About glucose, it is true that nearly half of these patients received glucose before they recovered, but not quite half. The interesting fact is that those who did *not* receive glucose received the *least* amount of Insulin, on the average, of any of the group. And although the severity index for the two groups was the same, the CO_2 in those not receiving glucose was 11, and in the others 15 volumes percent. Thus, if anything, those without glucose represented the more severe types of cases. In this series of cases, it was also evident that the Insulin was given early and that the objective in the treatment was to give the Insulin early; throughout, as the cases proceeded, that endeavor came more and more to the fore. We know from our own cases that more than ten times as many got well when they had Insulin given in the first three hours as when the same amount was given in our coma group the first twenty-four hours. *It is Insulin,* Mr. Lilly, in which you believe and in which I believe, which brought these patients out of diabetic coma, and we should never forget that fact.

But there is one more feature of these cases that involves every citizen, doctor or otherwise—that involves every medical student—that involves every doctor—but does not involve Mr. Lilly. These people had all the Insulin they wanted, didn't they? They all had the Insulin available, but when I asked Cynthia, the first patient (there were eleven that I interrogated), how long she had had diabetes before she was brought to the hospital in diabetic coma, her mother simply said, "I never knew she had diabetes until she reached the hospital." The diagnosis was first made there. Then I said, "Charles, how long did *you* have diabetes?" "I never knew it, Dr. Joslin, until I came to from unconsciousness in the Indianapolis City Hospital." "How long did you know, Robert?" "I *never knew* I had diabetes! I never heard of it until I became conscious at the Indianapolis City Hospital." And so it goes with Edna, and with Ada, and with Ellis, and with Anna, and with Mr. Alonzo, too. *Not one* had had the diagnosis of diabetes made until he reached the hospital in coma. I wonder if diabetics must wait until they become unconscious before they are given Insulin?

Last year I heard one of the most encouraging things of the present day; that is, in Cincinnati that problem has been recognized, and Dr. Mirsky has been deputed to solve it. Gentlemen, I thank you for your attention. I know that Mr. Lilly and I both feel that Insulin is the factor, as it was in these cases, in bringing to life patients in diabetic coma.

Recent Work on the Action of Insulin

SIDNEY P. COLOWICK, PH.D.

DR. G. T. CORI, who was originally asked to present this work, was unfortunately unable to do so because of illness. I have been delegated to summarize briefly for you the results of recent experiments on the action of Insulin which were carried out in the Department of Pharmacology of the Washington University School of Medicine, St. Louis. These results have already been published in preliminary form, [1, 2] and a detailed account is now in press.[3]

The important role of the pituitary gland and the adrenal cortex, as well as the pancreas, in the regulation of carbohydrate metabolism is now well recognized. Through the fundamental work of Houssay in Argentina, and of C. N. H. Long in this country, it was established that the diabetes produced in animals by removal of the pancreas could be greatly ameliorated either by hypophysectomy or by adrenalectomy. The site of action of the hormones elaborated by these glands has been the subject of considerable debate, but it is now generally agreed that both the production of carbohydrate by the liver and the utilization of glucose by the peripheral tissues

are subject to their influence. At the risk of oversimplification, the main effects may be summarized as follows.

Certain adrenal and pituitary hormones tend to increase carbohydrate formation from protein by the liver and to diminish the peripheral utilization of glucose, while the pancreatic hormone, Insulin, has an opposing action at both sites. The present report deals with an attempt to analyze in greater detail the effects of these hormones on the peripheral utilization of glucose.

Effects of Insulin on the utilization of glucose are readily demonstrable in many types of intact muscle preparations (eviscerated animals, isolated perfused heart, isolated rat diaphragm, etc.), the fate of the glucose depending on the particular preparation used. In some cases the glucose utilized under the influence of Insulin undergoes complete oxidation, while in other cases it is converted largely to glycogen. These are the two fundamental effects of Insulin on peripheral glucose utilization which any theory of the mechanism of its action must take into account.

It appeared possible that both of these effects of Insulin might be explained in terms of an increased activity of the enzyme hexokinase. This enzyme is now known to initiate the metabolism of glucose by catalyzing the reaction of the sugar with adenosine triphosphate to form glucose–6–phosphate, which may then be either oxidized or converted to glycogen.

Methods were developed for measuring hexokinase activity in cell-free extracts of muscle and other animal tissues. The addition of Insulin to such extracts had no effect on the rate of the hexokinase reaction. An effect of Insulin could be demonstrated, however, in the following way.

It was found that the rate of the hexokinase reaction could be depressed by the addition of certain protein fractions obtained from the pituitary gland. Inhibition of the reaction was much more marked when the pituitary fraction was supplemented with adrenal cortex extract. The latter had no inhibitory effect per se in these experiments with normal tissue extracts.

The inhibition of hexokinase by the pituitary fraction plus adrenal cortex extract could be completely released by the addition of Insulin. Insulin which had lost its physiological blood-sugar-lowering activity, as the result of mild treatment with alkali, also lost its ability to exert the above effect on the hexokinase system.

When hexokinase activity was measured in muscle extracts from diabetic animals (alloxan-treated rats), adrenal cortex extract alone was found to produce inhibitions as great as those obtained by addition of adrenal cortex extract plus pituitary protein to normal tissue extracts. Presumably, there is present in the diabetic extracts an inhibitory factor of pituitary origin in suf-

ficient amounts to give rise to marked inhibition in the Insulin-free system when adrenal cortex extract is added. Addition of Insulin to this system results, as with normal extracts, in complete release of the inhibition.

The pituitary factor, as obtained in diabetic extracts or in fractions isolated from the pituitary gland, is remarkably labile, and no procedure has yet been devised whereby it can be kept even for short periods of time at 0°C. without loss of its inhibitory power. Further progress will depend on the development of an improved method for its preparation.

The results described here are in harmony with the conclusion reached from earlier observations on experimental animals, namely, that Insulin is not essential for glucose utilization but serves rather to oppose the inhibitory action of anterior pituitary and adrenal cortex hormones on this process. Although no Insulin effect could be obtained in these experiments unless the inhibitory factors were present, there is no question that Insulin can also exert certain effects in the absence of pituitary and adrenal factors, since it has a profound hypoglycemic effect on adrenalectomized and hypophysectomized animals. The nature of this effect remains to be elucidated. It seems likely that other enzymes in addition to hexokinase will be found to be under endocrine control when in vitro experiments of the type described here are extended to other enzyme systems. In particular, it appears that some of the effects of hormones on carbohydrate metabolism in the liver cannot be explained in terms of regulatory effects on the hexokinase reaction.

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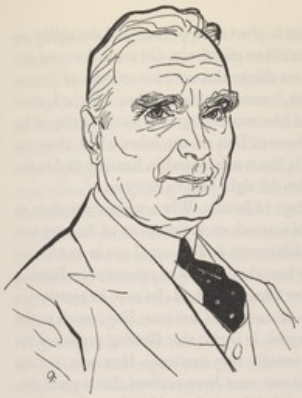

In Retrospect

GEORGE H. A. CLOWES, PH.D., D.SC. (Hon.)

IT IS A great treat after twenty-five years to see Dr. Joslin, Dr. Williams, Dr. Woodyatt, and Dr. Sansum, all members of the original committee that handled the problems involved in the development of treatment of patients in this country, and it is most particularly satisfactory that Dr. Best is also with us. Dr. Best was already a distinguished, highly trained young physiologist, having had five years of experience in this field, when he was selected by Dr. Macleod as the only person in the Department of Physiology at Toronto who was qualified to co-operate with Dr. Banting in his investigation.

First and foremost, I want to pay my tribute to Dr. Banting. All of us who knew him well and felt that we were his friends will feel that his spirit is with us on this great occasion. If it had not been for Banting, for his brilliant ideas and imagination, his great tenacity of purpose, and his indomitable spirit, I doubt that we should ever have had Insulin, and we should not have been celebrating the twenty-fifth anniversary of the event. Banting was a most unusual person. He was not merely a thinker. He was, in a sense, a visionary. He had imagination and intuition. In other words, he combined

both factors, the intuitive sense that is often so important and the ability to reason things out, which at times he did so effectively. On more than one occasion his great determination to see things through, his attitude of "come hell or high water, we are going on," saved the day, and no Martin Luther ever fought the devil or exorcised him more diligently than Banting if he felt that any injustice was being done to Best or any other of his close associates. He was a great man and his loss was irreparable. Banting died in the path of duty, but he left behind him the right man to carry on.

Now, to get back to the early days of Insulin. When we heard rumors to the effect that an important piece of research on the problem of diabetes was being carried out in Dr. Macleod's laboratory in Toronto, I got in touch with him and he advised me to be sure to hear the paper to be presented by Banting and Best in New Haven the day after Christmas, 1921. So as to be certain not to miss their paper, I left Indianapolis early on Christmas Day, much to the disgust of my family. I was well repaid. It is true that Banting presented his material somewhat haltingly and certainly very modestly. However, anyone who was at all cognizant of the subject must have realized that a great discovery had been made and that, provided the work could be brought to fruition, there was every prospect that an important means of treating diabetes would be developed.

Immediately after Banting and Best had presented their paper, I went to see Dr. Macleod. He introduced me to Banting and Best and we had several talks that day and the following day. I pointed out to them that before long their problem might well be one of large-scale production, in which case they would need the sort of help we could give them, represented by men like Rhodehamel, Walden, and Scott—men who were not just chemists but also chemical engineers. The Toronto group had at its disposal able physiologists and latterly biochemists and chemists, but chemical engineers were at that time great rarities in university groups and Toronto lacked that type of experience.

Dr. Macleod, who was the head of the department, was extremely cautious and hesitant about any sort of co-operation with any group outside Toronto University. However, after three or four months of negotiation, a plan of co-operation was worked out; then commenced, at least for me, the most strenuous, the happiest, and certainly the most interesting period of my life. I believe that all those who collaborated in the work on Insulin will have the same feeling. It should be said for Macleod that when he returned to Toronto in the fall and realized what had been accomplished during the summer by Banting and Best, he immediately threw himself heart and soul into

the project, brought Collip into it to work on certain very important biochemical aspects of the problem, and did everything in his power to accelerate the progress of the investigation.

By the time we came into the picture, in the spring of 1922, a crude product was being produced and some tests had been made on human cases. We concentrated our entire research effort on the problem of producing Insulin —purifying it, stabilizing it, and testing it. By July and August, 1922, we were already supplying Insulin to Banting and his associates, and shortly thereafter Insulin was being provided for Drs. Joslin, Williams, Woodyatt, Wilder, and other leading diabetes specialists in the United States.

At first we experienced considerable difficulty with deterioration of the product and consequently were never certain that material which had given a good test on animals in Indianapolis would give an effective result when tested clinically at some remote point. Cases of coma called for large amounts of material, and in the early days we were more than once compelled to interrupt important experiments or to deflect material from severe cases in order to take care of some emergency coma case.

It should be remembered that during a considerable part of this developmental period we were still dependent on the convulsive test in rabbits, carried out statistically. It was only later that the blood-sugar-lowering test in rabbits was introduced as a standard procedure and much later the convulsive test in mice, which is now so extensively employed. I believe that during the last six months of 1922 we employed over 100,000 rabbits in the conduct of these convulsive tests.

Meanwhile, during the months from August to December, 1922, the problem of Insulin production on a fairly substantial scale was worked out, step by step, as a result of close collaboration between the Toronto and Indianapolis groups. At the beginning of this period, I remember more than one occasion in the course of the summer when 100 to 200 units meant a great deal to Dr. Banting and his patients. A month or two later we were talking in terms of thousands of units, then later in terms of tens and hundreds of thousands, and finally, when Walden perfected his isoelectric fractionation procedure, it could be said that we were really "out of the woods." By the beginning of 1923 we were in a position to produce all that was needed for the clinical treatment, which was still restricted by the Toronto Committee to a limited number of institutions and individuals having experience in its use. The Toronto and Indianapolis groups were working on parallel lines, exchanging information freely and having conferences in Toronto or Indianapolis almost weekly, until finally both groups were able to "get their

heads above water" and were no longer compelled to restrict unduly the supply of Insulin sent out for clinical purposes.

In conclusion, in an attempt to sum up Banting's amazing accomplishments, I should feel inclined to express the opinion that much of the success that he and Best met with was due to the fact that they did not allow themselves to be deterred by adverse reports in the literature. Dr. Macleod was frankly skeptical about the whole enterprise, but fortunately was willing to waive his own opinions and to give Banting facilities to carry out the experiments which he outlined with the collaboration of Dr. Best. I think that this is most important. We should avoid undue regimentation of scientific research. In wartime it is obviously necessary. In peacetime the developmental stages of any problem may, of course, be regimented; but in the initial stage, the breaking open of a new field, such as the first development of Insulin or of penicillin, young investigators full of enthusiasm should be given the greatest possible free hand and should be allowed to carry out experiments as they see fit, in a manner that might not be approved in a thoroughly regimented system. Older men in charge of research laboratories should be extremely cautious about exerting a dominant position or dictating in any way to the younger men working with them. They should simply be advisers. I would venture a guess that had regimentation such as we have seen during the war been practiced at the time that Banting and Best were doing their epoch-making work, the discovery and development of Insulin might have been greatly delayed.

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