Advancement in Insulin Therapy

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THE object of this article is to give a brief review of some of the work which has been done in the past four or five years, for the purpose of advancing insulin therapy; to point out what improvements have been made, and, in some measure to give an explanation of these.

In 1922, Sir Frederick Banting discovered Insulin. After scientists had battled for many years, a disease against which no weapons were adequate, and with apparently no relief in sight, this discovery was hailed as an event which would go down in the history of medicine.

The advantages of this new cure for diabetes mellitus were obvious. They overshadowed to such a degree its few drawbacks, (which, however, really did merit consideration) that for thirteen years no great progress was made in improvements to overcome these deficiencies. However, in late 1935, came word from Hagedorn and his associates, working in Copenhagen, Denmark, that they had developed a compound of Insulin, which was, in many ways,superior to the Insulin Hydrochloride then in use. This latter, they concluded, was too rapidly absorbed into the circulation, causing wide fluctuations in the blood sugar, which therefore causes much subjective discomfort in Insulin therapy. Their object, then, was to slow down the rate of absorption, and to accomplish this purpose various methods were tried, such as: injection as a suspension, or emulsion in oil; injection together with vaso- constrictor substances; and injection of an insulin compound which was but slightly soluble in tissue fluid. These methods were tried for several years, but with no success until, while experimenting along some other line, the observation was made that a combination of Insulin and Nucleic Acid had a more acid isoelectric point than did insulin alone. This fact indicated the possibility of a combination of insulin with some basic substance to form a compound, the isoelectric point of which would be very close to the pH of the tissue fluids. (Here insulin would act as an acid, whereas, in Insulin Hydrochloride, it acts as a base. The resulting compound would then, on injection, be precipitated at its isoelectric point, and the rate of absorption would be retarded.

To seek confirmation of this theory, experiments were carried out with certain groups of compounds, thyrins, histones, globins and protamines, but it was found that only the latter group would combine with insulin to give a solubility which would justify experiment. Accordingly, clinical experiments were prepared, in which the effect of injection of insulin plus a mono-protamine* on the blood sugar would be observed. Extracts of various fish sperm were to be used (sconobrine, salmine,, clupeine, etc.) and since injection of fairly large amounts of these alone showed no ill-effects on patients, it was thought safe to try them.

*Protamines are divided into mono,- di-, and tri-protamines, according to the amount of basic constituents, lysine, histidine, and argenine, contained.

In the first experiment, an injection of insulin plus clupeine was made, the amount of protamine in solution being about one-tenth the weight of the insulin. The resultant effect was, disappointingly, no different from that of ordinary insulin. As this was probably due to the protein of the tissue fluid acting as a protective colloid, thus preventing precipitation, they tried readjusting to pH 7-3, and more encouraging results were obtained. The precipitate thus formed in the tissue fluid has a homogeneous insulin concentration, and, with slow absorption, this provides a steady, uniform supply of insulin which is available over a long period.

The results which were finally adopted were based on more than 15,000 blood sugar determinations, and 3,000 urine examination for sugar and ammonia (ammonia is used to indicate the fluctuations in acidosis). Tests were also made by comparing periods of protamine insulinate treatment, with other periods of treatment with unmodified insulin, in the same individual, all other conditions being identical. It was found that the sudden, abrupt lowering of the blood sugar, which occurs three or four hours after injection with insulin, was avoided by using the protamine insulinate, and the effect of the new compound lasted about twice as long as did that of ordinary insulin. If was not necessary to increase the number of daily injections, and the blood sugar fluctuations which generally occurred in unmodified insulin therapy were cut down considerably. Glycosuria was decreased or prevented, and the danger of hypoglycaemic reactions was reduced. Also, no ill-effects were observed, neither local nor protein reactions; failure of the blood sugar reducing power never occurred; and the effect was equally good in children and adults, whether ambulant or inactive. Subjective discomfort, due to high blood sugar and acidosis, was relieved, and when the infrequent hypoglycaemic reactions do take place, they come on very slowly, so that the patient has sufficient warning to combat them. In diabetes with complications, the protamine insulinate therapy was most satisfactory, especially so in neuritis and in enlargement of the liver, and in discharged patients, it is much more valuable in maintaining a steady blood sugar, than is the unmodified insulin. These results marked the first step in the advancement of Insulin Therapy.

At about the same time as the above work, Scott and Fisher[2], working at the University of Toronto, carried out experiments to determine the effect of certain metals on the action of Insulin. They concentrated mainly on zinc, because it is a normal constitutent of the pancreas, combines easily with insulin, and because it is nearly always found in crystalline preparations of insulin. They found that when zinc salts were added to insulin, the latter was reduced to 40% of its original activity, resulting in a prolonged blood sugar reducing effect, and reduced hypoglycaemic reactions and convulsions. Convulsions which took place occurred late in all tests, and were very mild, showing a delayed action of the insulin. The total effect of the insulin was not reduced in any way, and though the action was prolonged, the total amount of sugar metabolized was the same as it would be with insulin alone. Different zinc salts were used, but the actual

amounts of zinc in each case were equal, and since the results were invariably the same, they concluded that the change in effect was due to the zinc alone. Also, the effect of separate injections of insulin and zinc chloride was the same as that of insulin alone, which indicated that the combination of zinc and insulin took place before injection.

The next investigation[3] which these men undertook, was to determine if metals played any part in the action of Hagedorn's Protamine Insulinate. Again they used zinc (zinc forms complex salts with basic substances such as amines), and the protamines employed were obtained from the tests of the various types of salmon,—cohoe,—spring, steelhead,—and rainbow trout. The experiments were carried out on rabbits, and different combinations of insulin, protamine and zinc were used, to study their effects. These combinations were: (1) Insulin alone, as a control; (2) Insulin plus protamine; (3) Insulin plus zinc; (4) Insulin plus protamine plus zinc. The insulin and protamine used had a very low ash content, i.e., the amount of zinc, which they naturally contained was practically Insulin plus protamine; (3) Insulin plus zinc; (4) Insulin plus protamine both with a low ash content, was about twice that of the insulin alone, the most prolonged hypoglycaemic reaction was obtained by using insulin plus protamine plus zinc. Therefore, they suggest that zinc is responsible for the combination of insulin and protamine, and consequently, that zinc is the important factor in producing prolonged hypoglycaemia.

In further experiments [3] by Scott and Fisher, it was found that other substances, namely spermine and thymine, had the same power of prolonging the insulin effect. Again, zinc was necessary for the combination. The effect of spermine is of particular interest, because it is obtained from the pancreas, as also is zinc, and the effect of insulin plus spermine plus zinc may more nearly approach the normal physiological action of insulin. Although insulin is recovered from the pancreas as such, it is quite certain that this is not the form in which it acts.

In contrast to the results of Scott and Fisher are those of Altshuler and Leiser[4]. In their experiments they used four different preparations, as follows: (1) Standard commercial insulin, with a zinc content of 0.02 to 0.05 mgm. per 1000 units of insulin. (2) The first preparation with zinc added to make a content of 1.2 mgm. per 1000 units. (3) Crystalline insulin with 0.8-0.9 mgm. of zinc per 1000 units. (4) Crystalline insulin with a zinc content of .25 mgm. per 1000 units. The results show a better control of blood sugar with (3) and (4) and that (1) and (2) have the same effect, indicating that the prolonged action of crystalline insulin is not entirely due to the zinc content. However, the results of Scott and Fisher are the ones which have held under test.

It has been noted in some clinical observations [5], that occasional glycosuria may occur in protamine zinc insulin therapy. This has been explained as being due to (1) a sudden lowering of the renal threshold (about 60 mgm. glucose per 100 cc. blood) while the blood sugar is decreasing, and (2) an increase of the blood sugar after the ingestion of a meal. This second fact involves another point which may be dealt with

here. The normal pancreas is pictured as having two different secretions [6]: one, a slow, steady supply of insulin which is sufficient to control the endogenous blood sugar; and another, a quick, intermittent supply which regulates the metabolism of ingested sugar. It would seem then, that, although the protamine zinc insulin would simulate the first secretion, a supply of quick acting, unmodified insulin would be necessary to control the sudden rise of blood sugar after a meal. Again, some irregularity of results has been noted, owing to the fact that protamine zinc insulin is a suspension, and injections may vary in insulin content, and also that the rate of absorption from various sites of injection may differ.

To explain the absence of hypoglycaemic reactions even in the face of long periods of low blood sugar values, it is necessary to consider the action of adrenalin. When the blood sugar reaches hypoglycaemic levels, there appears to be a protective mechanism brought into play which returns it to a normal level. Studies of blood sugar and blood pressure [5] indicate that it is adrenalin, which favours glycogenolysis. In the use of unmodified insulin, the amount of *free* insulin available is too great to be counteracted by the small amount of adrenalin set free, whereas, in the case of protamine zinc insulin the free insulin supply is small, and the release of adrenalin is sufficient to combat it. Furthermore, the rate of fall of blood sugar is just as important in causing hypoglycaemic reactions as is its actual level [7], and protamine zinc insulin shows a more gradual falling off of the blood sugar than insulin, which has an abrupt blood sugar lowering effect.

The adrenalin mechanism is also important in infection [8], where large amounts are released due to overstimulation of the sympathetic nervous system, and the adrenalin, acting synergically with thyroxin, which is also produced, gives a high insulin resistance. The use of unmodified insulin, with its intense, rapid action, is preferable in such cases.

In addition to infection there are certain other contra-indications for protamine zinc insulin therapy [9], such as coma, emergency pre- and post-operative treatment, and all other conditions in which the rapid action of unmodified insulin is required, although a large enough dose of protamine zinc insulin causes a fairly rapid lowering of the blood sugar.

With regard to the prolonged effect of protamine zinc insulin, it has been suggested that this is due to its insolubility in the tissue fluid. But in experiments by Scott and Fisher [2], where this action was compared with that of insoluble crystalline insulin, the results showed that insolubility alone would not account for the prolonged blood sugar lowering effect, and they maintain that the question of chemical combination is also involved here.

Summary:

From the foregoing, certain facts are apparent in the treatment of diabetes mellitus by means of protamine zinc insulin, thus:

(1) The effect lasts at least twice as long as that of unmodified insulin.

(2) The blood sugar is maintained at more constant levels.
(3) Glycosuria is decreased or prevented.
(4) Hypoglycaemic reactions seldom occur, and when they do, there is sufficient warning to combat them.
(5) Subjective conditions are improved.
(6) Conditions which require a large, immediately available supply of insulin are contra-indicative of the use of protamine zinc insulin.

These observations have all been substantiated by clinical evidence.

In conclusion, it might be well to note some statistics [10], which are interesting, and especially valuable in refuting adverse criticism which one frequently hears, against the use of insulin. Comparison of a diabetic's expectation of life in two periods: one, 1897-1914, and the other, 1914-1929, have been prepared by the Metropolitan Life Insurance Company, and they show that at ten years of age the average expectation in the second period is 31.7 years, compared with 1.5 years in the first period; at thirty years of age it is 22.7 years compared with 4 .2 years; and at fifty years of age it is 13.2 years compared with 8.1 years. These were compiled before 1930, and make no allowances for any advances made since then. Furthermore, in diabetic doctors, men who have a knowledge of the disease, death caused by diabetic coma has dropped to 3% since 1922; the death rate of diabetic M.D.'s is far below that of ordinary patients, and in the diabetic patients of Elliott Joslin [10], doctors outlived the others by five years, the death ages being respectively 68 years and 63 years. Also, the diabetic who has a knowledge of his disease, and looks after it properly, can, and will, outlive his non-diabetic friends of the same age. In treatment with unmodified insulin, the diabetic could maintain a normal blood sugar for one-half to three-quarters of the 24 hours only. If the disease lasted for, say, twenty years, this would mean living a total of five to ten years with an abnormal blood sugar, and being subject to all the dangers to which this leads. With protamine zinc insulin therapy, the possibility of such dangers is eliminated, by the production of a constant blood sugar. The more nearly normal physiological state thus produced will probably raise bodily health to a level where the individual can effectively ward off such diabetic complication as infection, abnormal neurological and ophthalmological conditions, and vascular degeneration.

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