THE DURATION OF THE ACTION OF DIFFERENT INSULINS

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This investigation was undertaken to compare the different insulins under the same experimental conditions, and to ascertain the duration and, if necessary, the character of the action of new insulins brought on the market. The textbooks on insulin do not supply any data on the duration of action (Grevnstuk and Laqueur, 1925; Hill and Howitt, 1936; Geiling *et al.*, 1937; Jensen, 1938; Joslin, 1946).

Experimental Method

The experimental conditions are of great importance. In the first place, it would seem desirable to investigate the action of insulin in human beings, and so avoid the necessity of adapting the results obtained with test animals. Besides, the experimental conditions of this investigation would not be suitable for animals. It would, of course, have been advisable to use diabetic patients, but for various reasons this was not possible.

The subjects were young healthy student volunteers, whose blood sugar was investigated under standardized conditions, as follows. They were asked to lie down from 11 p.m. the night before—that is, seven hours previous to the experiment—and also during the experiment, in order to eliminate the influence of exercise on the blood sugar. From 6 a.m. onward on the day of the experiment they ate 50 g. of mashed potatoes (about 10 g. of carbohydrate) and drank 30 ml. of water every hour, until the experiment was finished—that is, until the average blood sugar had returned to its original level. Every hour the blood sugar was determined according to the Hagedorn-Jensen method. The values at 7 a.m. and 8 a.m. were used as a starting-point. At 8 a.m. 20 units of insulin was injected subcutaneously, a different brand of insulin being given to each group of test persons. One group which did not receive any insulin served as controls.

In all, 10 different insulins were tested. The accompanying Chart shows the number of persons in each group from whom the average was calculated. This method was chosen because it eliminates all other factors which might influence the blood-sugar level (Himsworth, 1934; Martin *et al.*, 1940). As fasting might decrease the blood sugar, food to a standardized amount was given each hour, to prevent the rise in blood-sugar level which would occur with a larger food intake and which might simulate the end of the action of the insulin. On this matter Peck (1942) states: "There have been only a few published reports of clinical investigations of protamine-zinc insulin which have taken advantage of the invaluable procedure of measuring the rate of insulin action against a known constant carbohydrate intake."

With the present method one finds a constant average blood-sugar level in healthy people, so that any decrease of this level may be attributed to the insulin. If diabetic patients are examined under these conditions a 24-hour rhythm is found in the blood sugar (Gerritzen, 1942). This makes it very difficult, if not impossible, to judge whether the decrease is in consequence of the 24-hour rhythm or whether it should be attributed to insulin. Besides, it is impossible to find enough diabetic subjects in whom to calculate the average blood-sugar level after the administration of insulin.

The hourly intake of 10 g. of carbohydrate diminishes the hypoglycaemia, and this would meet the objection that

Chart showing duration of action of various insulins.

with a strong decrease of the blood sugar a regulating mechanism, with adrenaline as its principal component (Drury and Greeley, 1939), might become active and thus render evaluation of the action of insulin impossible. During my experiments only minor hypoglycaemic symptoms occurred, and, as expected, without any close relation to the level of the blood sugar.

In addition to the duration of action of equal doses (20 units) of various insulins, it was my intention to find out whether there were any differences in the character of their action (de Jongh, 1924). While assuming that the resorption of insulin in diabetic persons does not in any way differ from that in normal individuals, I think that the differences in action in the latter may also apply to diabetics. In other words, that the order of succession in the duration of the action, as demonstrated in this experiment with healthy

individuals, may also be supposed to exist in diabetic patients. It is obvious, of course, that the actual figures characterizing the duration of the action of insulin in this experiment need not apply to diabetics. However, if, under the conditions of this experiment, it is found that regular insulin is effective for eight hours, this result would agree fairly well with the data in the literature (Scott and Dotti, 1932; Lawrence, 1938; Martin *et al.*, 1940; Peck, 1942; Colwell, 1947), and one would not be far out in accepting the figures as applying to diabetics.

Results

In the control group no significant deviation was found between the average blood sugar values at different hours. There was a significant deviation between the blood-sugar values of the different test persons, calculated according to the F-test (Davies, 1949), the standard error of which was 6.9 units if the blood-sugar values are expressed in percentages to the third place of decimals. In the group receiving insulin there was no significant increase of this standard error in comparison with the control group.*

Control Group.—The average blood-sugar level does not show any oscillations, and may be considered constant.

Regular Insulin (Organon).—The blood sugar reaches its lowest point after one hour and returns to its starting point in eight hours.

Protamine-zinc Insulin (Organon).—Contrary to the many data in the literature, I find for P.Z.I. a duration of action of 18 hours. No evidence was found of a duration of 48 hours or longer. The lowest point is reached in five to eight hours.

Di-insulin (Novo).—Duration of action, 16 hours. The greatest decrease in blood sugar is during the first three hours.

Iso-insulin (Novo).—Duration of action 17 hours. Rapid decrease of sugar in the first hour. The steep decrease within the first hour leads to the assumption that this preparation contains regular insulin, but, according to Hallas Möller, this is not the case.

Globin Insulin (Burroughs Wellcome).—Action is identical with that of di-insulin.

(1) *N.P.H. 50 (Organon)*.—Duration of action, 11 hours. Lowest point after four hours.

(2) *N.P.H. 50 (Lilly)*.—Duration of action, 11 hours. Lowest point after three hours.

Insulin Mixtures (Organon).—The impression is gained that the duration of action of the mixture depends on the regular insulin content only so far as the decrease during the first three hours is greatest with the mixture containing most regular insulin—the 3:1 mixture—and that thereafter the blood sugar takes longer to come back to the starting-point than with the 2:1 and the 1:1 mixtures. This would explain the longer duration of the action with a higher regular insulin content.

I am indebted to Miss S. P. I. Hoogveen for the accurate blood-sugar determinations, and to Mr. F. H. Stieltjes for the statistical calculations. I am grateful also to the 64 students who voluntarily served as test subjects. This experiment was made possible by a grant from Messrs. Burroughs Wellcome and Co.

*An analysis of variance is effected with the control group, globin insulin 1:1 mixture, 2:1 mixture, and 3:1 mixture according to Fisher (1937). The χ^2 test (Bartlett test 3), applied to all error variances, showed that the inclusion of the 1:1 mixture in the calculations led to very uncertain results; otherwise this experiment had a very low error variance. The χ^2 test was employed to find whether there was an individual variance of reaction to insulin, as this would only result in an increase of the error variance. There remains, therefore, a reasonable chance that there are no individual differences of reaction to insulin. This implies that there is a reasonable possibility that, by increasing the number of test persons, more definite results might be obtained.

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The Ministry of Health has reminded local authorities (circular 5/52 of January 21) that they have powers to find and pay for temporary homes for mental defectives when these patients cannot be looked after by their families—for example, during illness in the house, or in holiday periods.