Comparison of the Hyperglycemic Effects of Glucocorticoids in Human Beings

The Effect of Heredity on Responses to Glucocorticoids

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Fajans and Conn[1] recently called attention to a difference between the responses of persons with and without a family history of diabetes mellitus to glucose tolerance tests modified by prior administration of cortisone. Under the conditions of their experiment, the majority of subjects exhibited very slight impairment of glucose tolerance after cortisone. However, in a group with a positive family history of diabetes, 24 per cent of the subjects showed a substantial impairment of glucose tolerance after cortisone administration. This contrasted with a 3 per cent incidence of significant impairment in a group with no evident family history of diabetes. Hoet has reported investigations concerning the effect of cortisone on diabetes and "pre-diabetes." [2]

One of the purposes of these investigations was to evaluate the ability of a simplified glucose tolerance test as modified by prior administration of a glucocorticoid to establish the hyperglycemic potencies of glucocorticoids in human beings. Another purpose was to investigate the degree to which heredity, especially diabetic heredity, influences the response to glucocorticoids.

PROCEDURE

All subjects received a high (300 gm.) carbohydrate diet for at least three days prior to the test. Glucose was administered orally (1.75 gm. per kg. of ideal body weight). A dose of a glucocorticoid was administered by mouth eight and one-half hours and two hours before the oral glucose. The venous blood glucose was determined by the Somogyi-Nelson method[3] two hours after oral glucose. This test, then, is nothing more than a highly simplified glucose tolerance test except that a steroid is administered before the test.

The order in which the glucocorticoids were tested was not systematically randomized. However, a segment of those subjects tested consecutively to three steroids was systematically randomized (fourteen subjects, forty-two tests). Statistical analysis using the t test showed no effect of order of testing on the results. The specific dosages used in testing each of the steroids are indicated in the accompanying figures. It should be noted that the dose of the steroid was increased 25 per cent for each subject who weighed more than 160 lb. The "priming" dose of a steroid administered eight and one-half hours before the oral glucose was given in order that our data might be compared with those of Fajans and Conn. All subjects were healthy and ambulant.

RESULTS

Comparison of hyperglycemic potencies of glucocorticoids. Figure 1 shows the degree to which cortisone acetate, prednisone, and prednisolone modified glucose tolerance when given orally in a dosage ratio of 5:1:1 (50 mg. cortisone acetate, 10 mg. prednisone, and 10 mg. prednisolone). This group of thirty-nine healthy, nondiabetic subjects ranged in age from eighteen to forty-seven. Each subject had a glucose tolerance test, a cortisone-glucose test, a prednisone-glucose test and a prednisolone-glucose test.

The fairly wide variations in the responses to each of the steroids among individuals is evident. However, the responses of a particular individual to each of these three steroids were usually, although not invariably, of relatively similar magnitude. For example, a subject who exhibited a response to cortisone above the average for the group usually showed responses to prednisone and prednisolone which were also above average. Table 1 shows the data from which figure 1 was derived and is included to illustrate the variations in response to the drugs demonstrated by each test subject. It may be noted that the presence or absence of a family history of diabetes is indicated in this table.

The average responses of this group were 109 mg. per

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TABLE 1

Results of glucocorticoid-modified glucose tolerance tests

Subject No.	Age	Family history of diabetes	Sex	Two-hour blood glucose in mg. per cent: Control tests after oral glucose	Cortisone-glucose test	Prednisone-glucose test	Prednisolone-glucose test
1	24	No	M	71	104	146	136
2	24	No	M	84	114	132	128
3	21	No	F	74	146	120	116
4	24	No	M	71	96	180	114
5	30	No	M	74	108	124	104
6	25	No	M	76	80	100	96
7	24	No	M	80	142	88	96
8	23	No	M	84	80	76	84
9	23	No	M	92	88	132	108
10	27	No	M	100	80	156	84
11	26	No	M	52	100	124	124
12	28	No	M	80	80	100	76
13	23	No	M	92	114	120	124
14	25	No	M	88	92	116	116
15	21	No	M	100	104	72	116
16	43	No	M	100	146	180	142
17	28	No	M	71	48	114	84
18	37	No	M	76	92	120	108
19	38	No	M	52	96	76	74
20	33	No	M	76	114	172	84
21	31	No	M	71	192	88	132
22	35	No	F	60	146	114	200
23	24	Yes	M	65	75	100	120
24	23	Yes	M	79	142	124	96
25	23	Yes	M	60	66	74	84
26	18	Yes	F	108	172	208	192
27	32	Yes	F	80	114	200	280
28	36	Yes	F	76	160	204	187
29	40	Yes	F	80	132	100	120
30	28	Yes	F	74	108	104	176
31	30	Yes	F	71	76	132	108
32	24	Yes	F	71	114	187	142
33	39	Yes	M	104	88	166	128
34	40	Yes	F	92	88	100	116
35	18	Yes	F	104	108	180	172
36	28	Yes	M	84	124	136	120
37	21	Yes	M	76	84	76	120
38	21	Yes	F	54	114	88	56
39	47	Yes	F	100	124	120	172

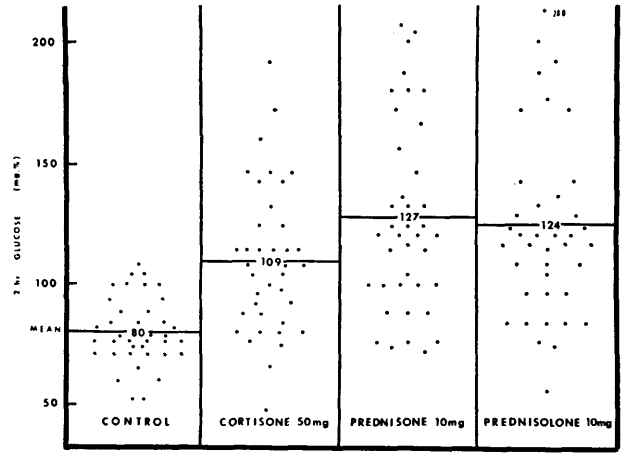

FIG. 1. The effect of glucocorticoids on glucose tolerance. Each dot represents a blood glucose value obtained two hours after ingestion of glucose. Each of thirty-nine subjects was tested on four occasions. In the first column marked "control" their responses to oral glucose alone are represented. In the remaining columns are presented the response of these same subjects to subsequent tests modified by prior administration of a glucocorticoid.

cent after cortisone, 127 mg. per cent after prednisone, and 124 mg. per cent after prednisolone. Statistical analysis indicates that the difference between the responses to prednisone and prednisolone was not significant ($p = 0.6$-0.7). The responses to both prednisone and prednisolone were significantly greater than the responses to cortisone (p value for prednisone $= 0.02$-0.01; p value for prednisolone $= 0.02$-0.025).

A clearer representation of the responses of this group to these steroids is obtained by expressing the responses as milligrams per cent above the control level. After cortisone plus oral glucose these subjects showed two-hour glucose values which were, on the average, 29 mg. per cent above the values after glucose alone. Prednisone and prednisolone caused elevations above the control levels which averaged 47 mg. per cent and 44 mg. per cent respectively. It may be noted that prednisone and prednisolone modified glucose tolerance to the same degree while both were decidedly more potent than cortisone even though the dose of cortisone acetate was five times greater. (Since 50 mg. of cortisone acetate contains approximately 44 mg. of cortisone, the steroids were actually compared in a ratio of 4.4 of cortisone to one to one.)

Figure 2 shows the blood glucose responses of fourteen subjects, each of whom had two glucose tolerance tests (blood glucose two hours after oral glucose), one after 40 mg. of hydrocortisone and another after 10 mg. of prednisone. The average two-hour glucose value after prednisone was 137 mg. per cent as compared to 145 mg. per cent after hydrocortisone. This difference is not of statistical significance, having a p value of greater than 0.7. These data suggest that the hyperglycemic

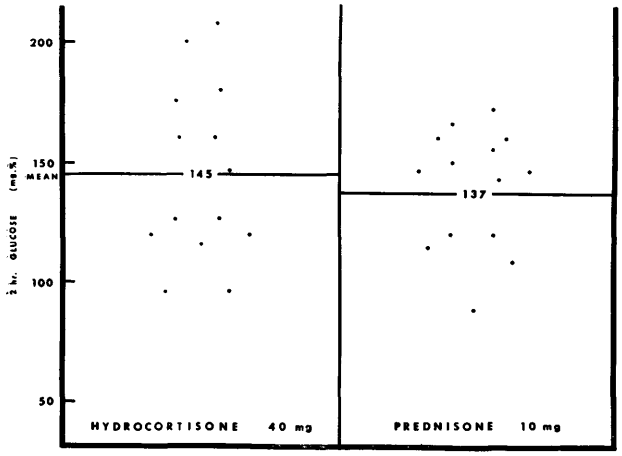

FIG. 2. Comparative responses (two-hour blood glucose) of fourteen healthy subjects each of whom had a hydrocortisone-modified and a prednisone-modified glucose tolerance test. See procedure for technic of testing.

potency of prednisone (and by inference prednisolone) is four times that of hydrocortisone. It may be noted that the mean response to prednisone of the subjects in figure 2 (137 mg. per cent) was greater than the mean response of those in figure 1 (127 mg. per cent). This was probably due to the fact that the results shown in figure 2 were from tests done on a group in which the majority of the subjects had a strong family history of diabetes (see figure 4). A control test (blood glucose two hours after glucose load without prior administration of a drug) was not done in each of these fourteen subjects. However, it was done in eleven instances and the mean value was 85 mg. per cent.

A group of seven subjects were given a glucose tolerance test after 2 mg. of 9-alpha-fluorohydrocortisone. None of the seven showed any hyperglycemic responses to this dose of the drug. This indicated that 9-alpha-fluorohydrocortisone was very definitely less than twenty times as hyperglycemic as hydrocortisone in humans.

The ability of this method to distinguish relatively small differences in hyperglycemic potencies was tested further by comparing the responses of healthy adults to 40 mg. hydrocortisone and 50 mg. of cortisone acetate. Since the response of each subject was greater than hydrocortisone the data were analyzed after six subjects had been tested with both drugs. The p value was less than 0.01 indicating a difference in potencies had been identified after only twelve blood glucose determinations.

An effect of age on steroid-modified glucose tolerance has not been previously reported. Our preliminary studies have indicated that older subjects are more likely to exhibit a "positive" response. Sufficient data are not available at present to describe clearly the effect of age on the response to such tests, since we have tested very few subjects over fifty or under twenty. In order to test the effect of age on the response to cortisone-glucose tolerance tests, all of the nondiabetic healthy subjects who had had this test were divided into two groups. The younger (below age thirty-one) showed two-hour glucose levels which averaged 17 mg. per cent above the control levels. The older subjects (above age thirty) had levels averaging 59 mg. per cent above control levels. Analysis of results in other groups concerning the effect of age on various types of steroid-glucose tolerance tests showed less significant differences but a positive correlation of some degree was consistently seen.

The influence of heredity on responses to glucocorticoids. The lack of influence of a family history of diabetes on responses of a small group of subjects to cortisone-glucose tolerance tests is illustrated in figure 3. The groups were carefully matched for age. The mean

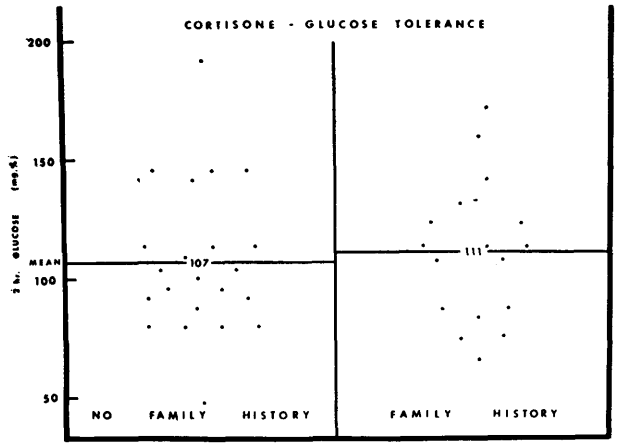

FIG. 3. A comparison of responses of twenty-two subjects having no family history of diabetes with those of seventeen individuals having diabetes in their families to a cortisone-glucose tolerance test. All subjects were nondiabetic.

FIG. 4. A further comparison of the response of subjects represented in figure 5 to glucose tolerance tests as modified by prednisone and prednisolone.

age for the group with a diabetic parent, sibling or child was twenty-nine (range 18 to 47), while the mean age of the subjects with no family history of diabetes was twenty-eight (range 21 to 43). The mean values were 107 mg. per cent for a group with no family history of diabetes and 111 mg. per cent for those with a family history of diabetes. Although cortisone-modified glucose tolerance test results were very slightly higher in the group with a diabetic parent, sibling or child, the difference between the groups is not statistically significant.

Our results resemble closely those reported by Fajans and Conn in that 20 per cent of the group with a family history had a two-hour glucose above 140 mg. per cent as compared to 24 per cent of their subjects with a similar family history. However, responses above 140 mg. per cent in their group with no family history of diabetes were rare (3 per cent), whereas our subjects with a negative family history exhibited values above 140 frequently (23 per cent). The results of investigations done by us more recently than those reported here suggest that the incidence of a "positive" response to cortisone in persons with no family history of diabetes may not be as great as was suggested by the results in this small group. Furthermore, the data of Fajans and Conn include a larger, and thus a statistically more reliable, sample.

Figure 4 shows that a group with a diabetic parent, sibling or child exhibited greater responses to both prednisone and prednisolone than did a group with a negative family history. The mean responses of the subjects with and without a positive family history were 135 mg. per cent and 120 mg. per cent after prednisone, while the mean values after prednisolone were 140 mg. per cent and 111 mg. per cent respectively. Statistical analysis showed that the group with a family history of diabetes exhibited a significantly greater response to both prednisone ($p < 0.02$) and prednisolone ($p < 0.01$) when compared to the control group.

It seemed important to study further the degree to which the response to glucocorticoids was conditioned by heredity, especially by diabetic heredity. For this reason preliminary studies are under way utilizing several special groups of subjects including (a) identical twins and (b) persons with a diabetic mother and father.

The responses of identical twins to steroid-glucose tolerance testing are strikingly similar. Results of testing carried out in ten pairs of identical twins (twenty subjects) were analyzed statistically with the following findings: (1) The responses of the pairs to the control test (to oral glucose alone) were significantly correlated ($r = 0.67$). (2) The responses of the pairs to prednisone were significantly correlated ($r = 0.58$; $p = 0.05$). This latter positive correlation is of great interest since effect on blood glucose produced by the glucose load per se was separated from the effect of prednisone. This was done by defining the response to prednisone as "glucose excess" produced by prednisone plus oral glucose above that produced by glucose loading alone. This suggests that heredity conditioned the response to the steroid itself.

The statistically significant correlations established for these twin pairs was not due to the equality in age of each twin and his twin. The method used in calculating this correlation took into account the effect of age in the total group before yielding the final correlation results.

The degree of similarity between the responses of twin

and twin was tested further in the following manner. First of all, the response of each twin to the corticoid was identified by determining the degree to which the drug elevated the blood glucose above the control value. The differences between the responses of twin and twin was compared and in the first pair found to be 4 mg. per cent, 4 mg. per cent and 10 mg. per cent for cortisone, prednisone and prednisolone respectively, whereas the differences between the responses of one of these twins (randomly selected) and ten randomly selected young adults were far greater. These averaged 27 mg. per cent, 50 mg. per cent, and 28 mg. per cent for cortisone, prednisone and prednisolone respectively.

The similarity between the second pair of twins tested was also analyzed in this manner. The differences between twin and twin were 10 mg. per cent, 16 mg. per cent and 2 mg. per cent. The difference between one of this pair and ten randomly selected young adults averaged 43 mg. per cent, 49 mg. per cent and 28 mg. per cent.

Studies on twins will not yield, of course, specific information concerning diabetic heredity until a sufficient number of identical twins with diabetic heredity are tested.

The ability of the steroid-glucose tolerance test to identify potential diabetics is being tested further by studying a group of subjects (four families) by glucose tolerance testing as modified by prednisone are shown in figure 5. It may be noted that although both of these groups exhibited greater responses to the test than did subjects with no family history of diabetes, the group whose mother and father were both diabetic showed no tendency toward greater responses than those subjects with a much less significant family history of diabetes mellitus. The responses of these two groups to cortisone-glucose tests were also comparable. If a larger number of subjects with two diabetic parents show no greater propensity to respond "positively" to steroid-glucose tolerance testing than has been exhibited by this small group then it is likely that such testing will have a limited specificity for identifying potential diabetes.

A nondiabetic subject who had exhibited a marked response to other glucocorticoids was given a glucose tolerance test after prior administration of 2 mg. of 9-alpha-fluorohydrocortisone. Even though seven previous subjects had shown no response, this subject had a two-hour blood glucose 44 mg. per cent higher after the fluorohydrocortisone-glucose test than after the glucose alone. This subject's mother (also nondiabetic) responded significantly to the same dose of fluorohydrocortisone. The latter two test results are described in order to cite examples of nondiabetic subjects who display unusual

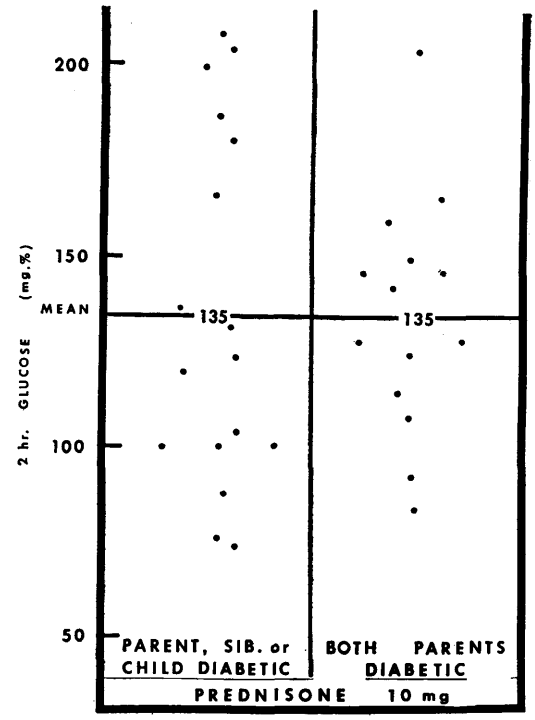

FIG. 5. A comparison of the results of prednisone-glucose tolerance testing in two groups with different degrees of diabetic heredity. Results in the first column are in subjects giving a history of diabetes in a child, sibling or single parent. In the second column, the results in subjects having two diabetic parents are presented.

sensitivity to small amounts of glucocorticoids. They also suggest that this tendency is to some degree conditioned by heredity (in this instance possibly diabetic heredity since there was a strong family history of diabetes).

DISCUSSION

The 50 mg. dose of cortisone acetate had been selected in order that these results might be compared with those of Fajans and Conn. The doses of the other glucocorticoids were chosen because their "organic metabolic-regulating potencies" were roughly similar to the potency of 50 mg. cortisone acetate and because tablets were readily available for oral administration at these dosages.

The ability of this method to identify small differences between the hyperglycemic potencies of 10 mg. prednisone and 50 mg. cortisone acetate on the one hand and between 40 mg. hydrocortisone and 50 mg. cortisone acetate on the other is impressive, especially since this difference was established by testing only a few subjects. A fairly large number of tests is necessary to establish accurately the hyperglycemic potency of a glucocorticoid if the potency is to be estimated by measuring the responses of a group of subjects to various doses of the

steroid. However, if the same subjects are tested with both a known and an unknown steroid, an accurate estimation of the hyperglycemic potency may be obtained after as few as twenty tests. When subjects are available whose sensitivities to a glucocorticoid of known potency (such as hydrocortisone) have already been established, it should be possible to estimate fairly accurately the potency of an unknown after administering it to a very few subjects. It was, for example, possible in one day to show that 9-alpha-fluorohydrocortisone was definitely less than twenty times as potent as hydrocortisone by testing seven subjects.

New glucocorticoids to be tested subsequently could conceivably exhibit a lack of correlation between their hyperglycemic potencies on the one hand and their anti-inflammatory and pituitary inhibiting potencies on the other. However, it may be noted that the hyperglycemic potencies of these glucocorticoids closely approximate their other "organic metabolic-regulating" potencies which were established previously by more elaborate methods. For example, these results suggest that prednisone and prednisolone are of equal hyperglycemic potency, and that both of these drugs are four times more potent than hydrocortisone. It is of interest that this same ratio has previously been established for the anti-inflammatory potencies of these three glucocorticoids. This type of procedure, then, may be a convenient way of estimating the "organic metabolic-regulating potency" of a glucocorticoid.

SUMMARY AND CONCLUSIONS

The carbohydrate metabolism regulating (or hyperglycemic) potency of a steroid may be quickly and easily estimated by testing the degree to which it modifies glucose tolerance (as measured by a single blood glucose determination two hours after glucose by mouth). The hyperglycemic potency of a steroid may be accurately estimated by testing only a few subjects if they are tested to both a known and an unknown steroid with similar characteristics of time-action.

Using such a method the carbohydrate metabolism regulating potencies of oral prednisone and prednisolone were measured and no significant difference found.

Under the circumstances of this experiment prednisone and prednisolone were more than five times more potent than cortisone acetate in their hyperglycemic effects. Prednisone and hydrocortisone were identical in effect when administered orally in a dosage ratio of one to four.

Older subjects showed a "positive" response to this steroid-glucose test more frequently than younger.

The strikingly similar responses of identical twins to glucose tolerance tests and to steroid-glucose tolerance tests suggest that heredity plays a significant role in conditioning the hyperglycemic response both to glucose loading and to glucocorticoids.

Nondiabetic subjects with a family history of diabetes showed significantly greater hyperglycemic responses to prednisone and prednisolone-glucose tolerance tests than a control group.

The responses to prednisone-glucose tolerance testing of fourteen subjects with a positive family history were no greater than the responses of subjects with a much less significant family history. These latter data are not adequate to permit a final conclusion but suggest that the steroid-glucose tolerance test has a limited ability to predict the eventual development of diabetes.

Although long-term follow-up of persons who exhibit both "positive" and "negative" responses to tests is planned, it is hoped that the degree to which the responses to steroids are conditioned by diabetic heredity may be elucidated at an earlier date by using the methods resembling those described above.

SUMMARIO IN INTERLINGUA

Comparation del Effectos Hyperglycemic de Glucocorticoides in Humanos: Le Effecto del Hereditate Super le Responsa a Glucocorticoides

Le potentia del effecto regulatori que es exercite per un steroide super le metabolismo de hydratos de carbon —i.e. su potentia hyperglycemic—pote esser estimate facile- e rapidemente per testar le grado a que illo modifica le toleration de glucosa (mesurate per medio de un sol determination de glucosa sanguinee duo horas post le administration de glucosa per via oral). Le potentia hyperglycemic de un steroide pote esser estimate accuratemente per testar solmente un parve numero de subjectos, providite que le tests es executate con duo steroides—le un cognoscite, le altere non—que possede simile characteristicas quanto al conditiones temporal de lor action.

Per medio de un tal methodo le potentia de regular le metabolismo de hydratos de carbon esseva mesurate pro prednisona e prednisolona in administrationes oral. Nulle differentia significative esseva trovate.

Sub le conditiones de iste experimento, prednisona e prednisolona se monstrava plus que cinque vices plus efficace que acetato de cortisona in lor effectos hyperglycemic. Prednisona e hydrocortisona esseva identic in lor effectos quando illos esseva administrate oralmente in doses del proportion de un a quatro.

Personas de etates plus avantiate monstrava responsas "positive" in iste test de steroide e glucosa plus fre-

quentemente que plus juvene subjectos.

Le frappantemente simile responsas de geminos identic a tests de toleration glucosic e a tests de toleration steroido-glucosic suggere que le hereditate es un factor de importantia in conditionar le responsa hyperglycemic a cargas de glucosa e etiam a glucocorticoides.

Subjectos nondiabetic con historias familial de diabete monstrava significativamente plus marcate responsas hyperglycemic a prednisona e prednisolona in iste tests que un gruppo de controlo.

Le responsas in tests de toleration a prednisona-glucosa in dece-quatro individuos con positive historias familial non esseva plus pronunciate que le responsas de subjectos con multo minus significative historias familial. Iste datos non suffice pro justificar le derivation de conclusiones final, sed illos suggere que le test de toleration steroido-glucosic es limitate in su poter de predicer le disveloppamento futur de diabete.

Ben que observationes consecutori a longe duration es planate con gruppos de personas que exhibi responsas positive o qui exhibi responsas negative, il es a sperar que le grado a que le responsas a steroides es conditionate per un hereditate diabetic pote esser elucidate plus rapidemente per medio de methodos simile a illos describite in le presente reporto.

ACKNOWLEDGMENT

The author would like to acknowledge the technical assistance of William Hood, Sandra Chiles and Richard Cahill.

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DISCUSSION

STEFAN S. FAJANS, M.D., (*Ann Arbor, Michigan*): I have been most interested in Dr. West's study on the effect of carbohydrate-active adrenal steroids on glucose tolerance in human subjects. Dr. West permitted me to see his slides yesterday evening. In general, Dr. West has found differences in response to adrenal steroids between the two groups of subjects, namely, those without a family history of diabetes as compared with those nondiabetic subjects with a family history of diabetes.

This would be in line with the findings that we reported before this society three years ago. As we mentioned at that time, the question of various time dose schedules had to be explored further; I am very happy to see Dr. West has done this. In order not to introduce further variables, we have not changed our technic so far. Now, Dr. West's study does bring out those differences in steroid dosages as far as response is concerned.

In our group of 100 controlled subjects without a family history of diabetes, we found three individuals who gave a positive response following cortisone. I mean in a two-hour blood sugar more than 140 mg. per cent.

Dr. West found a much higher incidence of positive responses in subjects without a family history of diabetes, if I remember correctly about five out of twenty-five.

The following differences between our studies have occurred to me. First of all, Dr. West's study indicated patients above age thirty gave a higher incidence of response than younger ones. In our series only one individual among a good number of controlled subjects above the age of thirty gave a positive response. Also in our subjects with a family history of diabetes there didn't seem to be much change in incidence of response by age; so there seems to be some disagreement in findings.

Another difference is that Dr. West's subjects received three courses of steroids on alternate days: cortisone, prednisone and prednisolone in various sequences.

Now, he has told me that as a group these responses to cortisone were the same irrespective of the order of administration of these steroids. This seems to be an important difference in our approaches when it comes to evaluating individual subjects rather than a group. Giving steroids daily and repeating the glucose tolerance test, we certainly get modifications in our responses.

Dr. West also mentioned the close parallelism between the increase in blood sugar following cortisone and prednisone and prednisolone irrespective of family history. If there was such a close parallelism, why did he get differences in response with prednisone and prednisolone, but not with cortisone?

One also wonders whether the patients with negative family history have the same incidence of familiarity with background in Oklahoma as in Michigan. This is something you certainly can't settle.

When one does only a two-hour blood sugar determination in a control test, one may include individuals who might under complete glucose tolerance test give a high peak, but have a normal two-hour blood sugar. Such individuals would not be called diabetic by us, but would certainly be eliminated from a control group.

Since he was dealing with four families, I don't believe the data on bilateral incidence of family history of diabetes quite justify any conclusion. In our series of

individuals with a family history of diabetes, we discovered previously undiagnosed diabetes in 20 per cent.

Dr. West did not mention anything about the incidence of previously undiagnosed diabetics which, of course, would have to be eliminated for testing.

DR. WEST: I should like to say I don't think this study in any way detracts from the value of the finding of Dr. Fajans, that a family history of diabetes affects the incidence of "positive" response to glucocorticoids. I think if we tested more persons to cortisone modified tests, that our results would be more nearly similar to those of Dr. Fajans. A somewhat greater percentage of our people tested to the cortisone modified test were over thirty, and that would explain a minor part of the so-called discrepancy between his figures and ours.

In regard to the order of testing, our routine was always to skip a day between the tests, and we felt with these small doses and with skipping a day, there would be little likelihood of any cumulative effect of diet or steroids in either a positive or a negative manner.

We tested this by systematically randomizing forty-two tests, and then we analyzed the control figure, the cortisone figure, the prednisone figure and prednisolone figure and found that the results were the same in each instance. So I believe, if there is a discrepancy, it is not because of the order of testing.

In regard to the appearance of previously undiscovered diabetes in each of the groups, our results were rather similar to those reported by Dr. Fajans. I am sorry I don't have the exact figures. My guess would be that in screening our subjects with a diabetic parent, sibling or child, we found about 15 per cent of these supposedly healthy people to have glucose tolerance impairment, and they were discarded from the study.

In screening the group with two diabetic parents, we found that approximately 20 to 25 per cent had diabetes, and, of course, they were not included in this study since we were testing for potential diabetes.

Diabetes and Insulin

The possible alteration of enzymes in animals with diabetes mellitus has long been considered. Foster in 1867 found no change in the blood amylase in six cases of diabetes. In commenting on his findings, he remarked that "the disease diabetes is due not to any excess but rather to some modified action of ferment." It is interesting that at such an early date Foster was already considering disease in terms of enzyme alterations. In subsequent years, and especially before the discovery of insulin, many other studies reported increases, decreases, or no change in the blood amylase level. Some of these works were cited by Roe, Smith and Treadwell. These studies confirmed Foster's original observation, in that there was no change in blood amylase in depancreatized or alloxan diabetic rats. The amylase activity in pancreas and salivary gland has also been studied in relation to insulin action. The so-called amylase activities of several other tissues are probably not referable to diastatic action.

Changes in concentrations have been found of enzymes whose function is related to carbohydrate metabolism. Such a change is that of glucose-6-phosphatase. Ashmore, Hastings, and Nesbett (Ashmore, J., Hastings, A. B., and Nesbett, F. B.: The effect of diabetes and fasting on liver glucose-6-phosphatase. Proc. Nat. Acad. Sc. 40:673-78, 1954) have come to the conclusion that:

". . . the increased concentration of glucose-6-phosphatase in diabetic liver could be returned toward normal by the in vivo injection of insulin. In vitro insulin was without effect. This suggests that liver glucose-6-phosphatase was elevated in the diabetic liver due to general metabolic changes that occur in diabetes.

"The nature of the changes in glucose-6-phosphatase that occur in diabetes suggests that this enzyme adapts in response to a decreased ability of the peripheral tissues to utilize glucose, thus bringing about an increase in the concentration of blood sugar. In fasting, glucose-6-phosphatase acts to release glucose arising from both glycogenolytic and gluconeogenic processes. In diabetes the increase in this enzyme offers evidence for the overproduction of glucose believed to occur in this disease."

From "Enzymatic and Metabolic Adaptations in Animals" by W. E. Knox, V. H. Auerbach, and E. C. C. Lin, in *Physiol. Rev.* 36:164-254, 1956.