An Artificial Endocrine Pancreas

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SUMMARY

In order to regulate the blood sugar in the intact depancreatized dog as precisely as that accomplished by its normal pancreas, specific equipment has been devised to deliver insulin or glucose continuously and establish normoglycemia both in the fasting and glucose-loaded states. A minicomputer was programmed to respond to the constantly monitored whole blood glucose by injecting appropriate insulin or glucose intravenously to maintain or restore the normal blood sugar.

Standardized glucose challenges consisting of uniform infusions of 10 mg. glucose per kg. min. for sixty minutes were applied to assess the performance of the artificial pancreas. Direct control which relates insulin dosage to the level of the circulating blood sugar results in a response to the challenge resembling mild maturity-onset diabetes both in the abnormally high blood sugar response to glucose loading and in the large amount of insulin required to effect a return to normoglycemia. In contrast, control based on projected (predicted) values of blood sugar not only prevents the abnormal rise but consumes in some cases only 10 per cent of the insulin used for the same glucose load. The performance of the system parallels that of the normal pancreas and lends support to the hypothesis that biphasic insulin responses to glucose challenges are essential for the economy of insulin and the precision of regulation seen in healthy subjects. DIABETES *23*:389-96, May, 1974.

The maintenance of constant normoglycemia in the diabetic patient is seldom achieved clinically by diet and insulin therapy, especially in the postprandial state. In spite of meticulous regulation of the factors concerned, if the high risk of hypoglycemia is to be avoided, then at certain intervals in the day hyperglycemia will have to be tolerated. One might postulate that the effect of this episodic insulin deficiency accumulating over a number of years might indeed be responsible for the ultimate degenerative sequelae of diabetes mellitus.[1] The normal pancreas secretes insulin as it is required within the normal range of blood sugars in order to prevent any changes beyond the physiological limits. A computerized control system has been devised which closely simulates this particular endocrine function of the pancreas and its performance will be described in this report.

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EXPERIMENTAL TECHNICS

A. *Apparatus*

A schematic diagram of the apparatus is shown in figure 1. A stream of venous blood is drawn continuously into the inner lumen of a specially constructed dual-lumen catheter and is fed into a standard glucose analyzer (Technicon Instruments Corp., Ardsley, N. Y.) modified[2,3] to measure glucose in samples of whole blood withdrawn at a rate of approximately 0.05 ml./min. A small inner lumen adaptor (Facets, Toronto, Canada) is situated inside the sheath of a cannula (Aloe Medical, St. Louis, Mo.) which provides a channel for the transportation of a solution 200 U./ml. of Heparin Sodium Injection (Connaught Laboratories, Toronto) in normal saline to the tip of the cannula where it mixes with the aspirated samples of whole blood preventing coagulation in the catheter and tubings which conduct the blood to the glucose analyzer. In the analyzer the stream of blood is further diluted with normal saline, segmented with air and dialyzed against alkaline potassium ferricyanide. Blood glucose and any other reducing sugars alter the color of the reagent whose optical density is then measured at 420 nm in a colorimeter and the value fed

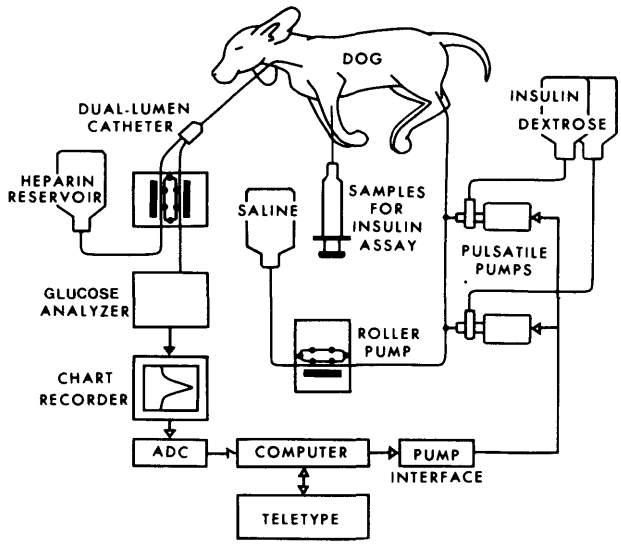

FIG. 1. Schematic diagram of apparatus used for monitoring and automatic regulation of blood sugar.

by a retransmitting slidewire to the analog to digital converter of a Nova minicomputer (Data General Corporation, Southboro, Mass.). The digital number corresponding to optical density is compared against a semilogarithmic calibration curve defined by previously analyzed standard solutions. Voltage signals from the chart recorder are sampled by the computer many times a second, glucose concentration computed and the values averaged for one minute. Elapsed time is printed by the teletype once every minute along with five variables: measured blood glucose concentration in mg. per cent, dextrose infusion rate in mg./min., insulin infusion rate in mU./min., total glucose infused in mg. and total insulin infused in mU. Two pumps controlled by the computer via the pump interface[4] deliver appropriately metered amounts of dextrose and insulin, which are rapidly carried into a peripheral vein by a steady infusion of normal saline (0.5 ml./min.).

The apparatus has an eight-minute measurement delay. This means that eight minutes elapse from the time a change in blood sugar enters the dual-lumen catheter to the time a change in optical density is registered on the chart recorder and the computer reacts by altering the dextrose or insulin infusion rates.

B. *Computer Algorithms*

The automatic control of blood sugar in diabetic subjects[5] is regulated by a computer[6] which in addition to computing the actual blood sugar concentration from the electrical analog of optical density, also calculates the rate of change of the blood sugar concentration. The computer uses these two important variables to determine the required rates of dextrose and insulin infusion. Such functions as signal filtering, operator controls and other engineering details will not be discussed in this article.

Blood sugar is regulated by the computer in accordance with two control algorithms which relate the respective rates of dextrose infusion R_d and insulin infusion R_i to the measured blood sugar level G and to the combination of measured blood sugar level and its rate of change. The graph in figure 2 illustrates several members of a family of curves described by the following mathematical relationships where subscripts d and i represent dextrose and

$$R_d = \tfrac{1}{2}M_d[1 - \tanh S_d\ (G - B_d)] \qquad (1)$$

$$R_i = \tfrac{1}{2}M_i[1 + \tanh S_i\ (G_p - B_i)] \qquad (2)$$

insulin, M, the maximum infusion rate, S, the slope, and B, the blood sugar level at which half maximum infusion rate is chosen to occur. Parameters M, S, and B are chosen by the operator and can be modified during the course of the experiment. The hyperbolic tangent function used here is similar to saturation control[18] at high blood sugar levels but differs significantly at normoglycemic levels.

Using the minute-to-minute changes of blood sugar level averaged over the preceding four minutes as an indication of the rate of change A of blood sugar, a projected blood sugar G_p can be computed by the addition of a difference factor DF to the current blood sugar level as follows:

$$G_p = G + DF \qquad (3)$$

$$\text{where} \quad DF = K_1[\text{EXP}(A/K_2) - 1] \qquad (4)$$

with K_1 chosen to adjust the magnitude of the difference factor and K_2 selected to establish its sensitivity to changes in A. One member of the family of curves represented by equation (4) is plotted in figure 3. Use of G_p for G in equation (2) results in the current insulin infusion rate being set on a projected blood sugar value.

C. *Subjects*

Subjects, nonobese beagles one to two years old, were fasted overnight. One-half hour after intramuscular injection of 1 ml. of Atravet (Ayerst

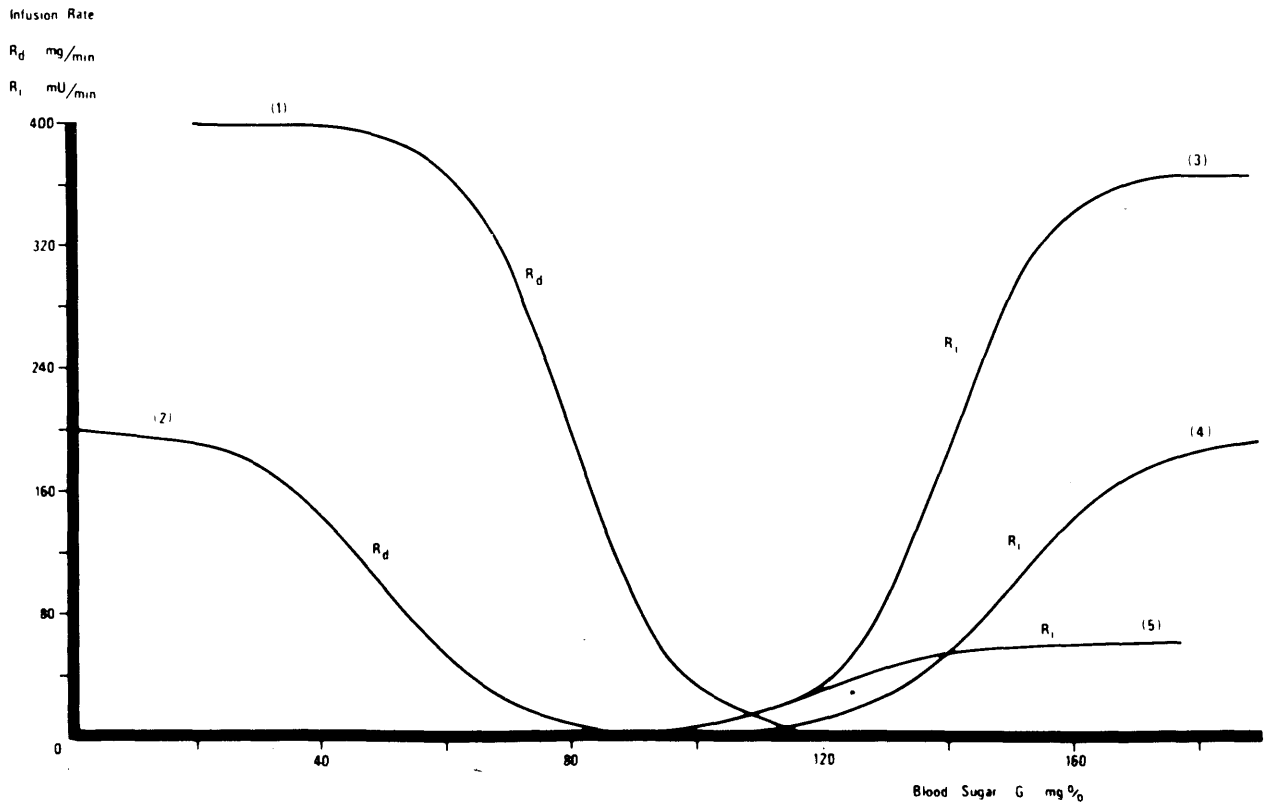

FIG. 2 Control algorithms relating insulin and dextrose infusion rates to projected blood glucose concentration. Curve parameters M, S, B are as follows: (1) 400, 0.06, 80; (2) 200, 0.05, 50; (3) 367, 0.06, 140; (4) 191, 0.05, 150; (5) 57, 0.05, 120.

Laboratories, division of Ayerst, McKenna & Harrison Ltd., Montreal) a light anesthetic, 0.1 cc. Nembutal Sodium Injection (Abbot Laboratories, North Chicago, Ill.) per kilogram body weight, was administered intravenously. The external jugular, a cephalic vein in the front paw and a saphenous vein in the hind leg were catheterized, the former one with a dual-lumen catheter for blood withdrawal and the latter two with single-lumen catheters, one for withdrawal of samples for insulin assay and the other for administration of insulin, dextrose and saline. At each experiment catheters were placed in these same blood vessels. Anesthesia was maintained by slow injection of Nembutal via the saphenous catheter as indicated. External warmth was also provided as needed.

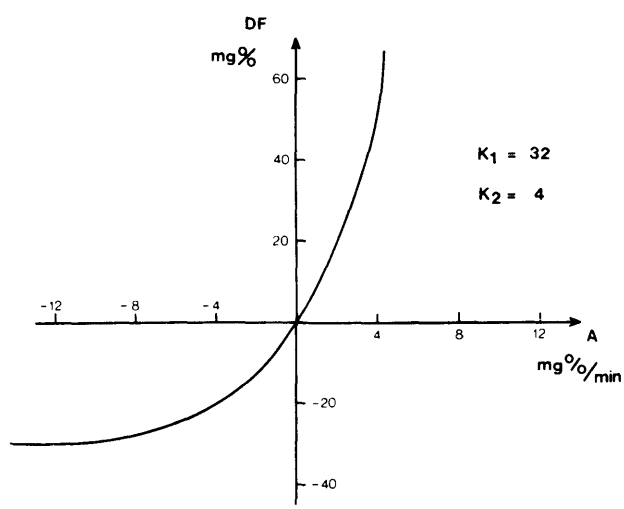

FIG. 3. Control algorithm relating the difference factor DF to the computed rate of change of blood sugar.

Pancreatectomies were performed under sterile conditions.[7] The abdomen was opened through midline incision from xiphoid process to umbilicus and the duodenum and pancreas were delivered into the wound. After inspection of the pancreas its removal was started at the uncinate process by dividing the vascular pedicle and cutting the mesentery on both sides of the pancreas. Where the pancreas was intimately connected to the second part of the duodenum, it was separated by blunt finger dissection. Blood vessels were not clamped or tied; bleeding was stopped by applying pressure with moist gauze. To remove the

splenic portion the mesentery was again cut and one or two of the large blood vessels ligated. The incision was closed in layers. Postoperatively the dogs are sustained with daily injections of eight units of Protamine Zinc Insulin (Connaught Laboratories Ltd., Toronto) and five capsules of Cotazyme (Organon, Montreal, Quebec) with each meal.

EXPERIMENTAL PLAN

Experiments were performed to compare the ability of a subject to accommodate a standardized glucose challenge both as a healthy control and then as a diabetic assisted by the computer 'pancreas.' In the first experiment, a healthy subject was prepared as described above under "Subjects" and challenged with a glucose loading test (GLT) consisting of a uniform infusion for sixty minutes of 10 mg./kg. min. dextrose (Baxter Laboratories of Canada, Ltd., Malton, Ontario). Blood samples were collected for subsequent insulin assay and the blood sugar response to the challenge was recorded by the monitoring system with the infusion pumps inactivated. At the end of this GLT, the blood sugar was allowed to return to its steady state value at which time pancreatectomy was performed. Insulin infusion under computer control was begun after the blood sugar had risen to about 150 mg. per 100 ml.

Under automatic, machine control blood sugar was then brought back to normoglycemic levels and the identical GLT was repeated, this time with the computer system acting to regulate blood sugar. Actually, two post-pancreatectomy GLT's were done, one based on direct blood sugar measurements and the second on projected blood sugars as defined above under "Computer Algorithms." The GLT procedure was repeated some weeks after surgery in order to evaluate the ability of the machine to control a chronic diabetic who had been supported by daily exogenous insulin.

RESULTS

Selected blood sugar patterns representative of all experimental results are plotted in figures 4 and 5 and the salient data from all experiments are listed in tables 1 and 2. The graphs in figure 4a show the time course of blood sugar and plasma insulin in a healthy subject challenged by a GLT of 10 mg./kg. min. for sixty minutes. Both variables were recorded during the initial fasting phase before the challenge, during the GLT itself, and finally during the re-establishment of the fasting level of the blood sugar.

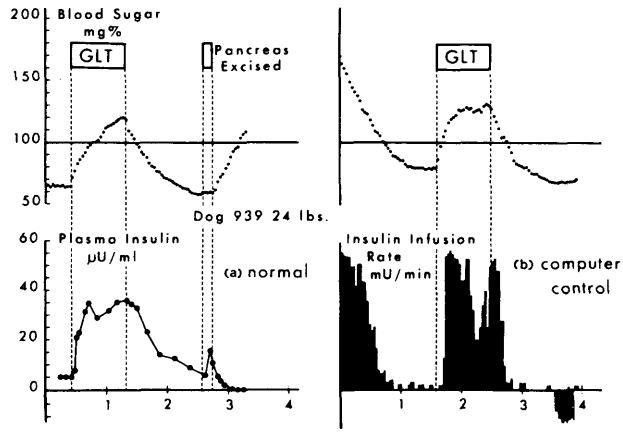

FIG. 4 (a) Blood sugar and plasma insulin responses to a glucose loading test in the normal subject.
(b) Blood sugar response to the same glucose loading test in a diabetic subject controlled by exogenous insulin and dextrose infusions. Control is based on projected blood sugars.

At this time the pancreas was excised and the blood sugar began to rise as the circulating insulin fell to zero. Manipulation of the pancreas caused a momentary rise in insulin level followed by rapid disappearance concurrent with the onset of surgical diabetes.[8]

Figure 4b shows the typical results of insulin infusion under computer control. Using the following parameters (M_i = 57, S_i = 0.05, B_i = 120, M_d = 200, S_d = 0.05, B_d = 50, K_1 = 32, K_2 = 4), the blood sugar was brought back to normal levels of 60 to 70 mg. per 100 ml. The standardized GLT was then applied and the blood sugar response was recorded along with the rates of insulin and dextrose infusion. After completion of the test the blood sugar returned to normoglycemic levels while under machine control.

The graphs in figure 5 compare blood sugar regulation when computer control is based on direct blood sugar measurement only (figure 5a) and on projected blood sugar values (figure 5b). This subject was maintained by daily insulin injections for two and one-half months. Then, under machine control its initial blood sugar (which was in excess of 250 mg. per 100 ml.) was brought back to normoglycemic levels. The parameters in the computer algorithm were (M_i = 367, S_i = 0.06, B_i = 140, M_d = 800, S_d = 0.06, B_d = 80, K_1 = 0) and insulin was infused at a rate corresponding to directly measured blood sugar. The rise in blood sugar which accompanies a 120-minute GLT and the subsequent return to normoglycemic levels is recorded in figure 5a along with the required

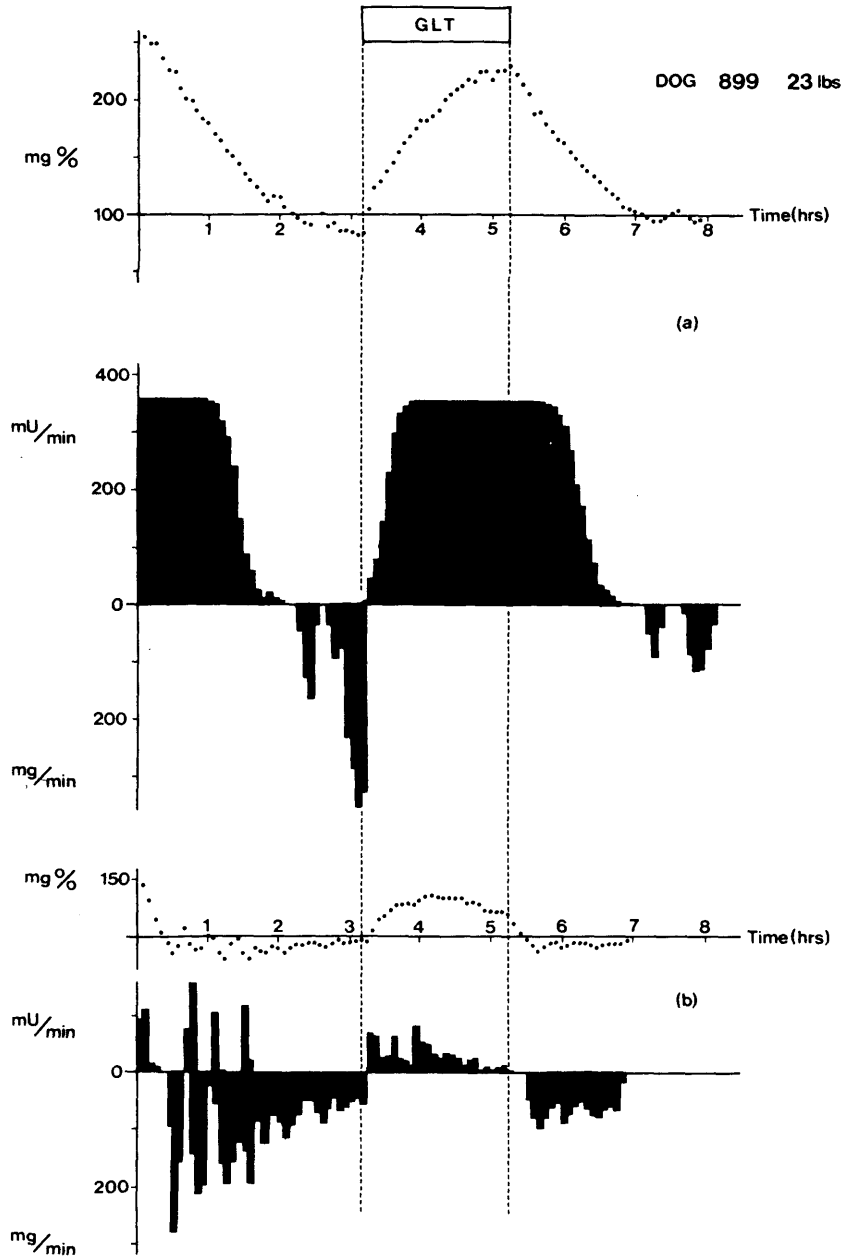

FIGURE 5

Blood sugar responses to a glucose loading test in a diabetic subject controlled by exogenous insulin and dextrose infusions, (a) when control is based on directly measured blood glucose levels and (b) when control is based on projected glucose levels. Due to the compressed scale, small but important time delays cannot be readily discerned.

TABLE 1

Data from three experiments
Direct control

Dog no.	Weight (kg.)	Blood sugar rise-healthy (mg. per 100 ml.)	Blood sugar rise-diabetic (mg. per 100 ml.)	Total insulin (U./kg.) per hour GLT
900	9	115	147	1.37
898	10	80	148	1.35
899	10	57	130	1.58
Average	9.7	84	142	1.43

insulin and dextrose infusions. Then the parameters of the computer algorithm were altered ($K_1 = 32$ and $K_2 = 4$) so that the machine based its current insulin infusion rates on projected blood sugar values. The rise in blood sugar which accompanies a GLT and the subsequent return to normoglycemic levels is recorded in figure 5b along with the required insulin and dextrose infusions.

In table 1 are the salient data from three experiments in which the method of control was based on

TABLE 2

Data from four experiments
Projected control

Dog no.	Weight (kg.)	Blood sugar rise-healthy (mg. per 100 ml.)	Blood sugar rise-diabetic (mg. per 100 ml.)	Total insulin (U./kg.) per hour GLT
899	10	57	42	0.16
897	11	53	67	0.22
939	11	55	52	0.15
940	11	88	68	0.16
Average	10.8	63	57	.17

directly measured blood sugar values. Similarly, in table 2 are the salient data from four experiments in which the method of control was based on projected blood sugar values. In both tables the results are tabulated according to (1) dog number, (2) body weight, (3) blood sugar rise in mg. per 100 ml. measured from the fasting level to the peak value during a GLT in the healthy dog, (4) in the diabetic dog and (5) total insulin in U./kg., the amount of insulin infused in response to a sixty-minute GLT, this value being normalized to one hour for purposes of comparison.

DISCUSSION

To ensure that the results reported here are statistically comparable the subjects represent random samples from the same population (pure bred beagles), who were within 10 per cent of the same body weight (10 kg.) and within six months of the same age (one and one-half years). Glucose loading tests were performed only after normoglycemic levels had been established and maintained for at least thirty minutes. This latter measure further ensured that all subjects were in similar metabolic states, in so far as blood sugar concentration is a measure of metabolic state. No real differences were found between post-pancreatectomy GLT's when the order of control modes was interchanged. Nor were there any significant differences noted in glucose control when the GLT procedures were repeated and compared in chronic diabetics who had been supported by daily exogenous insulin for periods of up to six months. Seven sets of experiments were done on six dogs.

In all six healthy subjects, the insulin response to a GLT was characteristically biphasic[9-11] and demonstrated a fast initial rise[12,13] and fall with rising and falling blood sugar, as demonstrated by the example in figure 4a. From the results of these experiments it is likely that this anticipatory action of the pancreas is instrumental in achieving the fine regulation seen in healthy subjects and perhaps in preventing an excessive reactive hypoglycemic response to glucose infusion. These attributes have been conserved in our system of automatic blood sugar regulation where, from figure 4b, it is clear that the initial fall in the rate of insulin infusion is rapid and similarly the rise and fall in insulin infusion rate with the following GLT are also rapid with the rising and falling blood sugar. This anticipatory action was accomplished by using projected blood sugar values. Projected blood sugar levels are computed according to equation 3 by the addition of a difference factor to the directly measured glucose value. This difference factor is heavily weighted by an exponential relationship to emphasize positive trends compared to negative trends as may be seen in figure 3.

In all newly diabetic subjects placed under machine control, normoglycemic levels were regained in approximately one hour's time without reactive hypoglycemia. With insulin infusion rates computed on the basis of projected blood sugar levels the response to a GLT was always similar to the response of the healthy animal, comparing figure 4b with figure 4a, and the recovery time to normoglycemic levels after the GLT was essentially the same in both cases. The action of the 'predictive' mechanism which enhances insulin infusion when blood sugar begins to rise, even at normoglycemic levels, may be seen in figure 4b, where at one and three-quarter hours there is a rapid initial infusion of insulin. When blood sugar reaches a plateau during the GLT at about two and a quarter hours, the difference factor of equation 3 is zero since the slope A of the glucose curve is zero in equation 4 and the rate of insulin infusion falls to the rate prescribed by equation 2 alone.

The sensitivity of the computer system to rising blood sugars is apparent in the second peak of the apparently bimodal graph of insulin infusion (figure 4b). This second peak is due to the small but significant rise in blood sugar just prior to the end of the GLT. As soon as the blood sugar falls, the rate of insulin infusion declines, essentially to fasting level requirements, as blood sugar approaches normoglycemia.

Regulating the diabetic by means of projected blood sugar values is the method of choice and has three advantages. First, by measuring the rate of blood sugar rise and infusing the appropriate amount of insulin while the values are in the normal range, a restraint against an unphysiological change is exer-

cised. In so doing some compensation is achieved for the inherent eight-minute delay in the monitoring system. With appropriately chosen parameters, this system will control blood sugar and restore homeostasis without reactive hypoglycemia. Second, when the predictive feature is in operation the excursion in blood sugar is always smaller and recovery is faster than when it is not present. Perhaps the most significant advantage is the third: less insulin is required. On the average only 0.17 units/kg.hr. of insulin were needed to control subjects challenged with a 10 mg./kg.min. glucose loading test when control was based on projected blood sugar levels. In contrast, 8.4 times as much insulin (1.43 U./kg.hr.) were less effective in controlling similar average subjects challenged with the same glucose load when control was based on direct blood sugar measurements alone. Referring to table 1 it may be seen that during a GLT with direct control in the average diabetic subject, the peak blood sugar rise is greater than that found in the same but healthy average subject while, under control based on projected blood sugars (table 2) the blood sugar rise is somewhat less than that found in the healthy average subject. In all the subjects controlled on the basis of projected blood sugar levels compared to those controlled on the basis of direct blood sugar levels, the blood sugar rise is smaller and the total insulin infused is much less. Better control is achieved with less insulin.

Following a glucose loading test the amount of glucose infused by the computer system when blood sugar is in the normoglycemic range depends a great deal on the parameters of R_d in equation 1. In figure 5b, these parameters were such that at a blood sugar of 80 mg. per 100 ml., the computer system would infuse dextrose at a rate of 200 mg./min.

Through the action of the system described in this paper, the glucose concentration of a surgically diabetic subject is confined to a 'well' (see figure 2) whose depth can be increased or decreased and whose width can be expanded or contracted simply by altering the parameters of two equations. Augmented rates of insulin infusion counter hyperglycemia and controlled dextrose infusions oppose hypoglycemia. In the normoglycemic range when blood sugar is constant, a balance is established between the rate of release of de novo glucose into the blood stream and its rate of uptake from the blood stream. The insulin required to sustain this dynamic equilibrium (homeostasis) is provided by the external artificial beta cell. Use of the hyperbolic tangent function as a control algorithm facilitates control because system gain is varied according to blood sugar level without a discontinuity in the normoglycemic range such as occurs in saturation control.[18] Decreasing the gain in the normoglycemic range greatly enhances the stability of the system when it is considered as a closed loop control system.

The engineering components of the closed loop system which regulated blood sugar must be reliable, stable and relatively free from interference. Clearly artifacts will be interpreted as rapid changes in blood sugar and unless these are detected and minimized, they can erroneously modify the rates of dextrose and insulin infusion. Also, the blood withdrawal subsystem must function reliably and without interruption due to clot formation or lumenal obstruction for long periods of time.

CONCLUSIONS

The computer system described in this paper is an artificial endocrine pancreas which simulates the blood sugar regulatory function of the normal endocrine pancreas. The reproducibility of the responses to uniform glucose loading tests observed in all the experimental work implies that the characteristics of computer controlled blood sugar are predominantly due to the selected parameters of the control algorithms. No Staub-Traugott effect[19] is noticed in repeated challenges. Choosing parameters for the control algorithms is an empirical process based partly on clinical experience, partly on background knowledge related to basal insulin secretion rates and total daily insulin requirements and partly on engineering constraints to ensure the proper operating point with the required dynamic operating range. The criterion for optimal control was to equal or exceed in the diabetic the measured characteristics of the healthy subject's response to a standardized glucose challenge. This was easily achieved within the system constraints of loop gain and total system delays.

Two forms of control were explored: direct control based on direct blood sugar measurement and control based on projected blood sugars. Control based on the latter is more effective and more economical of insulin than the former. Employing direct control in diabetic subjects produces a blood sugar response to a glucose loading test of 10 mg./kg. min. resembling that of mild maturity-onset diabetes. The addition of a difference factor to enable control based on projected blood sugar levels produces a response which is indistinguishable from that obtained in normal subjects. The latter mode of control helps compensate for the monitoring system delay of eight minutes.

We expect that smaller system delays, say of two to four minutes, will facilitate blood sugar control. We also anticipate that the use of projected blood sugar values in a system with minimal delay will result in a diminished insulin requirement, the elimination of reactive hypoglycemia and the absence of a need for controlled glucose infusion to maintain normoglycemia following a glucose loading test (see figure 5b).

A great deal of work has yet to be done in optimizing the artificial pancreas. The simulation of a biphasic insulin response to glucose challenges appears essential for the economy of insulin and the precision of regulation seen in healthy subjects. It has been postulated[14,15] that the initial release of stored granular insulin by the pancreas is a critical component of normal blood sugar control and that prediabetes and diabetes are characterized by a lack of this rapid phase, in spite of a competent second phase of insulin release. Our experimental results support this hypothesis.

Perhaps the physiological and anatomical relationships between liver, pancreas and the gastrointestinal tract are more than fortuitous. The pancreas momentarily secretes large pulses of insulin in response to changes in blood sugar level,[15] amino acid levels[16] and perhaps as a consequence of neural stimulation.[17] This anticipatory insulin release by the pancreas is conveyed directly to the liver, where the hormone is instrumental in quickly altering (switching) the net hepatic glucose balance: glycogenolysis and gluconeogenesis cease and glycogenesis begins. Since this initial phase of the biphasic insulin response acts in the normoglycemic range, we postulate that its absence in diabetes may account for the pharmacological amounts of insulin required in the treatment of the disease and that the ultimate microvascular complications may result from the cumulative effects of chronic episodic insulin deficiency and consequent inappropriate metabolic regulation.

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REFERENCES

[1]Lazarow, A.: Selective islet differentiation in organ culture and islet transplantation. Diabetes *22*(Suppl. 1): June, 1973.

[2]Weller, C., Linder, M., Macaulay, A., Ferrari, A., and Kessler, G.: Continuous in vivo determination of blood glucose in human subjects. Ann. N. Y. Acad. Sci. *87*:658-68, July, 1960.

[3]Brown, G. M., et al.: An improved technique for continuous in vivo analysis of glucose. Clin. Chim. Acta *14*:386-90, 1966.

[4]Ewart, T. G., and Albisser, A. M.: A versatile computer interface for direct digital control of infusion pumps. Toronto, International Electrical and Electronics Conf., October 1973, pp. 6-7.

[5]Kadish, A. H. et al.: Automation control of blood glucose homeostasis. 7th Int. Cong. Clin. Chem. Geneva/Avian, Vol. 1. Kanser, Basel, International Methods in Clinical Chemistry, 1970, p. 231.

[6]Ewart, T. G., et al.: A computer analog of the endocrine pancreas. Rochester, N.Y., Proc. American Physiological Society: Intern. Symp. of Dynamics and Controls in Physiological Systems, Aug. 1973, pp. 509-11.

[7]Markowitz, J., et al.: Experimental surgery. Baltimore, Williams and Wilkins, 1964, p. 242.

[8]Davidovac, Z., et al.: Continuous monitoring of blood glucose levels during pancreatectomy. Lac Beauport, Quebec, Fifth Winter Meeting, Canadian Physiological Society, Jan., 1973.

[9]Bergman, R. N., and Urquhart, J.: Prog. Horm. Res. *27*:583, 1971.

[10]Curry, D. L.: Insulin secretory dynamics in response to slow-rise and square-wave stimuli. Am. J. Physiol., *221*, No. 1, 324, 1971.

[11]Basabe, J. C., et al.: Insulin secretion studied in the perfused rat pancreas. Diabetes *20*:457, 1971.

[12]Grodsky, G. M.: A threshold distribution hypothesis for packet storage of insulin and its mathematical modeling. J. Clin. Invest. *51*:2047, 1972.

[13]Ibid: A threshold distribution hypothesis for packet storage of insulin, II. Diabetes *22*: Suppl. 2, 584, 1972.

[14]Cerasi, E., et al.: Decreased sensitivity of the pancreatic beta cells to glucose in prediabetic and diabetic subjects. Diabetes *21*:224, 1972.

[15]Grodsky, G. M., et al.: Effect of pulse administration of glucose or glucagon on insulin secretion in vitro. Metab. *16*:3, 222-33, March, 1967.

[16]Bergman, R. N., and Bucolo, R. J.: Nonlinear dynamic properties of the pancreas and liver. *In* Regulation and Control in Physiological Systems, Proceedings. A. S. Iberall, and A. C. Guyton, editors. Instrument Society of America, Pittsburgh, Penn., Aug. 1973.

[17]Frohman, L. A., et al.: Factors modifying insulin and glucose responses to hypothalamic stimulation. Diabetes *22*(Suppl. 1):298, 1973.

[18]Cahill, G. F., Jr., et al.: Practical developments in diabetes research. Diabetes *21*(Suppl. 2):703-12, 1972.

[19]Abraira, C., et al.: Absence of the Staub-Traugott effect (facilitated glucose disposal) in hypopituitarism. Diabetes *22*(Suppl. 1):300, 1973.