Effects of Environment on Diabetes

A study of partially-depancreatized rats exposed to cold

Dwight J. Ingle, Ph.D.
Robert C. Meeks, M.S.
Leo M. Humphrey, B.S.

KALAMAZOO, MICHIGAN

This is one of a series of studies of the effects of various stressors upon experimentally induced diabetes in the rat. It is shown that exposure of the rat to cold causes a rapid loss of weight, a rise in the level of urinary nonprotein nitrogen and suppression of the glycosuria. When the anmials were returned to room temperature these changes were reversed.

METHODS

Infection-free male rats of the Sprague-Dawley strain were partially depancreatized at a weight of approximately 275 gm. by the procedure described by Ingle and Griffith.[1] After the animals had recovered from the operation and had reached an average weight of approximately 330 gm., they were placed in metabolism cages and adapted to the force-feeding of a medium carbohydrate diet (Table I) by stomach tube each morning and late afternoon. The technic and diet were modified from those described by Reinecke, Ball, and Samuels.[2] During the period of adaptation to force-feeding, the amount of diet was increased gradually to prevent the development of food shock. The animals were brought to a full feeding of 26 cc. of diet per day on the sixth day. Twenty-four hour samples of urine were collected at the same hour each day (8:00 to 8:30 a.m.) and were preserved with toluene and citric acid (1 gm. per sample) to insure the acidity of the urines for nitrogen analysis. Urinary glucose was determined by the method of Shaffer and Williams[3] and the determination of urinary nonprotein nitrogen (NPN) was by the micro-Kjeldahl procedure.

The animals were maintained in a temperature cabinet which has a range of adjustment from 0° to 40° C.±1°. The cabinet has space for 12 rats to be maintained in metabolism cages, each in an individual compartment.

EXPERIMENTS AND RESULTS

Eight severely diabetic rats were maintained at 26° C. ±1° for a period of several weeks, the temperature was then lowered to 3°±1° for 2 weeks and was then raised to 26° for a second control period of 4 weeks.

During exposure to cold the rats lost an average of 55 gm., there was an accompanying rise in the excretion of urinary NPN and there was suppression of glycosuria. When the rats were returned to room temperature these changes were reversed. The changes from and the return to control values were gradual rather than immediate. The data are summarized in Figure 1.

DISCUSSION

Like many other investgiators, we have been concerned with the metabolic consequences of adrenal cortical activation during exposure to various stressors. It is known that exposure to cold causes activation of the adrenal cortices via the increased discharge of corticotropin from the anterior pituitary.[4] On this basis it might be expected that any nonspecific stressor should cause exacerbation of a diabetic state. Under the conditions of this experiment, exposure to cold brought about a suppression of the glycosuria. One of us[5]

Address communications to Doctor Ingle, Research Laboratories, The Upjohn Company, Kalamazoo, Michigan.

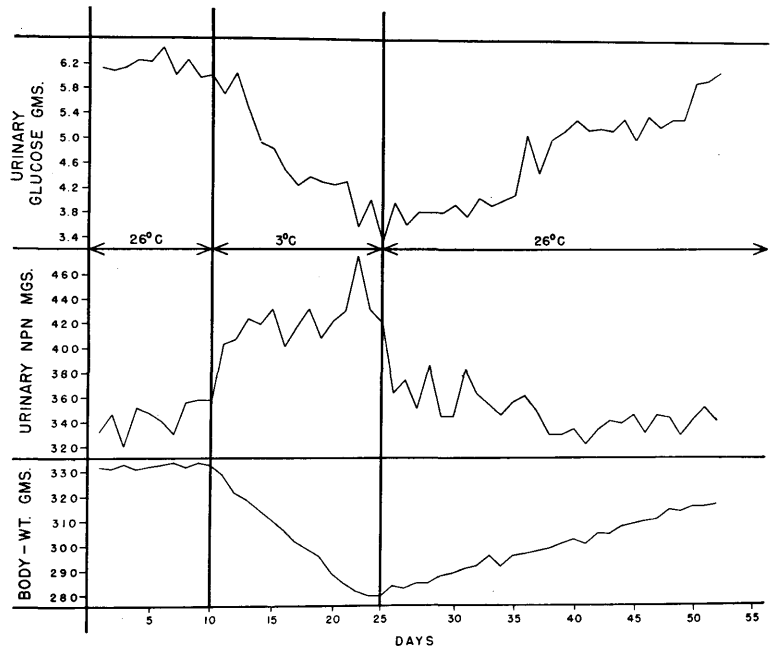

FIGURE 1 Effect of cold upon the partially depancreatized, force-fed rat. Averages for 8 male animals.

(D.J.I.) has reviewed the evidence from this laboratory that nonspecific stressors fail to cause exacerbation of diabetes in the rat. Such observations do not exclude the possibility that activation of the adrenal cortices during stress affects the metabolism of carbohydrate. The metabolic consequences of an increased secretion of adrenal cortical hormones may be masked by other metabolic adjustments during stress such as the increased oxidation of carbohydrate.

When homothermal animals are exposed to low temperature they must either produce more heat or die. Although the ability of the diabetic organism to oxidize carbohydrate for energy purposes is a subject of continuing debate, there is no doubt that these processes are not completely blocked in the partially depancreatized rat even when it is severely diabetic. It seems possible that the oxidation of carbohydrate can be accelerated during a vital need for increased energy in both the normal and diabetic organism. During exposure to cold the diabetic animal may utilize some of the glucose which is wasted into the urine under nonstressful conditions. One of us[6] (D.J.I.) has reviewed the indirect evidence that muscle work can accelerate the utilization of glucose in the diabetic rat. These changes coexist with an increased breakdown of protein and loss of weight which indicates that exposure to cold may also accelerate the mobilization of endogenous tissues for energy purposes. It is possible that gluconeogenesis is accelerated in these animals so that the extent of increased utilization of carbohydrate cannot be determined by measuring changes in the level of urinary glucose alone.

There are other considerations which may require a different interpretation of the results. It was anticipated that the full-blown effect of cold upon glycosuria and upon urinary NPN would be manifest immediately. On the contrary, these changes were gradual. Similarly, it was anticipated that when the temperature was increased to control values (26° C.), the changes in urinary glucose and nitrogen would be immediate, but they also were gradual. The assumption that the metabolic changes are determined by the immediate changes in the energy requirement of the animal may be difficult to reconcile with these data. The decrease in glycosuria became marked only after the animals had lost considerable weight and the glycosuria was restored to control values only after most of the weight had been regained. During recovery from the catabolic phase of the re-

TABLE 1 MEDIUM CARBOHYDRATE DIET

Constituent	Grams
Cellu flour (Chicago Dietetic Supply House)	60
Osborne & Mendel salt mixture	40
Dried yeast (Pabst)	100
Wheat germ oil	10
Cod liver oil	10
Vitamin K (2-methyl-1,4-napthoquinone)	100 mg.
Mazola oil	200
Casein (Labco)	160
Starch	200
Dextrin	190
Sucrose	200
Water to make total of	2000 cc.

sponse to cold, the animals were able to conserve some of the calories which were wasted as urinary glucose during the control periods. The rates at which the responses to cold wax and wane may be due to inertia in the basic mechanisms (unknown) involved. It can be suggested also that the suppression of glycosuria in rats which have become emaciated during exposure to cold may bear some relationship to the well known but poorly understood effects of emaciation caused by the starvation treatment of diabetes mellitus prior to the introduction of insulin.

SUMMARY

Eight partially depancreatized male rats were maintained on a uniform intake of medium carbohydrate diet by the technic of force-feeding. Following maintenance at 26° C. for several weeks the rats were exposed to a temperature of 3° C. for 14 days. During exposure to cold the rats lost weight and excreted more urinary nonprotein nitrogen. The level of glycosuria decreased during this period. When the temperature was raised to 26° C. for 28 days, the metabolic changes were reversed. The changes during the exposure to cold and during the recovery period were gradual rather than abrupt.

REFERENCES

[1] Ingle, D. J. and Griffith, J. Q.: Surgery of the rat. Chapter 16, The Rat in Laboratory Investigation, Philadelphia, J. B. Lippincott Co., 1942, p. 379.

[2] Reinecke, R. M., Ball, H. A., and Samuels, L. T.: High fat and high carbohydrate diets that can be fed to rats by stomach tube. Proc. Soc. Exper. Biol. and Med. *41*:44, 1939.

[3] Shaffer, P. A. and Williams, R. D.: Sugar determination by the ferricyanide electrode. J. Biol. Chem. *111*:707, 1935.

[4] Sayers, G., and Sayers, M. A.: The pituitary-adrenal system. Recent Progress in Hormone Research, Vol. II, New York, N. Y., Academic Press, 1948, p. 81.

[5] Ingle, D. J.: Some further studies on the relationship of adrenal cortical hormones to experimental diabetes. Diabetes *1*:345, 1952.

[6] Ingle, D. J.: Some studies on factors which influence tolerance for carbohydrate. Proc. Am. Diabetes Assoc. *8*:3, 1948.

Quackery in Weight Reduction

While quackery in conjunction with the employment of physical agents for the treatment of obesity is rampant, quackery is by no means limited to physical agents. Reducing pills, vitamin supplements, slenderizing creams, laxatives, candies to be taken just before eating, one-food diets, "Hollywood diets," seven-day and fourteen-day diets of bizarre ingredients have all been widely exploited.

Oliver Feld, in discussing "fooling the fat," has described creams, lotions, and bath powers, which were advertised as miracle agents for reducing weight. He has mentioned also various systems and salons where "rhythmic passive exercise" is claimed to give you the benefits of "active exercise without any of the tiring ill effects." One elaborate string of slenderizing salons" employs such equipment as the "Roaler Massager," the "Back Ring Roller," the "Leg Roller" and the "Modified Slendro Massager," as well as the "Rollo Massage Chair."

It can be commented that careful clinical observation by skilled physicians does not support the claim that any of these mechanical procedures will remove deposits of fat. There is no "easy way" to reduce fat and there is no scientific evidence whatever to indicate that adipose tissue can be made to disappear by massage or by any other means from one region of the body without its disappearing in comparable amounts from other regions.

Innumerable glib claims are made for procedures and devices which are supposed to be effective in "spot reducing" of adipose tissue . . . unscrupulous manufacturers of drugs, proprietors of beauty parlors and physical culture clubs, makers of "slimming creams," and manufacturers of chin straps, reducing belts, massaging devices, rollers, and electrical vibrators have produced almost unlimited claims concerning the value of their devices for "spot reducing."

While the value of nearly all of these devices is absolutely nil, some careful medical observers believe that massage may be of some value after much fat has been removed from the entire body, including the affected region, by appropriate dieting. At this time, massage may aid in restoring elasticity to the sagging skin. However, gentle manual massage is more satisfactory than any of the mehanical devices, rollers or vibrators. The only effective way of reducing fat in local regions of the body except when extremely large amounts of fat are occasionally removed by the heroic measure of surgical excision is to follow a program of general reduction of weight by remaining on a low-calorie diet.

—From *Physical Medicine and Obesity* by Frank H. Krusen, M.D. in *The Journal* of the American Medical Association January 24, 1953