# EFFECT OF EXCESSIVE GLUCOSE ADMINISTRATION ON LIPID LEVEL, RATE OF GLYCOLYSIS, AND OXYGEN CONSUMPTION BY TISSUES OF THE LIVER, HEART, BRAIN, AND AORTA

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Prolonged administration of an excess of glucose to rabbits was accompanied by accumulation of lipids in the liver and heart at the expense of cholesterol and triglycerides and by a simultaneous decrease in the rate of glycolysis and in the oxygen consumption. Succinate dehydrogenase activity also was low in the liver tissues 3 months after glucose administration. In the aorta a reduced rate of glycolysis and succinate dehydrogenase activity was accompanied by the formation of lipoid plaques. The oxygen consumption and total lipid content of the brain were the same as in the control animals, whereas the rate of glycolysis was reduced after the 1st month of the experiment.

Observations have shown an increase in the incidence of atherosclerosis in persons consuming large quantities of carbohydrates [6, 8, 10, 13], although in most investigations the level of the blood lipids has been taken as the sole criterion of atherogenicity.

The object of this investigation was to study the assimilation of carbohydrates and distribution of lipids in the organs of rabbits during the prolonged and excessive intake of glucose.

## EXPERIMENTAL METHOD

Experiments were carried out on 40 rabbits weighing from 2.5 to 3.5 kg. In addition to their ordinary diet the experimental rabbits received 3 g/kg body weight glucose daily by gastric tube for 3 months. All manipulations concerned with the taking of material and preparation of homogenates were carried out at 0-2°C and the total time until precipitation of the proteins in the last sample did not exceed 20 min. The tissue oxygen consumption was studied in a Warburg apparatus [11] at 37°C for 1 h in Krebs-Ringer buffer, pH 7.3. The level of preformed lactic acid and its increase were investigated under the same conditions but with the addition of 200 mg% glucose (the method of Barker and Summerson [5]). The total content of ether- and alcohol-soluable lipids in the tissues was determined gravimetrically, their phosphatide content was estimated as phosphorus by the method of Fiske and Subbarow in Braunshtein's modification [2], the cholesterol concentration was determined by the color reaction of Liebermann and Burchard [1], and succinate dehydrogenase (SDH) activity was estimated by Nachlas' method. Only the rate of glycolysis and the SDH activity were determined in the tissues of the aorta, and they were examined histologically after staining for lipids with Sudan III.

## EXPERIMENTAL RESULTS AND DISCUSSION

Administration of glucose to the animals was followed by hyperglycemia; the blood sugar reached a maximum 30 min after the beginning of administration, when it was 115% above the initial level (79.1 ± 1.37 mg%). After 1 h the blood sugar had returned to normal. After administration of glucose for 30 days the

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TABLE 1. Lipid Content in Heart Tissues (in percent of weight of dry tissue) during Prolonged Administration of Glucose (M± m)

| Experimental conditions | Number of animals | Ethereal fraction | | | | Alcoholic fraction | | Total content of lipids |
|---|---|---|---|---|---|---|---|---|
| | | lipids | cholesterol | | lecithin + cephalin | lipids | sphingomyelin | |
| | | | in tissues | in lipids | | | | |
| Control | 6 | 26,41±0,87 | 0,62±0,04 | 2,12±0,05 | 5,15±0,05 | 11,16±0,26 | 5,73±0,13 | 37,58±0,93 |
| After administration of glucose for 30 days | 5 | 31,18±2,10 | 0,81±0,04 | 3,04±0,15 | 6,16±0,21 | 14,42±0,40 | 5,72±0,16 | 45,60±2,01 |
| | | +18,0% | +26,5% | +43,4% | +19,6% | +29,2% | −0,02% | +21,3% |
| P | | >0,05 | <0,01 | <0,01 | <0,01 | <0,01 | >0,9 | <0,01 |
| Control | 5 | 25,30±0,42 | 0,63±0,02 | 1,69±0,03 | 5,12±0,11 | 11,66±0,31 | 5,76±0,05 | 36,86±0,76 |
| After administration of glucose for 60 days | 5 | 29,80±0,79 | 0,79±0,01 | 3,30±0,12 | 5,63±0,17 | 13,74±0,30 | 5,96±0,21 | 43,54±1,10 |
| | | +17,8% | +25,4% | +65,8% | +10,0% | +17,8% | +3,5% | +18,1% |
| P | | <0,01 | <0,01 | <0,01 | <0,05 | <0,01 | >0,6 | <0,01 |
| Control | 7 | 26,01±0,86 | 0,65±0,04 | 2,14±0,04 | 5,27±0,13 | 11,30±0,46 | 5,78±0,05 | 37,31±0,55 |
| After administration of glucose for 90 days | 5 | 32,16±0,77 | 0,77±0,02 | 3,20±0,11 | 5,97±0,26 | 13,04±0,78 | 4,49±0,13 | 46,10±1,30 |
| | | +23,6% | +18,4% | +49,5% | +13,3% | +23,4% | −22,3% | +23,6% |
| P | | <0,01 | <0,05 | <0,01 | <0,05 | <0,02 | <0,01 | <0,01 |

TABLE 2. Change in Rate of Glycolysis in Tissues during Prolonged Administration of Glucose (M± m)

| Experimental conditions | Number of animals | Brain | | Liver | | Heart | | Aorta | |
|---|---|---|---|---|---|---|---|---|---|
| | | level of preformed lactic acid* | increase (37°C, 1 h) | level of preformed lactic acid* | increase | level of preformed lactic acid* | increase | level of preformed lactic acid* | increase |
| | | Lactic acid concentration (in μg/100 mg fresh tissue) | | | | | | | |
| Control (healthy animals) | 7 | 0,88±0,03 | 4,62±0,08 | 0,82±0,05 | 0,79±0,02 | 2,64±0,06 | 2,16±0,04 | 0,76±0,04 | 0,51±0,03 |
| Day from beginning of glucose admin.: | | | | | | | | | |
| 30-th | 5 | 0,70±0,01 | 3,47±0,07 | 1,12±0,06 | 0,67±0,02 | 2,47±0,05 | 1,74±0,04 | 0,87±0,03 | 0,45±0,02 |
| P | | <0,01 | <0,01 | <0,01 | <0,01 | <0,05 | <0,01 | >0,05 | >0,05 |
| 60-th | 5 | 0,81±0,01 | 3,55±0,06 | 0,89±0,06 | 0,73±0,05 | 2,43±0,03 | 1,36±0,10 | 0,77±0,08 | 0,43±0,02 |
| P | | <0,05 | <0,01 | >0,3 | >0,2 | <0,05 | <0,01 | >0,9 | <0,05 |
| 90-th | 6 | 0,98±0,03 | 4,57±0,11 | 0,83±0,04 | 0,68±0,03 | 2,51±0,06 | 1,87±0,05 | 0,72±0,05 | 0,60±0,02 |
| P | | <0,05 | >0,7 | >0,8 | <0,1 | >1 | <0,01 | >0,4 | <0,05 |

*Lactic acid level before incubation (see "experimental method").

increase in the blood sugar when investigated in the fasting state was 6.4% (84.2± 3.06 mg%), while the maximum increase taking place during the next glucose loading, which also was observed after 30 min, was smaller (increase 76.2%). The duration of hyperglycemia was increased up to 2 h. After 3 months of administration the maximal increase in blood sugar, observed after 30 min, was only 43.6% of the initial level, while the duration of hyperglycemia was 2 h.

There was a significant increase in the total and ether-bound blood cholesterol levels after the 10th day of the investigation and in the free cholesterol level after the 50th day. The maximal increase in total cholesterol (by 61.0%) was observed after 90 days. The cholesterol concentration in the liver tissues was increased after 2 months by 33.0% (1.45± 0.10% in the experiment, 1.09±0.02% of the weight of dry tissue in the control), while after 3 months it was increased by 24.1% (1.34± 0.08% in the experiment, 1.08±0.05% in the control), while in the heart tissues the increases were by 25.6, 25.4, and 18.4% respectively. The content of total lipids, including both ether- and alcohol-soluble lipids, in the heart tissues was increased (Table 1). The rate of glycolysis in the heart was lowered after the 1st month (Table 2) and the oxygen consumption was lowered after the 2nd month (Table 3) of the experiment. The excess carbohydrate loads led to a decrease in the rate of glycolysis in the brain after 1 and 2 months but had no significant effect on the oxygen consumption of this tissue (Table 2). The level of total lipids in the brain under these conditions also remained constant, except for the sphingomyelin concentration, which was reduced by 23.2% after 2 months and by 46.7% after 3 months ($P < 0.01$). The rate of glycolysis in the tissues of the aorta was reduced after 2 months and increased toward the end of the investigations (Table 2). The SDH activity, on the other hand, was 34.7% lower by the end of the experiment than in the control (0.052±0.003 and 0.08± 0.001 mg tetrazolium/g fresh tissue). Histological investigation of the aorta revealed small plaques containing large quantities of lipids in the thoracic portion in two of the five rabbits receiving glucose for 3 months.

These results show that during prolonged administration of glucose the potential ability of the tissues of the heart, liver, brain, and aorta to assimilate this carbohydrate by glycolysis was diminished and the oxygen consumption of the tissues was reduced, thereby interfering with the conversion of glucose in the Krebs' cycle. This is confirmed indirectly by the results showing the decrease in SDH activity in the liver after 3 months from 0.28±0.01 to 0.17±0.03 mg tetrazolium. The excess of glucose can be deposited as glycogen or converted into lipids. Increased deposition of glycogen in the tissues has not been observed during excessive administration of sugars [7]. On this type of diet the activity of the enzymes of the Krebs' cycle was shown to be increased [3, 4, 9], with a consequent accumulation of $NADP \cdot H_2$ essential for lipid synthesis. It is also known [12] that hyperglycemia leads to crystallization of cholesterol and to its deposition

TABLE 3. Oxygen Consumption of the Tissues during Prolonged Glucose Administration (M± m)

| Experimental conditions | Number of animals | Liver | Number of animals | Heart | Number of animals | Brain |
|---|---|---|---|---|---|---|
| | oxygen consumption (in μg/100 g fresh tissue per hour) | | | | | |
| Control..... | 6 | 36,43±1,95 | 6 | 38,75±2,71 | 6 | 75,15±2,39 |
| After administration of glucose for 30 days | 6 | 36,11±1,49 | 6 | 36,43±1,25 | 6 | 72,80±2,80 |
| P | | >0,8 | | >0,4 | | >0,5 |
| Control..... | 6 | 36,03±0,85 | 6 | 38,00±1,04 | 6 | 75,48±1,33 |
| After administration of glucose for 60 days | 6 | 30,15±2,22 | 6 | 27,85±1,03 | 6 | 69,30±2,97 |
| P | | <0,05 | | <0,01 | | >0,05 |
| Control..... | 8 | 36,63±1,87 | 8 | 38,51±0,74 | 8 | 74,97±0,55 |
| After administration of glucose for 90 days | 8 | 32,47±0,76 | 8 | 26,56±0,94 | 8 | 73,01±2,02 |
| P | | >0,05 | | <0,01 | | >0,3 |

in the vessel walls. All these considerations suggest that the increased content of lipids in the heart and liver tissues of the rabbits was due to the prolonged and excessive administration of carbohydrates.

## LITERATURE CITED


1. V. S. Asatiani, Biochemical Analysis [in Russian], Part I, Tbilisi (1949), p. 212.
2. S. D. Balakhovskii and I. S. Balakhovskii, Methods of Chemical Analysis of Blood [in Russian], Moscow (1953), p. 286.
3. M. Nocheva, Probl. Endokrinol., No. 3, 20 (1970).
4. I. V. Sidorenkov, Vopr. Med. Khimii, No. 5, 535 (1967).
5. S. W. Barker and W. H. Summerson, J. Biol. Chem., 138, 535 (1941).
6. E. Cheraskin, W. Ringsdorf, A.T.S.H. Setyaadmodya, et al., Internat. Z. Vitaminforsch., 37, 449 (1967).
7. K. Held, G. Walpurger, and U. Gorrstein, Klin. Wschr., 46, 15 (1968).
8. P. T. Kuo and D. R. Basset, Ann. Intern. Med., 62, 1199 (1965).
9. H. F. Sasson, B. C. Johnson, and K. Moser, J. Nutr., 90, 275 (1966).
10. A.T.S.H. Setyaadmodya, R. A. Barrett, M. D. Cheraskin, et al., Angiology, 18, 224 (1967).
11. W. W. Umbreit, R. H. Burris, and J. F. Stauffer, Manometric Techniques and Related Methods for the Study of Tissue Metabolism, Minneapolis (1945).
12. J. A. Wilkens and L. H. Krut, J. Atheroscler. Res., 5, 516 (1965).
13. J. Yudkin and J. Morland, Am. J. Clin. Nutr., 20, 503 (1967).