INFLUENCE OF CHLORPROPAMIDE ON SOME BIOCHEMICAL COMPONENTS OF THE LIVER IN NORMAL ANIMALS AND IN ANIMALS WITH EXPERIMENTAL TOXIC HEPATITIS

(UDC 612.353.014.46:615.778.25-017.47+616.36-002-099-085.778.25-017.47)

S. M. Leites and N. Kh. Abdullaev

Department of Pathological Physiology (Professor S. M. Leites, Director),
Central Institute of Postgraduate Medical Training, Moscow
(Presented by Active Member, A. L. Myasnikov, Academy of Medical Sciences USSR)
Translated from Byulleten' Éksperimental'noi Biologii i Meditsiny, Vol. 61, No. 3,
pp. 55-59, March, 1966
Original article submitted September 17, 1964

Decreased content of glycogen and protein in the liver accompanied by increased fat are characterized by the pathochemical changes in toxic hepatitis. It has been established that when the liver is depleted of glycogen, insulin promotes glycogen deposition and inhibits infiltration by fat. Insulin also activates protein synthesis [2, 4].

Oral administration of the antidiabetic sulfonylureas stimulates pancreatic β-cell function and activates insulin secretion [1, 3].

Based on these facts we investigated the influence of long term experimental chlorpropamide administration on the concentration in the liver of certain biochemical components, and also assessed the ability of this preparation to eliminate the fundamental pathochemical changes in the liver during toxic liver damage.

Although insulin injection with glucose is used clinically in hepatitis treatment, medication with chlorpropamide per os would seem more closely physiological, inasmuch as this substance stimulates endogenous insulin secretion and thus elicits less intense compensatory reactions than does exogenous insulin given subcutaneously.

It has been reported that administration of tolbutamide exercises a favorable influence on patients with liver cirrhosis and also on dogs in which that condition is produced experimentally [7].

METHODS

Albino rats weighing 180-200 g were used as the experimental subjects. In the Series I experiments the intact rats were given chlorpropamide (Diabenese, Pfizer) per os daily at 5 mg/100 g body weight over a 50-day period. In Series II experimental chronic toxic hepatitis was produced in the rates by subcutaneous CCl_4 injection twice per week at a dose rate af 0.12 ml/100 g body weight over a 65-day period. One group of such animals served as controls,and the other group was treated with chlorpropamide each day at the dose rate indicated above, beginning at the 15th day from the start of the CCl_4 poisoning. The experimental and control rats were sacrificed in groups at the 25th, 35th, 45th, 55th and 65th days of CCl_4 intoxication. The livers were examined for their content of both lipids by extraction (after drying to constant weight) in the Soxhelet apparatus; liver glycogen was determined by the modified method of Seifter [6]; lipoproteins by the Burstein and Samaille turbidimetric method modified by Kellen and Link [5], total and residual nitrogen according to Folin (the level of protein nitrogen was calculated from the difference of these two quantities).

Series I included 40 rats and 75 were used in Series II.

RESULTS

Chlorpropamide in the intact rats produced increased liver glycogen starting at the 20th day of administration (Table 1). There was a parallel increase in the liver protein level. There was a decrease in the amount of residual

TABLE 1. The Effect of Oral Chlorpropamide Administration (5 mg/100 g daily) on Liver Total Lipids, Glycogen, β-Lipoproteins, Protein and Residual Nitrogen and on Serum Total Protein in the Rat (Mean Values in g %)

Substance administered	Period following the start of admin. (days)	Liver						Serum total protein
		total lipids*	β-lipoproteins	Glycogen	Protein nitrogen	Residual nitrogen	$\frac{\text{Residual nitrogen}}{\text{Protein nitrogen}} \times 100$	
Control	10	18,5 (18,0 — 19,0)	—	1,9 (1,8 — 2,0)	2,44 (2,35 — 2,51)	0,24 (0,2 — 0,29)	9,6 (8,1 — 11,5)	7,25 (7,03 — 7,57)
Chlorpropamide		19,0 (16,0 — 21,0)	1,29 (0,92 — 1,62)	2,02 (1,98 — 2,06)	2,64 (2,59 — 2,75)	0,16 (0,14 — 0,18)	6,0 (5,4 — 6,5)	6,5 (5,79 — 6,98)
Control	20	17,3 (16,0 — 18,5)	2,34 (2,36 — 2,32)	1,99 (1,85 — 2,1)	2,52 (2,48 — 2,58)	0,26 (0,23 — 0,29)	10,1 (9,2 — 11,2)	7,3 (7,03 — 7,57)
Chlorpropamide		17,1 (14,5 — 20,0)	1,36 (0,84 — 1,8)	2,62 (2,54 — 2,71)	2,93 (2,7 — 3,43)	0,25 (0,2 — 0,31)	7,8 (6,3 — 9,5)	7,5 (7,03 — 7,85)
Control	30	16,0 (15,5 — 17,0)	2,72 (2,32 — 3,26)	1,84 (1,62 — 2,1)	2,91 (2,3 — 3,56)	0,24 (0,2 — 0,29)	8,2 (8,0 — 8,6)	7,44 (7,03 — 7,65)
Chlorpropamide		20,4 (17,0 — 23,0)	1,94 (1,19 — 2,56)	3,23 (2,98 — 3,5)	4,05 (3,43 — 4,92)	0,33 (0,27 — 0,38)	8,1 (7,7 — 9,1)	7,24 (7,03 — 7,57)
Control	40	15,6 (14,5 — 16,5)	2,3 (2,16 — 2,58)	2,07 (1,8 — 2,47)	2,78 (2,62 — 3,48)	0,26 (0,22 — 0,29)	8,5 (6,8 — 10,0)	7,75 (7,57 — 8,06)
Chlorpropamide		22,4 (19,0 — 24,5)	2,01 (1,16 — 3,62)	3,18 (2,8 — 3,53)	4,22 (3,54 — 4,63)	0,36 (0,31 — 0,42)	8,4 (7,6 — 9,0)	7,46 (7,09 — 7,98)
Control	50	17,3 (17,0 — 18,0)	1,86 (1,74 — 2,08)	1,95 (1,8 — 2,1)	2,72 (2,58 — 2,96)	0,27 (0,24 — 0,29)	9,7 (8,1 — 11,2)	7,49 (7,09 — 7,9)
Chlorpropamide		17,1 (15,0 — 19,0)	1,8 (1,18 — 2,3)	3,22 (2,93 — 3,7)	4,25 (3,7 — 4,66)	0,36 (0,3 — 0,44)	8,4 (7,9 — 9,4)	7,91 (7,47 — 8,36)

Notes: 1. The control group contained 15 animals and each experimental group contained 5. 2. Limits of variation are shown in parentheses here and in Table 2.

*Calculated on the dry weight basis here and in Table 2.

TABLE 2. The Effect of Chronic Toxic Hepatitis Induced by CCl_4 poisoning, and of Chlorpropamide Administration Starting on the 15th Day of Intoxication, on the Liver Glycogen, Fat, β-Lipoprotein and Indices of Protein Turnover, and on Serum Protein in the Rat. Average Values in g %

Administered substances	Period after start of poisoning (days)	No. of animals	Liver						Serum total protein
			total lipid	β-lipoproteins	glycogen	protein nitrogen	residual nitrogen	$\frac{\text{residual nitrogen}}{\text{total nitrogen}} \times 100$	
CCl_4	25	5	33,0 (29.5 — 39,0)	0,73 (0,54 — 1,16)	0,93 (0,76 — 1,01)	2,05 (1,84 — 2,39)	0,21 (0,19 — 0,26)	10,7 (9,4 — 12,1)	7,19 (6,44 — 7,77)
CCl_4 + chlorpropamide	25	10	23,5 (21,0 — 25,0)	— —	2,31 (1,79 — 2,84)	3,68 (2,86 — 4,63)	0,31 (0,24 — 0,42)	8,3 (6,9 — 9,0)	7,72 (7,03 — 8,54)
CCl_4	35	5	30,7 (28,5 — 34,5)	0,65 (0,54 — 0,76)	0,64 (0,51 — 0,84)	2,15 (1,85 — 2,69)	0,26 (0,22 — 0,28)	12,3 (9,6 — 14,5)	6,35 (5,9 — 7,03)
CCl_4 + chlorpropamide	35	10	21,0 (17,0 — 22,5)	1,6 (0,8 — 2,1)	1,82 (1,16 — 2,01)	3,74 (2,96 — 4,3)	0,33 (0,29 — 0,36)	8,86 (7,0 — 9,7)	7,23 (6,44 — 8,65)
CCl_4	45	5	31,2 (29,0 — 34,0)	0,8 (0,64 — 1,08)	0,81 (0,54 — 1,02)	1,91 (1,83 — 2,02)	0,18 (0,17 — 0,19)	9,3 (8,8 — 10,2)	6,69 (5,95 — 7,03)
CCl_4 + chlorpropamide	45	10	22,5 (20,0 — 25,0)	1,45 (0,94 — 2,10)	1,84 (1,49 — 2,04)	3,18 (2,92 — 3,67)	0,31 (0,28 — 0,34)	9,8 (8,4 — 11,2)	7,05 (5,84 — 8,17)
CCl_4	55	5	33,0 (25,5 — 38,5)	0,53 (0,46 — 0,7)	0,53 (0,38 — 0,75)	1,97 (1,64 — 2,41)	0,23 (0,16 — 0,29)	11,3 (9,7 — 12,2)	6,95 (6,44 — 7,52)
CCl_4 + chlorpropamide	55	10	21,0 (18,5 — 24,5)	1,63 (1,16 — 2,32)	2,14 (1,65 — 2,55)	4,21 (3,63 — 4,9)	0,39 (0,32 — 0,45)	9,2 (8,8 — 9,8)	6,97 (6,31 — 8,17)
CCl_4	65	5	29,4 (26,5 — 33,5)	1,04 (0,7 — 1,74)	0,52 (0,46 — 0,64)	1,77 (1,6 — 1,81)	0,19 (0,19 — 0,21)	11,4 (10,5 — 12,5)	6,62 (6,34 — 7,03)
CCl_4 + chlorpropamide	65	10	21,0 (17,0 — 23,5)	1,6 (0,7 — 3,8)	2,44 (1,65 — 3,1)	3,98 (3,63 — 4,62)	0,38 (0,32 — 0,44)	9,0 (7,6 — 9,8)	7,37 (7,03 — 7,77)

nitrogen 10 days after the start of administering the preparation but after the 30th day this substance increased somewhat. The ratio of residual nitrogen to protein nitrogen (proteolysis coefficient) declined during the first 20 days and was low on the 50th day. The fat content was somewhat elevated on the 30th to 40th day after chlorpropamide dosage was instituted but in the other periods it appeared essentially unaltered. The β-lipoprotein content of the liver declined in the first 30 days and remained unchanged from the normal control at 40-50 days. The total serum protein remained practically unchanged throughout.

Thus, when chlorpropamide is administered chronically to rats the most characteristic effect is increased glycogen and protein nitrogen in the liver.

Chlorpropamide administration results in an increased body weight and a greater deposition of body fat. The weight increase in the control rats for 50 days averaged 67 g while in the animals receiving chlorpropamide the average gain was 124 g.

It may be seen from Table 2 that the giving of chlorpropamide, from the 15th day after the start of CCl_4 intoxication, regularly produced a significantly increased liver glycogen over that observed in animals receiving CCl_4 without chlorpropamide. The liver glycogen in the latter group, as expected, was much decreased. Parallel with the observed increase in liver glycogen in animals, receiving chlorpropamide, there was a significant decrease in fat content of the liver. The control rats under CCl_4 intoxication showed elevated liver fat.

Administration of chlorpropamide to animals with toxic hepatitis significantly elevated the liver protein nitrogen. In contrast, the control animals under CCl_4 intoxication showed a fall in this component.

Chlorpropamide administration to rats with toxic hepatitis lowered (except on the 45th day) the ratio of residual N to protein N (proteolysis coefficient). The liver β-lipoprotein content was lowered under the influence of chlorpropamide treatment; this component is known to be synthesized in the liver itself.

The elevation in liver protein produced by chlorpropamide in animals with toxic hepatitis is accompanied by increased levels of total serum protein.

Thus, stimulation of insulin secretion by specified chlorpropamide dosage, in animals with toxic hepatitis, eliminates the fundamental pathochemical changes in the liver; it restores the glycogen content, inhibits fatty infiltration, normalizes the protein and β-lipoprotein content.

Rats with chronic toxic hepatitis, if given chlorpropamide, make greater gains in body weight than do control toxic animals. By the 65th day of chronic hepatitis the body weight of the (otherwise untreated) rats had increased by 36 g (from 198 to 234 g) while those animals receiving the additional treatment with chlorpropamide increased their weighton the average by 100 g (from 180 to 280 g).

LITERATURE CITED

1. S. G. Genes, Oral Therapy in Sugar Diabetes. [in Russian], Kiev (1962).
2. S. G. Genes, Sugar Diabetes. [in Russian], Moscow (1963).
3. S. M. Leites, Pat. fiziol., No. 6 (1962), p. 76.
4. A. Fischer, Physiology and Experimental Pathology of the Liver. [in Russian], Budapest (1961).
5. I. Kellen and F. Link, Z. ges inn. Med., Bd. 17, S. 1125 (1962).
6. S. Seifter, et al., Arch. Biochem., 25 (1950), p. 191.
7. I. Singh, K. B. Sehra, and S. P. Bhargava, Lancet, 1 (1961),p. 1144.