Influence of Cernitin Extracts on Serum and Liver Lipids in Rats Fed on a High-Fat Diet

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Two products are obtained by AB Cernelle from the pollens: Cernitin T60 and Cernitin GBX. Cernitin T60 contains mainly water soluble and Cernitin GBX mainly fat soluble substances. There are present many important, from the biological point of view components such, as amino acids, vitamins, enzymes, coenzymes, sterols, minerals and trace elements [8, 9, 10, 11].

Cernitin reveals anti-inflammatory properties [5]. It is an effective substance for the therapy of prostatic and urethral conditions [1, 12, 14]. Considering the chemical composition of pollen, the influence of Cernitin on the metabolic processes can be supposed.

The purpose of the investigation was to study the effectiveness of Cernitin in the experimentally induced hyperlipidemia.

Material and Methods

Cernitin was kindly supplied by AB Cernelle (Vegeholm, Sweden). The study included 72 male Wistar rats on a standard laboratory diet with body weight ranging from 160 g to 200 g at the beginning of the investigation. The animals were divided into 6 equal groups: Group 1— controls, Group 2— received a high-fat diet (HFD), Group 3— rats were given HFD and Cernitin T60 175 mg/kg/day, Group 4— animals were fed on a HFD and Cernitin T60 50 mg/kg/day. Group 5— rats were administered HFD and received simultaneously Cernitin GBX 10 mg/kg/day. Group 6— was given HFD and Cernitin GBX 50 mg/kg/day.

The HFD consisted of hydrogenated coconut oil 6.0 g/kg/day, cholesterol 3.0 g/kg/day and cholic acid 0.2 g/kg/day. Cernitin substances were given orally. Experiment covered a period of 24 weeks.

The animals were fasted for 16 hrs prior to dissection. After a mild ether anesthesia the thorax of the animals was opened and blood was drawn from the ascending aorta. At the same time the liver was removed; it was washed immediately in ice-cold isotonic solution and homogenated.

In the blood serum and liver homogenate study on lipids was carried out. The level of total lipids was determined according to Postma and Stroes [13], triglycerides according to Eggstein and Kreutz [4], total cholesterol according to Blaszczyszyn [2], free fatty acids according to Duncombe [3], β-lipoproteins according to Kellen and Belaj [6] and lipid phosphorus according to King and Wootton [7]. Total lipids, triglycerides and total cholesterol were detected in liver homogenate by the same methods. The electrophoretic separation of lipoproteins was carried out on agarose. Glucose concentration in the blood serum was tested with the orthotoluidine method. The results were elaborated statistically using the Student's t-test.

Results

Table 1 and Table 2 show the effect of Cernitin on the serum lipid levels of rats fed on the HFD. Except for lipid phosphorus, all lipid fractions were elevated significantly in the HFD-treated rats: total lipids were increased by 19%, triglycerides by 39%, total cholesterol showed a rise by 53%, free fatty acids by 34% and *β*-lipoproteins by 106%. Therapy of HFD-treated animals with Cernitin T60 resulted in a reduction of serum lipids. The concentrations of the lipids do not drop to the normal levels of the controls.

In rats receiving HFD (group 2) *α*-Lipoproteins level was markedly decreased with simultaneous elevation of pre-β and β-
lipoproteines (Table 3). A-Lipoproteins level was increased in animals receiving Cernitin T60, as compared with group 2.

Table 1.Effect on Cernitin on mean (±SD) serum total lipids, triglycerides and total cholesterol level in rats fed on a high-fat diet (HFD)

Group	Treatment	Total Lipids g/l	Triglycerides (mmol/1)	Total cholesterol (mmol/1)
1	–	2.19±0.15	1.15±0.10	1.31±0.24
2	HFD	2.61±0.38	1.60±0.20	2.01±0.35
3	HFD + Cernitin T60 175 mg/kg	2.35±0.38	1.46±0.30	1.49±0.47
4	HFD + Cernitin T60 50 mg/kg	2.56±0.41	1.32±0.14	2.32±0.33
5	HFD + Cernitin GBX 10 mg/kg	2.80±0.42	1.66±0.18	1.99±0.40
6	HFD + Cernitin GBX 50 mg/kg	2.46±0.71	1.39±0.27	2.11±0.22
	P 1/2	<0.01	<0.001	<0.001
	P 2/3	>0.1	>0.2	<0.01
	P 2/4	>0.7	<0.001	<0.05
	P 2/5	>0.2	>0.4	>0.8
	P 2/6	>0.5	>0.05	>0.4
	P 3/4	>0.2	>0.2	<0.001
	P 5/6	>0.1	<0.02	>0.3

Table 2.Effect of Cernitin on mean (±SD) serum level of free fatty acids, *β*-lipoproteins and lipid phosphorus level in rats fed on a high-fat diet (HFD)

Group	Treatment	Free fatty acids (µmol/l)	*β*-Lipoproteins (g/1)	Lipid phosphorus (mmol/1)
1	–	440±120	0.30±0.04	0.061±0.005
2	HFD	590±130	0.62±0.15	0.066±0.009
3	HFD + Cernitin T60 175 mg/kg	480±150	0.51±0.08	0.061±0.013
4	HFD + Cernitin T60 50 mg/kg	570±160	0.51±0.06	0.068±0.008
5	HFD + Cernitin GBX 10 mg/kg	600±100	0.68±0.15	0.068±0.008
6	HFD + Cernitin GBX 50 mg/kg	520±120	0.57±0.09	0.072±0.013
	P 1/2	<0.01	<0.001	>0.05
	P 2/3	<0.05	<0.05	>0.2
	P 2/4	>0.6	<0.05	>0.6
	P 2/5	>0.7	>0.2	>0.3
	P 2/6	>0.1	>0.3	>0.1
	P 3/4	>0.1	>0.8	>0.8
	P 5/6	>0.05	<0.05	>0.5

Table 3. Electrophoretic separation of lipoproteins into fraction (mean±SD)

Group	Treatment	*A*-Lipoproteins	Pre-*β*-Lipoproteins	*B*-Lipoproteins
1	–	69.03± 7.35	19.19± 5.16	11.78±3.90
2	HFD	40.81±14.77	34.87±10.59	24.32±8.14
3	HFD + Cernitin T60 175 mg/kg	46.64± 6.90	40.22± 7.79	13.14±3.52
4	HFD + Cernitin T60 50 mg/kg	59.57± 9.50	20.86± 4.71	19.65±7.32
5	HFD + Cernitin GBX 10 mg/kg	38.86± 6.73	45.06± 6.51	18.07±2.74
6	HFD + Cernitin GBX 50 mg/kg	41.41± 7.73	38.51± 7.02	20.08±3.61

Table 4. Lipids concentration (mean±SD) in the liver homogenate (calculated per 1 g of the tissue)

Group	Treatment	Total lipids (mg)	Triglycerides (mmol)	Total cholesterol (mmol)
1	–	0.34±0.03	0.21±0.03	0.09±0.02
2	HFD	0.69±0.06	0.43±0.06	0.22±0.06
3	HFD + Cernitin T60 175 mg/kg	0.57±0.11	0.43±0.11	0.28±0.08
4	HFD + Cernitin T60 50 mg/kg	0.51±0.08	0.38±0.08	0.20±0.04
5	HFD + Cernitin GBX 10 mg/kg	0.60±0.09	0.37±0.06	0.20±0.03
6	HFD + Cernitin GBX 50 mg/kg	0.60±0.07	0.36±0.06	0.24±0.05
	P 1/2	<0.001	<0.001	<0.001
	P 2/3	<0.01	>0.9	<0.05
	P 2/4	<0.001	>0.1	>0.3
	P 2/5	<0.01	<0.02	>0.4
	P 2/6	<0.01	<0.01	>0.3
	P 3/4	>0.1	>0.2	<0.02
	P 5/6	>0.8	>0.5	>0.05

All lipid fractions examined in the liver homogenate were significantly elevated under the influence of HFD: total lipids by 103%, triglycerides by 105% and total cholesterol by 144% (Table 4). Administration of Cernitin products did not affect the lipids in homogenate, in comparison with animals of group 2.

The glucose blood level in rats fed on a HFD was higher by 34%. It was significantly dimished in animals treated with Cernitin T60 in a dose 175 mg/kg (Table 5).

The liver weight was found to be increased in those animals, that were administered HFD alone. The weight of this organ was decreased in group receiving Cernitin T60 (Table 6).

The body weight of all groups of animals was being elevated in the course of experiment (Table 7). Rats of group 4, group 5, and group 6 shows the highest increase in the body weight

Table 5. Glucose level (mean±SD) in the blood serum of rats receiving Cernitin

Group	Treatment	Glucose (mmol/l)
1	–	4.40±0.64
2	HFD	6.92±0.92
3	HFD + Cernitin T60 175 mg/kg	5.83±1.05
4	HFD + Cernitin T60 50 mg/kg	7.25±1.00
5	HFD + Cernitin GBX 10 mg/kg	6.57±0.64
6	HFD + Cernitin GBX 50 mg/kg	6.44±1.09
	P 1/2	<0.001
	P 2/3	<0.01
	P 2/4	>0.3
	P 2/5	>0.2
	P 2/6	>0.2
	P 3/4	<0.001
	P 5/6	>0.6

Table 6. Liver weight of rats (mean±SD) receiving Cernitin

Group	Treatment	Liver Weight (g/100 g body weight)
1	–	2.64±0.17
2	HFD	3.06±0.22
3	HFD + Cernitin T60 175 mg/kg	2.80±0.41
4	HFD + Cernitin T60 50 mg/kg	2.84±0.21
5	HFD + Cernitin GBX 10 mg/kg	3.05±0.20
6	HFD + Cernitin GBX 50 mg/kg	3.19±0.23
	P 1/2	<0.001
	P 2/3	>0.05
	P 2/4	<0.01
	P 2/5	>0.9
	P 2/6	>0.1
	P 3/4	>0.7
	P 5/6	>0.1

Table 7. Mean body weight of rats in the course of experiment expressed n percentage (initial body weight was taken as 100%)

Group	Weeks											
	2	4	6	8	10	12	14	16	18	20	22	24
1	115	124	120	129	129	144	145	148	153	158	160	166
2	104	94	122	132	137	145	143	150	149	158	159	167
3	94	110	119	129	137	142	147	150	154	162	164	164
4	114	129	145	152	156	167	168	178	173	180	183	194
5	109	125	139	149	153	161	166	173	172	181	184	188
6	108	124	133	139	152	160	159	170	167	177	180	186

Discussion and Conclusion

Our studies showed that Cernitin – the microbiologically prepared extract from the pollens of specially selected plant, reduced the disturbances in lipid metabolism caused by a HFD. Among the examined products, Cernitin T60 was active substance regarding affecting the metabolism. These results are to some degree in agreement with our previous investigations concerning the evaluation of effectiveness of bee-pollen as a raw material in experimental hyperlipidemia [15]. However Cernitin products seem to be less active, at least in the doses applied. Therefore it would be advisable to check the metabolic activity of the Cernitin products administered in various doses separately and in combination, both orally and intraperitoneally.

Another observation [15] confirmed in this study is an increase of body weight of animals treated with Cernitin. Significance of that fact may be considered in animals as well as in human beings. Our studies showed that Cernitin is able to improve lipid metabolism disturbances in animals.

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