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294 **ANTISTREPTOLYSIN "O" TITER IN PATIENTS WITH VIRAL HEPATITIS** 359

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Viral hepatitis appears to be a problem of utmost importance for infectious pathology in this country at present. The progress achieved of research studies in this respect in the past several years is not insignificant, but nevertheless, numerous aspects related to diagnosing remain unsolved. This is the explanation of the continuous strive for finding new tests.

Many authors report high antistreptolysin "O" titers in patients with affections of the liver. Köhler (6) states that raised titers in patients with hepatitis are met with, but invariably in the same percentage as elevated titers found in healthy subjects. Packellen, Oker-Blum (cited by 1) point out that the sera of hepatitis patients appear to be strongly inhibitory in terms of "O" streptolysin and furthermore, that this effect is due to the lipoproteins which differ from those found in normal sera by the cholesterol groups' content. A similar non-specific increase, due to the  $\beta$ -lipoproteins is reported by Hällén too (7). Tzonchev and assoc. (5) assume that high antistreptolysin values are brought about by hypercholesteremia which inhibits the hemolysis. Panayotov and assoc. (3) establish a rise of antistreptolysin "O" titers in patients with infectious hepatitis, in the early phases of the affection, and a fall — after the second week. Bearing in mind the discrepant and hardly convincing opinion hitherto expressed as regards high antistreptolysin titer in patients with hepatitis, further investigations were undertaken on the serum of newly affected in which the antistreptolysin titer was determined and attempts were made for clarifying the essence of high titers.

#### Material and method

Antistreptolysin "O" titer was found in 68 patients with infectious hepatitis. Fifty one of them were subjected to repeated investigation, the first one — on admission and the second — on dismissal. All patients undergoing investigations were found to be with proved infectious hepatitis on the basis of clinical and biochemical indices. The antistreptolysin titer is investigated according to the Köhler method (5) with streptolysin "O". In addition, lipoproteinograms were made of some of the sera and the height of antistreptolysin titer was determined in the single fractions of the lipoproteinogram. The lipoprotein fractions were suspended by means of electrophoresis (3). Several electrophoregrams were obtained from a single serum. One of them was developed in order to establish the

locality of lipoprotein fractions after Swahn (8), whilst the remainder were subjected to extraction. Some of them were extracted with ether, with subsequent evaporation on water bath, and others — with chloroform, which was evaporated in water bath under vacuum. The dilutions were prepared with physiological solution.

For quantitative determination of lipoprotein fractions, they were eluted from the developed lipogram with 20% acetic acid and 80% ethanol for one and a half hour duration. Photometration was performed employing photoelectric colorimeter with red filter.

Extraction of lipoproteins besides by means of electrophoresis, was carried out as follows: 0,25 ml serum was mixed up with 1 ml ether (chloroform respectively). The mixture was thoroughly stirred and the two liquids were separated. The antistreptolysin titer of the serum was determined. The ether (resp. chloroform) was evaporated. Restoration of the initial level (0,25 ml) was achieved by addition of physiological solution, and thereby, the level of the antistreptolysin titer was determined.

### Results

Of the 51 investigations performed repeatedly, a fall of the titer was noted in 40, increase — in 7 and in 4, the level was maintained unaltered. The results of the investigations of patients in whom a fall of the antistreptolysin titer was observed are illustrated in table 1.

Table 1

Number of patients during	Antistreptolysin "O" titer in units										Total
	bene-ath 125	125	166	250	333	500	625	833	1250	above 2500	
I investiga-tion	8	7	4	8	9	9	10	5	6	2	68
II investi-gation	22	10	7	4	1	1	4	1	1	—	51

It is evident that the titers at the first investigation of the patients are higher than those of the second. A shift is marked of the titers towards lower values. It seems that during the first investigation, merely eight patients had antistreptolysin titer beneath 125 U, whereas at the second investigation they are twenty two. The same holds true for the patients with titer 1250 U: at the first investigation they amount to six, and at the second — merely one. Antistreptolysin titer exceeding 2500 U is noted only at the first investigation, whereas titers above 250 U at the second investigation are found in isolated cases.

Table 2 demonstrates the dynamics of antistreptolysin titers during double investigations, displaying a fall of the titer.

The seven cases in whom the antistreptolysin titer revealed ascending dynamics were established as moderately severe and protracted. In them

Table 2

ANTISTREPTOLYSIN „O“ TITER IN UNITS												
I investigation	above 2500	1250	1250	833 833	625	625	500	333	333 333 333	250	166	125
II investigation	166	125	125	250	166 250	125	125	125	125 166 250	125	166	125
Number of patients	1	3	1	1	2	3	1	1	1	2	4	1

Table 3

№ of serum	Antistreptolysin „O“ titer of:				Antistreptolysin „O“ titer of lipoproteinogram fractions extracted with:								
	serum after extraction		chloroform		ether			chloroform			chloroform		
	with ether	with chlorof.	Albumin	Albumin	A	B	C	Albumin	A	B	C		
1	below 125	125	below 125	below 125	625	below 125	below 125	below 125	below 125	833	1250	625	
2	below 125	166	below 125	below 125	833	below 125	below 125	below 125	below 125	below 125	below 125	below 125	
3	below 125	333	below 125	below 125	166	below 125	166	below 125	below 125	1250	500	833	
4	below 125	250	below 125	below 125	250	below 125	below 125	below 125	below 125	below 125	below 125	below 125	
5	below 125	500	below 125	below 125	500	below 125	below 125	below 125	below 125	below 125	below 125	below 125	
6	below 125	333	below 125	below 125	333	below 125	166	below 125	below 125	500	166	below 125	
7	below 125	833	below 125	below 125	166	below 125	below 125	below 125	below 125	833	833	below 125	
8	below 125	166	below 125	below 125	166	below 125	below 125	below 125	below 125	166	below 125	below 125	
9	below 125	625	below 125	below 125	333	below 125	333	below 125	below 125	1250	166	250	
10	below 125	833	below 125	below 125	625	below 125	500	below 125	below 125	below 125	below 125	below 125	
Control Serum.					below 125	below 125	below 125	below 125	166	250	below 125	below 125	

the antistreptolysin titer was maintained higher even after the 30th day of the onset of the illness. Concomitant diseases or complications were not recorded in this group.

The first and second investigation of the antistreptolysin titer gave negative result in four of the patients. Slight acute forms were concerned, in which the clinical tests were, as a rule, weakly manifested.

The results of the investigations of the antistreptolysin titer of the lipoproteinogram fractions, as well as those obtained from direct extraction of sera with ether and chloroform, are presented in table 3.

In table 3 the albumin fraction of the lipoproteinogram exhibits the lowest titer, justifying the assumption that the latter is in the least dependent on the height of the antistreptolysin titer. The same holds true for the C fraction. Between fractions A and B, in all likelihood, the more important is the former for it displays higher antistreptolysin titers.

In addition to the extraction of the lipoproteinogram fractions, extraction was also carried out of the serum with ether and chloroform. Probably, these substances extract some of the serum components, exerting effect on the height of the antistreptolysin titer, for in most of the cases, the height of the latter after extraction was lowered.

The percentage content of the lipoprotein fractions A, B and C and the ratio B/A is presented in table 4.

Table 4

	№ of serum	AST	% of lipoproteins			Ratio B/A	
			A	B	C		
Serum from:	viral hepatitis	1	above 2500				
		2	2500	30,9	39,8	29,3	1,29
		5	250	32,2	47,7	20,1	1,48
		8	500	34,8	28,8	26,4	0,82
		9	166	41,2	31,7	27,1	0,77
		625	25,5	32,0	42,5	1,26	
	healthy	333	21,7	34,2	44,1	1,6	
	obstructive jaundice	above 2500	33,3	46,2	20,5	1,4	

It is evident from the table that no relationship exists between antistreptolysin titer and the separate fractions, nor between the antistreptolysin titer and the B/A ratio.

#### Inferences

The investigations carried out demonstrate that in patients with viral hepatitis a rise in antistreptolysin titers occurs in the initial stage of the disease and accordingly — fall during the convalescent period. With the

experiments described, carried out with the main purpose to clarify the high titers' essentials, it is furthermore established that the A-fraction accounts for inhibition of hemolysis.

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#### АНТИСТРЕПТОЛИЗИНОВЫЙ „О“ ТИТР У БОЛЬНЫХ ВИРУСНЫМ ГЕПАТИТОМ

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#### РЕЗЮМЕ

Антистрептолизиновый „О“ титр исследован у 68 больных вирусным гепатитом. У 40 из них исследование сделано двукратно — при поступлении и при выписке из больницы. Обнаружено снижение этого титра в период реконвалесценции. Из опытов, которые были проведены в целях выяснения сущности высоких титров было установлено, что А-фракция задерживает гемолиз.