THE COMPARATIVE STUDY ON SEROLOGICAL PROPERTIES OF ANTIGENS DERIVING FROM THE LARVAE OF TRICHINELLA SPIRALIS ISOLATED FROM MUSCLES OF RATS, RABBITS AND PIGS

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Introduction

The revision of reports on the matter under investigation leads to the conclusion that the value of the. *T. spiralis* antigen is influenced by the technics applied for its extraction, purification and sterilisation [1, 3-6]. It seems likely that the stage of the parasite development is also of some importance as well as the kind of animal from which larvae have been isolated to obtain antigen.

The purpose of this study was to verify whether there are or not any differences between the antigens prepared from the larvae of *T. spiralis* isolated from muscles of rabbits, rats or pigs.

Material and Methods

Antigens

Antigen of T. spiralis larvae isolated from rabbits (Rb).

Rabbits were infested with 0,2 ml. of the thick suspension of the T. spiralis larvae administered through gastric tube. The animals were killed by narcosis 6 weeks later. The meat containing considerable amount of the larvae was ground in a mill and digested with artificial gastric juice ($1^{0}/_{0}$ HCl + 0,7°/₀ pepsin) in Baermann funnel at 37° for 18 hours. Then larvae were rinced with physiological saline and with distilled water, dried in vacuum dessicator, ground in a mortar, and salt extract was prepared at the proporation of 1:100. The extract was then ten times freezed and wormed again, centrifuged, treated with some preserving agent and after titration was employed as antigen.

Antigen of the. T. spiralis larvae isolated from rats (Rt)

Fasting rats were infected by ingesting about 10 grams of meat containing many *T. spiralis* larvae. They were killed by narcosis 6-8 weeks later. Antigen was prepared as described above.

Antigen o fthe T. spiralis larvae isolated from pigs (P)

Trichinous porc meat obtained from sloughter-house was used to producte antigen. We failed to establish the time of infestation even approximatively, since no data concerning the killed animals were available. Antigen was prepared in the same way as the two described above.

Sera

The rabbit immune sera were obtained from infested animals at 5-8 weeks after invasion. High serum titres were found in all rabbits (1:512 and higher) and the presence of the T. spiralis larvae in diaphragm was detected in killed animals.

Sera of persons with trichinellosis were sampled from patients between 5th and 12th week of disease. The diagnosis was established in these subjects basing on their level of eosinophils in peripheral blood amounting over 10^{0} /o and on some typical symptoms such as periocular oedema, muscular pain and fever. The sera showing high titres of the complement fixation test (up to 1:1024) were used in this study.

The Ascaris, Cysticercus and Echinococcus immune sera derived from rabbits injected previously with increasing doses of the given antigen at 3-day intervalls. Then the animals were bled, sera separated and titrated.

Sera of persons in which the possibility of trichinellosis had been excluded for some recent years by means of anamnesis were used as the control ones.

A modification (3,4) of the complement fixation test after Bordet-Wassermann was applied to evaluate individual antigens.

Results and Discussion

Table 1 presents the results of studies on individual antigens as tested against the following sera: The rabbit immune sera, the sera of persons suffering from trichinellosis, the *Ascaris*, *Echinococcus* and *Cysticercus* immune sera and those deriving from healthy persons.

Fifty one rabbit immune sera out of 57 tested (89%) showed the

TABLE 1

Results of complement fixation test in relation to the different kind of T. spiralis antigen as tested against various sera

	No of exa-		Kin	d of posi	ture ant	igen	
Kind of sera	mined	Antiger	n "Rb"	Antiger	1 ,,Rt"	Antige	n "P"
	samples	No	%	No	%	No	%
Rabbit immune sera	57	52	91.0	51	89.2	50	87.0
Sera of persons vith trichinellosis	41	32	78.0	32	78.0	31	75.0
Sera of healthy persons	42	0	0	0	0	0	0
Immune sera of							
Ascaris	10	0	0	0	0	0	0
Echinococcus	10	0	0	0	0	0	0
Cysticercus	10	0	0	0	0	0	0

positive reaction toward the antigen "Rt", 52 did so toward the antigen "Rb" (84%) and 50 toward the "P" (79%).

The complement fixation test appeared to be positive in 41 sera of trichinous persons against antigen "Rb" in 32 such a sera against antigen "Rt", and in 31 when antigen "P" was used.

The same test was negative or only slightly positive (up to the dilution 1:8 only) when 10 Ascaris, Echinococcus or Cysticercus immune sera were examined against each of the antigens.

Tests performed with sera of healthy persons were always negative. The results were verified statistically using the χ^2 .

When antigen "Rb" was compered to antigen "Rt" the velue test $\chi^2 = 3.46$ was found.

It was $\chi^2 = 0.36$ for antigens "Rb" and "P" and $\chi^2 = 0.084$ for antigens "Rt" and "P".

The differences found may be considered as insignificant on the 95% probability level with 1 degree of freedom.

Table 2 presents the mean values of titres in 10 rabbits immune sera in relation to the kind of the employed antigen. It is seen that there wre no marked differences between the individual antigens in their titre values nor in the intensivity at which they react on the complement fixation test.

The mean intensivities of the complement fixation test found in 10 sera of trichinous persons when examined against various antigens

TABLE 2

Mean values of titres in 10 rabbits immune sera in relation to the kind of the employed T. spiralis antigens

											Tit	re (Titre of rabbit immune sera	bbi1	in	nwı	ne S	era										
Antigen		1:8	8			1:16	16			1:32	8			1:64	4			1:128	8			1:256	2			1:512	72	1 1
	+++++	++	+	1	++++	+	+	1	++++	++	+	i	+ + + + + - + + + + + - + +	++	+	_	++	+++	+	+	+++++++++++++++++++++++++++++++++++++++	+	+	+	+++	+	- 1	1
"Rb"	8		П	0	1 0 7 1 1 1			-	5	-	က	П	1 3 1 4 2 1 0	-23		0	3 1 2 4 3	П	- 7	4	က	2 0 5	0	Σ Ω.	7			9
"Rt"	10	0	0	0	0 0 0 10 0 0	0	0	0	ည	1 3 1	က	-	က	0 2 3	72	က		က	81	4	3 2 4 1. 2 1. 6 0	8	 	 6	0	83	က	ம
"Ł"	10	0	0	0	10	0	•	0	0 0 0 10 0 0 0 6 1 1 2			8	-	က	ည	-	3 5 1 0 3 5 5 0 2 2 6 0	က	ည	S.	0	8	63	9	0	0		2

TABLE 3

The mean intensivities of the complement fixation test in 10 sera of patients suffering from trichinellosis when examined against various T. spisalis antigens

											Tit	re	Titre of rabbit immune sera	bbit	ri in	nmı	ne s(ıra									
Antigen		1:8	8			-	1:16			=	1:32			-	1:64			1:128	80			1:256	9			1:512	2
	+++++		+ + +	ı	++	+	+	ı	+ + +	+	+	1	- + + + + + + - + + + + + + - + +	+	+	1	++++	+	+	ı	++	+++	+	+	+ + + + + +	+	1 +
"Rb"	∞	П	1 0 1	-	<u>-</u>			1 1 1	8		1 0 1	-	9	-	-	1 2	41	4	0	63	4 0 2 3 3 1 3	က		က	61	-	_
"Rt"	80	-	1 0 1	-	ω	-	-	1 0	2	-	-	-	2	0	0 2 3	က	5	0	0 0	ည	3 0	0	2	2	0	0	က
"P"	- ∞	0	0 1 1	-	ω	0	7	-	80			0	8 0 1 1 8 1 1 0 3 1 3 3 0 4 4 2 0 2 2 6	-	က	က	0	4	4	63	0	87	~~~	9	0	63	2 6

are gathered in table 3. Here, also no significant differences were found.

Fig. 1 was drown to demonstrate graphically the mean intensivities of the same test in 10 sera in relation to the kind of antigen.

Summary and conclusions

It might be concluded from the obtained results that no significant serological differences exist between the *T. spirails* antigens prepared from larvae deriving from muscles of rabbits, rats or pigs, when the methods applied in this study were used.

All the three antigens showed the simillar ability of the complement fixation when tested against the rabbit immune sera as well as against those of persons suffering from trichinellosis. The antigens behaved also in the simillar way towards sera of normal persons and towards Ascaris, Echinococcus and Cysticercus immune sera.

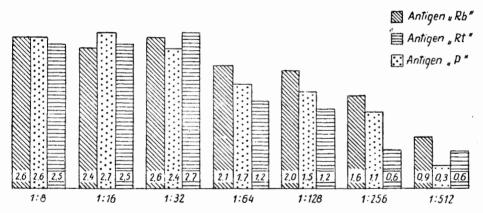


Fig. 1. The mean intensivities of the complement fixation fest in 10 serums of patients with trichinellosis when examined against various T. spiralis antigens.

The results of present experiments do not confirm Gaase's opinion, who suggested that antigen prepared from. *T. spiralis* larvae isolated from muscles of swine are superior to these obtained from muscles of rats (2).

Studies on larger material using other serological methods should be needed to drow out some more detailed conclusions.

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