1970

Translation of Galuzo, I. G., N. I. Sevast'yanova, M. N. Mel'nikh, A. E. Grigorashenko, U. D. Vustina, A. M. Krivkova, L. S. Bogdanyuk, and M. E. Bokova. 1970. Serologicheskaya kharakteristika shtammov toksoplazm, vydelennykh ot razlichnykh vidov zhivotnykh [= Serologic characterization of strains of *Toxoplasma* isolated from various species of animals]. *Voprosy Prirodnoi Ochagovosti Boleznei* [= *Contributions on the Natural Nidality of Disease*], Alma-Ata, Kazakhstan, USSR 3: 37-48

I. G. Galuzo
Academy of Sciences, Kazakh SSR

N. I. Sevast'yanova
Academy of Sciences, Kazakh SSR

M. N. Mel'nikh
Academy of Sciences, Kazakh SSR

A. E. Grigorashenko
Academy of Sciences, Kazakh SSR

U. D. Vustina
Academy of Sciences, Kazakh SSR

COLLEGE OF VETERINARY MEDICINE
UNIVERSITY OF ILLINOIS
URBANA, ILLINOIS

TRANSLATION NO. 28

Translated from Russian by Frederick K. Plous, Jr.
Edited by Norman D. Levine

Galuzo, I. G., N. I. Sevast'yanova, M. N. Mel'nikh, A. E. Grigorashenko, U. D. Vustina, A. M. Krivkova, L. S. Bogdanyuk and M. E. Bokova. 1970. Serologicheskaya kharakteristika shtammov toksoplazm, vydelennykh ot razlichnykh vidov zhivotnykh. [Serologic characterization of strains of Toxoplasma isolated from various species of animals.] Voprosy Prirodnoi Ochagovosti Boleznei. [Contributions on the Natural Nidality of Diseases.] Alma-Ata, Kazakhstan, USSR. 3:37-48.

While working with strains of Toxoplasma of differing virulence from many species of domestic and wild animals, and having in our collections strains from man as well, we conducted research into their serologic identities. The task consisted in determining the serologic resemblance between various strains distant in origin and differing as to virulence.

Materials and Methods

Material for research consisted of virulent and avirulent strains of Toxoplasma isolated at different times from domestic and wild animals in Kazakhstan (Table 1), sera from various species of wild and domestic animals artificially infected at the experimental base of the Institute of Zoology, Academy of Sciences, KazSSR with virulent and avirulent strains of Toxoplasma, or from spontaneously infected animals and man.

Antigens from virulent and avirulent strains were prepared at the Mechnikov Institute (Odessa) from the peritoneal exudate of infected white mice. The one-batch system was used; all material being prepared at one time, under the same conditions, by the same people. The sera were used in the experiment in their fresh, unstored form. The complement fixation reaction (CFR) was run in the same way in both Alma-Ata and Odessa when serologic studies of toxoplasmosis were being conducted. The controls consisted of antigens prepared from producer-strain RH at the Odessa Bacteriological Preparations Plant and strains isolated from humans in Alma-Ata, as well as sera from healthy animals.

Artificial immunization of animals was produced by infecting them with a massive dose of Toxoplasma of different virulent and avirulent strains.

Research Results

I. Group studies

1. Outcome of CFR with blood sera of humans and antigens from virulent strains V-XVIII. This experiment made use of sera from 58 patients suspected of having toxoplasmosis (Odessa) and 21 workers (Alma-Ata) who

had come into contact with toxoplasmosis-infected animals. Twenty-two of the thirty-four in the first group reacted positively at titers of 1:12, 1:14 and 1:16. At the same time the same sera were positive to the CFR with various antigens at rather high titers. The most active appeared to be antigen VI and the weakest antigen XI at titers for control-antigen XVIII. Six sera of 21 studies from persons in contact with infected animals were positive with antigens from virulent strain V.

It follows from the experiment that spontaneously infected carriers of Toxoplasma have antibodies in their blood which are positive to the CFR with antigens from virulent strains; all antigens of varied and distant origin yield in general equal titers, tho they are somewhat lower than the control, which was 37 (antigen XVIII) as against 22-34.

2. Outcome of CFR on human sera and antigens from avirulent strains I-IV. Using antigens prepared from avirulent strains (Table 1), we set up CFR's with the same sera from patients with clinical toxoplasmosis and from laboratory workers in contact with artificially infected animals. Out of 58 sera in the first group, 15 and 19, respectively, reacted with antigens III and IV, and 33 reacted with the control. In the second group antigens II and III were positive with 8 and 5 sera, respectively (control, 6). Antigens I and IV, which reacted with 16 and 19, respectively, in the first group, yielded negative results (Table 2).

This was caused by the fact that in the first case (Odessa) almost 3 times more sera were used, and the test was repeated several times, which gave a greater possibility of catching positively reacting sera with antigens I-IV.

From this series of experiments it is apparent that antigens prepared from avirulent strains work more or less in the same way as antigens from virulent strains (but only at significantly low titers--1:2).

3. Outcome of CFR on sera from spontaneously infected animals and antigens from virulent strains. We used sera from sheep, cattle, swine, goats and rabbits, among which the CFR with antigens from virulent strains turned up positively reacting sera with all antigens, but in varying titers (Table 3). This was apparently caused by the different amounts of antibody in the sera of the animals used.

For this same reason the antibodies prepared from avirulent strains were positive only with the rabbit sera. Of 7 sera studies, 4 and 6 reacted positively.

4. Outcome of CFR on sera from artificially infected animals and antigens from virulent strains of Toxoplasma. The results of these experiments (Table 4) are more clear-cut. Especially distinct are the reactions with sera from an ass, horses, rabbits, dogs and the urial. The sera of these animals yielded high CFR readings with all antigens at high titers. Before the experiment the animals were infected at different times and several times apiece (hyperimmunization) with different strains of Toxoplasma for the different animals. The sheep and goats taken for this experiment were artificially infected once in the two or three years prior

to the experiment; their reaction to the CFR study was of a different character. Only a small number of them (1-3 out of 14 sheep and 4-7 of the 31 goats studied) were positive with all antigens used in the experiment. It is characteristic that the control antigen gave identical titers in the experiments with sera from hyperimmune animals and somewhat different data with the sera from long-infected animals.

The sera from high-hyperimmune horses, the ass and the urial reacted positively to the CFR with all antigens obtained from avirulent strains.

II. Individual studies of sera with various antigens

1. Human sera. We studied 18 sera, each of which was used with different antigens (Table 5). They reacted positively with various antigens on the CFR. Twenty sera not shown in the table yielded negative results with all antigens.

2. Sera from spontaneously infected animals. The results from individual research on sera from animals spontaneously infected with various antigens showed that the sera from both cattle and from goats and rabbits gave generally identical titers with slight deviations on the CFR (Table 6). Especially indicative were the 16 cases of negative results in cattle, the 31 in swine and the 35 in goats.

In three cases there were divergences in the titers of various antigens with the same sera: from - to ++.

The sera from rabbits reacted identically with the antigens from both the virulent (V-XVIII) as well as the avirulent (I-IV) strains. It is characteristic that the antigens from the avirulent strains were not positive at all in CFRs with sera from other species of animals.

3. Sera from artificially infected animals. Seventeen sera from a horse infected repeatedly with virulent strain STG were positive (Table 7) with all antigens from virulent strains. The same thing happened also with a serum from one ass repeatedly infected with a virulent strain. Distinct positive titers were obtained with sera from rabbits infected with avirulent strains, as well as with sera from 4 goats, 2 dogs and 1 urial infected with a virulent strain. Identical negative results were obtained with sera from 4 sheep and 24 goats which had become sick during artificial infection more than 2 years earlier.

The sera from the horse, ass and urial, which had been hyperimmunized with a virulent strain, were positive in a CFR with antigens from both virulent strains (V-XVI) and the control (XVIII) as well as with antigens from avirulent strains (I-IV).

In addition, as in the previous experiments with sera from spontaneously infected animals, this experiment included instances in which the same sera yielded varying CFR readings. Ten rabbits infected with the virulent strains VFG and LEI were distinctly positive to a CFR with antigens from virulent strains (see Table 7).

From the foregoing results of the cross-application of CFR's with various antigens it follows that the toxoplasmas parasitizing various animals

are serologically identical and related to human Toxoplasma. The differences in serologic titer of the individual strains are caused by technical nuances in the arrangement of the experiment, the quality of the sera and the strength of the antigens in conjunction with the nature of the initial material.

Of some interest are the serologic identification reactions of a group of avirulent strains of Toxoplasma with virulent ones. In apparent disregard of the lesser activity of antigens prepared from avirulent strains of Toxoplasma, the human sera gave positive CFR's no less than the same sera with antigens from virulent strains. The same picture was observed also in a reaction with sera from spontaneously infected rabbits.

This permits consideration of the possibility that avirulent strains of Toxoplasma are, according to CFR data, related to virulent strains when human and rabbit sera are used. As confirmation we have the results of studies of sera taken from animals carrying avirulent strains and reacting with antigens prepared from virulent strains which steadily yielded positive complement fixation reactions.

It should be noted that sera obtained from man react positively with antigens prepared from virulent and avirulent strains of Toxoplasma isolated from animals, and vice-versa.

The results of the research conducted permit us to ask the following practical question: Is it obligatory to prepare an antigen for widespread diagnostic studies of animal and human toxoplasmosis from one so called ethalone strain RH? Might it not be possible to select from the number of tested strains isolated within the USSR a stable strain, no less active than RH, regardless of whether it is isolated from man or animals? And might it not be possible to prepare an antigen and an allergen from this strain? It is to this question that we now turn our attention in the course of further studies in this direction.

Table 1

Strains of Toxoplasma from which antigens were prepared

Antigens	Strain code and virulence	Strain isolated from:	Authors, year	Place
I	VFG avirul.	Silver-black fox (Vulpes fulva)	Galuzo Krivkova 1965	KazSSR
II	ALG avirul.	Polar fox (Alopex lagopus)	Galuzo Krivkova 1965	"
III	OCG avirul.	Domestic rabbit	Galuzo, Krivkova, 1966	"
IV	LEI avirul.	Hare (Lepus europaeus)	I. Jira, 1964	Czechoslovakia
V	STG virul.	Saiga (Saiga tatarica)	Galuzo, Golosov, 1962	KazSSR
VI	ODG virul.	Sheep	Galuzo, Golosov, 1961	"
VII	CFG virul.	Suslik (Citellus fulvus)	Galuzo, Golosov, 1963	"
VIII	SDK virul.	Swine	Kuzovkin, 1961	"
IX	CDN virul.	Dogs	Novinskaya, 1962	"
X	EAG virul.	Ass	Galuzo, Golosov, 1962	"
XI	RTK virul.	Northern reindeer	Kolychev, 1963	Komi ASSR
XII	ADP virul.	(Rakgifer tarandus)	Pak, 1964	KazSSR
XIII	LEL virul.	Hare	Levit, Vustina, 1962	"
XIV	VFN virul.	Silver-black fox	Novinskaya, 1965	"
XV	MEL virul.	Polecat (Mustela eversmanni)	Levit, 1959	"
XVI	HSS virul.	Man	Syrgabayeva, 1962	"
XVII	VFG virul.*	Silver-black fox	Galuzo, Krivkova, 1965	"
XVIII	RN virul.	Man	Sabin, 1939	USA

* This strain was at first avirulent, after which one of its substrains became virulent upon passage.

Table 2

Results of CFR study of sera of man with antigens from avirulent strains of Toxoplasma

Sera	Number	Titer	Antigens I	II	III	IV
From patients suspected of having toxoplasmosis (Odessa)	58	1:2	16	17	15	19
		1:12	Neg.	Neg.	Neg.	Neg.
From persons in contact with infected animals (Alma-Ata)	21	1:2	Neg.	8	5	Neg.

Table 3

Results of CFR's between sera from spontaneously infected domestic animals and antigens from virulent strains of Toxoplasma (Titer 1:15)

Sera	Number tested	Antigens V	VI	VII	VIII	IX	X	XI	XII	XIII	XIV	XV	XVI	XVII	XVIII
Sheep	10	2	1	3	3	not tested									
Cattle	22	2	3	1	2	1	2	4	6	3	2	1	2	2	1
Swine	37	2	4	5	5	2	1	1	3	2	3	2	2	2	2
Goats	39	3	4	4	4	4	4	4	4	4	4	4	4	4	4
Rabbits	7	6	6	6	6	5	5	5	6	4	6	5	4	6	6

Table 4

Results of CFR's between sera from artificially infected animals and antigens from virulent strains

Sera	Number of animals	Number of sera (titer 1:15)	Antigens V	VI	VII	VIII	IX	X	XI	XII	XIII	XIV	XV	XVI	XVII	XVIII (control)
Sheep	14	14	1	2	3	2	2	2	2	2	3	3	2	2	1	1
Goats	31	31	6	7	7	7	5	5	4	6	6	1	5	7	4	5
Horse	1	17	17	17	17	17	17	17	17	17	17	17	17	17	17	17
Ass	1	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2
Rabbits	10	10	10	10	10	10	10	10	10	10	10	10	10	10	10	10
Dogs	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2
Urial	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1

Table 5 - Results of individual studies of human blood sera in CFR's with various antigens

Serum No. or patient name	Antigens																	
	Avirulent				Virulent (1:.2)													
	I	II	III	IV	V	VI	VII	VIII	IX	X	XI	XII	XIII	XIV	XV	XVI	XVII	XVIII
Titer 1:2																		
38	++	++	++	++	++	++	++	++	++	++	++	++	++	++	++	++	++	++
	++	++	+	++	++	++	++	++	++	++	++	++	+	++	++	++	+	++
42	++	++	++	++	++	++	++	++	0	0	++	++	++	++	++	++	++	++
	++	++	++	++	++	++	+	+			+	++	+	+	++	++		++
47	0	++	++	++	++	++	++	++	++	++	++	++	++	++	++	++	++	++
		+	+		++	++	++	++		++	+	+	+		++	+	+	++
44	0	++	++	++	++	++	++	++	0	++	++	++	++	++	++	++	++	++
		++			++	++	+	+		++	++	++	++	+	++	+	+	++
49	++	++	++	++	++	++	++	++	++	++	++	++	++	++	++	++	++	++
	++	++	+	+	++	++	++	++	++	++	++	++	++	++	++	++	++	++
50	++	++	++	++	++	++	++	++	++	++	++	++	++	++	++	++	++	++
			+	++	++	++	++	++	++	++	++	++	++	++	++	++	++	++
St-ma	++	++	++	++	++	++	++	++	++	++	++	++	++	++	++	++	++	++
	+		+	+	++	++	++	++	++	++	++	++	++	++	++	++	++	++
45	++	++	++	++	++	++	++	++	++	++	++	++	++	++	++	++	++	++
	++	++	++	++	++	++	++	++	++	++	++	++	++	++	++	++	++	++
Titer 1:6																		
51	++	No	++	++	++	++	++	++	++	++	++	++	++	++	++	++	++	++
		test	+		++	+			+	+	+	+		++	++		+	++
Gr-ko	++	++	No test		++	++	++	++	++	++	++	++	++	++	++	++	++	++
					++	++	+	++	+	++	+		++	++	++	++	+	++
18		No test			++	++	++	++	++	++	++	++	++	++	++	++	++	++
					++	++	+	++	++	++	++	+	+	+	++	+	++	++
Ct-k		"			++	++	++	++	++	++	++	++	++	++	++	++	++	++
					+	++	++	++	+	++	++			+	++	++	+	++
10		"			++	++	++	++	++	++	++	++	++	++	++	++	++	++
					+	++	++	++	+	++	+		+	++	+	++	+	++
P-ko	++	++	No	++	++	++	++	++	++	++	++	++	++	++	++	++	++	++
			test		++	++	+	++	++	++	+	+	++	+	++	++	+	++
K-va	++	No	test	++	++	++	++	++	++	++	++	++	++	++	++	++	++	++
					+	++	++	++	++	+	++	+	+	++	++	++	++	++
P-va	++	++	No	test	++	++	++	++	++	++	++	++	++	++	++	++	++	++
					+	++	+	++	++	+	+	+	+	++	+	+	++	++
32		No test		++	++	++	++	++	++	++	++	++	++	++	++	++	++	++
					++	++	+	+	+	++	+	+	+	+	+	++	++	++
33		"		No	++	++	++	++	++	++	++	++	++	++	++	++	++	++
				test	++	++	+	++	+	++	+	++	+	+	++	++	+	++

Table 6 - Results of individual tests of CFR's between blood sera from spontaneously infected animals and various antigens

Sera from	Number of Sera Tested	Antigens: Avirulent I	II	III	IV	Virulent V	VI	VII	VIII	IX	X	XI	XII	XIII	XIV	XV	XVI	XVII	XVIII (control)
		Alma-Ata											Alma-Ata						
Cattle	1	0	0	0	0	++	+++	+	++	+	+	0	++	++	++	0	+++	++	+
"	1	0	0	0	0	+	++	+	+	+	+	+	++	++	+	0	0	+	+
"	1	0	0	0	0	+	+	+	+	+	+	++	++	+	+	+	+	+	+
"	1	0	0	0	0	+++	+++	+++	+++	+++	+++	+++	+++	+++	+++	+++	+++	+++	+++
"	1	0	0	0	0	+	+	+	+	+	++	++	++	0	+	0	+	+	+
"	16	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Swine	1	0	0	0	0	++	++	++	++	+	+	+	++	++	++	++	++	++	++
"	1	0	0	0	0	+++	+++	+++	+++	+++	+++	+++	+++	+++	+++	+++	+++	+++	+++
"	1	0	0	0	0	+	++	++	++	++	+	+	++	+	++	+	+	+	+
"	31	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Goats	1	0	0	0	0	++	+++	++	++	++	++	++	++	++	++	++	++	++	+++
"	1	0	0	0	0	+++	+++	+++	+++	+++	+++	+++	+++	+++	+++	+++	+++	+++	+++
"	1	0	0	0	0	+++	+++	+++	+++	+++	+++	+++	+++	+++	+++	+++	+++	+++	+++
"	1	0	0	0	0	+++	+++	+++	+++	+++	+++	+++	+++	+++	+++	+++	+++	+++	+++
"	35	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
		Odessa											Odessa						
Rabbits	1	+++	+++	+++	+++	++	++	++	++	++	++	++	+++	+++	++	+++	++	++	+++
					++	++	++	++	++	++	++	-			++		++	++	
	1	++	++	++	++	++	++	++	++	++	++	++	++	++	++	++	++	++	++
		++	++	++	++	++	++	++	++	++	++	++	++	++	++	++	++	++	++
	1	++	++	++	++	++	+++	+++	++	+++	++	++	++	+++	+++	++	++	+++	++
		++	++	++	++							++	++				++		++
	1	++	++	0	++	++	+++	+++	+++	+++	++	+++	++	+++	++	+++	+++	++	++
						++					++							++	++
		++	++	0	++	+++	++	++	+++	+++	+++	+++	+++	++	++	++	++	++	++
	1					++		++	++		++			++	++	++	++	++	++

Table 7

Results of CFR's between sera from artificially infected animals and antigens from virulent strains

Sera from:	Number of sera tested	Antigens V	VI	VII	VIII	IX	X	XI	XII	XIII	XIV	XV	XVI	XVII	XVIII	Note
Horse	17	++	++	++	++	++	++	++	++	++	++	++	++	++	++	Infected
		++	++	++	++	++	++	++	++	++	++	++	++	++	++	with STG
Ass	2	++	++	++	++	++	++	++	++	++	++	++	++	++	++	"
		++	++	++	++	++	++	++	++	++	++	++	++	++	++	
Rabbits	10	++	++	++	++	++	++	++	++	++	++	++	++	++	++	Avir. strain
		++	++	++	++	++	++	++	++	++	++	++	++	++	++	used
Sheep,	1	++	++	++	++	++	++	++	++	++	++	++	++	++	++	"
9141			+	+	+	+	+	+	+	+	+	+	+			
Sheep																
B/No.	4	-	-	-	-	-	-	-	-	-	-	-	-	-	-	Chronically
																sick
Goat	1	++	++	++	++	++	++	++	++	++	++	++	++	++	++	Viru.
B/No.		++	++	++	++	++	++	++	++	++	++	++	++	++	++	strain
Goat	1	++	++	++	++	++	++	++	++	++	++	++	++	++	++	"
3510		++	++	+	++	+	++	+	++	++	++	++	++	+	+	"
G.1029	1	++	++	++	++	++	++	-	++	++	++	++	++	-	++	"
G.1027	1	++	++	++	++	++	++	++	++	++	++	++	++	++	++	"
		+	+	+				+	+	+	+		+	+	+	"
G.1029	1	++	++	++	++	++	++	++	++	++	++	++	++	++	++	"
		++	++	++	++	++	++	++	++	++	++	++	++	++	++	
GoatB/No.	24	-	-	-	-	-	-	-	-	-	-	-	-	-	-Chronically sick	
Dog	1	++	++	++	++	++	++	++	++	++	++	++	++	++	++Infected with vi-	
Woman		++	++	++	++	++	++	++	++	++	++	++	++	++	++rulent strain	
Dog	1	++	++	++	++	++	++	++	++	++	++	++	++	++	++	"
1334		++	++	++	++	++	++	++	++	++	++	++	++	++	++	
Urial	1	++	++	++	++	++	++	++	++	++	++	++	++	++	++Hyperimmunized with	
1074		++	++	++	++	++	++	++	++	++	++	++	++	++	++virulent strain	