Prevalence of *Toxoplasma gondii* Antibodies Among Veterinary College Staff and Students, Iowa State University

SINCE THE INITIAL FINDING and description of Toxoplasma gondii from a North African rodent by Nicolle and Manceaux in 1908 (1), numerous studies have revealed the widespread distribution of Toxoplasma gondii in man. Feldman (2), investigating the problem in 18 population groups ranging from Alaska and Iceland to South America and Australia, observed prevalences ranging from none for Eskimos to 68 percent in Tahiti. Within the United States, studies have indicated prevalences of 4 percent for Navajo Indians (2), 28 percent for Alaskan natives (3), and 17 to 37 percent for several metropolitan areas (2, 4-6).

McCulloch and associates (7) reported the following prevalences among selected population groups in Iowa: wildlife workers, 19 percent; veterinarians, 20 percent in 1962 and 24 percent in 1965; and persons having premarital blood tests, 11 percent.

With the recognized distribution of the parasite in man and animals, increasing attention has been given to the role of animals in the spread of the disease. McCulloch and co-workers (8) found evidence that toxoplasmosis is associated with marked animal and soil contact. They skin tested 775 veterinary and medical students from Iowa, Illinois, and Minnesota during 1960-61. The overall prevalence rate was 17.8 percent; 19.4 percent for veterinary students and 14.9 percent for medical students. Rates for veterinary students at the three schools were: Illinois, 23.0 percent; Minne-

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sota, 20.1 percent; and Iowa State, 17.0 percent. Those students who had spent more than 70 percent of their lifetime on a farm had a significantly higher proportion of positive skin tests than did those who had spent more than 70 percent of their life in a city. Skin test positivity was significantly associated with moderate or marked contact with swine, horses, sheep, cattle, chickens, and turkeys.

Schnurrenberger and associates (9) found significant variations in Ohio between the prevalence of positive skin reactions for students of veterinary medicine (34.0 percent) and medicine (16.7 percent), between farm (35.6 percent) and city (22.3 percent) residents, and between persons with maximum (34.3 percent) and minimal (21.9 percent) animal contact. A possible relationship with consumption of raw eggs was indicated.

In a study of obstetrical patients in Minnesota, Kimball and associates (10) found that people who had spent their lifetime on farms had a significantly higher prevalence (44 percent) than those who had never lived on farms (21 percent). A significant relationship was demonstrated for contact with some farm animals, especially chickens, but not with household pets. Positive dye test reactions were significantly associated with consumption of raw beef.

French and associates (11) found a significantly higher prevalence of positive skin tests for agriculture workers in California than for cattle feed-lot employees or county employees. Discoveries since 1969 have indicated that a coccidia of cats is actually a stage of the life cycle of *T. gondii* and is capable of inducing the disease in man and animals (12-16). Peterson and associates (5), in Seattle, found a significantly greater prevalence of antibodies against *Toxoplasma* in persons who had a pet cat during their lifetime than for people who never had a cat. Sengbusch and Sengbusch (17) also established a possible relationship between toxoplasmosis and cat contact. A prevalence of 18 percent was obtained for 60 veterinarians and their staff from western New York who had almost daily contact with cats, while none of a similar group with no cat contact had positive test reactions. Only 1 of 12 veterinarians in the contact group had antibodies.

Other recent studies have not been able to determine this relationship between cats and toxoplasmosis in man. Comstock and Ganley (6) found a relationship between presence of cattle, horses, and chickens on premises and toxoplasmosis in human beings but found no relationship for the presence of cats and dogs. Rieman and coworkers (18), in checking students and personnel of veterinary colleges in California and Brazil, found a general relationship between prevalence of toxoplasmosis and animal contact but could not specifically implicate cats, other animal species, or food habits. The prevalence for second-year California students was 20 percent compared to 41 percent for Brazilian veterinary students.

The study reported here was an attempt to relate animal contact and other epidemiologic factors to toxoplasmosis in man. It is an adjunct to an epidemiologic study to determine the distribution, importance, and modes of transmission of toxoplasmosis in the closed-animal populations at the Veterinary Medical Research Institute, Iowa State University.

Materials and Methods

Serum samples were obtained from two groups at the College of Veterinary Medicine, Iowa State University: (a) 108 persons from the Veterinary Medical Research Institute (VMRI) and (b) 142 veterinary students. The VMRI group included professional staff, techni-

cians, caretakers, administrative support workers, and student assistants. Their serum samples were taken in the fall of 1972, 1973, and 1974. Also, a limited number of serum samples from a 1968–71 rabies vaccine evaluation program was made available. Serum samples were obtained from 58 of 79 members of the veterinary class of 1975 and 84 of 92 from the class of 1976 during a rabies vaccination program in 1972. The students were in the second and first year of veterinary school, respectively.

Questionnaires were completed by all veterinary students and by 94 of the 108 VMRI personnel. Information solicited included age, sex, primary lifetime residency (farm, village, or city), food habits as related to eating of meat products, the possibility of confirmed or suspected mononucleosis, and contact with various species of domestic and wild animals (little or no, occasional, or marked).

The indirect fluorescent antibody (IFA) test was used for serologic evaluation (19). Antigen slides prepared at the VMRI with the RH strain of *T. gondii* were used with commercial conjugates and control serums. A titer of 1:16 or greater was considered indicative of exposure to *T. gondii*.

Statistical analyses of data were made by the chisquare test and use of the Yates correction factor for small sample size. A probability of 0.05 or less was considered significant.

Results

Serologic evaluation. The initial serum samples from 65 (26.0 percent) of the 250 persons yielded a titer of 1:16 or greater by the IFA test. Of the 108 VMRI personnel 33.3 percent reacted, as did 20.4 percent of the 142 veterinary students. However, there was a marked difference in results within the latter group; the reactive rate was 31.0 percent for the 58 members of the class of 1975 and 13.1 percent for the 84 members of the class of 1976. The prevalence for members of the class of 1976 was significantly lower (P < 0.05) than the 31.0 percent for the class of 1975 or the 30,1 percent for the comparable group of VMRI personnel aged 15–34 years.

Of the 65 seropositive persons, 28 had initial titers of 1:16, and 16 reacted at the 1:32 level. Eight had titers of 1:64, seven had 1:128, four had 1:256, and two had 1:512.

Serum samples were taken at yearly or greater intervals from 55 of the VMRI personnel: 20 submitted 2 samples; 23, 3 samples; 7, 4 samples; and 5, 5 samples. In the multiple-sample group, 13 remained seropositive, 25 remained seronegative, and 17 showed serologic change. Many of the changes in reactivity were minimal and probably not significant; 9 of the 17 had a maximum titer of 1:16 and 3 others had a maximum of 1:32. One conversion and one reversion involved a titer of 1:64, one conversion reached 1:128, and another reached 1:256. The 1:256 conversion was in a 24-year-old female technician who was negative in 1972 and 1973 but converted in 1974 shortly after acquisition of a young cat. A 54-year-old technician was negative in 1968 and 1969 but then had titers of 1:64 in 1972, 1:16 in 1973, and 1:128 in 1974. One technician reverted from 1:32 in 1968 to 1:4 in 1973, then converted to 1:128 in 1974.

Epidemiologic characteristics. No definite relationships between the epidemiologic characteristics examined and the presence of antibodies to T. gondii could be established. No significant differences were noted for sex. age, primary residency, consumption of rare beef or smoked pork products, or suspected mononucleosis (table 1). The tabular data indicated a general, but erratic, increase for prevalence with age. The overall prevalence for the age group 15 to 34 years was 23.7 percent compared to 40.0 percent for persons 35 years or older. With respect to sex, the prevalence rate of 44 percent for the 9 women examined from the class of 1975 was significantly higher (P < 0.05) than the prevalence of 4.8 percent for the 21 women in the class of 1976. Comparison of other characteristics for the two classes also revealed significant differences between the classes for primary lifetime city residency. marked cat contact, marked dog contact, and consumption of little or no rare beef.

Similarly, there appeared to be little relationship between the extent of animal contact and development of serum antibody titers (table 2). For persons with marked contact with cats, the overall prevalence rate was 41.9 percent for women and 18.6 percent for men a nonsignificant difference. However, for the VMRI group only, the rate for women (58.8 percent) was significantly higher (P < 0.05) than the rate for men (14.3 percent).

By occupational groups, the VMRI personnel had the following prevalence rates: animal caretakers (7 of 12), 58.3 percent; technicians (15 of 40), 37.5 percent; professionals (6 of 19), 31.6 percent; student laboratory assistants (3 of 12), 25.0 percent; student animal caretaker assistants (4 of 18), 22.2 percent; and administrative support personnel (1 of 7), 14.3 percent. When men aged 35 years or over in the VMRI sample were considered, the 75 percent prevalence rate for animal caretakers was significantly higher at the P < 0.05 level than the 18.2 percent rate for other men in this age category. Research using cats is not normally carried out at the VMRI. With the exception of a cat maintained for toxoplasmosis research during 2 weeks in 1974, no cats have been used at the VMRI during the past 20 years; the animal caretakers had no contact with that animal. Therefore, the high rate for the animal caretakers may be related to prior background or other unknown factors.

Some of the veterinary students were from other States. Of the 58 members of the class of 1975 from whom blood samples were obtained, 48 were from Iowa and 16 of these (33.3 percent) were seropositive; 10 were from 5 other States, and 2 of these (20.0 percent) were seropositive. All 11 of the positive serums from students of the class of 1976 were from the 64 members of Iowa origin. The 20 out-of-State members, representing 10 States, were negative.

Accidental laboratory infection. I acquired a mild toxoplasmosis infection when a needle, previously used for intraperitoneal inoculation of mice, accidentally pierced my thumb. The inoculate was of swine origin and produced an acute infection in mice with 100 percent mortality in 5 or 6 days.

The infection was devoid of many of the common signs of toxoplasmosis including lymphadenitis. Primary signs included a low-grade temperature elevation persisting intermittently from days 13 to 29, slight respiratory involvement, malaise, and occasional profuse nighttime sweating. The standard triple sulfa-pyrimethamine-folinic acid therapy was initiated on day 30 and continued for 30 days.

Serologic conversion by the IFA test was evident. The results of serologic testing before and shortly after the accident were negative. The titer rose to 1:64 on day 15, 1:256 on day 34, then declined to 1:64 on day 41. This was followed by a slow rise to 1:512 on day 154 and 1:2,048 on day 203. The titer then slowly declined to 1:64 at 32 months' postinfection.

Discussion

Three basic observations on the toxoplasmosis problem can be made from the results of this study:

• *Toxoplasma* antibodies are common in the human population of Iowa.

Table 1. Relationship of toxoplasmosis antibodies to epidemiologic characteristics, excluding animal contact, of personnel from the Veterinary Medical Research Institute (VMRI) and veterinary students of the classes of 1975 and 1976, lowa State University

Characteristics		· .	I.S.U. veterinary students					
	VMRI personnel '		Class of 1975		Class of 1976		Total	
	Number examined	Percent positive	Number examined	Percent positive	Number examined	Percent positive	Number examined	Percent positive
Sex								
Male	60	30.0	49	28.6	63	13.9	172	24.4
Female	48	37.5	9	44.4	21	4.8	78	29.5
Primary residency								
Farm	31	51.9	30	33.3	43	18.6	104	30.8
Village	19	22.2	8	25.0	10	20.0	37	21.6
City	44	36.6	20	30.0	31	3.2	95	23.2
Food habits Ate rare beef:								
Never	30	46.4	16	43.8	27	7.4	73	30.1
Infrequently	35	45.7	25	20.0	41	19.5	101	28.7
Often	29	33.3	17	35.3 [·]	16	6.3	62	25.8
Ate smoked pork products:								,
Never	14	46.2	4	0.0	9	11.1	27	25.9
Infrequently	49	38.3	32	28.1	52	13.5	133	25.6
Often	31	34.6	22	40.9	23	13.0	76	27.6
Mononucleosis								
Diagnosed	8	42.9	6	0.0	2	0.0	16	18.8
Suspected	7	50.0	3	66.7	3	0.0	13	38.5
Unknown	79	37.0	49	32.7	79	13.9	207	26.1
Age group (vears)								
15–19	11	18.2			. 2	0.0	13	15.4
20–24	30	33.3	50	32.0	67	13.4	147	23.8
25–29	21	28.6	7	28.6	13	15.4	41	24.4
30–34	11	36.4	1	0.0	2	0.0	14	28.6
35–39	9	55.5					. 9	55.5
40–44	6	33.3					. 6	33.3
45–49	3	0.0					. 3	0.0
50–54	10	20.0					. 10	20.0
55–59	4	75.0					. 4	75.0
60–64	3	66.7					. 3	66.7

+ Except for sex and age, data available for only 94 VMRI personnel.

Table 2. Relationship of toxoplasmosis antibodies to animal contact of personnel from the Veterinary Medical Research Institute and veterinary students of the classes of 1975 and 1976, Iowa State University

Animal contact and study group	Little or no contact		Occasional contact		Marked contact	
	Number examined	Percent positive	Number examined	Percent positive	Number examined	Percent positive
Cats	54	31.5	108	22.2	74	28.4
VMRI ¹	28	39.3	35	28.6	31	38.7
Class of 1975	8	37.5	32	25.0	18	38.9
Class of 1976	18	16.7	41	14.6	25	8.0
Cat litter	155	27.1	50	22.0	31	29 .0
VMRI	61	36.1	18	27.8	15	40.0
Class of 1975	36	30.6	16	31.3	6	33.3
Class of 1976	58	15.5	16	6.3	10	10.0
)oas	31	38.7	79 [.]	24.1	124	25.0
VMRI	27	40.7	26	26.9	39	38.5
Class of 1975	2	0.0	21	23.8	35	37.1
Class of 1976	2	50.0	32	21.9	50	6.0
wine	105	22.9	61	27.9	70	30.0
VMRI	50	32.0	20	35.0	24	41.7
Class of 1975	15	26.7	19	36.8	24	29.2
Class of 1976	40	10.0	22	13.6	22	18.2
attle	89	25.8	69	24.6	78	28.2
VMRI	52	34.6	23	30.4	19	42.1
Class of 1975	10	20.0	22	36.4	26	30.8
Class of 1976	27	11.1	24	8.3	33	18.2
heep	163	23.9	53	26.4	20	45.0
VMRI	64	31.2	24	37.5	6	66.7
Class of 1975	35	28.6	12	25.0	11	45.5
Class of 1976	64	14.1	17	11.8	3	0.0
orses	119	24.4	77	31.2	40	22.5
VMRI	57	28.1	28	46.4	9	44.4
Class of 1975	19	36.8	22	31.8	17	23.5
Class of 1976	43	14.0	27	14.8	14	7.1
oultry	143	24.5	66	31.8	37	16.2
VMBI	53	35.8	29	37.9	12	25.0
Class of 1975	30	23.3	21	38.1	17	17.6
Class of 1976	60	15.0	16	12.5	8	0.0
Vild animals	138	29.0	81	22.2	17·	23.5
VMRI	65	33.8	24	37.5	5	40.0
Class of 1975	29	34.2	24	25.0	5	40.0
	44	40.0	L-7	20.0	ž	-0.0

Data available for only 94 VMRI personnel.

• The prevalence rates can vary significantly for basically similar population groups.

• The toxoplasmosis problem is complex and probably involves a multiplicity of factors, some of which may be still unrecognized.

The overall prevalence of 26.0 percent for Toxo-plasma antibodies obtained in this study, when considered with the data of McCulloch and associates (8) for selected population groups a decade earlier, indicates that toxoplasmosis is widespread in the human population of Iowa. Although the sample in this study also was selected, the basic lifetime residencies of the Iowa-reared students and many VMRI personnel were

distributed throughout the State. There also was a broad diversity in background factors such as food habits, rural verus urban residency, and degree of animal contact. The primary biases would be age and an inherent interest in animals. However, there was no current work- or school-related contact with living cats. Serum samples had been taken from the students before they were exposed to clinical medicine.

The finding of a significant difference between the prevalence rates of 31.0 percent for the class of 1975 and 13.1 percent for the class of 1976 was surprising because previous studies in the north-central United States had indicated no differences in rates between

veterinary classes (8.9). The epidemiologic characteristics for the two classes were quite similar, as shown in the tables. Men comprised 84.5 percent of the 1975 class sample and 75 percent of the 1976 sample. About 51 percent of each group had farm backgrounds and about 35 percent had primary residencies in cities. Marked exposure to cats was indicated for 31.0 percent of the class of 1975 and 29.7 percent of the class of 1976. Most of the other characteristics examined for the two classes also were similar. The primary difference noted was in regard to consumption of meat products-29.3 percent of the participants from the class of 1975 reporting eating rare beef often as compared to 19.0 percent of those from the class of 1976. Similarly, 37.9 percent of the class of 1975 ate smoked pork products often as compared to only 27.4 percent of the class of 1976.

With the similarity in characteristics, it is difficult to explain the significant differences for overall rates for the classes, for women class members, and for such characteristics as primary lifetime residence and marked cat and dog contact. The members of the class of 1975 had about 1 more year of exposure to the Ames and Iowa State University environment than the class of 1976. Whether this could account for much, if any, of the significant difference in rates is questionable. A more refined, longer-term study would be necessary to give a clearer determination of the primary factors affecting these prevalence rates.

The difficulty in making comparisons between the two academic classes only stresses the difficulty in pinpointing specific factors influencing rates in the overall study. Although cat contact (5, 17) and ingestion of improperly prepared meats are generally considered to be the most logical modes for dissemination of the disease to man, this study did not indicate any definitive relationships between *Toxoplasma* antibodies in the study group and these epidemiologic factors.

Several studies (6, 8, 9, 18) have indicated a potential relationship between prevalence and degree of contact with large animals. *T. gondii* is common in domestic animals of Iowa. McCulloch and associates (20)tested 105 swine serum samples for *T. gondii* antibodies during 1961-63 and found 6.7 percent with a titer of 1:16 or greater by the Sabin-Feldman dye (SF) test. Followup of the reacting herds gave 40 percent reactors, indicating herd involvement. Two other swine herds had 8.7 percent and 85.7 percent reactors, respectively. Two groups of sheep serum samples had prevalences ranging from 12.5 to 36.8 percent. No reactors were found in 129 cattle and 32 horses tested. The prevalence was 16 percent for 112 dogs and 4.8 percent for 21 cats.

Even with the distribution of T. gondii in Iowa animals, the only indication of a relationship of prevalence and large-animal contact was in the VMRI group; a significant difference was noted between animal caretakers and other men 35 years of age or older. However, the caretaker-versus-others relationship was not valid in the overall study.

The inconclusiveness of this study, as well as others, indicates the possibility of one or more potentially important, but yet unrecognized, modes of transmission. The overall problem can be defined only with more intense studies. Instead of a single serum sample and questionnaire type study such as this one, there is a need for longitudinal, in-depth studies with frequent taking of serum samples and continual epidemiologic monitoring and followup. The studies should include children because they are the most susceptible age group. Only through such studies can the modes of transmission be defined and adequate control measures formulated.

Although this study does not give evidence indicating a role for pork in the spread of Toxoplasma, certain comments should be made regarding the potential of pork as a vehicle of transmission. The current prevalence of Trichinella spiralis in U.S. swine is 0.125 percent (21); about 0.06 percent in Iowa. In some of my unpublished studies on toxoplasmosis in slaughter swine of Iowa, IFA testing revealed a prevalence of 23.0 percent. T. gondii were isolated from 15 of 100 swine diaphragms by passage in mice; with the use of this finding as a minimal base, T. gondii is about 250 times as prevalent as T. spiralis in Iowa swine. Thus, when this relationship is considered along with an ageadjusted prevalence of 2.2 percent for T. spiralis in the U.S. population and an estimated yearly acquisition of the parasite by 150.000-300.000 people (22), it is difficult not to assign a major role in the transmission of T. gondii to pork. In the study reported here, 30.4 percent of the persons ate smoked pork sausage often, and 53.2 percent ate it occasionally. No information was requested regarding consumption of fresh pork products.

One facet pertaining to the accidental infection that I incurred leads to speculation. The strain of T. gondii that initiated the infection, although originally isolated from a field pig, had become highly pathogenic to mice during passage for 26 months. The strain was passaged into 5 swine approximately 3 months after my infection. One pig died from acute toxoplasmosis 11 days after infection; the other four developed mild symptoms and a marked serologic response with peak IFA titers of 1:4,096, 1:2,048, and 1:1,024, respectively. The titers persisted at moderate levels until the animals were sacrificed-two at 5 months postinfection and the other two at 7 months. Passage of 10 tissues from each pig into mice gave negative results. The same finding has been reported for the RH strain in swine (23,24). Therefore, the possibility of either nonencystment or tissue clearance also may exist for laboratory acquired human infections induced with acute strains of the parasite. Thus, the hazard may be related to the acute phase of the disease rather than any long-term effects.

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SYNOPSIS

ZIMMERMANN, W. J. (Iowa State University of Science and Technology): Prevalence of Toxoplasma gondii antibodies among veterinary college staff and students, Iowa State University. Public Health Reports, Vol. 91, November–December, 1976, pp. 526–532.

Serum samples obtained from two groups within the College of Veterinary Medicine, Iowa State University, were examined for *Toxoplasma gondii* antibodies. Serum samples from 65 (26.0 percent) of the 250 persons examined yielded a titer of 1:16 or greater by the indirect fluorescent antibody test. A prevalence of 33.3 percent was obtained for 108 Veterinary Medical Research Institute (VMRI) personnel. For 142 veterinary students, the reactive rate was 20.4 percent. There was a marked difference of results within the student group—the reactive rate of 31.0 percent of 58 members of the class of 1975 was significantly higher (P < 0.05) than the 13.1 percent for 84 members of the class of 1976. Studies on multiple serum samples were carried out on 55 VMRI personnel: 17 showed a change in serologic reactivity but only 5 had titers of 1:64 or greater.

No definitive relationships could be determined between the epidemiologic characteristics examined and the presence of *T. gondii* antibodies. Characteristics examined included age, sex, primary residency, food habits (eating of rare beef and smoked pork products), and extent of animal contact.