Evaluation of the Suessenguth-Kline Test for Trichinosis

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TRICHINOSIS affects an estimated 16 per cent of the population of the United States, according to examinations of muscle tissue for trichinae larvae (*1*). The majority of infections are subclinical, but even those with clinical symptoms frequently escape recognition. The high prevalence, the manner of transmission, the frequent severity, and occasional fatal outcome of the disease make trichinosis a matter of public health concern.

Clinical diagnosis of this disease is difficult (*2*). Some laboratory examinations are helpful only if positive, while others show conflicting results (*1, 3–5*). A simple and rapid flocculation slide test for the disease was reported by Suessenguth and Kline (*6*) and subsequently improved (*7*). Because of encouraging results in the previous studies, it was decided to determine the value of the test for public health and diagnostic laboratory purposes. The test was studied for simplicity of performance, for reproducibility, for sensitivity and specificity, and in comparison with one other immunological method of testing.

Simplicity of Performance

It had been found in an earlier study (*6*) that an alkaline aqueous extract of freeze-dried trichinae larvae possesses the property of coating cholesterol crystals so that, when such coated crystals are dispersed in physiological saline, an antigen emulsion is formed. When used in the slide test, the antigen emulsion is sensitive and specific for trichinosis. The freeze-dry method has been found invariably satisfactory for drying larvae. Properly stored larvae used over a period of 9 years showed no loss of sensitivity or specificity when compared with recently prepared larvae.

In performing the test, 0.5 cc. of the serum to be tested is placed on a ringed slide, one capillary drop of the antigen emulsion is added, and the mixture is rotated at 120 r.p.m. for 4 minutes. The paraffin wall of the ring effectively retains the ingredients. Results are read microscopically by the degree of flocculation of the coated crystals. The technique is essentially that of the Kline test for syphilis (*8*).

The test method is simple and rapid. It does not require highly trained personnel for performance. When refrigerated, the antigen emulsion is satisfactory for a period of at least 8 months.

Reproducibility

The reproducibility of the test was determined by examining duplicate portions of 1,216 serum specimens in two different laboratories. The specimens were from suspected trichinosis cases and from random samples of blood sub-

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mitted for routine tests for syphilis. The tests were performed over a period of 3 years; thus, several lots of antigen emulsion were used.

Each serum specimen was tested in both the division of laboratories of the Ohio Department of Health, Columbus, and the department of laboratories, Mt. Sinai Hospital, Cleveland. When quantity of the specimen permitted, the titer of each positive serum also was determined in duplicate.

The results of the study of reproducibility of the test showed excellent agreement (table 1). The duplicate tests of 97.1 percent of the specimens agreed completely (96 positive, 44 weakly positive, and 1,041 negative in both), and 1.1 percent showed partial agreement (positive in one laboratory and weakly positive in the other).

Quantity permitted duplicate titration of 42 positive serums. We considered 38 of these in agreement since they showed no more than a 1-tube difference. The complete results of duplicate titration of these serums were as follows:

	Number of serums
Same titer	23
1-tube difference	15
2-tube difference	4
More than 2-tube difference	0
Total	42

Sensitivity and Specificity

While simplicity of performance and reproducibility of a test are important, sensitivity and specificity are the factors determining its usefulness in medical practice. By questionnaires and investigations of trichinosis epidemics, pertinent clinical and laboratory data were obtained for the evaluation of positive test results (sensitivity) and negative test results (specificity).

Table 1. Comparison of results of duplicate tests for trichinosis on 1,216 serum specimens using the Suessenguth-Kline flocculation slide test

Reactions at Mt. Sinai laboratory	Reactions at Ohio Department of Health laboratory		
	Positive	Weakly positive	Negative
Positive	96	9	2
Weakly positive	4	44	5
Negative	0	15	1,041

The division of laboratories of the Ohio Department of Health tested 306 serum specimens for this portion of the study. These specimens were taken from patients who either showed some clinical symptoms, had presumptive laboratory findings, or were suspected of having trichinosis because they were members of family groups suffering epidemic trichinosis. Of the 306 specimens tested, 108 (35.3 percent) showed some degree of positivity, and 198 (64.7 percent) gave negative test results.

From questionnaires to attending physicians and investigations of epidemics, data were available for analysis of 62 cases for both sensitivity and specificity. The test had shown some degree of positivity for 27 and negative results for 35 of these 62 cases.

Of the 27 cases showing positivity, 25 were diagnosed cases of trichinosis. The 27 cases giving positive results on the test are described below:

	Number positive
Diagnosed as trichinosis	25
Larvae in meat ingested	17
Positive clinical findings and eosinophilia	8
Diagnosed as other than trichinosis	2

One of the two cases not diagnosed as trichinosis was diagnosed as *Strongyloides stercoralis* infection. *S. stercoralis* was found in duodenal drainage; there was an eosinophilia of 50 percent; and the flocculation slide test was weakly positive. This was probably a nonspecific, weakly positive reaction. The other case not diagnosed as trichinosis was given a final diagnosis of "chronic brain syndrome associated with convulsive disorder," although the diagnosis is questionable considering the history and laboratory findings. The patient was a cook and had had trichinosis in 1946; the eosinophil count was 1.2 percent; a skin test for trichinosis was positive; and a complement fixation test was negative. In this case the reaction may or may not have been nonspecific.

None of the 35 cases which had shown nega-

Table 2. Comparison of results of testing 122 [1] serum specimens for trichinosis by both S-K flocculation slide and complement fixation methods

Reactions to S-K flocculation slide test	Reactions to complement fixation test		
	Positive	Weakly positive	Negative
Positive	14	0	21
Weakly positive	1	0	6
Negative	1	0	72

[1] 7 specimens were anticomplementary in the complement fixation test. 2 of these were negative in the slide test, 1 was doubtful, and 4 were positive.

tive test results was diagnosed as trichinosis. In two of these cases there had been exposure to trichinous meat, but clinical symptoms were not found. Pertinent data on these cases are summarized below:

	Number of cases
Muscle pains	11
Vague or positive clinical findings	10
Eosinophilia	5
Had trichinosis 3–10 years before	3
Mental symptoms	2
Other parasitic infection (1 *Leptospira;* 1 *Taenia saginata*)	2
Exposed to trichinous meat; negative clinically	2
Total	35

Although the number of suspected cases on which data were available for analysis was not large, results of this analysis corroborate the high degree of sensitivity and specificity previously reported (*6*) for the S-K flocculation slide test for trichinosis.

Comparison With CF Test

To compare results of the S-K flocculation slide test with those of another immunological test method, 122 specimens were examined by both the flocculation slide and the complement fixation tests. Portions of the specimens were sent to the Communicable Disease Center, Public Health Service, for the complement fixation tests.

Results of the two tests are compared in table 2. Seven specimens were anticomplementary by the complement fixation technique; 87 (71.4 percent) showed complete or partial agreement; and 28 specimens (22.9 percent) showed complete or partial disagreement.

Since disagreement was largely between positive or weakly positive reactions in the slide test and negative reactions in the complement fixation test, it was decided to determine whether test reactions were specific or nonspecific by examining case histories.

Case histories of 57 patients, accounting for 78 of the 122 specimens (for some cases more than one specimen was examined), were available for study. Table 3 compares test results for these 57 cases by type of case as determined by the diagnoses reported in the case histories.

Both tests gave negative results for 30 of the 31 cases diagnosed as diseases other than trichinosis; the remaining case was negative by flocculation slide and anticomplementary by complement fixation. The other 26 of the 57 were cases of diagnosed trichinosis—6 sporadic and 20 epidemic. Of these 26 trichinosis cases, 11 were positive and 2 were negative in both tests, 2 were positive in the slide test and anticomplementary in the complement fixation test, and 11 were positive in the slide test but negative in the complement fixation test.

Table 3. Comparison of reactions to S-K flocculation slide and complement fixation tests for trichinosis of serum specimens representing 57 clinically diagnosed cases

Type of case and reaction to S-K flocculation slide test	Reaction to complement fixation test		
	Positive	Negative	Anticomplementary
Other than trichinosis (31):			
Positive	0	0	0
Negative	0	30	1
Trichinosis—sporadic (6):			
Positive	3	2	1
Negative	0	0	0
Trichinosis—epidemic (20):			
Positive	[1] 8	[2] 9	1
Negative	0	2	0

[1] Specimens from 4 cases were negative 1 month and positive subsequently on the complement fixation test.
[2] Specimens from 2 cases were negative 1 month and positive subsequently on the flocculation slide test.

On the basis of this comparison of test results with case data, it may be concluded that the S-K flocculation slide test is more sensitive for trichinosis than the complement fixation test. An additional advantage of this flocculation slide test is the absence of anticomplementary reactions.

Further studies of the S-K test and its antigen are in order, together with additional comparison with complement fixation tests and also with a recently described flocculation test (*9*) utilizing bentonite particles.

Summary

Because of frequent difficulty in clinical and laboratory diagnosis of trichinosis, there is need for a simple, rapid, and reliable diagnostic test for the disease. The Suessenguth-Kline flocculation slide test for trichinosis is simple and rapid. An antigen emulsion made of cholesterol crystals coated with an alkaline extract of lyophilized trichinae larvae is mixed on a slide with the serum to be tested and rotated for 4 minutes. The result is read microscopically by the degree of flocculation of the coated crystals.

The reproducibility of the flocculation slide test was shown by the 97.1 percent complete agreement and 1.1 percent partial agreement of the results obtained in two laboratories where duplicate tests were performed on 1,216 serum specimens.

An analysis of 62 cases showed the test to have a high degree of sensitivity and specificity. Of 27 positive test results, 25 were for cases diagnosed as trichinosis; for 1 of the other 2 cases, the diagnosis can be regarded as inconclusive. Of 35 negative test results, none were for cases diagnosed as trichinosis; although there had been definite exposure to trichinous meat in 2 cases, no clinical symptoms of the disease were evident.

In a comparison of the S-K flocculation slide test with the complement fixation test, the flocculation slide test showed greater sensitivity. It gave positive results for 11 diagnosed cases of trichinosis for which the complement fixation method gave negative results.

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