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***Brief Communication***

# A NEW TECHNIQUE FOR DEMONSTRATION OF TRICHINELLA SPIRALIS LARVAE IN SUSPENSIONS OF DIGESTED MUSCLE TISSUE

Up to now, a sedimentation technique has been used for demonstration of Trichinella spiralis larvae released by artificial digestion (*Henriksen* 1973, *Skovgaard* 1975, *Thomsen* 1976, 1977, and others).

The following alternative method is based on visual observation of larvae retained and subsequently stained on disposable sieves.

*Equipment* (cf. Fig. 1, I—III)

Sieve 1 ($S_1$), made by fastening a 7—8.5 cm wide, circular piece of nylon netting, mesh width 350 µ between 2 6.5 cm wide, slightly tapering rings (f. inst. the cut-off brims of 2 disposable drinking cups) and sealing the slit between the inner ring and the netting with paraffin wax (p).

Sieve 2 ($S_2$), made in the same way as Sieve 1, using a nylon netting, mesh width 20 µ.

Funnel 1 ($F_1$), an ordinary glass funnel.

Funnel 2 ($F_2$), a disposable drinking cup with the bottom cut off.

Four Petri dishes: A, with iodine solution (2 g J + 4 g KJ in 100 ml $H_2O$).
B, — saturated solution of sodium thiosulfate.
C, — 5—7 layers of filter paper.
D, — 1—2 — — — —

*Procedure* (cf. Fig. 1, II—III)

$S_1$ is placed in $F_1$ and $S_2$ fixed just below by means of $F_2$. The digested suspension is passed through $S_1$ and $F_1$ down into $S_2$. The sieving process through $S_2$ may be accelerated by tapping gently on the side of $F_2$. When the sieving is completed the sieves

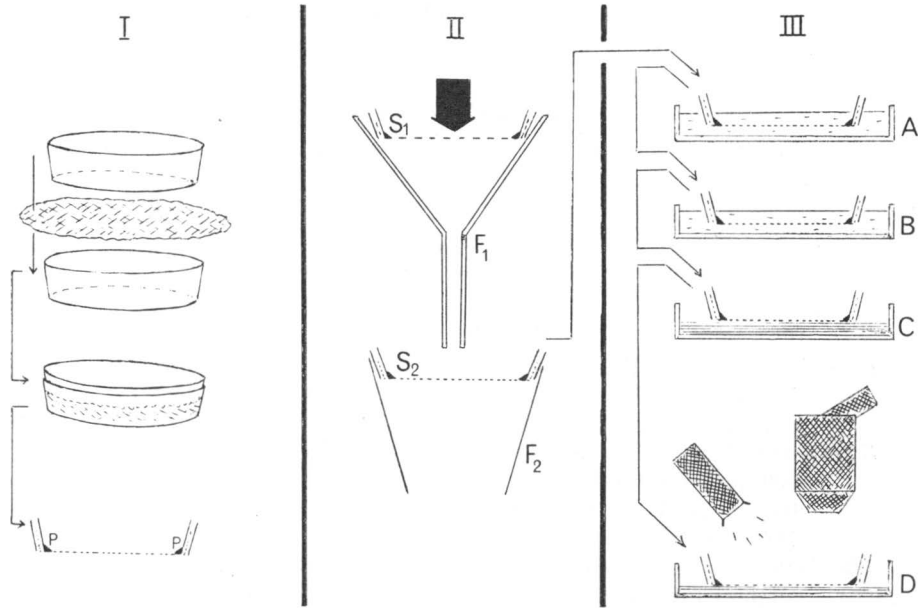


**Figure 1.** Technical equipment and procedure (schematically).

are rinsed with 50—100 ml of water. $S_2$ is then successively transferred to dish A (3 min., for staining), dish B (10 sec., for decoloration), and dish C (20 sec., for removal of excess fluid). Finally, $S_2$ is placed in dish D and examined in a stereo-microscope under incident light. Larvae of Trichinella spiralis are clearly distinguishable, presenting themselves as red-brown nematodes on a bright background.

When known numbers of larvae were added to digested suspensions of muscle tissue from trichina-free swine, a recovery rate of 100 % was obtained in 10 successive trials with the method described. This means that all the larvae added to the suspensions had passed $S_1$ but been retained on $S_2$. The suspensions used in the trials were produced by digestion of 25 g of minced muscle tissue in a Stomacher blender (*Thomsen* 1976). Digestion temperature: 38—42°C. Digestion fluid: 10 g pepsin (1:3000) + 10 ml conc. HCl in 1000 ml $H_2O$. Ratio of muscle tissue (g) to digestion fluid (ml) was 1:14.

The described method is apparently suitable for detection of larvae of Trichinella spiralis in suspensions obtained by digestion

of muscle samples from slaughter pigs. The applicability of the technique for examination of muscle samples from other species of animal remains to be tested.

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