

Differentiation of *Trichinella spiralis spiralis* and *Trichinella spiralis nativa* Based on Resistance to Low Temperature Refrigeration

H.J. Smith*

ABSTRACT

A refrigeration technique to differentiate the subspecies, *Trichinella spiralis spiralis* and *T. spiralis nativa* is described. *Trichinella spiralis spiralis* trichinae in musculature do not survive 48 hours post-refrigeration at -32°C while *T. spiralis nativa* will survive 72 hours and longer at the same temperature.

Key words: Trichinosis, *Trichinella spiralis spiralis*, *Trichinella spiralis nativa*, subspecies differentiation, low temperature refrigeration, resistance.

RÉSUMÉ

L'auteur décrit une technique destinée à différencier les sous-espèces *Trichinella spiralis spiralis* et *T. spiralis nativa*. Les larves de la première ne survivent pas 48 heures dans la musculature, après la réfrigération à -32°C, tandis que celles de la deuxième survivent 72 heures ou plus, à la même température.

Mots clés: trichinose, *Trichinella spiralis spiralis*, *Trichinella spiralis nativa*, différenciation des sous-espèces, réfrigération à basse température, résistance.

In Canada, trichinosis is known to exist in two forms; sylvatic tri-

chinososis in arctic mammals and urban trichinosis in rats with extension to swine in temperate regions. Recently, there have been reports (1, 2, 3, 4, 5, 6) which indicate that trichinae found in arctic mammals survive low temperatures for long periods of time as opposed to trichinae found in swine (7, 8).

Since 1980, the author has been carrying out comparative studies on two strains of *Trichinella spiralis* isolated from animals in Canada. To date, five isolates of the urban subspecies *T. spiralis spiralis* and four isolates of the arctic subspecies *T. spiralis nativa* have been compared. The *T. spiralis spiralis* isolates were all from porcine field cases in Nova Scotia while the *T. spiralis nativa* isolates were from a wolf in Labrador, a wolf in Alberta, a polar bear in Manitoba and a black bear in Quebec.

During refrigeration trials at low temperatures, it was observed that viable larvae were still present when musculature, containing *T. spiralis nativa* trichinae, was digested in a 0.6% pepsin-0.8% HCl mixture three days post-refrigeration at -32°C. On the other hand, viable larvae never were recovered at 48 h post-refrigeration from musculature containing *T. spiralis spiralis* trichinae held at the same temperature. These observations suggested that resistance of trichinae in musculature to refrigeration at -32°C could be used as a simple and rapid method to differentiate between *T. spiralis*

spiralis and *T. spiralis nativa*.

The technique developed consists of placing three samples of musculature of equal size (usually 10 to 20 g depending upon number of larvae/g of tissue) in Whirl-Pak polyethylene bags (Canadian Laboratory Supplies Ltd., Montreal, Quebec) and refrigerating at -32°C. At 24 hour intervals, a package is removed, thawed at room temperature and digested as previously outlined (8). If viable larvae are recovered after 72 hours refrigeration at -32°C, they are identified as *T. spiralis nativa*. If viable larvae are not recovered or recovered only after 24 h refrigeration, they are identified as *T. spiralis spiralis*. It should be noted that larvae which are killed by refrigeration are also digested. If samples are examined prior to completion of digestion, partially digested larvae (ghosts) occasionally may be observed.

This test has proved to be a valuable technique enabling a rapid, initial identification of the subspecies of *Trichinella spiralis* in musculature from field cases. The technique has also been applied to single and/or composite muscle (masseter, diaphragm, intercostal, abdominal, psoas, adductor, etc.) samples from foxes, black bear, ferrets and rats infected experimentally with *T. spiralis spiralis* and *T. spiralis nativa* on nine and 15 occasions respectively with comparable results. It is presented so that others working in this field may find it useful in their investigations as well.

*Agriculture Canada, Animal Pathology Laboratory, P.O. Box 1410, Sackville, New Brunswick E0A 3C0.

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