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# A RAPID TECHNIQUE FOR IDENTIFICATION OF TAENIOID CESTODES USING UNSTAINED SCOLICES

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A rapid technique for identification of three taenioids from bobcats, based on hook morphology, using a simple system of microprojection and unstained scolices is described. Three species of *Taenia* from bobcats of Nebraska were collected. The rostella were removed; placed between two microscope slides; flooded with fixative, 70% ethanol, or tapwater; and pressed by means of two clamps to spread the hooks. The wet mount was then placed on the stage of a simple microprojector equipped with a calibrated ocular micrometer. The hooks were drawn and measured simultaneously.

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#### INTRODUCTION

Helminthological surveys and epidemiological studies of commercially important fur-bearers frequently involve frozen and poorly preserved carcasses. The helminths may be macerated and identification based on conventionally stained and cleared specimens is difficult or impossible. Riser (1956) identified taenioid cestodes from felids based on rostellar hooks, and Verster (1969) revised the genus *Taenia* Linnaeus, 1758, finding hook number and size to be reliable taxonomic criteria. A technique has been developed for rapid identification of taenioid species of bobcats.

#### MATERIALS AND METHODS

During the 1977 and 1978 trapping seasons, carcasses of 76 bobcats (*Lynx rufus* Schreber) were obtained from furbuyers, taxidermists, trappers, and game biologists in Nebraska. The carcasses had been frozen and thawed repeatedly between capture and examination. More than one taenioid species was found in the intestines, but the specimens were

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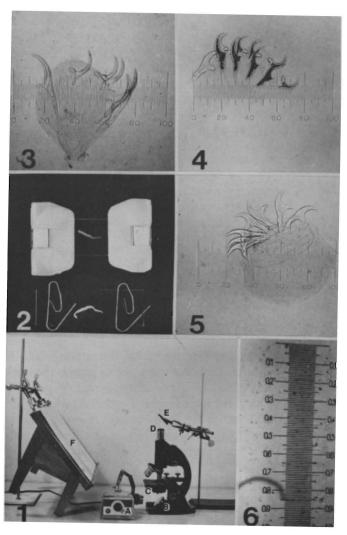
usually macerated and nuclear stains yielded unsatisfactory preparations.

The strobilae were removed from the intestines and disentangled in water. Care was taken not to disturb the rostellum while large pieces of debris were removed. Worms to be permanently mounted for a reference collection were fixed in AFA or 10% formalin for several hours. The rostellum was then removed from the scolex, positioned dorsoventrally, laterally, or in an en face view between two microscope slides and flooded with fixative, 70% ethanol, or tapwater. Two spring clamps made from paper clips or snap clamps from garment hangers were attached to both slides on opposite sides of the rostellum (Fig. 2) and pressure was adjusted so that the hooks were separated but not crushed. The slide was then placed on the stage of a microprojector. For investigators without access to a conventional microprojector, an adequate system (Fig. 1) may be devised using a light source (A), microscope mirror (B), substage condenser (C), calibrated eyepiece (D), reflecting mirror (E), and drawing table (F).

When the system is properly aligned, images of the hooks and a calibrated ocular micrometer are projected onto the drawing table. The hooks can be drawn and measured simultaneously by rotating the ocular micrometer and adjusting the slide with a mechanical stage. Hooks can be drawn, the specimen removed, and the paper rotated on the drawing table while measuring with the ocular micrometer; or the hooks and scale can be drawn and measurements recorded later.

#### RESULTS AND DISCUSSION

Figures 3-5 illustrate the rostella of taenioids found in the intestines of bobcats collected in Nebraska. Figures 3-6



FIGURES 1-6. Equipment and specimens of *Taenia* identified using the rapid technique. 1. Simple microprojector: (A) illuminator, (B) microscope mirror, (C) substage condenser, (D) calibrated eyepiece, (E) reflecting mirror, and (F) drawing table. 2. View of slides with scolex and both spring clamps and snap clamps in place. 3. *En face* view of *T. macrocystis* with ocular micrometer scale superimposed. 4. *En face* view of *T. pisiformis* with ocular micrometer scale superimposed. 5. *En face* view of *T. rileyi* with ocular micrometer scale superimposed. 6. Stage micrometer with ocular micrometer superimposed and photographed at the same angle and magification as figures 3-5.

were taken with a 35 mm, slr camera mounted directly above and at the same angle as the reflecting mirror. The scale present in the three figures is that of the ocular micrometer; each unit being equivalent to 73  $\mu$ m. The image of a stage

micrometer with the ocular micrometer superimposed is shown in figure 6.

In revision of the genus *Taenia*, Verster (1969) found that variations in the size of the cirrus pouch, primary branches of the uterus, size of the eggs, number of testes, shape and size of the individual strobilae, and the presence or absence of a neck region are subject to intraspecific variation and in some cases, artifacts of fixation. She concluded that the number and morphology of the rostellar hooks are reliable taxonomic characters and that other characters should be used in conjunction with hooks when data on hooks alone are not definitive. The taenioids from Nebraska bobcats possessed three distinctive sets of rostellar hooks and were identified as *Taenia macrocystis* Diesing, 1850, *T. pisiformis* (Bloch, 1780) Gmelin, 1790, and *T. rileyi* Loewen, 1929.

Although Verster (1969) stated that the number of hooks was a reliable character, macerated material frequently lacks the total number of hooks. Hook morphology is also a reliable taxonomic character. Rostella with hooks missing provide better views of the hooks because there is greater room to spread and less overlap.

The microprojector is preferred to a microscope equipped with a camera lucida because the full intensity of the illuminator may be used to penetrate the tissue.

Rapid identification based on rostellar hooks may be made either by mounting the rostella in water as soon as they are obtained from the host or by taking them from any preservative. Identical results were obtained by using either fresh or preserved specimens. The addition of a small amount of glycerine to the 70% ethanol on the slide will prevent dessication from evaporation. Specimens to be mounted permanently should be prepared according to usual procedures before mounting them in the final mounting medium.

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