## Second Instar (Overwintering) Larvae

•-	Three methods are used to evaluate overwintering spruce budworm populations: (1) Soak out procedure, which involves washing foliage with hot sodium hydroxide (Miller and McDougall, 1968; Miller et al, 1971; Sanders 1980), (2) Forced emergencepaper cone method (Sanders, 1980), and (3) Forced emergenceenclosed box method (Miller, 1958). Because mean-variance relationships for determining numbers of sample branches are similar for each of the three methods described, a table giving required number of branch samples based on numbers of larvae found is included at the end of this section of overwintering larval survey techniques (Table 8). Likewise, a table predicting infestation level and
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## **METHOD 1: Soak-out Procedure**

Equipment Needed:

1. **Field:** Extension pole pruners, tape measure, heavy paper sacks to hold branch samples, hand pruners, marking pen.

- 2. Lab: Refrigerator or cool storage space (<50°F), data sheets
  - a. For washing foliage: 10-liter plastic pails, 400 ml collecting jars, 2 sets of tags that indicate plot, tree, and branch number (so that washing pail and collecting jar can be tagged),

C).

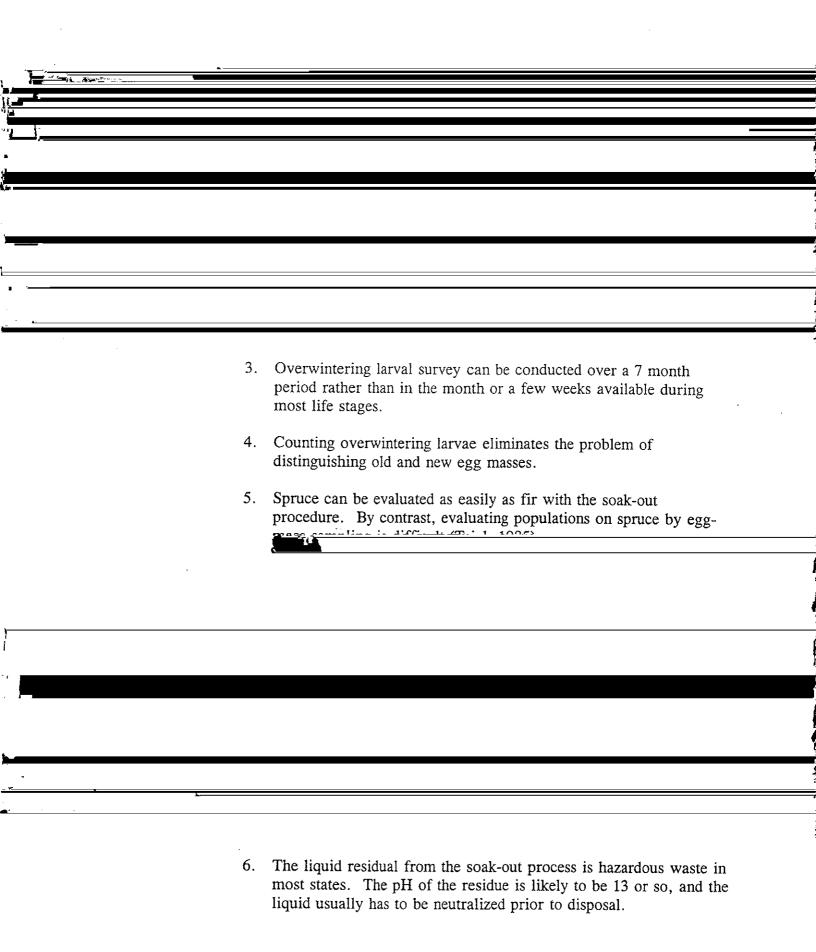
- b. For processing the foliage: large sink or 90 x 150 x 9 cm deep tray, preferably with a corrugated bottom, fitted to a sewer outlet, two interlocking sieves (soil sieves)--top sieve should be about 0.8 mm mesh, bottom sieve 0.25 mm mesh, wire basket made of hardware cloth, 5 mm mesh, with a "false" bottom (wire basket should fit inside a 40 cm x 20 cm tub or container).
- c. For separating larvae from debris: 500 ml separating funnel, hexane.

	Spruce Budworni-Overwintering Larvae Survey - Soak Out Procedure
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	balsam fir, red/black spruce (they hybridize in New Brunswick) or white spruce. Where fir and spruce are both abundant, fir is selected as the indicator species (L. Hartling, pers. comm.). Though whole-branch samples are taken by workers in some regions, others standardize sampling units as 75-cm branch tips
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	75-cm branch tip correlate very well to counts expressed per 10 sq.
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	b.	Processing the foliage: Pour the liquid contents of the pail
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		Pour the remaining contents of the pail into the wire basket.  Rinse the pail and pour the rinse through the sieves. With a
		flexible hose, wash the foliage thoroughly in the wire basket
		inside the tub and discard. Pour the contents of the tub  through the sieves, being careful not to overflow the sieves
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		Ringe the fish and nour through the ciaves. Wash the contents
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Spruce Budworm-Overwintering Larvae Survey - Soak Out Procedure larvae could be missed when one is checking under the microscope. Addition of methyl blue helps in detecting the larvae. It is also helpful to have the filter paper gridded to a

bodies. Note: Careless handling of foliage samples can result



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