December 2009 Adapted By Allison Kanoti, Maine Forest Service

These guidelines have been adapted for use in Northern New England from guidelines provided by Mausel *et al.*, and have been amended to include information for sampling for *Sasajiscymnus tsugae*. *S. tsugae* may be referred to as St and *Laricobius nigrinus* may be referred to as Ln.

Season to Sample

Note: Adjust sampling days to avoid heavy wind, heavy rain, or temperatures below freezing.

To sample species simultaneously plan field work for late April and late October

Laricobius nigrinus

- First sampling fall following release (spring_y release 1st sample fall_y, fall_y release 1st sample fall_{y+1}.
- Mid to late Oct.(after aestivation broken), ideally, if negative results in Oct. resample in mid Nov.
- Mar./Apr. (ovisacs 2-3 mm dia.), record larvae and adults.

Sasajiscymnus tsugae

• Apr. through Oct. (Note: in ME we have retrieved adults when sampling for Ln in early November).

Post-release Monitoring Data Form

These data are to be collected each time the site is monitored in the fall and spring. Fill out as completely as possible. Enter zero if no beetles are found.

Office Information (You may want to create a mail merge to access your database for this information.)

- 1. **Release Location:** Indicate the site name and the state where the release was made.
- 2. First Release dates: first release of each species monitored at the date of sampling.
- 3. **UTM:** Set the datum to NAD83, record the zone, northing, and easting at the center of the release trees.
- 4. **Monitoring Contact Information:** Name, phone number, organization, email for the person collecting the post-release information.

Field Information

- 5. Curr. Date: date of sample
- 6. **Current weather:** Temperature, wind speed (none, low, medium, high), precipitation (none/light rain/heavy rain/snow), cloud cover (none, low, medium, high).
- 7. **Stand Condition:** Record overall hemlock health, overall HWA densities, and note any changes or disturbances to the site.

Health:

Good: foliage has normal color and density (transparency), overall appearance is good.

Fair: foliage somewhat off-color and/or some trees have thinning crowns, overall appearance is fair.

Poor: most trees stressed, foliage chlorotic and/or thinning crowns common, overall appearance poor.

Dead: Most trees dead.

HWA Density:

Low: most trees uninfested and/or have <10% infested branches

Medium: ≥ 50% trees infested, most infested trees have 10-50% branches infested.

High: Most trees infested and often >50% of branches infested.

8. **Condition of Release Trees:** Record tree health (categories as defined above) and HWA density. **HWA Density:**

Low: <10% branches on tree infested

Medium: 10-50% branches on tree infested

High: >50% branches on tree infested

- 9. **Notes:** Record any notes you feel may be appropriate.
- 10. Predator Recovery: Record # branches per sample tree/cluster, species recovered, number of indivuals/sample, life stage, generation (if possible), host (release tree or not), distance to nearest release tree, time recovered, location, sampling method (beating/visual/crown samples) and

identification method (visual/genetic and identifier). If no recoveries, enter 0 in No. and leave rest of data cells blank.

General Monitoring Instructions for Sampling of Biocontrol Agents

- 1. Sample 2 times/year for at least 3 years post release.
- 2. If possible, include release tree in samples. Regardless, record release tree data in the space provided. If HWA populations or tree health declines in the release area, look for the closest area where HWA appears to be healthy to sample for adults.
- 3. If possible, complete sampling between 10am and 4pm.
- 4. Avoid brushing or otherwise disturbing branches of a sample tree before beginning sampling. Focus on branches with healthy HWA as much as possible. Tap the branch smartly with the stick multiple times above the beat sheet such that the branch does not hit the beat sheet.
- 5. Sample 8 branches per tree or nearest neighbors if 8 are not available on a single tree. Distribute sampled branches to all sides of the primary sample tree. If all the branches are on one side, then sample those branches. Strive to maximize the number of infested branches that are sampled.
- 6. Quickly scan the beat sheet for introduced predators. Inspect twigs and needles on the beat sheet for biocontrol agents because they often hide under debris. Collect specimens in a labeled vial if unsure about identifying beetles.
 - a. *Note:* L. rubidus is occasionally found on hemlocks and can be easily confused with Ln because they are the same shape and size. Ln is all black while Lr has a lighter rust/reddish color with a darker line down where the wing covers meet together. If you are uncertain, collect specimens in alcohol for later confirmation.
 - b. Suspect Ln specimens can be sent to the Virginia Tech Laboratory for confirmation. Carrie Jubb or Ashley Lamb 216 Price Hall, VA Tech (MC 0319), Blacksburg, VA 24061
 - c. Suspect St specimens can be mailed to Dr. Carole Cheah at the Connecticut Agricultural Experiment Station.
 - d. You may be able to rear questionable larvae to adulthood in captivity on infested foliage.
- 7. Using the same protocol, sample at least 10 primary trees with healthy HWA near the release trees. Sample 80 branches if time and adelgid populations permit. Otherwise maximize the number of infested twigs sampled. If fewer than 80 are sampled, record approx no. sampled in notes.

Sources:

Mausel, D.L., T.A. Dellinger, A.B. Lamb, S.M. Salom, L.T. Kok. 2008. Field Instructions for release of *Laricobius nigrinus*, a biological control agent for the hemlock woolly adelgid in the eastern U.S. USFS/ Virginia Tech. Dept. of Entomology.

Dr. Carole Cheah, Personal Communications.

Approximate life cycles of hemlock woolly adelgid and its introduced predators

	Stage	Ja	an	Fe	eb	M	ar	A	pr	M	ay	Jı	ın	Jı	al	A	ug	S	ер	О	ct	No	OV	Dec
Hemlock Woolly Adelgid (HWA): Target Species	HWA Egg						?	_		?	_			_										
=likely present =may be present =aestivating	HWA Crawler									?						?								
	HWA Nymph				?	?	?	?	?		_		_	_		A	A	A	A	A				
	HWA Adult	?	?	?	?										?									
Sasajiscymnus tsugae (St): Larvae and adults feed on	St Larvae																							
all HWA stages	St Adult																						?	
==likely present ==may be present	St Not Feeding																						?	
Laricobius nigrinus (Ln): Larvae and adults feed on all	Ln Larvae																							
HWA stages present at active time	Ln Adult																							
=likely present	Ln Not Feeding																							
		Ja	an	Fe	eb	M	ar	A	pr	M	ay	Jı	ın	Jı	ul	A	ug	S	ер	О	ct	No	OV	Dec

Much of the data come from studies which were in places with warmer climates than the Northern New England States.

St Description:







St Adult, larva, and pupa (photos: Carole Cheah, Connecticut Agricultural Experiment Station)

Adults: oval, 1.5x long as wide (~1.6 mm x 1.0mm), entirely black exc. antennae slight brown, covered in silvery white pubescence. Even eye and appendages have setae. 9-seg. antennae with 4 seg. club.

Mature Larvae: elongate, fusiform, ~3mm long by ³/₄ mm wide, gray or reddish brown with light coating of waxy fuzz. Legs and head dark brown. *Laricobius* larvae will also be wax covered, but the wax is more dense and blanket-like and they have larger, three segmented antennae (as opposed to 1-seg. in St).

For more see: http://www.nysaes.cornell.edu/ent/biocontrol/predators/pseudoscymnus tsugae.html.

From: http://na.fs.fed.us/fhp/hwa/pubs/proceedings/2002 proceedings/recognition.pdf and Sasaji, H and M.S. McClure. 1997. Description and Distribution of *Pseudoscymnus tsugae* sp. nov.(Coleoptera: Coccinellidae), an Important Predator of HemlockWoolly Adelgid in Japan. Ann. Entomol. Soc. Am. 90(5): 563-568.

Ln Description:







Ln Adult, egg and larva (photos: Bugwood.org: adult, egg: Ashley Lamb, Virginia Polytechnic Institute and State University, larva: Gabriella Zilahi-Balogh, Agriculture and Agri-Food Canada)

Adults: ~2-3 mm long, black and shining on dorsal surface; body covered with fine erect hairs; elytra striate with 10 rows of oval punctures; antennae 11-segmented with three-segmented club. NOTE: *L. rubidus* has a lighter rust/reddish color with a darker line down where the wing covers meet together.

Eggs. within ovisacs, bright yellow; shiny; oval; <0.5 mm.

<u>Mature Larvae.</u> elongate, slightly fusiform, with scattered short setae, ~ 1.5 to 5.5 mm; yellow-green to brown; movement looper-like; feed within woolly ovisacs; covered in waxy blanket.

For More See: http://web.ento.vt.edu/ento/People/814879/L nigrinus.morph Can.Ent 06.pdf

From: http://www.na.fs.fed.us/fhp/hwa/pubs/proceedings/2002_proceedings/morphological.pdf

Introduced HWA Biocontrol Monitoring Form—Northern New England. Ver. 2010.1										
<u>Location Name/Desc:</u>		Condition of Release Trees (#:)								
			Tree	Tag ID	Tree Health	HWA Density				
Curr. Date:	1st Rel. Date:	1		GFPD	LMH					
Weather		Monitoring Contact Information	2		GFPD	LMH				
temperature: °C/°	PF	name:	3		GFPD	LMH				
wind: n/a, l, m, h	1	phone:	4		GFPD	LMH				
ppt: n/a, lt. rain, hv	v. rain, snow	org:	5		GFPD	LMH				
cloud: n/a, l, m,	h	e-mail:	6		GFPD	LMH				
GPS Location Datus	<u>m:</u>	Type: Lat/Long, UTM, Other:	7		GFPD	LMH				
Coord:			8		GFPD	LMH				
Stand Conditon: Ove	rall Health: Good/Fa	9		GFPD	LMH					
Recent Changes/Distur	rbances:	10		GFPD	LMH					
Notes:			Notes:							

	Predator Recoveries (enter 0 if no recoveries)												
Sample	Species	No.	Stage	Gen.	Tree Type	Dist.	Time	Location	Location	Sample type	ID type		
sample # /# branches	(Ln/St)	count	e,l,p,a	F1, F2	release/non rel.	to rel. tr.	military	type: (eg. latitude)	type: (eg. northing)	beat/visual	vis./gen. identifier		
1/						m/ft							
2/						m/ft							
3/						m/ft							
4/						m/ft							
5/						m/ft							
6/						m/ft							
7/						m/ft							
8/						m/ft							
9/						m/ft							
10/						m/ft							
Notes:		•			•		•	•	•	•			