

Egg Mass Survey Protocols for Oregon Spotted Frogs and Red-Legged Frogs 2015

Updated Feb 2015 by MMPearson, BALANCE ECOLOGICAL

This document anticipates multiple site visits by potentially different observers, and will ensure that all observers are following the same protocols.

Materials:

- Polarized sunglasses
- GPS unit, set to NAD83/UTM Zone 10
- Clipboard, data forms, pencil
- Camera (polarized lens if you have one)
- Flagging tape, if planning to return to the site (use yellow in locations with public access)
- Sharpie marker
- Thermometer
- Depth-measuring device (folding extensible ruler)
- Waders
- Canoe / kayak, if necessary to move effectively in the habitat.

GPS settings:

Set GPS to collect data in NAD83/UTM Zone 10, and to **track your movements**.

Turn the GPS on when you begin your search, and mark the time in your notes. This will allow us to track effort. *Note: When using your GPS for other activities during the breeding window, please ensure that other tracks are not recorded in the same file as your OSF search tracks.

'Mark' waypoints in your GPS, and transcribe the waypoint number into your notes along with the UTM coordinates. **Please write the UTM coordinates AND waypoint # in your notes.** Include your initials in your waypoint (eg. MMP-001).

When all surveys are completed, provide your GPS files to the data processor in .xls and .gpx format, along with your digitized field notes (or scanned / original forms).

Single-Visit Protocols:

At locations not known to have Oregon spotted frogs (OSF) on-site, single visits will be made to count red-legged frog (RLF), northwestern salamander (NWS) egg masses and search for OSF egg masses. These visits will begin once OSF egg masses have been seen in all known locations, ensuring that breeding season has begun.

RLF egg mass locations may be grouped into logical landscape units (i.e. # of egg masses in a pond or section of stream). If using this method, ensure that the marked GPS location is central within the landscape unit. Please DO NOT provide a range (eg. 20 – 30 egg masses), instead, count or estimate number

Please fill in each field on the spreadsheet for every waypoint.

- In your notes:
 - Time
 - Waypoint #
 - UTM coordinates
 - Comments - Please make a note of pictures taken, and submit them with your data. Photographs of overall habitat conditions are also very useful.
 - Species (use species code – see below)
 - Water temperature

If you suspect you have found OSF egg masses in an unexpected location, collect all data (water temperature, conductivity, Gosner stage, est % dead eggs), flag the location of the egg mass and alert Monica Pearson immediately to arrange for confirmation.

Multiple-Visit Protocols

At locations known to have OSF on-site, multiple site visits will be made to ensure that all egg masses are counted. The following instructions are intended to help observers track changes more easily.

When OSF egg masses are identified, ensure that each **cluster** has a unique waypoint.

- In your notes:
 - Time
 - Waypoint #
 - UTM coordinates
 - Species (use species code – see below)
 - Water temperature
 - Gosner stage
 - Estimate % dead eggs
 - Photo number and drawing of the cluster showing individual egg masses.
 - Comments
 - Access details if necessary
- In your GPS waypoint file:
 - Note in the waypoint file (in the ‘notes’ section) how many egg masses are in each cluster.
- In the field:
 - Mark each cluster in the field with flagging tape / pin-flag as close as possible to the cluster
 - Write your initials, the waypoint #, date, and # egg masses on the flagging tape.
 - Install a second flag at a visible location (in yellow if in a sensitive area)

When returning to an egg mass, take the same information in your notes, and add # egg masses / cluster to your GPS file. Compare new data to flagging tape. Add updated information to flagging / pin. If this is your last field visit to the site, please remove flags.

Species codes

Common name	Species code	Scientific Name
Oregon Spotted Frog	RAPR	<i>Rana pretiosa</i>
Red-Legged Frog	RAAU	<i>Rana aurora</i>
Northwestern Salamander	AMGR	<i>Ambystoma gracile</i>
Pacific Chorus Frog	PSRE	<i>Pseudacris regilla</i>
Long-Toed Salamander	AMMA	<i>Ambystoma macrodactylum</i>

OSF HABITAT - SIDE B

Waypoint	#	EM	Drawing	Waypoint	#	EM	Drawing
NOTES:				NOTES:			
NOTES:				NOTES:			
NOTES:				NOTES:			

Oregon spotted frog (RAPR) / Red-legged Frog (RAAU) Egg Mass Differentiation

Updated Feb 2012 by MMPearson

For Oregon spotted frog (*Rana pretiosa*, RAPR) and Red-legged frog (*Rana aurora*, RAAU), identification of egg masses can be tricky. Tightly clustered egg masses are always RAPR, however single egg masses are more difficult to identify.

Table 1 attempts to clarify the differences. Egg masses described are the 'typical' mass; in the field there will be a wide range from the 'typical RAAU' to the 'typical RAPR' with a grey zone in between. The following observations are the author's, and are not supported by statistical analysis. The most reliable indicator for differentiation appears to be attachment to vegetation, as is embryo size. Uncertain egg masses should be revisited to determine speciation at the hatchling stage, when tadpoles can be distinguished from one another.

Table 1. Observations regarding the differentiation of RAPR and RAAU egg masses in the field.

	RAPR – Oregon spotted frog (Figure 1)	RAAU – Red-legged frog (Figure 2)
Clustering	Communal oviposition site with multiple egg masses laid directly beside or on top of one another. Individual eggs appear crowded with smaller ova and jelly than RAAU.	Oviposition often close by to other egg masses but not tightly clustered. Common to find many egg masses within a pond or along the same shoreline.
Attachment	Never attached to grass or vegetation. If uncertain, tug gently on any vegetation that touches the egg mass; all vegetation should pull away, leaving the egg mass in place.	<u>Almost</u> always attached to vegetation. If uncertain, tug gently on vegetation that touches the egg mass; one piece of vegetation will cause the egg mass to move with it.
Water depth	Under 15cm; often sitting on substrate with upper egg mass exposed. Water level fluctuations may cause egg mass to float away from oviposition site.	Over 20 cm; rarely exposed above water level; often be laid below water surface. Attachment to vegetation reduces likelihood that egg mass will float away during high water, but may cause egg mass to be held out of water if water levels drop.
Older egg mass	Egg mass becomes less solid and spreads out just below the water line. Often has a greenish tinge. (Not for identification purposes)	Egg mass becomes less solid and spreads out just below the water line. Often has a reddish tinge. (Not for identification purposes)
Egg size	Embryo ~ 2.3 mm diameter. Jelly layer is thinner than RAAU, and eggs more densely packed. NOTE – egg size increases with age of female.	Embryo ~3.0 mm diameter. Jelly layer is thicker than RAPR, and more loosely packed. NOTE – egg size increases with age of female.
Hatchling stage	Hatchling tadpoles migrate to the centre of the egg mass. This stage can clearly identify RAPR from RAAU, but does not happen in every egg mass, especially if the egg mass has been disturbed.	Hatchling tadpoles exit the egg mass and remain in the surrounding vegetation as the mass dissolves.



Predation

If you find egg masses that look like this, mark it as predated. (take a photo and coordinates, plus other relevant information eg. Base of post / how far from breeding location?)

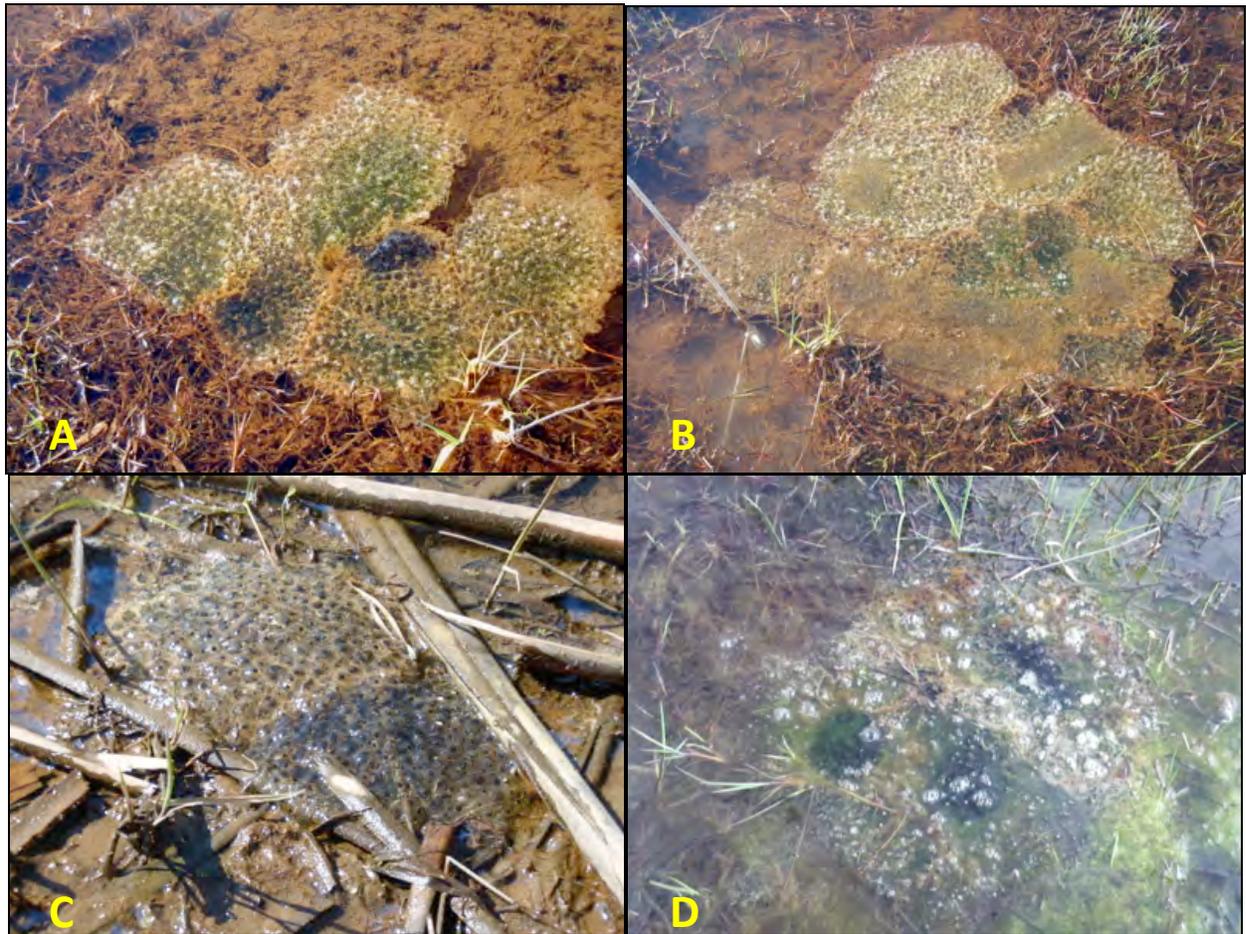


Figure 1. Clustered Oregon spotted frog egg masses. **A:** 6 egg masses within one group; notice the difference in freshness between the middle two egg masses and the outer four. **B:** 12 or 13 egg masses laid together. As development progresses differentiation of individual egg masses within the larger mass becomes more difficult. **C:** Egg masses laid in very shallow water that has receded. **D:** RAPR egg masses at the free-swimming tadpole stage: tadpoles cluster in the centre of the individual egg mass to continue development. Photos: Monica Pearson.

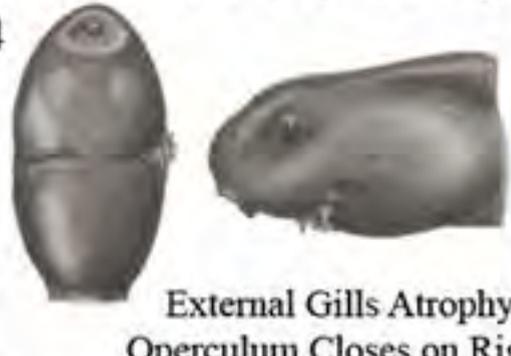


Figure 2. Red-legged frog egg masses. **A:** Three separate egg masses laid in close proximity. These are in shallow water, but each is attached to a blade of grass. **B:** Adhesion to twigs has caused these RAAU egg masses to lift out of the water following a drop in water levels. Photo A: Monica Pearson, B: Mike Pearson.

EMBRYOS

1  Fertilization	2  Gray Crescent	3  2-Cell	4  4-Cell	5  8-Cell
6  16-Cell	7  32-Cell	8  Midcleavage	9  Late Cleavage	10  Dorsal Lip
11  Yolk Plug	12  Late Gastrula	13  Neural Plate	14  Neural Folds	15  Elongation, Rotation
16  Neural Tube, Gill Plates	17  Tail Bud Adhesive Gland	18  Muscular Response Olfactory Pits	19  Heart Beat Gill Buds	

HATCHLINGS

20  Gill Circulation, Tail Elongation	21  Cornea Transparent, Mouth Opens	22  Tail Fins Transparent, Fin Circulation
Operculum, Oral Disc, and Pigmentation		
23  Labia and Teeth Differentiate Operculum Covers Gill Bases	24  External Gills Atrophy Operculum Closes on Right	25  Mouthparts Obvious Spiracle Forms on Left