FWF 493/560:
"Amphibian Sampling"

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Goal of the Lecture

To familiarize students with common techniques used to capture, measure, and mark amphibians.

Lecture Structure

Amphibian Sampling

I. Capture Techniques

II. Marking Techniques

III. Breeding Call Surveys

IV. Malformations

Amphibian Capture Techniques
Aquatic Sampling & Area Searches

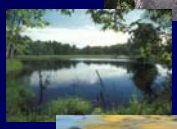
- Area-constrained Searches
- Dip and Seine nets




- Funnel Traps
- Enclosure Sampling

Aquatic Sampling
Types of Areas Sampled

- Small bodies of water
  - Tree holes, sink holes, puddles
    - Sample with dip-net or small seine net repeatedly
      - 10 sweeps
- Ponds
  - Stratified sampling techniques
  - Transects at varying depths
- Streams
  - Sample riffle, runs, and pools
  - Riffle and run = 1 minute samples
  - Pools = 30 second sweeps


Aquatic Sampling
Seine Nets

- Seines
  - Mesh size and width
    - 1.5 to 7 mm and 1 to 1.5 m wide
      - Larger and small sizes can be used
    - Pre-hung lead weights
    - Dowels
  - Methods
    - Walk directly across water
    - Seine parallel to shore
    - Plant seine in vegetation


Aquatic Sampling
Dip-nets and Enclosure Sampling

- Dip-nets
  - Size depends on sampling situations
    - D-nets
    - Wire mesh sieves
  - Standardize
    - Number of sweeps or time
    - Length or depth of sweep
- Enclosure Sampling
  - Fixed box size
  - Sampled
    - Fixed amount of time
    - Fixed number of sweeps



Amphibian Sampling
Pitfall Trapping

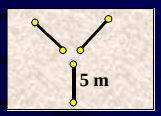
•Movement
•Interception

•Redirected Movement
•Capture

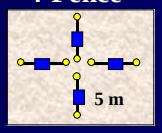


Drift Fence and Pitfall Designs
Straight-line Arrays

3-Fence

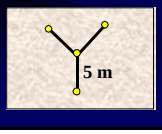


4-Fence

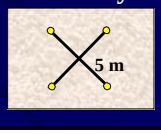


Forested Sites

Y-Array

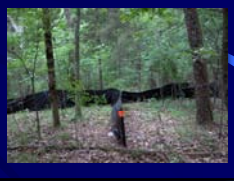


X-Array



Pitfall Traps

X-Array

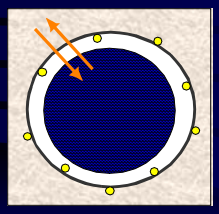


Goal: Estimate Population Size and Composition

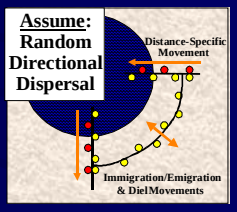
Drift Fence and Pitfall Designs

Continuous and Partial Drift Fences

Complete



Partial



Goal: •Estimate Population Size and Composition

•Estimate Directional Movement

Drift Fence and Pitfalls

Materials and Costs

- **Fence** ($0.35-$1.50 per meter)
  - Aluminum Flashing
  - Hardware Cloth
  - Plastic/Cloth Erosion Fence

24" stakes

- **Pitfalls** ($2.50-$5.00 per bucket)
  - Plastic Buckets (8- or 19-liter) w/ Lids
  - #10 Tin Cans (2 fastened = 8-liter)
- **Shade Covers/Sponges** ($0.25-$1.50 per bucket)
  - Wood or Pegboard Planks with Legs
  - Synthetic Foam or Sponges

Drift Fence and Pitfalls
Installation

Fence Placement: Stratified Random or 5 m above anticipated HWL

Pitfall Placement: Every 10 m and Adjacent to Fence

- Hoe, Mattock, or Ditch Witch ($150/day)
- 12-inch Auger ($75/day)
- Shovels, 3-5 lb Sledges, Tape Measure, Flags ($100)
- Personnel (4 people: 300 m/1-2days [$250/day])



Drift Fence and Pitfalls
Installation

STEP 1: Measure and Distribute Materials


STEP 2: Dig Holes and Install Buckets (top flush w/ ground)


Drift Fence and Pitfalls
Installation

STEP 3: Remove Vegetation and Dig Trench (3-5 inches)


STEP 4: Install Fence

STEP 5: Bury Fence

Continuous Drift Fence

Completely Set Up

5 m Leads

Drift Fence and Pitfalls

Program Mark

Censor Buckets

Maintenance **$200/month**

Weather

- Precipitation
- Wind
- Sun

Animals

- Livestock
- Rodents

Drift Fence and Pitfalls

Operation

- **Pitfalls should be checked daily (before 1200 hrs)**
  - Reduce Probability of Predation (snakes, raccoons), Desiccation, Drowning, or Ammonia Toxicity
- **Processing time is capture frequency dependent**
  - 15 minutes (0 captures) to 15 hours (14K) for 350 m
- **Processing should be continuous**
  - Reduce Probability of Density-Induced Movement
- **Handling can enhance desiccation**
  - Rehydrate prior to release
- **Closing Buckets (sample alternate days)**
  - Reduce probability of immediate recapture
  - Increases temporal independence

Drift Fence and Pitfalls

Considerations

Pitfalls: Yes or No?

- **Research Question** (Are pitfalls necessary?)
- **System/Terrain** (Is it realistic?)
- **Funding** (What are the costs & benefits?)

Species-specific Biases

- **Differential Capture Rates**
  - **Climbing, Jumping, Digging Ability**
- **Differential Trespass**
  - **Can be quantified**
- **Location of Fence and Pitfalls**

Other Sampling Methods

- **Visual Encounter Survey (VES)**
- **PVC pipes**

Amphibian Marking Techniques

Biological Processing

General Information

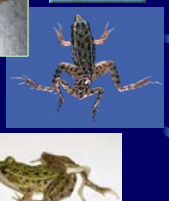
- Species, Age, and Gender
- Snout-vent Length (SVL)
- Weight (50, 100, 250 g Pesola®)
- Abnormalities
- Malformations, Tumors, Sores, Parasites

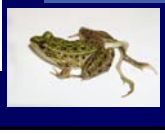


Malformations

- Hypotheses
  - Genetic
    - 1-5% Malformation rate
  - UV-B rays
  - Chemical Pollution
  - Trematodes





Clearing Techniques

- Trematode prevalence
  - Malformed individuals opportunistically collected
  - Malformation classified using USGS Field Guide to Malformations of Frogs and Toads
  - Humanely euthanized via transdermal exposure to benzocaine hydrochloride
  - Fixed in 10% buffered formalin and Cleared
  - Light microscopy used to detect presence of encysted trematode metacercariae






Mass Marking Techniques

Batch

- Florescent Elastomers
  - Injectable Liquid Elastomer (4 colors)
  - $1000 Kit (1000 individuals)

- Florescent Dyes
  - Water resistant Dyes
  - Poweder and Shake-and-Bake
  - Ultraviolet Light Sensitive
  - $12/lb (1lb/100 individuals)

- Toe-Clipping
  - Mass-mark or uniquely ca. 2,000 individuals
  - Rapid and Inexpensive

Batch or Individual

Toe Clipping Protocol

Steps

STEP 1: Sterilization
- 0.01 % Chlorhexidine diacetate

STEP 2: Clip
- First Joint of Toe

STEP 3: Stop Bleeding
- Silver Nitrate Sticks

STEP 4: Store DNA
- Ice Bath
- Store <-70C

Individual Marking Techniques

Individual-Specific

- Coded Wire Tag
  - Injectable Stainless Steel Tag
  - Etched Binary ID Code
  - 1.1 x 0.25 mm, $15/$6000 wand

- Alpha-numeric Tag
  - Injectable Visible Tag
  - Alpha-numeric Code
  - 1.1 x 2.5 mm, $1 each/$100 injector

- Passive Integrated Transponder (PIT) Tag
  - Injectable Electromagnetic Tag
  - Transmit ID Code
  - 11.5 x 2.1 mm, 0.06 g
  - $4.50 each / $500 scanner

PIT Tagging Protocol

Steps

STEP 1: Sterilization *(0.01% Chlorhexidine diacetate)*

- Tags, Needles, and Injection Point

STEP 2: Injection

- Inject Tag Subcutaneously left of Midventral Vein

STEP 3: Seal

- Seal Puncture Wound w/ Cyanoacrylate or Glue

Non-Capture Methods

Breeding Call Surveys

http://www.state.tn.us/twra/tamp.html

Begin: ≥ 30 minutes after sunset **End:** 1:00 a.m.

Duration: 5 minutes

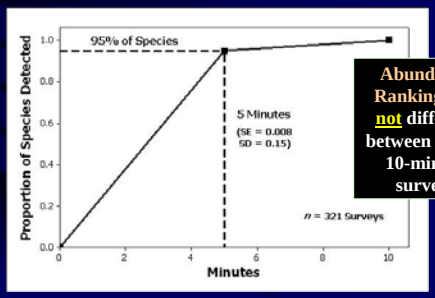
Abundance: (By Species)

0 = none heard
1 = individuals can be counted
2 = calls overlap but individuals can be distinguished
3 = calls overlap and individuals cannot be distinguished (full chorus)

Non-Capture Methods

Breeding Call Duration

Burton et al. (2007)



Abundance Ranking was not different between 5- and 10-minute surveys