SAFE PRACTICES
RNA Preservation Card Sampling of Dead Birds for West Nile Virus Testing

A Tutorial for Local Agencies

Presented by: The California Department of Public Health Vector-Borne Disease Section West Nile Virus Dead Bird Surveillance Program
RNA preservation card sampling of dead birds for West Nile virus testing

This tutorial explains and illustrates safety protocols for obtaining dead bird oral swab samples.

By following these protocols, you can minimize your risk of potential exposure to West Nile virus.
There is a risk of West Nile virus (WNV) transmission when working with dead birds:

- Sharp beaks can puncture skin
- Fluids from the mouth or cloaca may carry virus
- Virus may be on the feathers and body
- Virus can be aerosolized and potentially inhaled
Safety Precautions

• Avoid touching the carcass with bare hands.
• Pick up the dead bird by inverting a plastic bag over your hand, or wear gloves.
• Double-bag the bird and tie or seal.
Where to work?

If your lab does not have a biological safety cabinet (BSC), the bird can be handled in the field with appropriate protection.

If your lab has a BSC, transport the bird to the laboratory to work in the BSC.

OR

Work in BSC

Work outdoors wearing N-95 mask
OUTDOOR SAMPLING

To sample birds for RNA preservation cards in the field, you must wear an N-95 mask and work in a well-ventilated area.

1. You will need: **N-95 mask, disposable gloves, sterile swabs, RNA preservation cards, and a permanent pen or pencil.**

2. Label RNA preservation card with dead bird #. Put on mask and gloves. Isolate the bird’s head, and partially unwrap the polyester swab. Pry open the bird’s beak and hold open with one hand. With other hand, remove the swab from its wrapping. Aggressively swab the oral cavity and throat area.
Outdoor Sampling cont’d

3. Roll and press the swab onto RNA preservation card within the sample ring area, over the pre-punched discs.

4. Place the swab and used gloves in the bag with the bird. Seal the inner and outer bags and throw the bird away in an outdoor trash can.

5. Discard the N-95 mask. Wash hands with soap and water.

6. Do not touch the card’s sampling area. Allow sample to dry (2 hours). A clipboard can be used to hold the dead bird form and open card.

7. After drying 2 hours, close the card’s flap. The card is ready to be mailed to DART (UC Davis Arbovirus Research and Training Laboratory) for analysis.
Example of an outdoor sampling station. The fold-out shelf is convenient, and the metal surfaces can easily be sterilized.
LABORATORY SAMPLING

You can also work in a biological safety cabinet to sample dead birds.

1. You will need: **disposable gloves, sterile swabs, RNA preservation cards, metal spatula, permanent pen, 10% bleach, and 70% alcohol in spray bottles.**

2. Label RNA preservation card with dead bird number.

3. Wear disposable gloves and change gloves between birds.

4. Isolate only the bird’s head from the bag.

5. Swab the bird’s oral cavity and press on RNA preservation card as described previously.
Laboratory Sampling cont’d


7. Sterilize surfaces and instruments with 10% bleach followed by 70% alcohol.

6. Allow the card(s) to dry in the back of the cabinet for 2 hours.
Tips for Sampling

A metal spatula can be used to help pry open the bird’s beak.

Note: Samples may be dry. It is not necessary to soak the filter paper or cover the entire sample area. Vigorously swabbing and firmly pressing the swab onto the card should ensure a good sample. Blood may also be present. WNV has been detected in both dry and bloody samples.
Proper Technique

Work in a BSC. Sterilize surfaces and change gloves after each bird.

Swab bird’s oral cavity. Only head is removed. Avoid sharp beak.

Press and roll swab onto card.
Carcass conditions suitable for sampling

- Not desiccated
- Intact body
- Died recently (within 48 hours)
- Not heavily infested with maggots or ants (covering less than 25% of body)
Species accepted for RNA preservation card testing*

- Corvids (e.g. American crow, Common raven, Western scrub-jay, Steller’s Jay, Yellow-billed magpie)
- Other passerines (American robin, Northern mockingbird, sparrows, finches, blackbirds, flycatchers, warblers) (to name a few)
- Hawks, owls, raptors
- Egrets, parrots, and other uncommon species will be accepted too

*A field guide is recommended for identification
Purchasing Information

RNA preservation cards
RNASound ReadyPunched™ cards can be ordered in packages of 10 or 25 online at the Fortius Bio LLC website.

Polyester swabs
Swabs must be synthetic (not cotton), with a long handle. For example: Fisherbrand® product #22-029-682.
Where to mail cards

Ship overnight, or mail cards by USPS signature required mail to DART (UC Davis Arbovirus Research and Training Laboratory):

Dr. Ying Fang  
University of California  
One Shields Avenue  
Vet Med: PMI Room 3336  
Vet Med 3A  
Davis, CA 95616

For UPS Shipments only:  
Dr. Ying Fang  
VM://PMI 3336  
Vet Med 3A  
1285 Veterinary Medicine Mall  
University of California, Davis  
Davis, CA 95616

Or: to save costs, seal all cards in another ziplock bag and add to your mosquito box if you send mosquitoes to DART for WNV testing.
RNA preservation cards will be tested by RT-PCR at the UC Davis Laboratory in batches of 10 to 12.

Results are entered into Gateway. You should receive an automatic email with test results as soon as they are entered.

Turnaround time is typically 2-7 days during the regular season and ~3 weeks in winter.
The WNV website’s “Response Plans and Guidelines” section contains more information, including the oral swab sampling protocol.