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.

DEAD BIRDS CAN CARRY WNV

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

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.

Work in BSC

OR

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.**

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.

A partially unwrapped swab

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.

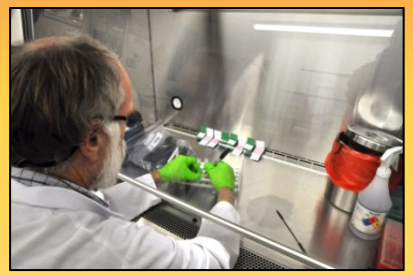
5. Discard the N-95 mask. Wash hands with soap and water.

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 CVEC (Center for Vectorborne Diseases at UC Davis) for analysis.

Press and roll swab onto card

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

6. Dispose of the swab in the bag with bird. Double-bag, seal, and put in trash.

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

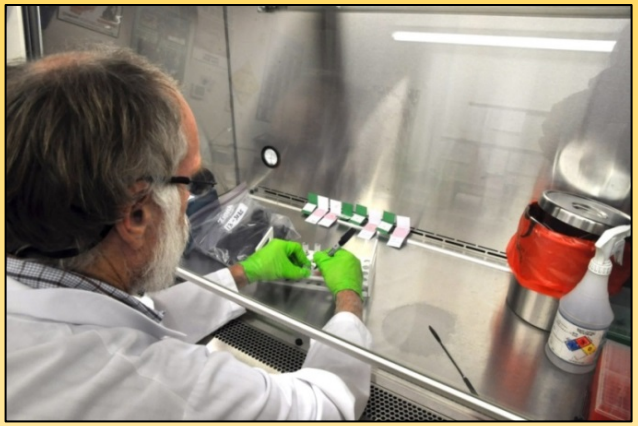
8. Allow the card(s) to dry in the back of the cabinet for 2 hours.

Tips for Sampling

A **metal spatula** can be used to help pry open the bird's beak. In the field, **wooden sticks** are a disposable alternative.

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 24 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 + swabs
RNASound ReadyPunched™ cards can be ordered online at:
http://www.fortiusbio.com/RNA_Sampling_Card.html
Use the code "CDPH" when ordering to receive the $140 price (regular $200).

Polyester swabs are included with the cards, but if you need more:
Swabs must be synthetic (not cotton), with a long handle. Fisherbrand® product #23-400-116.

Where to mail cards

Mail cards in an envelope by signature required to:

Dr. Ying Fang
University of California
One Shields Avenue
Vet Med: Pathology, Microbiology, & Immunology
3336 Vet Med 3A
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.

Test Results

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.

MORE INFORMATION

The WNV website's "Response Plans and Guidelines" section contains more information, including the oral swab sampling protocol: http://www.westnile.ca.gov/resources.php