SUBMISSION OF ANIMALS FOR RABIES TESTING

When the decision is made either to euthanize an animal or submit a dead animal for rabies examination, several procedures must be followed to obtain quick and accurate results. Many times rabies prophylaxis of an individual is dependent on these results. Contact Los Angeles County Veterinary Public Health (323-730-3723) for information on appropriate sample handling and to arrange for specimen pick-up. The rabies specimens will be transported to the Public Health Laboratory.

Animals that bite humans

Wild carnivores & bats

In California, wildlife account for close to 99% of the rabid animals. Any wild carnivore that bites a person should be immediately tested for rabies. The four most common terrestrial reservoir species harboring rabies in the U.S. are: raccoons, skunks, foxes and coyotes. In addition to these terrestrial, indigenous rabid bats have been found in every state except Hawaii.

Healthy cats and dogs

A healthy dog, cat, or ferret that bites a person should be confined and observed for 10 days. Such animals should be evaluated by a veterinarian at the first sign of illness during confinement. If signs suggestive of rabies develop, the animal should be euthanized, its head removed, and the head shipped under refrigeration (not frozen) for examination of the brain by the Public Health Laboratory.

Collection and submission of animal specimens for rabies testing

This information is supplied to facilitate the handling and to provide the most accurate and rapid diagnosis of specimens submitted for rabies testing. The Public Health Laboratory accepts specimens for rabies testing from private veterinarians or animal shelters who adhere to the following guidelines.

- A. Listed below are animals that will be tested for rabies.
- 1. Domestic mammals that bite/expose humans (except fully-vaccinated dogs, cats, and caged animals raised indoors).
- 2. All wild carnivores that bite humans. Signs of rabies in wildlife cannot be interpreted reliably.
- 3. All bats should be tested regardless of human exposure. The bite of a bat is so minuscule that often the individual is not aware of it. Also, because rabies is

endemic in bats, we test them for surveillance purposes to determine the prevalence of bat rabies.

- 4. Ill animals with signs of rabies.
- B. Listed below are animals that will NOT be accepted for rabies testing:
- 1. Cage raised pets will NOT be tested. These include all gerbils, guinea pigs, and hamsters.
- 2. Rodents and rabbits will NOT be tested, except when a Veterinary Public Health Veterinarian deems that extraordinary circumstances exist which indicate rabies infection is likely.
- 3. Baby bats submitted with the mother will NOT be tested regardless of exposure history. Only the mother will be tested if an exposure has occurred.
- 4. No live animals will be accepted for testing.

Guidelines to Ensure Proper Rabies Testing of Specimens

- 1. Avoid damage to the brain by shooting or other traumatizing procedures.
- 2. Have a qualified person separate the suspect animal's head from the body immediately after death. Submit only the head of the animal.
- 3. If only the brain is submitted rather than the entire head, be sure to include parts of the cerebellum, hippocampus, and brain stem. Specimens that do not include at least two of these three areas of the brain will be considered unsatisfactory due to a lack of sufficient material. DO NOT FORMALIN FIX BRAIN TISSUE.
- 4. Pour chloroform over the specimen or use flea spay in sufficient quantity to kill any fleas which may be present. Wrap the specimen in several layers of newspaper. Place wrapped specimen in a plastic bag, seal the bag and attach completed lab transmittal form to the outside of the plastic bag. Place this entire package inside a second plastic bag to protect the lab slip. Seal the bag.
- 5. Keep the head refrigerated until time to deliver to the laboratory. Do not freeze.
- 6. Brains submitted from larger animals such as livestock nominally should include labeled portions from the hippocampus, cerebral cortex, cerebellum and brain stem from both sides of the brain.
- 7. Bats should be submitted whole.

Routine and emergency laboratory processing of rabies specimens

The processing of rabies specimens is performed daily. Results are telephoned or faxed to the submitter. Positive results are also telephoned to Acute Communicable Disease Control (ACD) and Veterinary Public Health.

Routine Processing:

Processing is initiated on all specimens submitted to the laboratory by 1:00 p.m. of that day, smears are made and fixed overnight for direct immunofluorescent antibody (IFA) staining the following morning. Results are ready before noon of the day following submission.

Emergency Processing:

Specimens deemed to be of an emergent nature are processed including staining on the same day of submission. These results are ready within six hours of specimen receipt in the laboratory. Special arrangements must be made for emergency testing by contacting Veterinary Public Health (323-730-3723) before the laboratory can proceed with testing. All IFA results done by six hour fixation are considered to be preliminary, therefore retesting of brain material will be performed on the next regular laboratory work day, using slides that had at least an overnight fixation.

Questions on Laboratory Testing for Rabies

What animals will be tested by the Public Health Laboratory (PHL)?

In general, the following animals will be tested.

- 1. Mammals that bite/expose humans (except fully-vaccinated dogs, cats, ferrets, and caged animals raised indoors).
- 2. Ill animals with signs of rabies.
- 3. All bats.

What test is performed for rabies at the Public Health Laboratory (PHL)?

The test of choice for detecting rabies in animals is the direct immunofluorescent antibody (IFA) test. A slide containing fresh brain tissue is reacted with fluorescein labeled anti-rabies virus antibody. When viewed under a fluorescence microscope, rabies virus antigen will fluoresce. Any animal excreting virus in its saliva should have detectable virus in its brain.

What types of specimens are required for IFA testing?

Rabies IFA testing requires fresh, unfixed brain tissue.

What does PHL accept for testing?

No living animal will be accepted for rabies testing. For testing of bats, the entire animal should be sent to the PHL. For other species, just the animal's head and upper neck should be submitted.

Euthanizing a bat that needs to be tested: Captured bats should be euthanized before being shipped to the PHL. Many local veterinarians or animal shelters can safely and humanely euthanize bats.

Who should remove the head for testing?

Because the brain, spinal cord, salivary glands, and saliva may contain rabies virus, only veterinarians, animal control officers, or others who have been appropriately trained (and adequately vaccinated) should remove animal heads. This work should be done in a properly ventilated area using adequate protective gear.

How should specimens be shipped to the PHL?

Wrap the specimen in several layers of newspaper. Place wrapped specimen in a plastic bag, seal the bag and attach completed lab transmittal form to the outside of the plastic bag. Place this entire package inside a second plastic bag to protect the lab slip. Seal the bag. Keep the head refrigerated until time to deliver to the laboratory.

What are the possible outcomes of the testing?

Rabies IFA testing at the PHL can result in the following outcomes:

Positive: A "positive" specimen is one in which rabies virus antigens have been detected. Any animal with a confirmed positive rabies IFA test is considered capable of transmitting rabies. The IFA test is highly specific for rabies (other diseases do not cause a positive test).

Negative: A "negative" specimen is one in which adequate brain tissue was examined yet no rabies antigens were detected. A negative IFA test performed with an adequate specimen reliably rules out the possibility that the animal tested was capable of transmitting rabies.

Unsatisfactory: An "unsatisfactory" test is one in which the specimen submitted was inadequate for testing.

In addition, non-specific or indeterminant fluorescence patterns are found rarely. In such instances, additional studies will be performed.

What is the turnaround time for test results?

Processing is initiated on all specimens submitted to the laboratory by 1:00 p.m. of that day, smears are made and fixed overnight for IFA staining the following morning. Results are ready before noon of the day following submission.

How are the test results disseminated?

Results are telephoned or faxed to the submitter. Positive results are also telephoned to Acute Communicable Disease Control (ACD) and Veterinary Public Health.