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MAKING DIAGNOSIS OF RABIES IN DOGS EASIER

A new technique enables ACCURATE DIAGNOSIS of canine rabies in the Philippines and beyond.

A rapid, accurate and low-cost test for rabies in dogs, which doesn't require a laboratory or technical equipment, has been developed by researchers at Oita University in Japan.

A deadly viral disease that can infect humans and other mammals, rabies is mostly transmitted via bites and scratches from dogs carrying the rabies virus. In people it causes inflammation of the brain and spinal cord and is almost 100% fatal once symptoms, such as confusion, agitation and fear of water, appear.

▲ A greater focus on rabies vaccinations for dogs could help reduce cases of rabies in humans.

According to the World Health Organization (WHO), 95% of human rabies cases occur in Africa and Asia, and lead to an estimated 59,000 deaths globally each year, about 50% being children under 15 years old.

Each year, more than 29 million people receive a post-bite rabies vaccine. Alongside immediate wound treatment and administration of immunoglobulin — an antibody-boosting medicine to fend off the virus — the shot can prevent the disease from progressing. These measures prevent hundreds of thousands of deaths annually.

But post-exposure vaccines and treatments for humans don't attack the problem at its source.

SIMPLE BRAIN SAMPLING

"The most effective way to end rabies is to implement proper canine rabies control," says Akira Nishizono, a microbiologist and professor at Oita University, in Japan. "Without it, the number of casualties will not decrease."

In the Philippines, for example — where domesticated and stray dogs often roam freely — a lack of vaccinations for dogs hampers anti-rabies efforts. This is exacerbated by the fact that post-exposure vaccines for people have been prioritized over implementing more stringent animal controls, notes Nobuo Saito, an associate professor at Oita University.

Identifying and mapping canine cases is the first step towards rabies control, and it can help build an effective vaccination strategy for dogs in regions where rabies is prevalent. But this step is often not implemented.

A study authored by Nishizono and his colleagues in Japan and the Philippines, revealed that canine rabies cases on Luzon, the largest island in the country, are most commonly reported in the area surrounding the regional animal laboratory, despite many human cases being reported in areas of the island far from the laboratory.

This is likely because there is no active rabies surveillance in animals. Furthermore, there is no sample submission or transportation system for rabies testing. Consequently, rabies surveillance is limited to areas surrounding animal diagnostic laboratories and many rabid animals further afield go undetected. The researchers suspect this is also the case in other regions of the country[1].

As diagnosing rabies is not easy, there are very few animal diagnostic laboratories that are capable of it. The standard method for detecting rabies in animals is the direct fluorescent antibody test (dFAT), which involves opening the skull of a deceased animal, taking brain samples from specific regions with high viral content — such as the brain stem — staining them with a fluorescent antibody and examining them under a microscope.

This complex process limits the accessibility of rabies control in rural regions without special facilities and trained experts.

A simpler technique, using lateral flow devices (LFDs), can detect rabies virus nucleoproteins within 15 minutes from brain tissue samples mixed with solution. But its accuracy varies, and the need to collect brain samples is still a challenge.

To overcome this, Nishizono and his international team have combined an LFD with a simpler sample collection technique called straw sampling[2].

Straw sampling, which is endorsed by the WHO, does not require opening the deceased animal's skull. Samples still have to be collected from the brain, but they can be collected by inserting a plastic pipette or straw into the foramen magnum — a round opening at the base of the skull. The foramen magnum can be accessed by bending the neck of the animal and cutting the skin and neck muscles to expose the atlanto-occipital joint.

To test the accuracy of an LFD test using straw sampling, the researchers compared this technique against dFAT and the skull-opening method on 97 rabies-suspected animals.

The team collected samples using a plastic straw, mixed it with a solution, and applied it to the strip of an LFD kit jointly developed by Oita University and the Japanese pharmaceutical company ADTEC.

The researchers concluded that LFD with straw sampling can provide a relatively high accuracy, with 97% sensitivity and 100% specificity in detecting rabies in animals.

To further evaluate the feasibility and accuracy of LFD with straw sampling, the technique is being used at 17 animal laboratories across the Philippines. The team is collecting performance data on in various settings with different resource availabilities.

BEYOND DIAGNOSIS

Furthermore, the researchers hope to use the LFD kits to transport samples for genetic analysis on the rabies virus. Unlike the transport of brain samples which require biosafety containers and refrigerated systems, the LFD kits containing genetic information of a local virus is perfectly safe for transport at room temperature, permitting further studies to be conducted on rabies viruses at Luzon's central animal laboratory in Quezon City. This technique may enable wide-scale molecular analysis to be performed throughout the Philippines. The test kit will also be useful in other rabies-endemic countries besides the Philippines.

"We'll be able to understand the habitat and species of host animals, and how rabies spreads in a certain geographical area or cross-species," says Nishizono.

▲ Members of Nishizono's team talking to members of the public in Luzon, The Phillipines.

A data-sharing system developed by the research team — alongside officials at the Philippines' Department of Agriculture and Bureau of Animal Industry — can visualize and share information on rabid animals confirmed by LFD with straw sampling, in real time.

The system enables a 'one health approach' — connecting vets, clinics, the local council, residents and the national government — and gathers data to identify high-risk areas and facilitate prompt action, as well as raise awareness in the community.

"This not only makes rabies diagnosis possible anywhere, but it also helps to visualize where, and to what extent, rabies is prevalent, so that the data can be used for policy makers to secure a budget for dog vaccinations," says Saito.

Nishizono hopes their efforts will contribute to the global target to end human deaths from dog-transmitted rabies by 2030. "The main victims of rabies are the same around the world: children and people without access to medical care," he says. "Our mission is to open a passage towards medical care for those people."

The project is supported by Japan's Science and Technology Research Partnership for Sustainable Development (SATREPS). Joint research between Japan and developing countries under SATREPS tackles global challenges including environmental issues, disaster risk management and infectious disease control. ■

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