



Instituto Valenciano de Microbiología

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**Laboratory authorized by the European Union
for serological testing to monitor the efficacy of
rabies vaccines**

Rabies. Information about rabies antibodies quantification test in vaccinated animals.

The method used is recommended by the reference laboratory of the European Community of Nancy -ANSES- (France) (Laboratoire de la Rage et de la Faune Sauvage de Nancy). The test quantifies the rabies antibody by a virus neutralization procedure (FAVN: Fluorescent Antibody Virus Neutralization) using a quantized rabies virus CVS-11 strain suspension.

This procedure is complex and costly because: 1) using a quantified rabies virus suspension (titrated), against to the sera; 2) employs cultured BHK-21 cells for evidence of virus neutralization by antibodies; 3) using immunofluorescence with specific antibodies against rabies virus labeled with fluorescein, to demonstrate replication (viability) of the virus in the absence of antibodies, or otherwise neutralizing if any antibodies is present in inoculated cell cultures; 4) the serum must be tested at several dilutions, to quantify the amount of antibodies contained, and everything must be done in quadruplicate; and 5) must be done by comparing with an international standard reference serum, having a known amount of rabies antibodies, to give the result in International Units (IU/mL). To consider the animal is protected, you must have a serum antibody titre equal or higher than 0.5 IU/mL.

Recommended sample:

Serum separated blooded animal (0.5 to 1 mL), taken in dry tube or vacutainer tube type serum separator. The serum sample must be kept refrigerated until shipment to the laboratory. If they were to take more than 3 to 4 days before received in the laboratory must be frozen. Alternatively, if the blood comes to the laboratory the day after extraction, can be sent a type vacutainer tube whole blood (5 to 10 mL), and the serum would be separated in our laboratory. The blood sample must be drawn at least 30 days after completion of the vaccination to allow time for antibodies generation, and the time required by the European regulation currently admitted (EU Regulation No. 576/2013). When the animals come from other third countries and are introduced into the European Union, the sample must be taken at least 3 months before the pet being moved, by a registered veterinarian and for certain countries before 3 or 6 months (see Rules in corresponding section).

Delivery time for results:

The time for issuing results is **9 to 10 business days** (Monday to Friday), counting from receipt of the sample, if no repetition is required, and the test payment has been received. The report with the results, in Spanish and English, is sent by email

with a digital signature, together with the paid invoice, provided that the test has been paid by deposit or transfer to the bank account number that appears at the bottom of the order form. Payment by bank check, credit card or bizum are not accepted.

Cost of the test:

121.- € (one hundred and twenty-one euros) for samples from any origin (taxes included when it corresponds and service with our concerted courier from points of the Spain main land having courier office delegation, and that do not involve extra payment of mileage). The courier is not included for shipments from Balearic Islands, Canary Islands, Ceuta, Melilla or countries other than Spain. For samples sent from other countries, from Balearic Islands, Canary Islands, Ceuta or Melilla, the sender can make use of his choice, and should take charge of their cost, and expenses that may involve customs and inspection services veterinary seaports or airports

Other issues:

- See "Clarifications on the management of rabies antibody test for samples sent **from Spain**".
- See "Clarifications to frequently asked questions requesting information for sending samples to test rabies antibodies **from countries other than Spain**."

Sending samples

For sending samples of serum or whole blood, our center has a courier service (see the limitations in the previous section cost of the test (only in Spain main land, but not for Balearic Islands, Canary Islands, Ceuta, Melilla or countries other than Spain). To require our concerted courier send an email to ivami@ivami.com with the name and full address (including ZIP code), telephone, contact person and timing availability. If after reading the document you have any questions, you can call us at 96-169 1702 (our hours are from 8.30 am to 15.00 pm and from 16.00 to 17.30 pm Monday to Thursday; 8:30 am to 15:00 pm on Friday). July and August 8:00 am to 15:00 pm.

It must send the tube with the serum, or whole blood inside a plastic container sealing, preferably biological safety, and placed inside an insulated container (Styrofoam), preferably cooled with a pack frozen. Include the completed order form attached full-filled in the computer or clearly written (upper case), inside a plastic envelope to prevent wetting during transport. At the end of the order form there is a section called Billing, to tell us the name, full address and tax codes, which must appear on the invoice. Fridays shipment should not be done because we do not have courier delivery on Saturdays.