Laboratory support provided to countries in the region, opportunities for potential collaboration (WOAH Rabies Reference Laboratory, Onderstepoort)

Claude Sabeta, PhD 20 February 2023

Make today matter



UNIVERSITEIT VAN PRETORIA UNIVERSITY OF PRETORIA YUNIBESITHI YA PRETORIA

### Faculty of Veterinary Science

Fakulteit Veeartsenykunde Lefapha la Diseanse tša Bongakadiruiwa

# Rabies is still a neglected tropical disease

Under-reporting of animal and human rabies cases due to:

- Limited laboratory capability
  - **Misdiagnosis** may result with the presentation of fever and coma in children because of confusion with other diseases, such as cerebral malaria.
- Qualified personnel
- Medical and veterinary infrastructure
- Lack of reagents (biological conjugate)
- No accredited tests (for primary rabies diagnostic tests.



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Example #	Situation	Remarks
1	Brain and central nervous samples previously reported to be negative (on initial diagnosis), amplified in PCR reactions. Highly positive samples easily read under UV fluorescence, whilst low positive samples were reported as negative.	Microscope filters
2	Previously recorded (positive) samples were negative on a retest.	Phosphate buffered saline (appeared to contain detergent) and biological conjugate (shelf life)
3	100% glycerol used as mounting fluid	Glycerol concentrations >20% affect the antigen binding capacity of some antibodies.

Example #	Situation	Remarks
4	Microscope – non-functional	Introduce and adhere to ISO17025 practices
5	Positive control not stained (under ultraviolet fluorescence)	Fixation step influenced by quality of acetone.
6	False positive tests obtained in a proficiency test	Use of counterstain (Evans Blue)



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### Regional training (theory and bench) – ARC-OVI, University of Pretoria and Anses (Nancy)











### Questionnaire to identify gaps

Survey Item	Yes	No	Comment
1. Is the Rabies laboratory adhering to accreditation bodies? If yes list them.		x	New laboratory to be established but the AHI is accredited for some test methods with Ethiopian Accreditation services. The rabies diagnostic methods will be accredited once they are adopted to the laboratory especially FAT.
2. Does the Rabies laboratory have the Quality manual in place?		x	The AHI has one quality manual for all test methods, which would be amended to include the rabies laboratory. New laboratory to be established, but AHI have a quality manual.
3. Does the Rabies laboratory have a Standard operating procedure for the FAT method?	x		Still being developed.
4. Is the maintenance, servicing and calibration schedule available?	yes		The AHI has calibration schedule and critical equipment such as incubators and fluorescent microscope for earmarked for rabies laboratory are not on the list.
5. Are the Rabies laboratory personnel vaccinated against Rabies?		x	Not yet vaccinated. Waiting for the vaccine to be imported.
6. Are the activities in the Rabies laboratory performed in separate rooms?		x	So far there is a room dedicated for rabies sample receiving, processing, incubating and testing. Plans underway to demarcate the room to separate activities. Dark room available as separate room
7. Does the institute have a working incinerator?	x		
8. Equipment check list (indicate number under comment)			

**Standardise** the direct fluorescent antibody test throughout all diagnostic laboratories:

- **Fixation** of the slides left for 12/24 hours in the freezer at -20°C.
- Use of Evans blue as counterstain



SADC Harmonized SOP of the rabies fluorescent antibody test (FAT)

#### Edition 1.0

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Training – rabies diagnosis, National Veterinary Research Institute (Nigeria)



2009 – cell culture based training

2013 – PCR based training







## **Current activities of the Reference Laboratory**

- Twinning project between the ARC (Republic of South Africa) and the Animal Health Institute (Ethiopia), 2021-2022:
  - Capacity development towards standard rabies diagnosis in Ethiopia (2021-2022).
- Establishing core cells for a future South African rabies laboratory network (2022-2026):
  - Target countries are Lesotho, Eswatini, Malawi, Mozambique and Zimbabwe.
  - Will include collaborative activities and proficiency testing.
  - Understanding rabies epidemiology of rabies in the region.
- FLI (Germany)
  - Target countries Angola, Botswana, Namibia and Zambia
  - Training on dFAT and PCR conducted in Botswana on 20-23 September 2022.
- Increase the capability to diagnose rabies in animals and humans in Eritrea:
  - Support to improve diagnostic capabilities for rabies at the NAPHL in Asmara, Eritrea (2022-2023).



### **Potential for collaboration - 1**

- Mapatse et al., 2022. Seroprevalence of rabies in dogs in Limpopo National Park and phylogeny of rabies viruses in Mozambique. Pathogens. 11(9):1043. doi: 10.3390/pathogens11091043. PMID: 36145475; PMCID: PMC9506193.
  - ARC, Central Veterinary Laboratory (Mozambique) and Eduardo Mondlane University.
- Eze et al., 2020. Molecular detection of rabies lyssaviruses from dogs in South-eastern Nigeria: evidence of transboundary transmission of rabies in West Africa. Viruses. 12, 134. Doi.10.3390/v12020134.
  - National Veterinary Research Institute (Nigeria), ARC-OVI (Pretoria) and the University of Nsukka.
  - MOA signed between University of Nsukka and the ARC-OVI.
- Eze et al., 2020. Economic and feasibility comparison of the dRIT and DFA for decentralized rabies diagnosis in resource-limited settings: The use of Nigerian dog meat markets as a case study. Plos Neglected Tropical Diseases. 14(2), <u>https://doi.org/10.1371/journal.pntd.0008088</u>.
  - National Veterinary Research Institute (NVRI), ARC-OVI, University of Pretoria (Rabies Group) and the University of Nsukka.



## **Potential for collaboration - 2**

- Evaluation of Immunogenicity of a Dog Rabies
  Vaccine in young Naive Dogs in Kogom, Chaha
  and Chaha Road Communities, Vom, Nigeria.
  - NVRI and the University of Pretoria.





### Thank you for your attention



