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Laboratory Proficiency Testing For Rabies Diagnosis

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Presentation Outline

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What is Proficiency Testing

A method used to demonstrate competency and validate a laboratory's diagnostic process by comparing results of a participating laboratory to the results of a reference laboratory and those of other participant laboratories.

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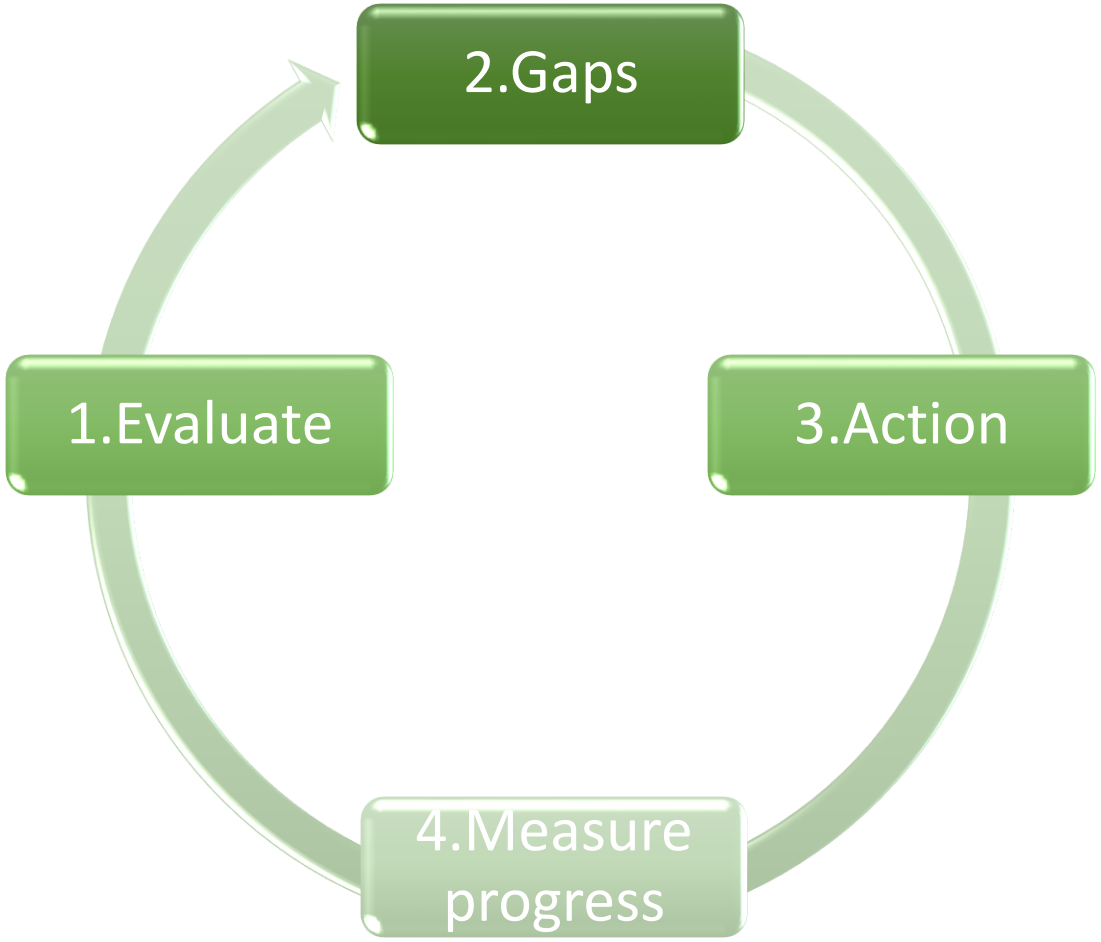
Objectives of the Proficiency testing



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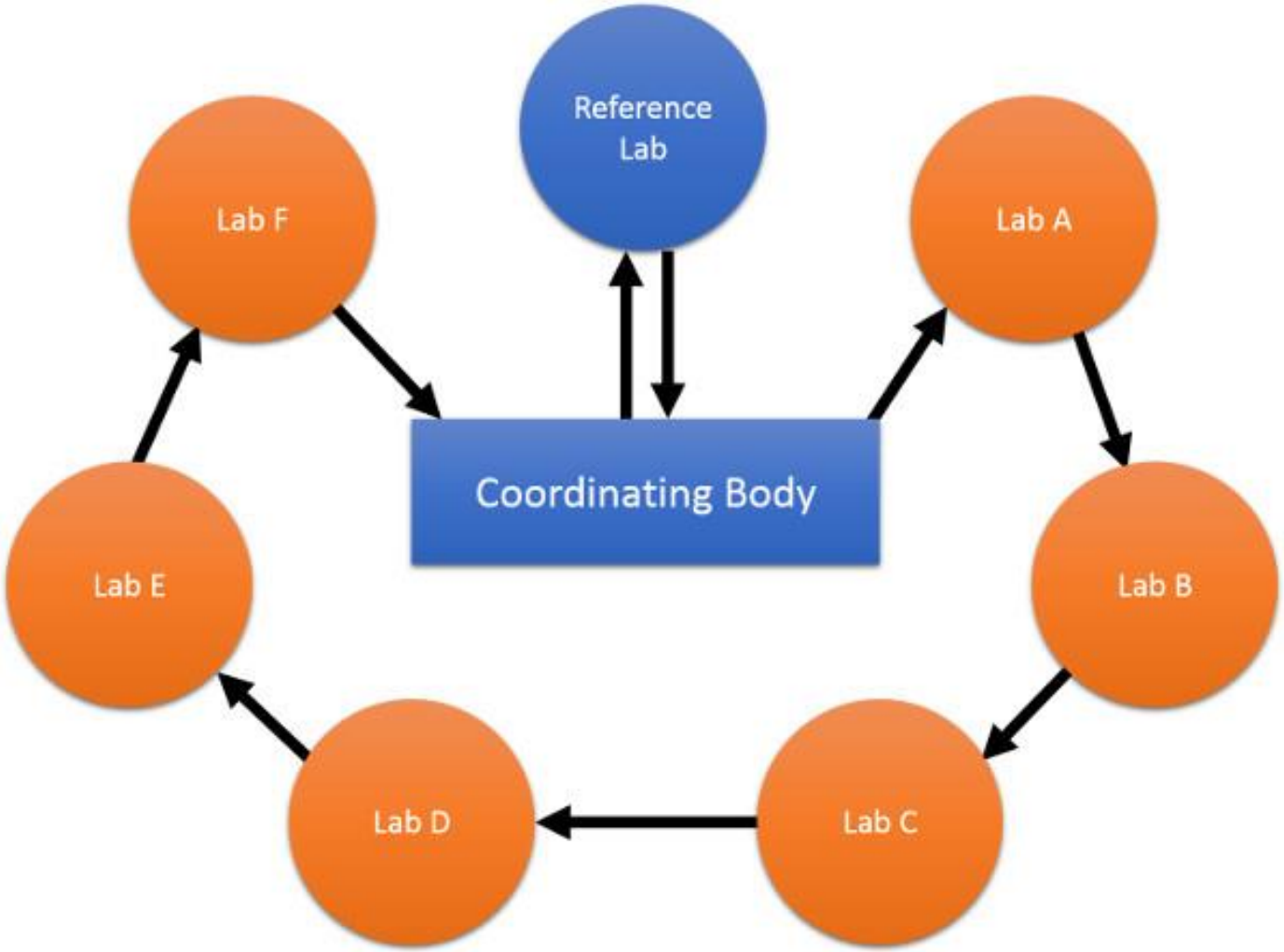
Components of a Proficiency Testing scheme



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Why is Proficiency Testing Important for Effective Rabies Prevention Detection and Control?

- Enables participating laboratory to demonstrate competency in producing accurate and reliable results

- Can also be used to validate
  - Procedures
  - Technical capacity of personnel;
  - Reliability of chosen test procedure under prevailing local conditions
  - Estimates of measurement uncertainty

- A requirement for lab accreditation

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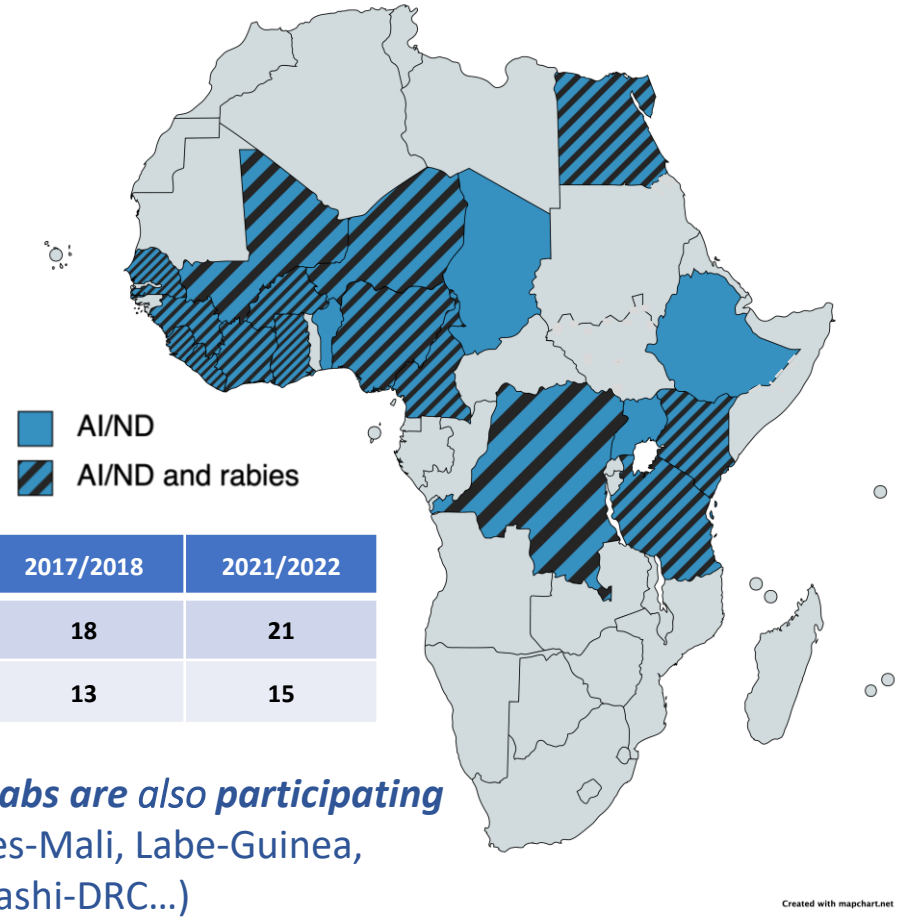
Recent Efforts to Support PTs – African schemes in 2021/2022

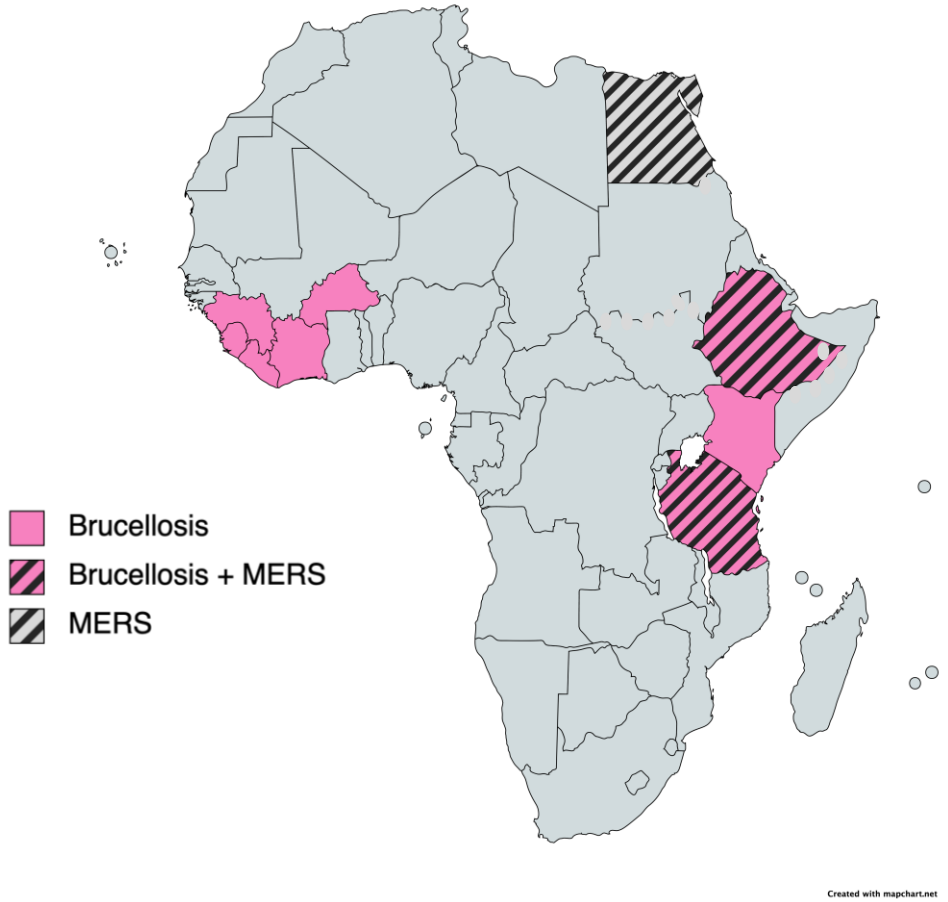
AI/ND and rabies PT schemes



	2017/2018	2021/2022
AI/ND	**18**	**21**
Rabies	**13**	**15**

District labs are** also **participating
(Eg: Kayes-Mali, Labe-Guinea, Lubumbashi-DRC…)

New PT schemes: Brucellosis and MERS



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Scope of the PT Schemes

FAO reference Centers for Animal Influenza/NDV and Rabies
WOAH and National Reference Laboratory for Avian influenza
(IZSVe- Italy)

Hong Kong University

WOAH and National Reference Laboratory for Brucellosis (IZSAM-Italy)

AI/NDV

- Serology (ELISA/HI test)
- Molecular testing (conv RT-PCR and/or Real time RT-PCR)

Rabies

- DFA
- Molecular testing (conv RT-PCR and/or Real time RT-PCR)

Mers-CoV

- Molecular testing (real-time RT-PCR)

Brucellosis

- Serology (Rose Bengal Test (RBT)
- Complement fixation test (CFT)

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Rabies PTs results in Africa 2022

Table 5. Summary of submitted results from testing the standard PT panel for Rabies PT 2022.

Laboratory code	DFA test		Conventional RT-PCR		Real-time R-PCR	
	Success rate for positive samples	Success rate for negative samples	Success rate for positive samples	Success rate for negative samples	Success rate for positive samples	Success rate for negative samples
L01	6/7	0/6	NO REPORTED RESULTS		NO REPORTED RESULTS	
L02	4/7	3/6	NO REPORTED RESULTS		NO REPORTED RESULTS	
L03	6/7	5/6	9/9	4/4	NO REPORTED RESULTS	
L06	5/7	5/6	NO REPORTED RESULTS		NO REPORTED RESULTS	
L08	7/7	5/6	8/9	4/4	NO REPORTED RESULTS	
L11	7/7	4/6	NO REPORTED RESULTS		NO REPORTED RESULTS	
L12	7/7	5/6	9/9	4/4	NO REPORTED RESULTS	
L13	7/7	6/6	NO REPORTED RESULTS		NO REPORTED RESULTS	
L14	5/7	3/3	NO REPORTED RESULTS		NO REPORTED RESULTS	
L15	NO REPORTED RESULTS		9/9	2/4	NO REPORTED RESULTS	
L17	7/7	1/6	9/9	2/4	NO REPORTED RESULTS	
L20	7/7	6/6	9/9	4/4	NO REPORTED RESULTS	
L21	6/7	6/6	NO REPORTED RESULTS		NO REPORTED RESULTS	
L22	NO REPORTED RESULTS		8/9	4/4	NO REPORTED RESULTS	
L26	7/7	6/6	7/9	4/4	NO REPORTED RESULTS	
L28	6/7	4/6	8/9	4/4	NO REPORTED RESULTS	
L29	6/7	3/6	7/9	2/4	7/9	2/4
L32	6/7	5/6	NO REPORTED RESULTS		NO REPORTED RESULTS	
Overall statistical analysis						
Concordance (%)	79.81		90.00			
Overall sensitivity (%)	88.39		92.22			
Overall specificity (%)	69.79		85.00			
Fleiss' kappa	0.40		0.60			

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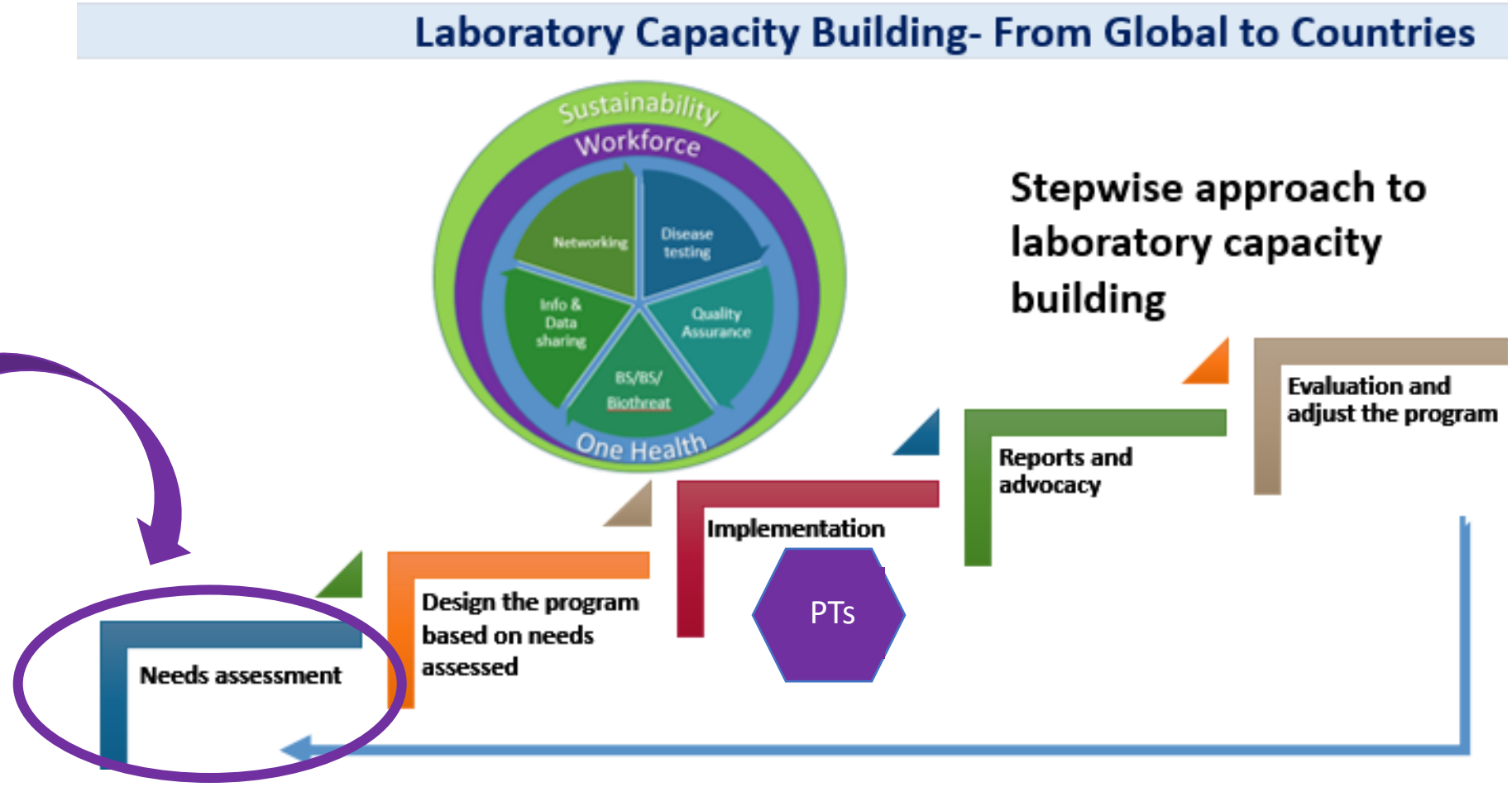
Application of PT results in stepwise approach to laboratory capacity building



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Activities Implemented After PT Results

- **Laboratory interviews:**

2.Identify gaps

- Evaluation of the results obtained
- Interview with the participating labs and discuss root cause
- Agree on the main gaps identified

3.Propose corrective actions

- Agree on the main actions and plan for next steps

Comments from IZSVe	Comments from FAO	Comments from the lab
Excellent skills in molecular testing: 3 FN, the IZSVe recommend to test these samples again.	Were the samples retested?	Due to high diagnostic activity in the lab, the staff has not had the chance to test the samples yet.
Issues with the use of inadequate molecular diagnostic reagent kits	FAO online service available to assist with primer and probes supply	Problem in the supply of the appropriate diagnostic kits (amplification kits, extraction kits….). For this reason, the change of kits is often done according to what is available. The last of couple of years, the lab managed to always analyse the samples. The same is applicable to primers and probes. To place order, the authorization of the Chief executive so that purchases can be performed.
All positive samples were detected by molecular testing, main problem is the presence of 5 FP (1 for M gene and 4 for AIV-N1)	- Cross contamination for AIV-N1 with another PT sample is not possible. - Contamination of kits or equipment (like pipettes). - Specificity of the protocol used to be assessed **Action:** The panel should be retested for M gene and AIV-N1 protocols	• probable source of contamination: centrifuge • Protocol use for N1 detection: duplex for H5N1 • CT value of the false positive results + amplification plots: to be provided by the laboratory by 12/03/2021 • H5N1 used as positive control **Action:** 1) decontamination the PCR room + equipment (SOPs to be shared with the lab – (trough regional BS/BS network via the whatsapp group) 2) Study of the data and have a following discussion on the contamination 3) re-test the samples.
Is this exercise relevant for your needs? Scope of the test, techniques used for the test, disease		The lab would request the PT AI/NDV and Rabies for the next year **Action :** FAO ECTAD country to insert these activities in the WP

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Observed Challenges

Gaps identified

- **Capacities in laboratories at the time of the PT exercise :**
  - Stock out of reference material/reagents , expired reagents/kits in some countries
  - Lack or Non functional key equipment (district laboratories), Lack of maintenance and calibration of key equipment
  - Test not routinely carried out
  - High turnover of staff proficient in the technique
  - Contamination
  - Inadequate protocols
  - Conflicting with other tasks in the lab
  - Power cut during the period of PT exercise

Corrective actions

- Provision of reference material/reagents together with the PT panels
- Assistance for procurement/servicing of equipment
- Repeat of the PTs internally
- Training, including lab directorate recalling the retired resource persons to train new staff,
- Remote assistance/troubleshooting on diagnosis testing
- Updating of SOPs, Sharing of protocols from FAO ref labs/updated SOPs
- Lab strategy for better planning with other tasks in the lab
- Install Back up generator, inverters and regulators, solar kit for securing the cold chain

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Success stories

Niger

Engagement of the government of Niger to support the national laboratory (LABOCEL) based on their high score obtained during the 2021 PT exercise.

- 100% correct results along with the confirmatory results of HPAI outbreaks by LABOCEL :

✓ Results used to advocate fundraising from Government of Niger

✓ **Additional budget allocated by vet services to the LABOCEL in 2022**

Kenya

Accreditation ISO 17025: 2017

Tanzania ISO 17025: 2017

Lab	Lab section	Procedure
CVL	Molecular	Standard Operating Procedure for detection of Avian Influenza using real time RT-PCR
CVL	Molecular	Standard Operating Procedure for detection of **Rabies** virus by using Real Time rt-PCR
CVL	Virology	Standard Operating Procedure for Direct Fluorescent Antibody Test (dFAT) for Detection of **Rabies** virus Antigen

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Thank You