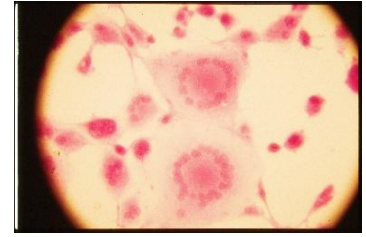


# « Diagnostic techniques for Peste des petits ruminants »



Credit photo : Abdallah Traoré



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23 to 30 November 2011

1 week

Scientific expert :  
Geneviève Libeau

## Introduction to the problem

Peste des petits ruminants is a highly infectious viral disease of small wild and domestic ruminants. In rearing zones affected by endemic PPR, the disease inflicts high economic losses due to the high morbidity and mortality rates. PPR affects nearly one billion small ruminants around the world.

The etiologic agent is PPRV belonging to the *Morbillivirus* genus, *Paramyxoviridae* family. Serological diagnosis is classically realized using competitive ELISA (cELISA). As virus isolation remains particularly tricky within a lag time of three weeks, rapid identification of the virus directly from field samples is possible using other methods including genome-based amplification, that are highly sensitive and specific. These are the conventional reverse transcription-PCR (RT-PCR) and the real time RT-PCR for quantification of viral loads. Conventional RT-PCR provides a template for sequencing and subsequent phylogenetic analysis.

## Pedagogic objectives

The training aims at providing knowledge about different methods classically implemented for serological and molecular diagnosis of peste des petits ruminants. This training will include harmonized protocols and procedures as well as equipment and diagnostic guidelines. The specific objectives are the following:

- Training in serological diagnosis of PPRV
- Knowledge about OIE reference protocols for serology (i.e. virus neutralisation and cELISA)
- Training in molecular diagnosis of PPRV
- Knowledge about different procedures of RT and rRT-PCR
- Information about molecular sequencing and phylogenetic analysis of PPRV

At the end of the training, participants should be able to establish a diagnosis in their own laboratory, reproducing the methods learned, as well to provide support to laboratory staff.

**«Cirad is a reference centre for OIE for diagnosis and control of animal diseases in tropical regions. Organizing training sessions in this field is part of the mandate»**

## **Programme**

The course will be organized and coordinated by the CIRAD-Bios, UMR15, TA A-15/G, Campus International de Baillarguet, 34398 Montpellier, France.

The programme will alternate theoretical presentations and practical training (serology, virus titration, conventional and real time PCRs).

## **Conditions**

Participants' number is strictly limited to 8.

Participants must be actively involved in animal diseases diagnosis and with experience in molecular biology techniques. A minimum of theoretical knowledge in PCR is required. **Please fill out the enclosed test : "Practical evaluation"**.

## **Training costs**

Pedagogic : 1 300 €  
Travel towards Montpellier : to be determined by participant  
Housing expenses (about) : allow a minimum of 80 € a day

If necessary, a customized estimate can be established upon request.

## **Note :**

CIRAD does not deliver fellowships or travel grants. It is your responsibility to fill a request as soon as possible to:

- national authorities dealing with livestock and/or grants ;
- Cooperation and Culture Services of French embassies and consulates ;
- other foreign countries embassies ;
- international organisms (FAO, UNPD, European Union, IAEA, IDB, ...) ;
- development projects or non governmental organizations.

## **Application procedure**

Applications, consisting of a detailed resume, a motivation letter and details about the organization managing your grant, must be sent as much as possible before **October 1st, 2011**, to the course secretary at the following address :

**CIRAD Enseignement en Elevage et Médecine Vétérinaire Tropicale  
TA A-15/B  
Campus international de Baillarguet  
34398 MONTPELLIER Cedex 5  
France**

Tel : 33 (0) 4.67.59.39.02  
Fax : 33 (0) 4.67.59.37.97  
E-mail : [marie-caroline.estienne@cirad.fr](mailto:marie-caroline.estienne@cirad.fr)

**PPR TRAINING:  
QUESTIONNAIRE FOR PRACTICAL EVALUATION <sup>1</sup>**

I- Do you have any skill in Molecular Biology?

- YES
- NO

If yes, time of experience (in months): .....

II- Which type of equipment are you used to?

- Brand and type of machine: .....
- No

III- Are you regularly using PCR for diagnosis?

- YES
- NO
- If Yes,  
For which disease? .....
- Which gene(s) do you amplify? .....

IV- Give one of the principal advantages or drawbacks of conventional PCR?

- Drawback: .....
- Advantage: .....

V- Paul received in the laboratory 4 samples suspected to be contaminated by *Mycoplasma pneumoniae*. He prepared a PCR mix and implemented the PCR according to the following Table:

<b>PCR MIX for 1 reaction</b>		<b>Conditions of PCR reaction</b>		
PCR Buffer 10X	5µl	94°C	2min	1 cycle
Mix dNTP Mix containing 10mM of each dNTP):	0,5µl	94°C	30sec	} 30 cycles
Specific forward primer 20µM	1µl	55°C	30sec	
Specific reverse primer 20µM	1µl	72°C	30sec	
Taq DNA polymerase enzyme	0,5µl			
DNA	2µl	72°C	7min	
H2O	40µl	4°C	over night	

After migration of 10 µl of the PCR reaction, results remain negative, including the positive control. In place of Paul what would be your first hypothesis to explain this unexpected result?

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<sup>1</sup>Delete or fill if necessary