



Validation report **IDvet BLV AGID**

Agar gel immunodiffusion (AGID) test for the detection of BLV anti-gP51 antibodies in bovine serum

- **Bright, distinct precipitation lines facilitate test interpretation**
- **Standardised against the European Standard E05 serum**

Introduction

Bovine leukemia virus (BLV), an oncogenic retrovirus, is widely distributed and endemic in many cattle herds. The most important disease caused by BLV is enzootic bovine leukosis, essentially a form of malignant lymphoma (ML). ML develops in only a small percentage of cattle infected with BLV, but it is a fatal disease characterized by lymphomatous involvement of multiple organs. Horizontal transmission of BLV is probably the primary means by which cattle become infected, although both vertical and vector transmission can also occur.

The agar gel immunodiffusion (AGID) test is an antibody detection method widely used for detection of BLV antibodies in serum samples. It is simple and easy to perform, and has proven to be highly useful in eradication schemes.

The **IDvet BLV AGID** uses a BLV glycoprotein gP51 antigen to detect anti-gP51 antibodies in bovine serum.

Kit components	
BLV gp51 antigen	2 ml
Positive control	3 ml
Agar gel*	200 ml

* IDvet also offers a kit without agar gel, product code: BLV-AGID-NOGEL.

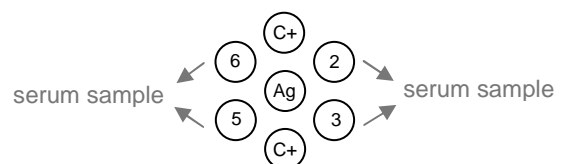
Test Principle

The agar gel immunodiffusion (AGID) test is a method whereby antigen and antibody diffuse toward each other in a semi-solid medium to a point where the optimum concentration of each is reached. A band of precipitation occurs at this point.

In this test:

- Agar gel is cast and equidistant wells are cut out in agar.
- BLV antigen is placed in the central well, positive control and samples to be tested are placed in the peripheral wells (see schema).
- After diffusion, the complex BLV antigen – anti-gP51 antibodies leads to the formation of precipitates (a whitish line visible to the naked eye).

After 24-48 hours, plates are examined but no final results can be obtained before 72 hours. Samples are considered positive if a precipitation line forms with the antigen.



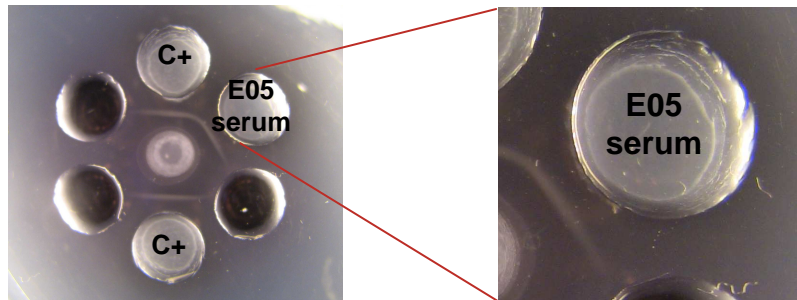
Storage conditions:

The antigen and the positive control are freeze-dried and may be stored:

- at 5°C (±3°C) in the dark until the expiry date
- or reconstituted for 3 days at 5°C (±3°C) or 1 year at ≤ -16°C.

Analytical sensitivity

The test is standardised against the OIE reference serum E05^{1,2}.



Results (Figure 1):

- ▶ The E05 serum standard was found positive each time it was tested.

Sensitivity

25 bovine sera from infected herds in Italy (Peruggia) were tested. These sera gave positive results with the ID Screen[®] BLV Competition ELISA.

Results:

- ▶ All sera gave positive results and showed clearly precipitation.

Specificity

220 bovine sera from disease-free certified herds from France (Hérault and Brittany) were tested.

Results:

- ▶ All sera were found negative.
- ▶ Measured specificity = 100% (CI_{95%}: 98.28-100%), n=220.

Conclusion

The **IDvet BLV AGID**:

- ▶ is standardized against the European Standard E05 serum
- ▶ demonstrated high sensitivity
- ▶ shows excellent specificity on disease-free sera

References

- (1) OIE Manual of Diagnostic Tests and Vaccines for Terrestrial Animals 2012. Chapter 2.4.11: Enzootic Bovine Leukosis.
- (2) European Commission 2009. Commission Decision of 15 December 2009 amending Annex D to Council Directive 64/432/EEC as regards the diagnostic tests for enzootic bovine leucosis (2009/976/EU): Official Journal of the European Union L 336, 36-41.