Instruction Manual

VDPro® BLV AGID Reagent

CAT. NO. RB-BLV-41

1. INTRODUCTION

Agar gel immunodiffusion (AGID) reagents for the Bovine Leukemia Virus (BLV) is for the detecting antibody in serum samples from BLV-infected cattle. We used BLV antigen derived from persistently BLV-infected cell lines.

2. CONTENTS

Reagents		100 tests
1)	BLV AGID Antigen (Lyophilized)	1.0㎖ X 1
2)	Agar Gel	100㎖ X 1
3)	BLV Positive control (PC)	4.0㎖ X 1
4)	User Manual	1 copy

3. MATERIALS

1) A Glass with 90mm diameter or a Plastic Petri dish
2) Agar cutter:
   4mm central well of one well and 6mm hexagonal arrangement of six wells (Refer to Diagram 1)
3) Micropipette (10-100 ㎕)

4. AGAR GEL PREPARATION

1) The agar gel bottle is melt by heating in 100 ℃ water bath to completely dissolve it.
2) The dissolved agar gel bottle is allowed to 56 ℃ water bath for 30 minutes.
3) Pour into the 90mm Petri dish on the horizontal testing stand and the Agar gel each at 19 ㎖ so the height is 2.6mm.
4) The poured plates are allowed to cool at 1 hour in 4~5℃ for about 1 hour so the agar gel is completely solidified.
5) Make 7 wells by using the Agar cutter for the testing.

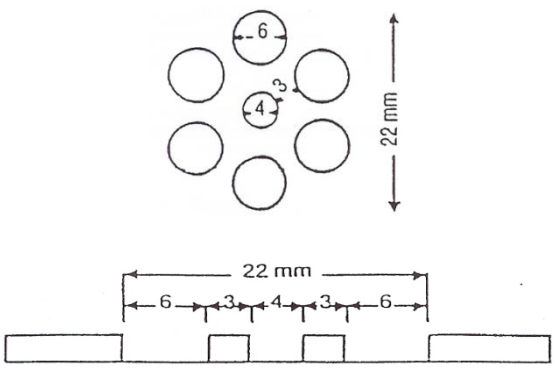


Diagram 1. BLV AGID OIE Standard Manual

5. PREPARATION OF ANTIGEN

Add 1.0 ㎖ of distilled water into the lyophilized BLV AGID antigen vial and the antigen is completely dissolved before the tests.

- ❖ After dissolved, store the dissolved antigen in conditions that are lower than -20℃ .
- ❖ Lyophilized antigens can be stored in refrigerating conditions.

6. TEST PROCEDURE

Mark the Gel Plate like the below Diagram 2 image and afterwards, use the micropipette to put in the antigen and samples.

- Put in 73㎕ of serum in the No. 2, 3, 5 and 6 wells.
- Put in 73㎕ of Positive Control (PC in No. 1 and 4 wells.)
- Put in 32㎕ of BLV AGID antigens in the center Ag well.

In the humidity box, put in a Gel plate and have it react at least for 72 hours to conduct the decode. (20℃ ± 5 ℃)

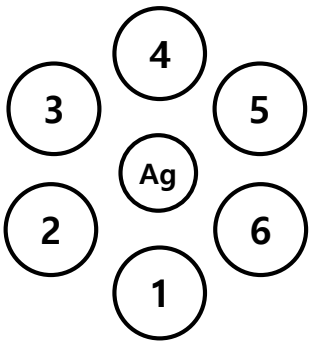


Diagram 2. BLV AGID Testing Diagram

Caution

1) After the BLV AGID antigen is dissolved and have it stored in a less than -20℃ freezer.
2) If freezing and thawing is repeated, the antigen titer can easily decline.
3) Agar Gel plate made right before the test or use it at least it within 24 hours.

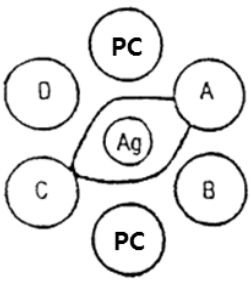
8. INTERPRETATION

1) By the OIE Standard Manual, the results can be read after 24 hours and results can be confirmed after 48 hours, which is before 72 hours.
2) If the positive control precipitation line shows to coincide and the tested serum show a white precipitation line, it can be judged the tests were positive.
3) If the positive control precipitation line is shown but not founded in the tested serum's precipitation line, the test can be judged to be a negative.
4) If a precipitation line cannot be found in the positive control, the testing will need to be repeated.

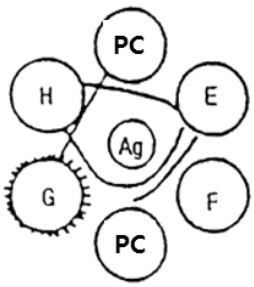
❖ Using a bright light and putting the Agar plate on a black-colored paper can make the reading process more easy.

❖ Refer to the BLV AGID reaction samples

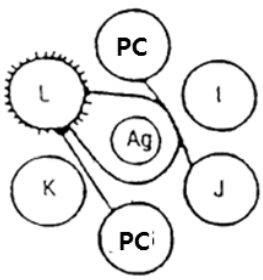
Reference Data: Samples of reading BLV AGID Results



PC: One precipitation line is founded in the positive control serum
A: Negative serum (-)
B & D: Positive serum (+)
C: Weak positive serum (weak positive)



PC: One precipitation line is founded in the positive control serum (gp51)
E: Doubtful serum
F: Positive serum (2 precipitation lines were founded, P24 and gp51)
G: Positive serum
H: Negative serum (Nonspecific precipitation line that has no relations with the positive control)



PC: One precipitation line is founded in the positive control serum(gp51)
I: Positive serum (2 precipitation lines were founded. P24 and gp51)
J: Positive serum
K: Positive serum (1 precipitation line)
L: Negative serum

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