

## DEVELOPMENT AND APPLICATION OF LATEX AGGLUTINATION TEST FOR DETECTION OF INFECTIOUS BURSAL DISEASE VIRUS ANTIGEN

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### ABSTRACT

The agar gel precipitation test (AGPT) is the most useful and recommended test for detection of viral antigens in bursal tissues. Though AGPT is simple to perform, however, results can only be read after about 48 h of incubation. Therefore, it was used as a reference test and compared with latex agglutination test (LAT) developed in the present study for viral antigen detection in experimentally infected broiler birds, natural cases suspected for infectious bursal disease as well as for detection of cell culture adapted infectious bursal disease virus (IBDV). Day-old broiler chicks in the IBD-vH, IBD-vI and IBD-F groups were inoculated with hot IBDV vaccine strain, intermediate IBDV vaccine strain, and chicken embryo fibroblast culture adapted field isolate of IBDV, respectively. LAT was found to detect IBDV antigen in bursa in IBD-vH and IBD-F groups as early as on 6<sup>th</sup> day post inoculation. IBDV antigen in bursa could be detected up to day 22<sup>nd</sup> in IBD-vI and IBD-vH bird groups. All the three IBD virus inoculated groups were positive for antigen detection in bursa by LAT on 10<sup>th</sup> and 15<sup>th</sup> day post inoculation and by AGPT on 15<sup>th</sup> and 22<sup>nd</sup> day. Out of 46 field bursa samples, AGPT detected 23 (50%) as positive, whereas LAT was able to detect 27 (58.7%) as positive for IBDV antigen. The sensitivity and specificity of LAT as compared to AGPT were 91.30% and 73.91%, respectively. The positive predictive value, negative predictive value, diagnostic accuracy of LAT were 77.78%, 89.47%, and 82.61%, respectively. The sensitivity of LAT was to detect up to 2.69 log<sub>10</sub> TCID<sub>50</sub> titre. The coated latex beads were stable up to 9 months when stored at 4°C in a refrigerator.

**Key words:** Latex agglutination test, infectious bursal disease virus, agar gel precipitation test

Infectious bursal disease (IBD), caused by infectious bursal disease virus (IBDV), is one of the most important diseases of poultry in more than 95% countries of world including India. Presumptive diagnosis of acute clinical outbreaks of IBD can be made on the basis of age of the birds and gross lesions. Laboratory diagnosis can be made by isolation of IBDV from bursa and spleen in chicken embryo fibroblast (CEF) or BGM 70 cell cultures or embryonated chicken eggs by yolk sac or chorioallantoic membrane (Hitchner, 1970). Detection of IBDV is performed by virus neutralisation, immunofluorescent test, agar gel precipitation test (AGPT) (Hirai *et al.*, 1972), enzyme linked immunosorbent assay (ELISA) (Snyder *et al.*, 1988), dot-ELISA and counter immuno electrophoresis (Gowri *et al.*, 1997a), coagglutination test (Shakya and Joshi, 1997). OIE (2004) has recommended AGPT or ELISA for detection of IBD viral antigens in bursal

tissues. Although AGPT is simple to perform, however, results can only be read after 48 h of incubation and the sensitivity is low. Other techniques have high sensitivity but are expensive, cumbersome and time consuming. The present study describes the development of latex agglutination test (LAT) and its comparison with AGPT for detection of IBDV antigen.

### MATERIALS AND METHODS

**Viruses:** Commercial Intermediate IBDV vaccine strain (IBD-vI), hot IBDV vaccine strain (IBD-vH) and La Sota strain of New Castle Disease virus (NDV-L) were purchased from the local market. A field isolate of IBDV (IBD-F) isolated from field outbreak was obtained from Head, Department of Veterinary Microbiology, PAU, Ludhiana and was passaged further in chicken embryo fibroblast culture before use.

**Antiserum:** Hyper immune serum raised in chickens against IBD-vI was used in the present

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