SEROLOGICAL METHODS FOR INFECTIOUS BRONCHITIS IN LAYING HENS

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Summary

Measurements of infectious bronchitis virus (IBV) antibody titres by IDEXX IBV antibody ELISA were compared with the results of agar-gel precipitation (AGP) and serum neutralisation (SN) tests. The percentage of samples testing positive by AGP and SN increased as the ELISA IBV antibody titre increased. Although it is not clear at what antibody titre level birds are protected against intercurrent infection, a mean IBV antibody titre measured by IDEXX ELISA of 439 correlated with a high level of protection against exposure to T-strain IBV. A comparison of two different IBV antibody ELISA kits showed a significant linear correlation although individual samples did not always correlate closely.

I. INTRODUCTION

Infectious bronchitis virus (IBV) and its deleterious effects were first recognised in the United States in the early 1930s (Cavanagh & Naqi, 1997) and the presence of this virus in Australian flocks was first identified by Cumming (1963) as the infectious agent associated with respiratory disease and uraemia. All layer flocks are vaccinated against IBV although it is recognised that drops in egg production and reduced internal quality and egg shell quality may occur as the result of exposure of vaccinated flocks. It is essential that the Australian egg industry has tools for surveillance of layer flocks for effects of IBV. The level of protection afforded by vaccination is usually measured in the form of IBV antibody titres. The most common method of doing this is to use commercially available IBV antibody ELISA kits. There are currently three available: KPL ProFlok; IDEXX and TropBio. The TropBio kit is the only one that was developed specifically for Australian conditions. However, the other two kits are also in common commercial use, especially IDEXX. Other methods of measuring IBV antibody titres include serum neutralisation (SN), agar gel precipitation (AGP) and haemagglutination inhibition although these tests are not always readily available commercially and are more time-consuming and expensive than ELISA. A number of studies have compared different methods for suitability. Monreal et al. (1983) concluded that ELISA was more suitable for monitoring antibody responses to vaccination than HI and AGP because of its higher sensitivity. A preference for ELISA, as compared with other tests, has been expressed by a number of other authors (Mockett & Darbyshire, 1981; de Wit et al., 1992). However it is important to correlate the IBV antibody titres obtained using ELISA with other ways of measuring IBV antibody levels and also with the level of protection of the bird to IBV challenge.

This study compared the IBV antibody titres obtained using an IDEXX IBV antibody ELISA kit (quantitative) with the positive/negative results obtained from AGP and SN. It also compared the results of two ELISA kits, IDEXX and TropBio.

II. MATERIALS AND METHODS

Plasma samples were obtained from a trial as reported previously (Sulaiman et al., 2003). Samples were taken from the same 70 birds on 12 occasions over the laying life of the flock. Some samples were negative for IBV antibodies (unvaccinated birds at weeks 4 and 6).

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whereas the samples taken at 79 and 80 weeks of age followed challenge with T-strain IBV at 77 weeks of age. Although one group of birds had not been revaccinated regularly during lay, all birds were vaccinated with VicS strain IBV at 62 weeks of age, 15 weeks prior to the exposure to T-strain IBV.

IBV antibody titres were measured using an IDEXX IBV antibody ELISA kit and were conducted by Birlong Avian Laboratories. AGP tests, which give a positive/negative result, were conducted after the method of Woernle (1966) and Chubb and Cumming (1971). SN tests were conducted after the method of Fabricant (1951) to identify positive or negative only.

A series of plasma samples, again ranging from negative for IBV antibodies to high positive, were obtained from a separate experiment in which unvaccinated birds were exposed to challenge viruses. The IBV antibody titres were determined on these samples, by the International Avian Health Group at the University of Melbourne, using both IDEXX and TropBio ELISA kits. The data were then compared.

III. RESULTS

The IBV antibody titres obtained for all the plasma samples of the main experiment are shown in Figure 1 and are separated into those from birds that were revaccinated regularly during lay and those that were not. The comparison of IBV antibody titres measured by IDEXX IBV antibody ELISA and the positive/negative results obtained from AGP and SN tests is shown in Figure 2. The ELISA antibody titres have been grouped to allow for the calculation of percentage positive results for AGP and SN. The sample sizes for the groupings are: 0-100, 33 samples; 101-200, 34 samples; 201-500, 48 samples; 501-1000, 38 samples; 1001-2000, 35 samples; 2001-4000, 30 samples; 4001-6000, 21 samples; 6001-8000, 19 samples; 8001-15000 12 samples. There is a general increase in the percentage of samples testing positive for AGP and SN, as the ELISA antibody titre increases. The mean IBV antibody IDEXX ELISA titre of the birds, just prior to exposure to T-strain IBV was 439. This titre level correlated with a high level of protection of the birds (see Sulaiman et al. in this volume).

A comparison of the results of IBV antibody titres obtained, using both IDEXX and TropBio ELISA, on the same samples, is shown in Figure 3. Although there is a statistically significant linear relationship between the two sets of titres (P=0.0003), the fit is not close (R^2=0.301) and there is not always a good match for individual samples.
IV. DISCUSSION AND CONCLUSIONS

The different methods of measuring IBV antibody levels actually measure different types of antibodies. Therefore, it is not surprising that the results obtained are not identical. However, it is very important for the egg industry to have a reliable means of determining if flocks are adequately protected against the possibility of an IBV intercurrent infection. At
the present time, it is difficult for producers to interpret the IBV antibody titres obtained from ELISA testing. There is also some uncertainty about which ELISA kit is most appropriate under what circumstances. Further information is required to address the need for better surveillance against IBV in layer flocks.

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REFERENCES