How to Use a Quantitative Turbidimetric Immunoassay Assay to Determine Immunoglobulin G Concentrations in Neonatal Foals

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1. Introduction

Newborn foals are born immunocompetent but without circulating immunoglobulins. It is crucial that the newborn foal receives immunoglobulins from the mare’s colostrum to prevent infectious disease during the first few months of life. Healthy foals ingesting good-quality colostrum will typically have immunoglobulin G (IgG) levels >800 mg/dl at 24 h of life, which is considered adequate passive transfer of immunity. Failure of passive transfer (FPT) is commonly defined as an IgG level <400 mg/dl after 24 h of age. Partial failure of passive transfer (PFPT) is defined as an IgG level between 400 and 800 mg/dl at 24 h of age.

The incidence of FPT in foals is 10–20%, and the incidence of PFPT in foals 17–19%. Risk factors for FPT include ingestion of colostrum with a low level of immunoglobulin, failure to ingest a sufficient quantity of colostrum, and failure to absorb colostral immunoglobulins from the gastrointestinal tract.

Success of passive transfer should be measured early in life so that it is possible to intervene if needed. An ideal screening test would be fast, quantitative, accurate, and inexpensive. Screening tests available for evaluation of passive transfer are single radial immunodiffusion (SRID), glutaraldehyde coagulation, zinc sulfate turbidity, latex agglutination, enzyme immunoassay, turbidimetric assay (TIA), and serum total protein.

The TIA is an excellent technique for screening foals for FPT. The TIA uses antibodies directed against an antigen to form immune complexes, which, in this case, is equine IgG. The amount of anti-equine IgG antibody in the assay is constant, and therefore, the number of immune complexes that form is proportional to the concentration of antigen (equine IgG) in the foal blood sample. A TIA can yield quantitative results by evaluation of the turbidity of a test sample using a calibrated spectrophotometer.

The objective of this paper is to describe the use of a new quantitative turbidimetric immunoassay for measuring concentrations of serum or plasma IgG in neonatal foals in veterinary practice.
2. Materials and Methods

Equipment

Application of this assay requires (1) a calibrated spectrophotometer and software, (2) pipetter, and (3) a test kit (Fig. 1). The spectrophotometer used for this assay was originally designed to estimate sperm concentrations and was later adapted for estimation of IgG. Consequently, this devise may already be available at many breeding farms and veterinary clinics.

Accuracy of the Assay

The turbidimetric immunoassay has a high correlation to the single radial immunodiffusion test, which is considered the gold standard for IgG determination.18 At an IgG concentration of 400 mg/dl, TIA sensitivity was >90%, specificity was 99.1%, positive predictive value was 98.1%, and negative predictive value was 96.4%.18 At an IgG concentration of 800 mg/dl, TIA sensitivity was >90%, specificity was 70.5%, positive predictive value was 71.5%, and negative predictive value was 91.1%.18 The assay reports values up to 2000 mg/dl. Samples with higher IgG levels are reported as >2000 mg/dl.

How to Implement the Assay Into Your Practice

A blood sample should be collected for analysis of adequate passive transfer as part of a foal wellness examination that is performed when the foal is 12–24 h old. Almost all tests used to evaluate foal IgG concentrations are able to give satisfactory results when the values are >800 mg/dl. However, the TIA stands out by being able to quantitatively differentiate IgG concentrations <800 mg/dl. The TIA allows practitioners to make fast, accurate, and informed decisions about how to treat FPT and PFPT.

3. Results

The turbidimetric immunoassay is used at our clinic to measure foal IgG levels at 12 and/or 24 h of life. The measurement of a foal’s IgG level at 12 h of age allows for the ability to supplement with frozen-thawed colostrum through a bottle or nasogastric tube. This has decreased the need for administration of IV hyperimmune plasma to foals for FPT or PFPT. Any foal at 12 h of age with an IgG concentration <400 mg/dl is routinely administered frozen-thawed colostrum or an equine IgG product by nasogastric tube. Foals with an IgG level between 400 and 800 mg/dl are also often administered frozen-thawed colostrum or an equine IgG product by nasogastric tube.

The use of the turbidimetric immunoassay in our clinic has helped to improve the care for the newborn foal by being able to accurately diagnose and treat FPT. A comparison of the results of TIA and SRID tests is seen in Table 1. A foal born at our clinic was examined at 12 h of age during a routine wellness exam. From the TIA, it was observed that the foal had PFPT (472 mg/dl). Frozen equine colostrum and an equine IgG product were administered by nasogastric tube. A second blood sample collected at 24 h of age showed a blood IgG value of 871 mg/dl. All blood samples were subsequently submitted for SRID test.

4. Conclusion

The turbidimetric immunoassay is a rapid, quantitative, and accurate technique to evaluate foal IgG levels. The use of the turbidimetric immunoassay in our clinic has led to a higher level of care for newborn foals through early detection and more sensitive monitoring of FPT or PFPT.
Table 1. Comparison of Turbidimetric Immunoassay and Single Radial Immunodiffusion

<table>
<thead>
<tr>
<th>Clinical Case</th>
<th>TIA</th>
<th>SRID</th>
<th>Comments</th>
</tr>
</thead>
<tbody>
<tr>
<td>12 h of age</td>
<td>472 mg/dl</td>
<td>495 mg/dl</td>
<td>Administration of oral frozen colostrum and equine IgG substitute</td>
</tr>
<tr>
<td>24 h of age</td>
<td>871 mg/dl</td>
<td>847 mg/dl</td>
<td></td>
</tr>
</tbody>
</table>

Results of TIA versus SRID at 12 and 24 h of age. The foal was supplemented with oral frozen colostrum and equine IgG substitute at 12 hours of age.

References and Footnotes


*Foal IgG Test, Animal Reproduction Systems, Chino, CA 91710.
ARS densimeter Model 590a or 591 with Foal IgG software, Animal Reproduction Systems, Chino, CA 91710.
ARS 180 ml positive displacement pipette, Animal Reproduction Systems, Chino, CA 91710.