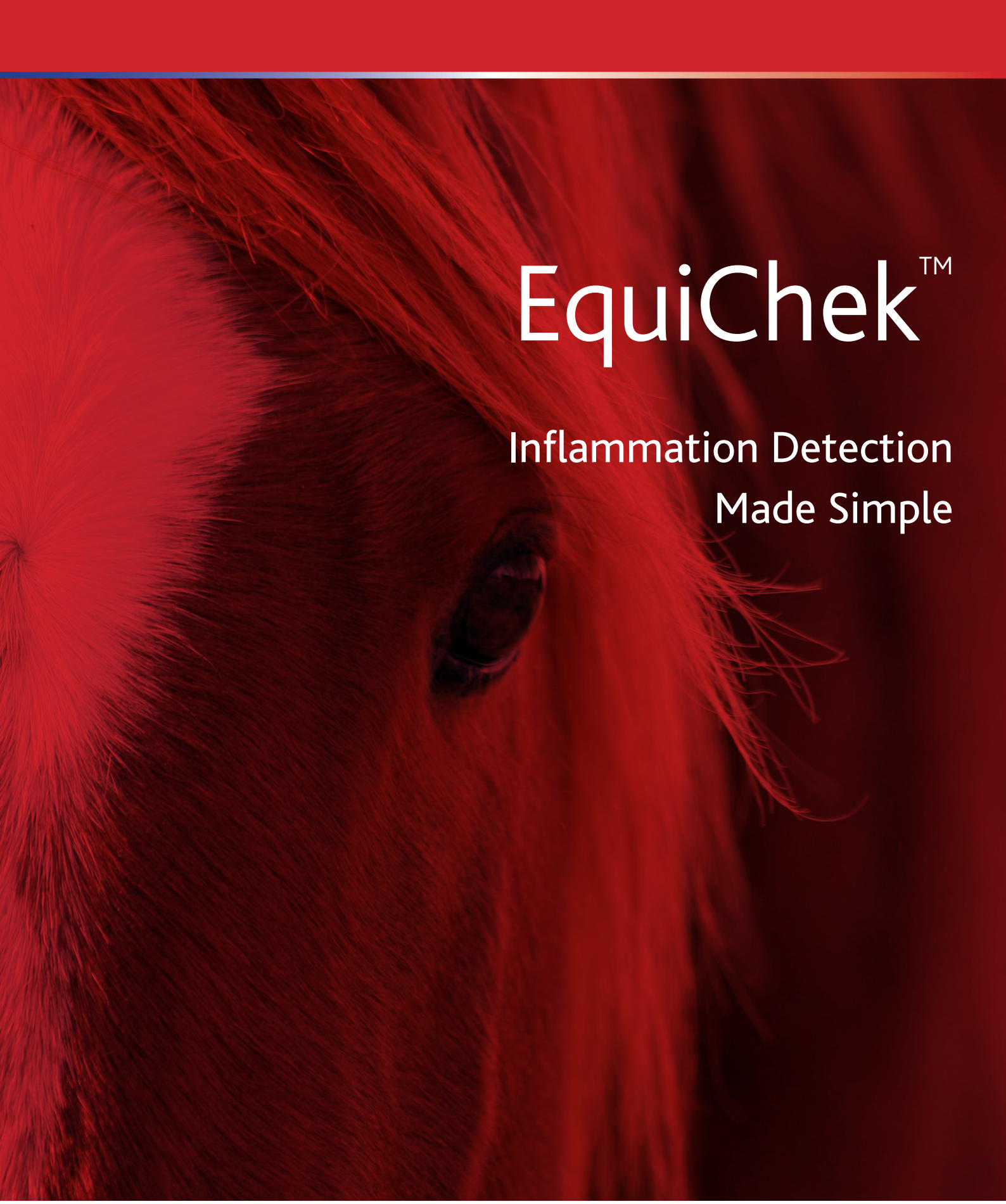


EquiChek™

Inflammation Detection Made Simple

INVICTA ANIMAL HEALTH

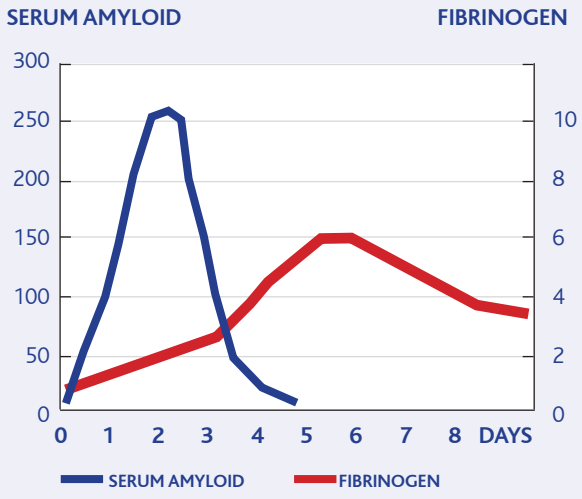
Testing for Inflammation in Horses

The role of Acute Phase Proteins (APPs)

The use of APPs to aid diagnosis of equine conditions is well documented[1,2,3]. However, the need for investment in laboratory equipment and the time taken to gain results have restricted their use for the ambulatory practitioner.

Major or Minor?

APPs can be classed as either major or minor depending on the level and timing of response. A major APP can often show elevated levels before clinical signs are observed.



Serum Amyloid A

Serum Amyloid A (SAA) is a major acute phase protein of inflammation in horses. Very low levels are seen in normal healthy conditions but it increases within hours of a problem to levels 100-1000 fold above normal. The rise in SAA during an inflammatory response is unambiguous. SAA increases are high and rapid. Many clinical and subclinical conditions will result in a significant SAA response making the diagnosis clear cut.

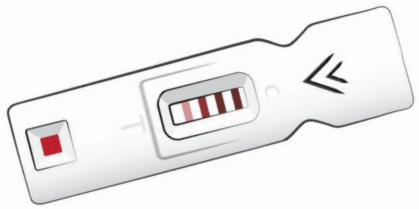
In contrast, fibrinogen is a minor acute phase protein in horses, with reference levels in most labs ranging between 100-400[4] or 200-400 mg/dl[5,6] and only rising 0.5-2 fold above normal. The increase in SAA is substantially higher, and hence more reliable. A recent publication[3] concluded that the relatively wide reference interval for fibrinogen concentrations in healthy horses and lengthy response period after an inflammatory stimulus, have rendered fibrinogen a fairly insensitive indicator of inflammation. Hence, a horse with an active inflammatory condition can have an increase in fibrinogen but still be within the normal range given the small increases that occur.

A recent study[1] by a specialized equine lab in Miami, in 212 horses, demonstrated a clinically significant and very distinct increase in SAA for horses with active inflammatory conditions.

Importantly SAA alone indicated several horses with sub-clinical conditions and these horses all developed clinical symptoms in following days, neither fibrinogen, nor white blood cells were raised. The reliability of fibrinogen was further compromised by high levels in some normal healthy horses.

The Road to Recovery

Monitoring recovery requires knowing when to administer and when to cease administration of therapeutics. With inflammation, it is essential to know when the horse has recovered to avoid unnecessary use of therapeutics. With fibrinogen the levels remain elevated well after the resolution of a problem, as long as two weeks after the horse has returned to normal. A very significant advantage of SAA is the rapid fall in levels to normality following successful intervention[7,8], giving a clear signal of recovery.



EquiChek™ SAA Test

The test is simple to use with results in less than 15 minutes. A semi-quantitative visual readout indicates whether the horse has normal, moderate or clinically significant inflammation.

- Confirming the presence of an active inflammatory condition in the field or at the stable
- Detection of sub-clinical inflammation where there is a suspicion something is wrong
- Real time monitoring recovery of disease activity
- Real time monitoring following therapeutic intervention
- Pre-breeding check for inflammatory status (e.g. detecting endometritis; high SAA is strongly correlated with high incidence of early embryonic loss)
- Detection of sub-clinical inflammation due to trauma as a result of over training
- Monitoring health status before an event to assess the horses ability/potential to perform at peak level
- Assessing whether an event horse is suffering from inflammation
- Assessing horse health prior to or after transportation
- An indicator to prompt for more detailed diagnostic testing

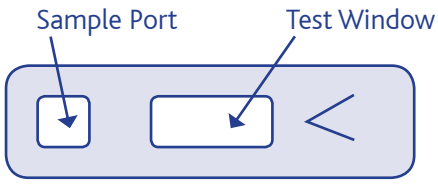
EquiChek™ uses a novel competitive assay format to detect SAA in whole blood. The test delivers a semi-quantitative visual read out to help distinguish between normal and mild to clinically significant inflammation. The test is based on competition between SAA present in a sample and SAA printed onto the test strip with antibody coated gold nanoparticles.

Instructions for use:

1. ***Remove the EquiChek kit from foil pouch:***

2. ***Take blood sample:***
The test is designed for use with freshly taken whole blood. Blood should be taken from the jugular vein, with a blood lancet or blood can be collected using a syringe. Touch the blood applicator to the blood and it will be drawn up into the applicator (do not squeeze). Stop when the blood reaches the black line on the applicator.

3. ***Apply sample to the Sample Port:***
Squeezing the end of the blood applicator will deliver the appropriate amount of blood to the EquiChek kit.



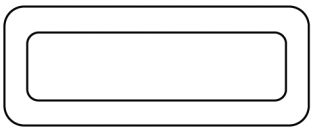
4. ***Add 2 drops from the Diluent bottle to the Sample Port.***

5. ***Leave 15 minutes and you will see the result in the Test Window.*** It is recommended that the test is read after 15 minutes, however, a normal result can be seen within 5 minutes.

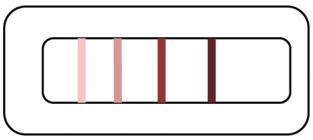
Interpretation of Results

The intensity of the colour on each line will vary depending on the level of SAA in the blood sample.

Invalid test

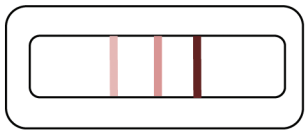


Normal



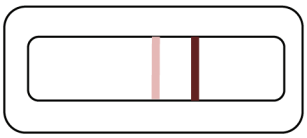
Normal or very low levels of SAA will give 4 lines.

Mild Inflammation
(Guide 20-75µg/ml SAA)



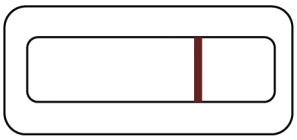
In a horse with mild inflammation, SAA in the sample prevents colour developing on the weakest test line so that only 3 lines will be visible.

Moderate Inflammation
(Guide 75-200µg/ml SAA)



As the severity of the inflammation increases further, so will the level of SAA. The second test line will get progressively weaker in colour so that only two lines become visible indicating the horse has moderate inflammation.

Clinically Significant
Inflammation
(Guide >250µg/ml SAA)



With clinically significant inflammation, only one line (the control line) will be visible.

EquiChek kit storage: 6°-30°C. Avoid direct sunlight.

Precautions: Use only horse whole blood. Do not use EquiChek kit for any other purpose. Use only the diluent proved and add to the Sample Port or the kit will be rendered ineffective. All specimens should be considered potentially hazardous and handled as an infectious agent. Do not use this beyond the expiry date. Do not use if foil pouch is damaged, already opened, or has been exposed to moisture before use. Single use only.

References

1. Assessment of Serum Amyloid A testing and its clinical application in a specialized equine practice. In press, JAVMA 2013.
2. Dynamics in serum of the inflammatory markers Serum Amyloid A, haptoglobin, fibrinogen and alpha 2 globulins during induced non infectious arthritis in the horse. Equine Vet J, 34: 699-704, 2002.
3. Blood proteins and inflammation in the horse. Vet Clin of North America; Equine Practice 24;285-297, 2008.
4. Why perform the CBC, and how can the information be used to manage cases. AAEP Proceedings, 45: 285-290, 1999.
5. Measurement of equine fibrinogen using the Abaxis VetScan VSpro analyzer. Special Equine section, 14-22, 2012.
6. Serum Amyloid A as an aid to management of infectious disease in the foal: comparison with total leukocyte count, neutrophil count and fibrinogen. Equine Vet J. 34:693-698 , 2002.
7. Use of serum amyloid A and other acute phase reactants to monitor the inflammatory response after castration in horses: a field study. Equine Vet J, 37:552-556, 2005.
8. Acute Phase Response to Surgery of Varying Intensity in Horses: A Preliminary Study; Veterinary Surgery 38:762–769, 2009.

WHERE INFLAMMATORY STATUS IS BORDERLINE, HORSES WITH EVIDENCE OF LOW GRADE INFLAMMATION SHOULD BE RETESTED WITHIN 24 HOURS TO ESTABLISH INFLAMMATORY STATUS. AS WITH ALL DIAGNOSTIC TESTS, A DEFINITIVE CLINICAL DIAGNOSIS SHOULD NOT BE BASED ON A SINGLE RESULT, BUT SHOULD ONLY BE MADE AFTER ALL CLINICAL AND LAB FINDINGS HAVE BEEN CONSIDERED.

Further information is available on request from:
Invicta, Unit 6 Shiprods Farm, Bashurst Hill, Itchingfield, Horsham, West Sussex RH13 0PD
T: 01403 791313 F: 01403 790062
E: mail@invictavet.com www.invictavet.com

INVICTA ANIMAL HEALTH

EQK_BRO3_00