Guide to Getting the Most Value Out of SAA Testing

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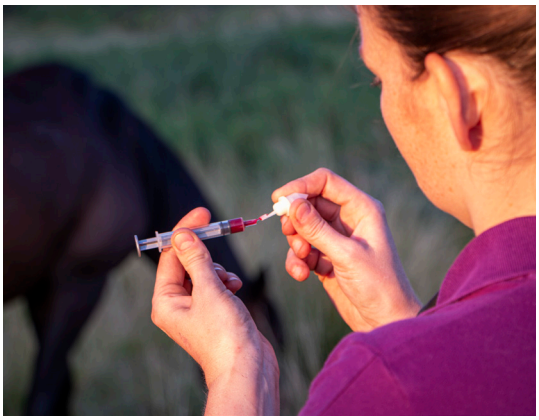
MAY
2021

Serum amyloid A (SAA) testing is a valuable tool for equine practitioners, whether they work in ambulatory practice or a referral hospital. Understanding the nature and dynamics of SAA allows testing to be utilized to its best potential, and proper case selection is important. Interpreting results in light of other clinical information is also vital.



Collecting the right samples at the right time and using the correct technique for testing ensures that results will be useful and consistent. When measuring a single value, exact numerical results are less important as long as they are in the same clinical range (i.e. lead to the same clinical decision). However, **consistency in technique becomes more important for effective monitoring**.

Important factors for patient monitoring

When testing multiple samples for comparison over time, it is important to keep these variables as consistent as possible:

- ☑ **Good test technique**
- ☑ **Sample type**
- ☑ **Test Lot**
- ☑ **Correct calibration card**
- ☑ **Environmental conditions**
- ☑ **Time from sample collection to test**

Samples should also be timed appropriately relative to the disease process and treatment (see Monitoring article)

SAA is particularly valuable for differentiating infection

SAA is most valuable in cases where infection causing acute, systemic inflammation is of concern. Although bacterial and viral infections both cause elevated SAA, it tends to be much more pronounced with bacterial. Importantly, SAA can help differentiate between acute infections (such as bacterial pneumonia) and other chronic or allergic etiologies (such as equine asthma) that may have similar clinical presentation. This facilitates **appropriate and judicious administration of antimicrobials** or other therapies.

Results should of course be combined with a good physical exam, history, and other appropriate diagnostics to determine the nature of infection. It is also important to recognize that SAA will not increase significantly with chronic or localized issues. Potential confounding factors, such as recent vaccination, should be considered when evaluating results (see Unexpected Results article).

Results from EDTA blood can remain consistent up to 7 days after collection

Fresh blood should be tested immediately, before it begins to clot. Anticoagulated blood samples can also be collected and tested later. We have demonstrated clinically consistent results with the VMRD SAA test up to 7 days after collection using EDTA blood samples. Blood samples can be stored refrigerated or at room temperature but should not be frozen. If blood has been stored, it should be resuspended thoroughly but gently prior to testing to break up rouleaux formation.

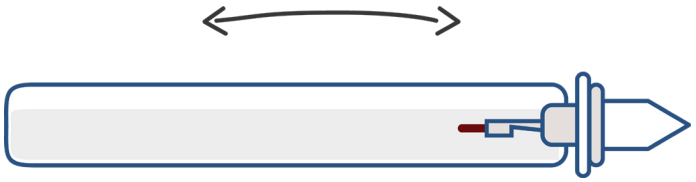
The necessary **dilution for serum/plasma differs from whole blood**, therefore the appropriate testing protocol and sampling materials should be used. Although hematocrit does not appear to affect SAA results, there can be slight differences between fresh blood, anticoagulated blood, serum, and plasma due to other unknown factors.

Diluted samples are stable for at least 30 minutes

When collecting blood using the capillary, it is best to avoid bubbles in the capillary tube and avoid transferring extra blood on the outside of the tube. However, neither of these situations should change the results drastically. Once diluted, the sample should be **stable in the diluent for at least 30 minutes**. After 1-2 hours the value may decrease slightly but should still remain relatively accurate up to 24 hours for all but the lowest SAA concentrations.

Vigorous horizontal shaking mixes samples thoroughly

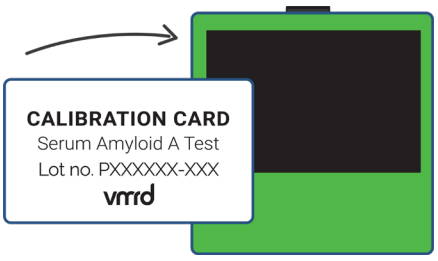
Tests should be run on a flat surface to ensure even flow of the sample through the test membrane. Particularly in high humidity environments, the test cartridge should be used within 10 minutes of opening, as exposure to moisture in the air can also affect flow.



Samples should be **mixed thoroughly** by holding the assembled dropper **horizontally and shaking vigorously**. Although this may cause the formation of some bubbles, they should not interfere with the test. When dispensing the diluted sample, discard several drops prior to applying the solution to the sample well to ensure there is no plain diluent or bubbles in the dropper channel. Drops should be applied holding the dropper vertically. If less than 3 drops are applied the test may not run properly but adding more than 3 drops should not cause any significant issues. In essence, **if drop count is in doubt, add another drop**.

Using the lot-specific calibration card enhances accuracy of results

Using the correct calibration card for the test lot being used will ensure that results are as accurate and consistent as possible. That being said, if the calibration card lot number does not match the test lot number, results will likely be clinically accurate but not as precise.



Results should be **read at exactly 10 minutes** as they may change gradually, usually increasing slightly over time.

Consistency is important for patient monitoring

Keeping sample type consistent when monitoring a patient over time avoids any variation that could occur due to slight sample type differences. If possible, it is also best to test in similar environmental conditions and within a similar time frame after sample collection for best consistency.

Whenever possible, the **same lot of tests** should be used for monitoring an individual patient, although the effect of lot-to-lot variation is minimized by the lot-specific calibration card that standardizes test performance over time.

For guidance on appropriate timing of sample collection for patient monitoring, see our article focused on this topic (Monitoring article). During antimicrobial treatment, SAA can be used to evaluate efficacy and help determine the necessary duration of therapy. SAA will continue to increase for 2-4 days after an acute inflammatory insult but should begin to decrease within a few days of instituting effective therapy.