How to Interpret Serum Amyloid A Concentrations

Rose Nolen-Walston, DVM, DACVIM

Author's address: New Bolton Center, School of Veterinary Medicine, University of Pennsylvania, 382 West Street Road, Kennett Square, PA 19348; e-mail: rnolenw@vet.upenn.edu.

1. Introduction

The role of the clinical veterinarian encompasses many features of medical practice, but in many cases starts with a simple dichotomization: is this animal normal or abnormal? This can include the question, "is it lame or sound," but often also includes, "is it sick or well?" A thorough history and physical examination will often reveal how to stratify the patient, but in cases of subtle disease especially for horses in high level competition, mild and early signs of infection and inflammation may be occult yet significant. A reliable test for infection or inflammation, therefore, can have an extremely valuable place in the clinician's armamentarium. Good tests allow for some degree of quantification both to allow the practitioner to assess the severity of the process and also to follow and document its response to therapy and track its resolution over time. The earliest of these tools was rectal temperature, in which fever signified a secondary indicator of increased cytokines such as tumor necrosis factor (TNF)-α and interleukin (Il)-1.[1] However, over the past century, blood analysis has allowed us to quantify multiple inflammatory markers including the acute phase proteins such as fibrinogen, haptoglobin, α1-acid glycoprotein, C-reactive protein (mainly in humans), serum amyloid A (SAA), and many others, as well as secondary indicators such as white blood cell count and serum iron levels.[2] Of these, fibrinogen has probably been the most heavily relied on for horses. It can be easily and inexpensively measured, but may be confounded by in vitro preanalytical microclot formation. However, its concentration only slowly increases in the 24 hours after induction of inflammation and often does not peak until 48 hours. In addition, there is often only a small increase (often only a 1–2-fold difference) from baseline,[3] and thus mild inflammation cannot reliably be distinguished from normal values. Nonetheless, any method that detects inflammation in the horse probably must outperform fibrinogen in one or more of these factors: accuracy, ease of interpretation, cost, and ease of use.

SAA is the major acute-phase protein of the horse (and most other mammals), and is produced predominantly by the liver as a systemic manifestation of the body's response to inflammation. It exists in equine plasma as one of three isoforms of apolipoprotein and is complexed to high-density lipoprotein in circulating blood.[4] First investigated in horses in the 1980s,[5,6] its clinical use as a marker of inflammation is probably eclipsed by fibrinogen as a function of assay availability rather than diagnostic inferiority. The advantages of SAA over fibrinogen include that it has both low/undetectable constitutive expression in normal animals but reaches levels of 100-1000-fold baseline values in clinical disease

NOTES

states.[3] In addition, its rapid increase in concentration over 6–12 hours combined with a 30–120-minute half-life[7] means that serum values track disease severity closely,[3] and subsequent relapse or secondary infections result in similar response to primary infections.[8] SAA is stable both at room temperature and refrigerated,[9] can be measured using a relatively inexpensive stallside test[9] or a variety of laboratory-based assays,[10] can be performed using plasma as well as serum,[11] and may be assessed using noninvasive samples such as saliva.[12] Although there is some difference in precision and accuracy between assays, most available tests seem to be accurate enough within clinically relevant ranges to be acceptable to the practitioner.[9,10,13]

2. Materials and Methods

A review of the literature reveals many publications that evaluate the use of SAA as a tool for distinguishing healthy horses from those with local or systemic inflammation, and as a diagnostic and monitoring tool for specific conditions. To maximize utility of this compilation of clinical equine veterinary publications, they are presented by body system or disease process, and the review focuses on the most clinically relevant studies. The astute reader will notice that these references refer to SAA in mg/L, μg/mL, and ng/mL; the first two of these units are equivalent and the third represents one thousandth the concentration of the first. The authors' original units are maintained throughout.

3. Results

SAA to Determine Infectious/Inflammation Versus Normal

Horses that are "not quite right" or performing poorly are often diagnostic challenges, and identifying mild inflammation and distinguishing it from noninflammatory differential diagnoses before its clinical signs declare themselves can stymie even the most meticulous clinician. A recent large study evaluated the SAA concentrations of hospitalized horses that had either local inflammation (gastric ulceration, abscesses, *Streptococcus equi* subsp *equi* infection), systemic inflammation (disease accompanied by fever, tachycardia, leukopenia/leukocytosis) or were otherwise healthy or had noninflammatory conditions.[14] Patients with systemic inflammation had significantly higher SAA (mean, 1583 mg/L; range, 688–4000 mg/L) than horses with local or no inflammation, which had mean SAA concentrations of 343 mg/L (range, 37–1609 mg/L) and 5.6 mg/L (range, 1.8–14.5 mg/L) respectively. This discrimination was more distinct than that of fibrinogen, in which the mean values of the three groups were 224, 181, and 128 mg/dL, respectively. Using receiver operator curve analysis, SAA had the highest accuracy for diagnosing inflammation (Fig. 1), but predictive modeling failed to generate useful algorithms.[14] A similar study[15] dichotomized horses into "clinically normal" and "clinically abnormal," the latter of which included conditions as diverse as pneumonia, cholangiohepatitis, *Streptococcus equi* subsp *equi* infection, meningitis, enterocolitis, various forms of colic and neoplasia, and orthopedic infections. The clinically normal horses had a mean SAA of 6.8 mg/L (range, 0.1–26.6 mg/L), whereas the clinically abnormal horses had a mean SAA of 71.7 mg/L with a range of 0.1–3,800. In the same cases, mean fibrinogen values (ranges) were 349 mg/dL (100–800 mg/dL) and 514 mg/dL (100–1200 mg/dL), respectively. For discrimination of clinically normal horses from clinically abnormal horses, SAA had sensitivity of 53% and specificity of 94% (diagnostic accuracy, 75%), whereas using white blood cell count, and plasma fibrinogen concentration and mean albumin:globulin ratio, accuracy ranged from 59 to 62%. The authors also showed data from six cases comparing the resolution of inflammatory markers over time (Fig. 2) and concluded that "SAA concentration can provide valuable information regarding the clinical state of horses and may be more useful for patient monitoring and as a prognostic indicator than are traditional markers of inflammation."[15]

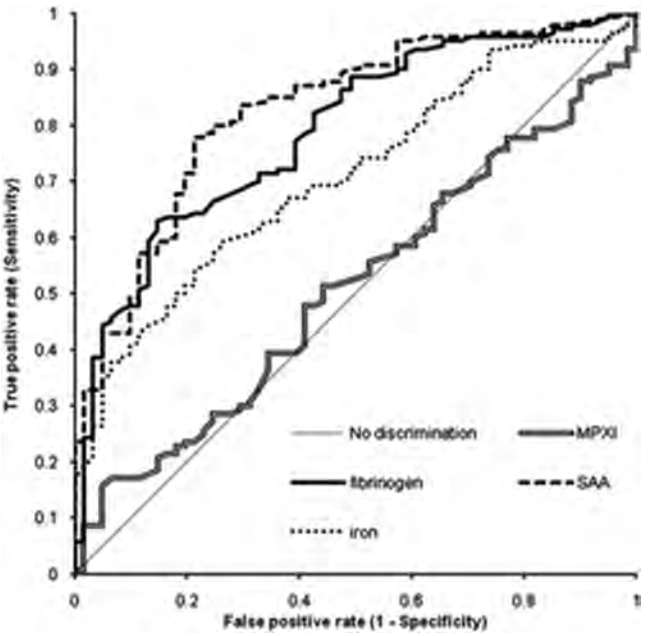

Fig. 1. Receiver-operating characteristic curves for myeloperoxidase index, fibrinogen, SAA, and iron for discerning horses with from those without inflammatory disease. Reprinted with permission by Hooijberg EH et al.[14]

Foals have been shown to have similar baseline values of SAA compared with adults, the kinetics of its rise and resolution seems grossly similar,[16–19] and SAA is higher in animals with bacterial infections than in those with nonbacterial or uncertain diagnoses.[18] In the largest study looking at SAA in foals, 226 healthy Thoroughbred neonates had me-

Horse	Clinical diagnosis	Time of blood sample collection	SAA (mg/L)	Plasma fibrinogen (mg/dL)	WBC count (X 10^3 cells/μL)	A:G ratio
1	Pleuropneumonia	Admission	1,220.0	600	10.2	0.57
		Day 2	1,084.4	600	7.4	0.62
		Day 5	299.5	400	8.6	0.67
		Day 20	1.7	200	8.8	0.98
2	Enterocolitis	Admission	1,426.8	900	8.1	0.91
		Day 3	1,214.8	800	9.0	0.70
		Day 9	1.0	900	18.8	0.81
3	Portal hepatitis and bile duct hyperplasia	Admission	3,628.0	800	6.4	0.54
		Day 2	3,049.0	1,200	6.5	0.54
		Day 6	341.0	500	11.0	0.63
4	Rectal mass	Admission	494.6	100	8.8	1.01
		Day 2	341.6	600	5.8	0.95
		Day 6	1.03	300	6.8	0.89
5	*S equi* subsp *equi* infection	Admission	1,346.9	800	23.4	0.47
		Day 2	1,921.0	800	23.2	0.62
		Day 6	176.6	200	20.5	0.59
		Day 16	4.2	800	14.1	0.69
6	Enterocolitis	Admission	800.8	800	6.4	0.70
		Day 1	1,269.0	600	5.8	0.70
		Day 6	345.3	400	20.1	0.78

See Table 3 for key.

Fig. 2. Data obtained for 12 clinically abnormal horses in which SAA concentration was considered abnormal (n = 6) or not abnormal (n = 6) at the time of hospital admission to illustrate the variation in that variable and other markers of inflammation (plasma fibrinogen concentration, total white blood cell count, and albumin:globulin [A:G] ratio) depending on signalment, diagnosis, and duration of clinical signs. Reprinted with permission by Belgrave RL et al.[15]

dian SAA concentrations of 0.9, 4.5, and 2.5 mg/L on days 1, 2 and 3 of life, with the values on day 2 being significantly higher than baseline.[17] In 136 foals with clinical disease, median SAA concentrations of cases with focal infections such as omphalitis were 195 mg/L and those with septicemia higher still at 280 mg/L. Foals with noninflammatory abnormalities such as failure of passive transfer and noninfectious disease had low SAA concentrations at 5.1 and 3.1 mg/L, respectively.[17] These data suggest that SAA can be used in foals even at young ages as an indicator of infectious or inflammatory processes, especially given that its quicker rise allows abnormalities to be identified in the first few days of life. Increased fibrinogen levels in neonatal foals often indicate intrauterine inflammation; whether SAA rises during prepartum exposure to infectious or inflammatory agents is not known.

SAA and Gastrointestinal Disease

The main applications for the use of SAA in colic would be to assist in the ability to distinguish surgical vs nonsurgical disease, to identify infectious complications, and to gauge prognosis and response to therapy. A study assessing SAA in colic cases admitted to two university teaching hospitals found that concentrations of SAA were significantly higher at admission in horses with colic attributable inflammatory causes (e.g., enteritis, colitis, peritonitis, or abdominal abscesses). This was most useful in separating enteritis cases (median, 65.5 μg/mL; interquartile range, 3–500 μg/mL) from strangulating obstructions (median, 4.8 μg/mL; interquartile range, 0.3–58.6 μg/mL). A significant difference was also seen in the SAA value between horses that survived the colic episode (median, 1.4 μg/mL) and nonsurvivors (median, 10.8 μg/mL).[20] Looking at SAA levels in the peritoneal fluid of horses with abdominal pain, cases with various etiologies of colic had a mean SAA concentration of 249 mg/L in serum and 97 mg/L in peritoneal fluid, compared with less than 1 mg/L in both samples obtained from a healthy control population.[21] Furthermore, SAA was elevated in horses with equine grass sickness (median, 50 mg/mL) compared with surgical lesions and noninflammatory colics (median, ~0 mg/mL).[22]A small experimental study found that inoculation with equine coronavirus resulted in SAA levels that mirrored clinical disease; although all three challenged horses shed large quantities of virus, only those that showed clinical signs of diarrhea, fever, and anorexia had elevated SAA, which peaked at 200–400 μg/mL.[23] Interpreting these data is tricky given that there is not clear consensus on the value of SAA in colics. However, based on these studies and clinical experience, SAA does not offer clear guidance in making the decision if a colic is surgical, nor should it be relied upon for prognosticating survivability when euthanasia is being considered. However, colic cases that are admitted with SAA well above the reference range (> 20 mg/L) should have inflammatory gastrointestinal lesions such as enteritis and colitis higher on the differential diagnosis, and SAA values in the hundreds should prompt a search for an infectious etiology.

SAA and Respiratory Disease

SAA has been investigated as a diagnostic tool for both noninfectious inflammatory airway disease and viral and bacterial infectious diseases. Horses with severe bacterial pneumonia often present with SAA values well into the thousands,[15] and even road transportation, the primary risk factor for pneumo-

nia in horses, increases SAA anywhere from ~30 to 500 μg/mL for 24 to 48 hours after shipping healthy Thoroughbreds 1200 km by road over 26 hours. This increase was abrogated by administration of antimicrobials,[24] and shorter-distance shipping (4 h) did not have an effect on SAA concentrations.[25] In equine influenza, serum SAA increases during the first 48 h of clinical signs and returns to baseline values in 11 to 22 days barring secondary infections, with maximum values of ~450 mg/L during the acute phase.[26] In ponies experimentally infected with EHV-1, serum levels peaked at 100 to almost 1000 mg/L in the week after inoculation.[5]

Various methods have been evaluated to screen foals for sub-clinical *Rhodococcus equi* pneumonia including sonographic examination, hematology, and physical examination, but each have been found to be imperfect with regard to either diagnostic accuracy or cost effectiveness. Acute phase proteins, especially fibrinogen, have been used as moderately effective screening tools for foals on endemic farms. It would seem that with its greater sensitivity and more labile kinetics, SAA would be a better biomarker for this disease that often exhibits an extended preclinical latency where detection is often difficult. Two recent studies have investigated SAA as a possible predictor of *R. equi* pneumonia in at-risk populations. The first of these studies used a large, well-selected population of affected foals and age-matched controls from endemic farms. No predictive value was found in SAA levels in 212 foals 7 to 14 days and 196 foals 21 to 28 days of age, nor at the onset of clinical signs of pneumonia.[27] The authors conclude that "monitoring concentration of SAA is not useful as a screening test for early detection of *R. equi*."[27] A subsequent smaller study also assessing weekly screening with SAA to identify pre-clinical *R. equi* infections found similar results. SAA concentrations did not show an association with the development of sonographic evidence of lung abscessation, and of six foals on an endemic farm diagnosed with *R. equi* pneumonia, only two demonstrated elevated SAA concentrations and this was around the time that the disease became clinically evident.[28] These results are surprising given that *R. equi* generally results in a robust increase in fibrinogen and leukocyte count, which for other inflammatory diseases tend to be less sensitive than SAA. In conclusion, one must wonder why SAA is not a better aid in early identification of *R. equi* pneumonia; no completely satisfactory explanation is apparent for these results.

Recurrent airways obstruction ("heaves") is a disease of the small airways characterized by neutrophilic exudate within the alveoli and bronchi. Although it is not associated with explicit signs of inflammation such as fever or peripheral neutrophilia, SAA has the potential to be more sensitive for subtle alterations. A prospective, observational study used six healthy and six heaves-affected horses challenged with hay and straw to examine a variety of acute-phase proteins.[29] Although haptoglobin concentrations were higher in the heaves horses both before and during an exacerbation, the SAA did not reliably increase, although there was a small but significant difference between heaves-affected horses and controls on day 7 of exacerbation (15.75 vs 3.22 μg/mL, respectively).[29] Nonetheless, probably the main use of SAA in horses with recurrent airways obstruction is distinguishing them from pneumonia cases, in which the SAA is likely to be much higher.

SAA and Surgery

However much attention is paid to careful tissue handling and correct technique, surgery is an inflammatory stimulus and this fact is revealed by elevations in SAA after even minor, uncomplicated procedures.[4,5,30,31] Therefore, its more useful application is probably identifying postoperative infections both earlier and with more accuracy than other methods. A study looking at standing castrations, for example, found that all horses had elevations of SAA to the 400–600-mg/L ranges at day 3 postoperatively, but those that went on to develop infections (as evidenced by fever, swelling, serohemorrhagic or purulent discharge) still had SAA values in this range at the eighth day whereas horses recovering without complication were in the ~200-mg/L range by this point. The increased SAA values associated with infection were not reliably reflected by increases in rectal temperature, leukocyte count, or fibrinogen, suggesting that SAA was a superior marker for infection.[31]A subsequent study found that perioperative treatment with penicillin reduced the SAA in horses undergoing castration from a mean of 708 to 543 mg/L at day 3 postcastration, and from 515 to 125 mg/L on day 8,[32] supporting the idea that even mild infections result in appreciable differences in SAA concentration. Looking beyond orchidectomy, the effect of minor surgical procedures on SAA shows that levels of 100 to 400 mg/L that peak at approximately day 3 after surgery can be expected in cases uncomplicated by infection. These include tibiotarsal arthroscopy and osteochodrosis fragment removal, laryngoplasty, and ventriculectomy (peaked at 50–150 mg/L at d 2; normal by d 7)[30] carotid exteriorization and flexor tendon division (peaked at 100–400 mg/L at d 2; normal by 7–14 d)[5] and a variety of elective procedures including minor airway and orthopedic surgeries (peaked at 16.4 μg/mL at 24 h, 15.5 on d 2).[16] SAA was also significantly lower in elective (defined as noninflamed) vs nonelective (pre-existing inflammatory foci) cases[16] as well as being able to delineate differing levels of surgical trauma based on the invasiveness of the procedure.[30] In several of these studies[16,30] SAA response was found to be a more sensitive indicator of inflammation than a variety of other acute-phase protein or leukocyte responses, and decreased more quickly in response to resolution than fibrinogen (Fig. 3). This is particularly useful to the practitioner who must decide whether hematologic ev-

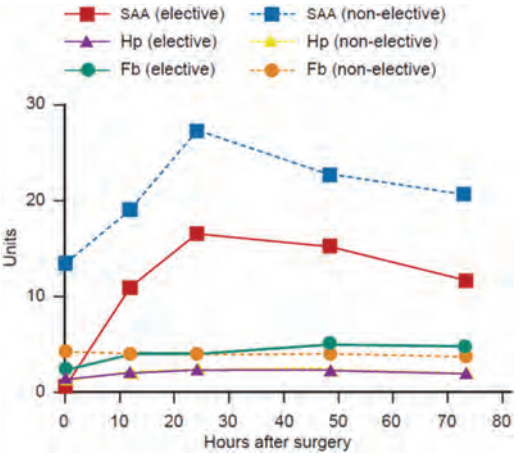

Fig. 3. Mean concentrations of SAA (μg/mL), haptoglobin (Hp; mg/mL) and fibrinogen (Fb; g/L) in 19 horses undergoing elective surgery and eight horses undergoing nonelective surgery. Reprinted with permission by Pollock PJ et al.[16]

idence of inflammation is simply a holdover from the effects of surgery or indicative of postoperative infection that requires further diagnostic evaluation or treatment.

SAA and Joint/Synovial Disease

SAA seems to be a sensitive marker of septic arthritis and tenosynovitis in adult horses. Healthy control horses have serum and synovial concentrations of SAA that are generally less than 1 mg/L. Repeated arthrocentesis (which increases cell count and total protein) and intra-articular amikacin injection do not affect SAA, and this may be one of its important uses in these cases given that the effects or repeated sampling can confound assessment of treatment efficacy and resolution.[33,34] Although much of the SAA found within synovial fluid may be an ultrafiltrate from plasma, a joint-specific isoform of SAA is also produced by synoviocytes.[35] As in other diseases, bacterial infection of joints and other synovial structures seems to be the most potent stimulant of SAA production. SAA concentrations in horses with bacterially contaminated (but not necessarily septic) synovial structures ranged from <1 to 402 mg/L (serum) and 94.5 mg/L (synovial fluid), whereas those with confirmed septic arthritis or tenovaginitis had varied SAA values that were possibly confounded by prior treatment; the highest value in these horses was a serum SAA of ~1700 mg/L and a synovial fluid SAA of ~1100 mg/L. In horses with noninfectious arthropathies such as osteoarthritis, SAA values were not different from control horses.[33] In experimentally induced chemical arthritis (amphotericin B, midcarpal joint), serum SAA peaked at day 2 with values of ~50–300 mg/L and returned to normal between days 7 and 14.[4,36] Although septic arthritis is rarely a diagnostic challenge, SAA may provide the clinician with supporting information regarding degree of inflammation and allow monitoring of resolution of time in conjunction with traditionally measured variables such as synovial fluid analysis.

SAA and Laminitis/Endocrinopathy

The question of how SAA changes in laminitis is complicated by the myriad inflammatory as well as noninflammatory etiologies of laminitis, as well as its varied chronicity and severity. A review of the literature in this area does not show a consistent story. For example, although SAA mRNA is increased in laminitic hoof tissue samples,[37] SAA levels in previously laminitic ponies currently in remission showed no elevation from baseline.[38] In obesity (which is associated with metabolic diseases and laminitis), increases in SAA were correlated with higher body condition score and higher plasma insulin.[39] However, all of the horses had SAA levels that would fall within the reference range of the clinical assay, with highest values obtained topping out at 3845 ng/mL, or 3.8 mg/L. However, even at these low values, the authors concluded that "SAA concentration is a better marker of obesity-associated inflammation and laminitis [and] … is possible that SAA is a component of laminitis pathophysiology."[39]

SAA and Exercise

In Standardbred trotters and Arabian racehorses, SAA levels were unchanged in response to racing.[40,41] However, significant findings have been identified looking at the acute phase response of horses undergoing long-distance endurance riding competitions and training. Although horses evaluated before an endurance ride all had SAA values within normal limits, SAA was lower in horses that successfully finished a 120–160-km competition compared with those that were eliminated based on intra- and postrace lameness and metabolic examinations (411.7 ng/mL in finishers vs 5809.5 ng/mL in eliminated horses; N.B. these units are equivalent to 0.4 and 5.8 mg/L). This study found that "serum SAA level was the only laboratory parameter that indicated most (66.6%) of the eliminated horses before entering the competition."[42] After long-distance (> 100 km) but not shorter (30–60 km) rides, a 10-fold or greater increase in SAA to ~13,000 ng/mL (13 mg/L) was found,[42] although other acute phase proteins such as C-reactive protein and haptoglobin remained unchanged.[43] Looking beyond the massive efforts of competitions at this level, there were no changes in SAA concentrations after race and endurance training sessions in experienced horses. Interestingly, SAA values also increased during training levels for inexperienced, early-career endurance horses (from ~1 to ~3.5 mg/L), but not in experienced competitors.[41] This suggests that using SAA after training sessions may be a method of determining preparedness for advanced competition, but this has not yet been explicitly evaluated.

SAA and Reproductive Disease

There is conflicting evidence on SAA in the peripaturient period of healthy mares. One study found that SAA levels were low in the 8 weeks prepartum although increased slightly in some mares in the week preceding foaling, whereas other studies have found no change in SAA associated prior to normal parturition.[44,45] At 12 and 36 hours postpartum, mean SAA increases to 62 mg/L (range, 0.7–305 mg/L) and 189 mg/L (range, 0–1615 mg/L) and returns to basal concentrations by 60 hours.[45] In mares with experimentally induced placentitis, SAA values peaked between 274 and 4385 mg/L within 2 to 6 days after intracervical inoculation with *Streptococcus equi* subspecies *zooepidemicus*; mares generally aborted within 2 to 6 days of the increase in SAA.[44,45] Abortion was more likely in mares with high SAA compared with mares where the SAA remained within the reference range,[45] and values increased steadily until abortion, when they then decreased rapidly.[44] In comparison, fibrinogen and white blood cell count were not found to be useful markers of placentitis.[44]

SAA and Parasites

In a study using horses experimentally infected with both small and large strongyles, acute-phase proteins were monitored over 161 to 164 days. Although haptoglobin, iron, and albumin/globulin ratios were associated with strongyle burden, SAA was not and remained low throughout the study.[46] In addition, no significant change in SAA was seen after anthelminthic treatment in a group of heavily parasitized horses.[47] This provides the practitioner with useful information as larval cyathstomiasis is a diagnosis difficult to make antemortem, and low SAA may steer the differential diagnosis away from inflammatory causes of colopathy.

SAA and Vaccination

After vaccination using two different influenza and tetanus toxoid products, horses showed variable acute-phase responses with six of 10 developing SAA concentrations greater than ~5 mg/L which peaked at 48 hours after vaccination. Maximum values ranged from ~30 to 175mg/L, and increased white blood cell counts and fibrinogen concentrations, and decreased serum iron were also noted. By 96 hours, SAA levels were returning to normal but had not quite reached baseline values.[48]

4. Discussion

SAA is a sensitive predictor of early inflammation, and due to its rapid onset and short half-life, tracks the course of disease closely. In most studies it outperforms the common variables of both fibrinogen and white blood cell count, and also often the other measured acute-phase reactors of haptoglobin, C-reactive protein, and serum iron. In a general sense, SAA can be used in most situations to obtain early identification of an inflammatory process, to assess the effectiveness of a chosen antimicrobial or other treatment, to monitor the rate of improvement, and to mark resolution of disease. SAA should be considered as an adjunct to diagnosis of a wide variety of inflammatory conditions in the horse, and may well replace fibrinogen in coming years as the main acute phase protein monitored in clinical medicine. However, it is not useful to diagnose specific diseases, and should not replace careful physical examination and identification of the etiologic causes of the inflammatory response. Table 1 shows a compilation of approximations of SAA values for common scenarios encountered in both primary care and referral equine practice.

The main disadvantages of SAA at this time are a lack of complete standardization of assay techniques, which imposes on the practitioner the need to commit to the same assay throughout evaluation of a particular case; values obtained from different assays may not be completely interchangeable. Although there are differences in results and precision between laboratory-based and in-clinic or stall-side tests, studies comparing the different forms of assay have found that the results given by each are clinically comparable.[9,10,13] A newer stall-side assay[a], can be read visually for semiquantitative measurement or using a handheld reader for a quantitative results. No peer-reviewed data have been published demonstrating the accuracy of this kit.

Although SAA has many advantageous characteristics, it is still not a diagnostic panacea. It seems to have no validity in assessing foals for *R. equi* pneumonia, although it often increases to extremely high concentrations in pleuropneumonia in adults and can be very valuable in assessing response to treatment in these cases. SAA does not reliably distinguish surgical from nonsurgical colic cases, although it is probably superior to fibrinogen in identifying postoperative infections because it follows a standard increase-and-decrease pattern of peaking at day 2 postoperatively and then returning to normal after approximately 1 week. Any deviations from a steady decrease after the first 2–3 days after surgery might prompt a search for infectious complications.

Although published reference ranges of SAA vary somewhat, the vast majority of normal, healthy horses tested in all these studies had SAA concentrations of 0 mg/L, or at the most, low single digits; control horses that had SAA levels greater than this may have been suffering from subclinical conditions that were not evident on physical examination. Most instances of infectious or inflammatory disease show SAA levels greater than 50 mg/L, which leaves a gray area between where interpretation may be difficult. SAA is still a relatively new technique compared with other measured indices of inflammation, so less is known about how to interpret these values than older, more familiar parameters. Studies correlating SAA concentrations to a large number of diseases have been published every year over the last decade, and currently the body of liter-

Table 1. Approximate Peak SAA Values Seen in Common Equine Diseases With Their Time to Peak and Resolution

Disease State	Approximate Peak Value, mg/L	Time to Peak; Time to Resolution
SAA in normal horses and foals	0–7 mg/L[a]	NA
SAA, inflammatory colic (enteritis, colitis, coronavirus)	50–500	Depends; decreases in tandem with disease resolution
SAA, surgical colic	5–50	NA
SAA, grass sickness	~50	NA
SAA, major bacterial infections (peritonitis, pleuropneumonia)	Up to 3000–5000 depending on severity	Depends; decreases in tandem with disease resolution
SAA, viral lung disease	500–1000	Peaks in first wk; 11–22 d
SAA, *R. equi* pneumonia	Variable; not reliable indicator	NA
SAA, recurrent airway obstruction ("heaves")	≤15	Increased > baseline only during acute exacerbation
SAA, castration	400–700	2–3 d; to < 200 by 8 d if no complications
SAA, elective surgery	50–400	2 d; 7–14 d
SAA, septic arthritis	Up to 1500 in serum, 1100 in synovial tissue	Not reported
SAA, chemical arthritis	50–300	48 h
SAA, laminitis	No change in chronic, metabolic etiologies	NA
SAA, post-race (STBs, Arabians)	No change	NA
SAA, long-distance endurance ride (> 120 km)	~13	No data; no data
SAA, shorter-distance endurance ride (> 40 km)	No change	NA
SAA, before normal parturition	No change	NA
SAA, after normal parturition	50–600 at 12–36 h postpartum	36 h; 60 h
SAA, placentitis	250–4500	Rises until abortion; rapidly drops after abortion
SAA, parasitism	No change	NA
SAA, after anthelminthic treatment	No change	NA
SAA after vaccination	30–175	2 d; probably between 4–7 d

Abbreviations: STB, Standardbred.
[a]Suggested reference ranges vary from <1.3 and <20 mg/L.

ature is expanding rapidly in ways that assist veterinarians in the application of this parameter. Overall, practitioners should feel comfortable using SAA in lieu of fibrinogen for most cases in which infectious or inflammatory disease is suspected, although ordering both for a period of time is probably wise until the clinician develops the experience necessary to interpret the wide range of values seen with this marker.

Acknowledgments

Declaration of Ethics

The Author declares that she has adhered to the Principles of Veterinary Medical Ethics of the AVMA.

Conflict of Interest

The Author declares no conflicts of interest.

References and Footnote

1. Netea MG, Kullberg BJ, Van der Meer JW. Circulating cytokines as mediators of fever. *Clin Infect Dis* 2000;31 Suppl 5:S178–S184.
2. Borges AS, Divers TJ, Stokol T, et al. Serum iron and plasma fibrinogen concentrations as indicators of systemic inflammatory diseases in horses. *J Vet Intern Med* 2007; 21(3):489–494.
3. Eckersall PD, Bell R. Acute phase proteins: Biomarkers of infection and inflammation in veterinary medicine. *Vet J* 2010;185(1):23–27.
4. Hultén C, Tulamo RM, Suominen MM, et al. A non-competitive chemiluminescence enzyme immunoassay for the equine acute phase protein serum amyloid A (SAA)—A clinically useful inflammatory marker in the horse. *Vet Immunol Immunopathol* 1999;68(2–4):267–281.
5. Pepys MB, Baltz ML, Tennent GA, et al. Serum amyloid A protein (SAA) in horses: Objective measurement of the acute phase response. *Equine Vet J* 1989;21(2):106–109.
6. Husebekk A, Husby G, Sletten K, et al. Characterization of amyloid protein AA and its serum precursor SAA in the horse. *Scand J Immunol* 1986;23(6):703–709.
7. Tape C, Kisilevsky R. Apolipoprotein A-I and apolipoprotein SAA half-lives during acute inflammation and amyloidogenesis. *Biochim Biophys Acta* 1990;1043(3):295–300.
8. Crisman MV, Scarratt WK, Zimmerman KL. Blood proteins and inflammation in the horse. *Vet Clin North Am Equine Pract* 2008;24(2):285–297, vi.
9. Hillstrom A, Tvedten H, Lilliehook I. Evaluation of an in-clinic Serum Amyloid A (SAA) assay and assessment of the effects of storage on SAA samples. *Acta Vet Scand* 2010; 52:8.
10. Christensen M, Jacobsen S, Ichiyanagi T, et al. Evaluation of an automated assay based on monoclonal anti-human serum amyloid A (SAA) antibodies for measurement of canine, feline, and equine SAA. *Vet J* 2012;194(3):332–337.

11. Howard J, Graubner C. Comparison of paired serum and lithium heparin plasma samples for the measurement of serum amyloid A in horses using an automated turbidimetric immunoassay. *Vet J* 2014;199(3):457–460.
12. Jacobsen S, Top Adler DM, Bundgaard L, et al. The use of liquid chromatography tandem mass spectrometry to detect proteins in saliva from horses with and without systemic inflammation. *Vet J* 2014;202(3):483–488.
13. Jacobsen S, Kjelgaard-Hansen M, Hagbard Petersen H, et al. Evaluation of a commercially available human serum amyloid A (SAA) turbidometric immunoassay for determination of equine SAA concentrations. *Vet J* 2006;172(2):315–319.
14. Hooijberg EH, van den Hoven R, Tichy A, et al. Diagnostic and predictive capability of routine laboratory tests for the diagnosis and staging of equine inflammatory disease. *J Vet Intern Med* 2014;28(5):1587–1593.
15. Belgrave RL, Dickey MM, Arheart KL, et al. Assessment of serum amyloid A testing of horses and its clinical application in a specialized equine practice. *J Am Vet Med Assoc* 2013; 243(1):113–119.
16. Pollock PJ, Prendergast M, Schumacher J, et al. Effects of surgery on the acute phase response in clinically normal and diseased horses. *Vet Rec* 2005;156(17):538–542.
17. Stoneham SJ, Palmer L, Cash R, et al. Measurement of serum amyloid A in the neonatal foal using a latex agglutination immunoturbidimetric assay: determination of the normal range, variation with age and response to disease. *Equine Vet J* 2001;33(6):599–603.
18. Hultén C, Demmers S. Serum amyloid A (SAA) as an aid in the management of infectious disease in the foal: Comparison with total leucocyte count, neutrophil count and fibrinogen. *Equine Vet J* 2002;34(7):693–698.
19. Chavatte PM, Pepys MB, Roberts B, et al. Measurement of serum amyloid A protein (SAA) as an aid to differential diagnosis of infection in newborn foals, in *Proceedings*. 6th International Conference on Equine Infectious Diseases 1991;33–38.
20. Vandenplas ML, Moore JN, Barton MH, et al. Concentrations of serum amyloid A and lipopolysaccharide-binding protein in horses with colic. *Am J Vet Res* 2005;66(9):1509–1516.
21. Pihl TH, Andersen PH, Kjelgaard-Hansen M, et al. Serum amyloid A and haptoglobin concentrations in serum and peritoneal fluid of healthy horses and horses with acute abdominal pain. *Vet Clin Pathol* 2013;42(2):177–183.
22. Copas VE, Durham AE, Stratford CH, et al. In equine grass sickness, serum amyloid A and fibrinogen are elevated, and can aid differential diagnosis from non-inflammatory causes of colic. *Vet Rec* 2013;172(15):395.
23. Nemoto M, Oue Y, Morita Y, et al. Experimental inoculation of equine coronavirus into Japanese draft horses. *Arch Virol* 2014;159(12):3329–3334.
24. Endo Y, Tsuchiya T, Omura T, et al. Effects of pre-shipping marbofloxacin administration on fever and blood properties in healthy Thoroughbreds transported a long distance. *J Vet Med Sci* 2015;77(1):75–79.
25. Casella S, Fazio F, Giannetto C, et al. Influence of transportation on serum concentrations of acute phase proteins in horse. *Res Vet Sci* 2012;93(2):914–917.
26. Hultén C, Sandgren B, Skiöldebrand E, Klingeborn B, Marhaug G, Forsberg M. The acute phase protein serum amyloid A (SAA) as an inflammatory marker in equine influenza virus infection. *Acta Vet Scand* 1999;40(4):323–333.
27. Cohen ND, Chaffin MK, Vandenplas ML, et al. Study of serum amyloid A concentrations as a means of achieving early diagnosis of *Rhodococcus equi* pneumonia. *Equine Vet J* 2005;37(3):212–216.
28. Passamonti F, Vardi DM, Stefanetti V, et al. *Rhodococcus equi* pneumonia in foals: An assessment of the early diagnostic value of serum amyloid A and plasma fibrinogen concentrations in equine clinical practice. *Vet J* 2015;203(2):211–218.
29. Lavoie-Lamoureux A, Leclere M, Lemos K, et al. Markers of systemic inflammation in horses with heaves. *J Vet Intern Med* 2012;26(6):1419–1426.
30. Jacobsen S, Nielsen JV, Kjelgaard-Hansen M, et al. Acute phase response to surgery of varying intensity in horses: A preliminary study. *Vet Surg* 2009;38(6):762–769.
31. Jacobsen S, Jensen JC, Frei S, et al. 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* 2005;37(6):552–556.
32. Busk P, Jacobsen S, Martinussen T. Administration of perioperative penicillin reduces postoperative serum amyloid A response in horses being castrated standing. *Vet Surg* 2010; 39(5):638–643.
33. Jacobsen S, Niewold TA, Halling-Thomsen M, et al. Serum amyloid A isoforms in serum and synovial fluid in horses with lipopolysaccharide-induced arthritis. *Vet Immunol Immunopathol* 2006;110(3–4):325–330.
34. Sanchez Teran AF, Rubio-Martinez LM, Villarino NF, et al. Effects of repeated intra-articular administration of amikacin on serum amyloid A, total protein and nucleated cell count in synovial fluid from healthy horses. *Equine Vet J Suppl* 2012;(43):12–16.
35. Jacobsen S, Thomsen MH, Nanni S. Concentrations of serum amyloid A in serum and synovial fluid from healthy horses and horses with joint disease. *Am J Vet Res* 2006; 67(10):1738–1742.
36. Hultén C, Grönlund U, Hirvonen J, et al. Dynamics in serum of the inflammatory markers serum amyloid A (SAA), haptoglobin, fibrinogen and alpha2-globulins during induced noninfectious arthritis in the horse. *Equine Vet J* 2002; 34(7):699–704.
37. Noschka E, Vandenplas ML, Hurley DJ, et al. Temporal aspects of laminar gene expression during the developmental stages of equine laminitis. *Vet Immunol Immunopathol* 2009;129(3–4):242–253.
38. Menzies-Gow NJ, Wray H, Bailey SR, et al. The effect of exercise on plasma concentrations of inflammatory markers in normal and previously laminitic ponies. *Equine Vet J* 2014;46(3):317–321.
39. Suagee JK, Corl BA, Crisman MV, et al. Relationships between body condition score and plasma inflammatory cytokines, insulin, and lipids in a mixed population of light-breed horses. *J Vet Intern Med* 2013;27(1):157–163.
40. Kristensen L, Buhl R, Nostell K, et al. Acute exercise does not induce an acute phase response (APR) in Standardbred trotters. *Can J Vet Res* 2014;78(2):97–102.
41. Cywinska A, Witkowski L, Szarska E, et al. Serum amyloid A (SAA) concentration after training sessions in Arabian race and endurance horses. *BMC Vet Res* 2013; 2013;9:91.
42. Cywinska A, Gorecka R, Szarska E, et al. Serum amyloid A level as a potential indicator of the status of endurance horses. *Equine Vet J Suppl* 2010;(38):23–27.
43. Cywińska A, Szarska E, Górecka R, et al. Acute phase protein concentrations after limited distance and long distance endurance rides in horses. *Res Vet Sci* 2012;93(3):1402–1406.
44. Canisso IF, Ball BA, Cray C, et al. Serum amyloid A and haptoglobin concentrations are increased in plasma of mares with ascending placentitis in the absence of changes in peripheral leukocyte counts or fibrinogen concentration. *Am J Reprod Immunol* 2014;72(4):376–385.
45. Coutinho da Silva MA, Canisso IF, Macpherson ML, et al. Serum amyloid A concentration in healthy periparturient mares and mares with ascending placentitis. *Equine Vet J* 2013;45(5):619–624.
46. Andersen UV, Reinemeyer CR, Toft N, et al. Physiologic and systemic acute phase inflammatory responses in young horses repeatedly infected with cyathostomins and *Strongylus vulgaris*. *Vet Parasitol* 2014;201(1–2):67–74.
47. Nielsen MK, Betancourt A, Lyons ET, et al. Characterization of the inflammatory response to anthelmintic treatment of ponies with cyathostominosis. *Vet J* 2013;198(2):457–462.
48. Andersen SA, Petersen HH, Ersbøll AK, et al. Vaccination elicits a prominent acute phase response in horses. *Vet J* 2012;191(2):199–202.

[a]StableLab SAA assay, StableLab, Ballinode, Sligo, Ireland.