

REVIEWS

Serum amyloid A as a sensitive marker of disease activity in rheumatic diseases

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Abstract

Inflammation is known to play a major role in rheumatologic disorders. Hence quantitating the degree of inflammation has become essential to tailor the treatment strategy, especially with the discovery of potent biologicals. CRP and ESR are the currently used predominant inflammatory markers for rheumatologic disorders but they have several limitations. Various studies have highlighted the superiority of serum amyloid A over these markers in quantitating inflammatory and treatment responses. With the widespread availability of ELISA kits, it has become necessary to implicate its use in day to day clinical practice.

Introduction

Most of the rheumatic diseases are characterized by chronic inflammation with acute exacerbation of inflammation and sometimes a spontaneous regression of disease activity. Excessive and uncontrolled inflammation causes significant body damage at the site of inflammation and in areas remote to the site due to spillover effect. Additionally, increasing insights into the mechanism of inflammation further adds to the need for quantifying it.¹

Acute phase response, induced by pro-inflammatory cytokines, predominantly includes synthesis of acute phase proteins by liver. The plasma concentration of acute phase protein (APP) either increases (positive acute phase proteins) or decreases (negative acute phase proteins) by at least 25% during inflammation. These changes are largely due to variations in the production of hepatocytes.² Diverse positive and negative acute phase proteins found in RA patients are given in table 1.

Ever since the discovery of APP in early 1930s, erythrocyte sedimentation rate (ESR) and c-reactive protein (CRP) have dominated the realm as markers of inflammatory activity.³⁻⁵ Although cost-effectiveness and ease of use at primary healthcare units have further enhanced the adoption, their clinical use as inflammatory markers have several limitations (Table 2). Their sensitivity and specificity for measuring inflammation in rheumatologic disorders are debated by several

studies.⁷ The advent of potent biological therapy, which has revolutionized the management of autoimmune and immunological disorders, has further accentuated the quest for newer biomarkers of inflammation. These markers should also benefit in monitoring the treatment and early identification of relapse.⁸

Although several studies have validated the use of serum amyloid A (SAA) protein as a biomarker, it has not been extensively used in clinical practice. This review focuses on the current status of SAA as a marker of common inflammatory conditions.

SAA: A marker of disease inflammation

SAA is a newer acute phase reactants belonging to the family of circulating apolipoproteins synthesized by hepatocytes, adipocytes, macrophages, and fibroblast-like synoviocytes.⁹ In addition, it is produced by cells involved in inflammation, reflecting both local and systematic spill (Figure 1). SAA acts through toll-like receptor (TLR) 2, activation of nuclear factor- κ B and binding to the G-protein coupled formyl peptide receptor like-1 (FPRL1).¹⁰ This in turn induces the release of various cytokines (granulocyte-colony stimulating factor (G-CSF), IL-1, IL-6, TNF- α) and stimulates angiogenesis, tissue factor, and matrix metalloproteinases.^{11, 12} Within 2 days of inflammatory insult, hepatic production of SAA may increase the serum levels up to 1000 fold from a normal level of less than 10 mg/ml. The protein also plays a role in activating humoral

immunity by acting as an opsonin for gram-negative bacteria and it has been shown to possess anti-viral properties.¹³ It could also be useful in determining the risk for secondary amyloidosis. A previous study has shown that median survival was significantly higher (95%) in patients with lower levels of SAA (<10 mg/ml) compared to those with higher level (40%). The recent diagnostic advances have contributed the development of monoclonal antibody-based ELISA test kits for measuring SAA.⁹⁻¹¹ The present review focuses on highlighting the key findings of studies that have evaluated the role of SAA as a pro-inflammatory marker.

Ankylosing spondylitis and spondyloarthropathies

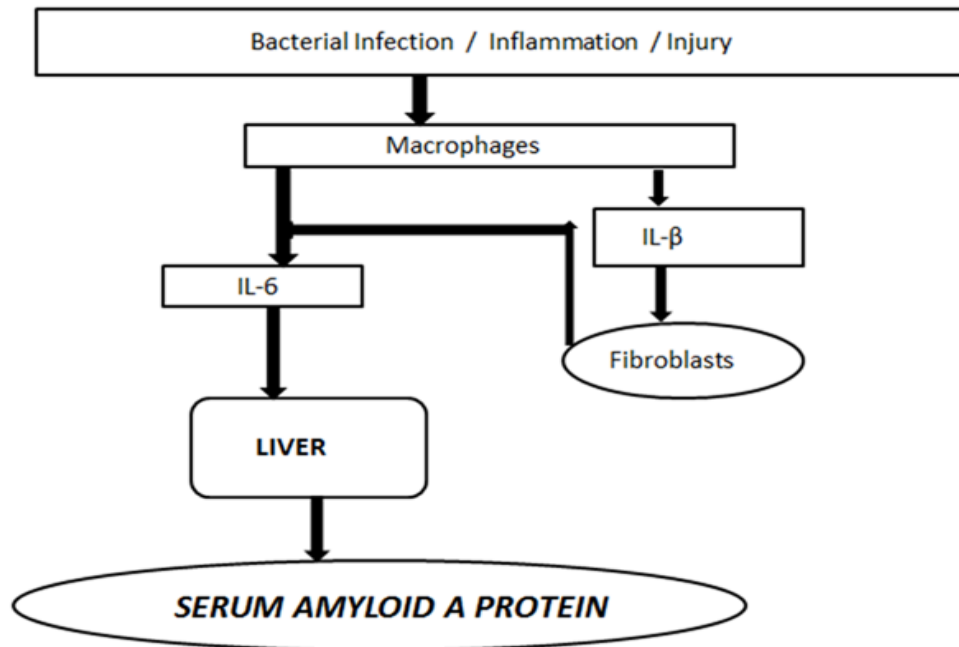
Ankylosing spondylitis (AS) and spondyloarthropathies, the chronic inflammatory rheumatic diseases that cause major functional disabilities, mainly affect the axial skeleton. Since spine and sacroiliac joints are not amenable to clinical examination, it is impossible to monitor the physical signs of inflammation. Traditionally used measures to assess disease activity in AS are ESR, CRP and Bath Ankylosing Spondylitis Disease Activity Index (BASDAI).⁷ A study by Spoorenberg *et al.* demonstrated that both ESR and CRP

levels correlated poorly with disease activity assessment by BASDAI and patient global assessment, especially in patients with exclusive axial disease.¹⁴ The study has raised concerns on the superiority of ESR and CRP in assessing disease activity. Sang *et al.* have suggested the use of SAA as a valuable indicator of disease activity in AS. The study results showed that SAA values were significantly higher in SAA patients (mean = 9.52±7.49 mg/dl) than healthy controls (mean = 2.73± 1.57 mg/dl) (P <0.05). SAA levels also showed a statistically significant linear positive correlation with BASDAI score (r = 0.431, P = 0.007) and ESR (r = 0.521, P = 0.001). Mean BASDAI score of patients with increased levels of SAA were higher than patients with normal levels (P <0.05). Additionally, SAA levels showed a trend towards positive correlation with BASDAI in patients with normal ESR and CRP. Out of 20 patients with increased levels of SAA, 16 had normal ESR (80%) and 9 had normal CRP levels (45%). The patients also had increased scores of BASDAI and the corresponding mean SAA levels noted were 13.7± 6.2 mg/dl and 10.9± 5.2 mg/dl. The study concluded that SAA levels may be elevated, even in patients with mild disease activity.¹⁵

Table 1: Positive and negative acute phase proteins

Positive acute phase proteins	
Complement system	C3 C4 C9 C4b-binding protein
Coagulation and fibrinolytic system	Fibrinogen Plasminogen Tissue plasminogen activator Protein S Plasminogen-activator inhibitor 1
Anti-proteases α1-Protease inhibitors	Anti-chymotrypsin Pancreatic secretory trypsin inhibitor
Transport proteins	Ceruloplasmin Haptoglobin Hemopexin
Participants in inflammatory responses	Phospholipase A Lipopolysaccharide-binding protein Granulocyte colony-stimulating factor
Others	C-reactive protein Serum amyloid A α1-acid glycoprotein
Negative acute phase proteins	
Albumin Transferrin Alpha-fetoprotein Thyroxin-binding globulin Insulin-like growth factor I Factor XII	

Fig.1 : Production of SAA in response to infection/ inflammation / injury



Vries and colleagues evaluated the usefulness of ESR, CRP, and SAA for patient selection and to monitor the anti-TNF therapy. The study conducted in 155 patients, treated with infliximab and etanercept, demonstrated that ESR, CRP and SAA levels were significantly associated with BASDAI scores. All markers decreased significantly after 3 months of treatment ($P < 0.0001$), most notably SAA. Normal baseline levels of CRP and SSA (48%) were significantly associated with non-response to treatment and the combination of the two at baseline yielded the highest predictive value (81%) for Assessment in Ankylosing Spondylitis response (ASAS). Sensitivity of CRP was 69%, while for SSA was 72%. The study concluded that SSA may facilitate patient selection and monitoring the efficacy of anti-TNF treatment in AS and could be added to response criteria.¹⁶ Similarly, the study by Lange and colleagues recommended the use of SAA as a marker of inflammation in AS. In 72 recruited patients, SAA levels correlated significantly with parameters such as ESR, CRP, and BASDAI ($P < 0.001$).¹⁷

Rheumatoid arthritis

Association of SAA with rheumatoid arthritis is more pronounced and extensively studied than AS. The primary study of SAA, CRP, and α 1-acid glycoprotein levels done by Chambers and group (1983) in 185 RA patients reported that SAA is a more sensitive marker of inflammation

than CRP.¹⁸ Though both SAA and CRP correlated with disease activity, rise in SAA levels were more prominent than CRP. The protein levels were nearly normal in 40% population, but SAA levels were considerably high.

Similar to these findings, Grindulis and colleagues also concluded on the use of SAA as a sensitive indicator of inflammation when compared to ESR and CRP. The study conducted in 19 RA patients showed that the SAA, CRP, and ESR were high in all the subjects before starting the therapy.¹⁹ But after the treatment, only 7% of the values of SAA were normal compared to 38% of CRP and 32% of ESR. This occurred in patients with persisting disease activity and relapses, and compared to all the three markers, the first increase was noted for SAA levels.

The study by Hara and coworkers concluded that progressive joint damage noted in RA patients is a direct result of SAA-induced effects on cartilage degradation.²⁰ Arthroscopic synovial biopsy conducted by the research team demonstrated the SSAmRNA expression in all subjects ($n=8$) and in cultured RA synoviocytes. Another study by Connolly *et al.* reported the correlation of serum SAA levels with ESR, CRP, matrix metalloproteinase (MMP)- 1,3,9, 13, tissue inhibitor of metalloproteinases 1 (TIMP-1), vascular endothelial growth factor (VEGF), and type-1 collagen generated biomarkers, C1, 2C and C2C (measured at 0-3

Table 2: Advantages and disadvantages of ESR, CRP, and SAA as inflammatory markers

Acute phase reactants	Advantages	Disadvantages
ESR	<ul style="list-style-type: none"> • Reflects overall health status • Much relevant literature evidence • Easily measurable • Cost effective 	<ul style="list-style-type: none"> • Affected by age and gender • Affected by hematologic disorders such as anemia • Responds slowly to inflammation • Requires fresh sample • Affected by proteins other than acute phase proteins • Affected by commonly used drugs such as glucocorticoids
CRP	<ul style="list-style-type: none"> • Rapid response to inflammation • Unaffected by age and gender • Reflects values of single acute phase proteins • Measured in stored samples 	<ul style="list-style-type: none"> • Wide range of clinically relevant values • Lackspecificity, as the levels increase in many associated non-rheumatologic disorders and tuberculosis • Some studies show racial differences in values⁶
SAA	<ul style="list-style-type: none"> • Rapid response to inflammation • Unaffected by age and gender • Easily measurable using ELISA • Not affected by other drugs 	<ul style="list-style-type: none"> • Lack of much literature evidence • Costly when compared to other tests

months of biological therapy). The study conducted in 62 patients with rheumatoid and psoriatic arthritis concluded that SAA-induced joint destruction is mediated through the activation of MMP induction and collagen cleavage. The research group found that baseline SAA levels correlated well with 28 swollen joint counts, but not with CRP and ESR.²¹ The available evidence indicates that the ability to regulate inflammation by SAA-mediated signaling pathway could pave the way for new therapeutic strategies.

Polymyalgia rheumatica and giant cell arteritis

Polymyalgia rheumatica is an inflammatory condition presenting with pain and stiffness particularly around neck, shoulders, and hip. Giant cell arteritis is an inflammatory disease mostly involving large and medium sized arteries of head particularly temporal artery. Both the diseases are thought to be inter-related as many of patients with polymyalgia also had giant cell arteritis.²² A prospective clinical study carried out by Hachulla *et al.* in 23 patients with giant cell arteritis (GCA) and/or polymyalgia rheumatica (PMR) assessed the markers of the disease activity (ESR, CPR, and SAA levels) during induction of disease remission following prednisone therapy and disease recurrence. They found that the SAA measurement to be more sensitive than the CRP in determining disease activity (97% and 61%, respectively). Furthermore, the specificity of SAA was greater than ESR in determining inactive disease (86% and 77%, respectively). The researchers concluded that SAA

measurement in combination with clinical features may be more useful than ESR and CRP in the management of GCA and/or PMR.²³ These results are consistent with the conclusion from another study by Yamane and group. The researchers determined the usefulness of measuring SAA levels in patients undergoing treatment with prednisolone for PMR. The study findings demonstrated that the mean SAA levels were significantly higher in the group with persistence of symptom (n = 6; 137.8 µg/mL) when compared to those with the symptom disappeared (n = 4; 21.8 µg/mL). Moreover, a positive correlation between CRP and SAA (r = 0.77) was noted. The researchers also observed increased sensitivity of SAA than CRP level and suggested its use as a parameter of PMR activity.²⁴

Other rheumatologic disorders

Sarcoidosis is an inflammatory disease that involves the formation of abnormal lumps or nodules (granulomas) in multiple organs. It is speculated to be caused by an immune reaction to an infection or to an antigen, which continues even after the initial infection or the clearance of the antigen.²⁵ Bargogli *et al.* conducted a proteomic analysis of sarcoidosis patients to evaluate the crucial pathogenetic role of SAA. Serum concentrations of SAA were found to be substantially higher in sarcoidosis patients than controls (P <0.001) and inversely correlated with forced expiratory volume (FEV1). Furthermore, SAA levels were significantly higher in patients with sub-acute onset requiring prolonged and multiple steroid treatments (class 6 sarcoid clinical

activity classification) than in patients with sub-acute onset not requiring therapy (class 4 sarcoid clinical activity classification) ($P < 0.001$). The study suggested that SAA could serve as an inflammatory marker of sarcoidosis.²⁶

Juvenile idiopathic arthritis (JIA), encompasses all forms of arthritis that begin before the age of 16 years. The disease usually persists for more than 6 weeks and it is characterized by inflammation of the synovium and the peri-articular tissues.²⁷ Cantarini *et al.* concluded that SAA is a more sensitive laboratory marker than ESR and CRP for evaluating the presence and number of active joints. The research team evaluated the association between circulating levels of SAA protein and disease activity in 41 patients with juvenile idiopathic arthritis. SAA, ESR, and CRP levels were also measured in these patients and 26 healthy controls. The study demonstrated a significant increase in SAA levels in JIA patients as compared to controls ($P < 0.001$). Significant positive correlations were also seen between SAA and the presence of active joints ($P < 0.05$), the number of active joints ($P < 0.05$), ESR ($P < 0.05$) and CRP ($P < 0.05$) in JIA patients. However, no significant correlations were noted between ESR and the presence of active joints ($P = 0.225$) or between ESR and the number of active joints ($P = 0.520$) in JIA patients. Furthermore, no significant correlations were demonstrated between CRP and the presence of active joints ($P = 0.855$) or between CRP and the number of active joints ($P = 0.859$) with a strong positive correlation between SAA level and JIA disease activity.²⁸

Conclusion

ESR and CRP are widely used in rheumatology clinical practice to assess the severity of inflammation and as a guide to diagnose and manage various rheumatologic disorders. But several limitations of these two markers have prompted rheumatologists across the globe to search for newer inflammatory markers. In the last decade, SAA has been added to the armamentarium of diagnostic tools for rheumatologic disorders. With widely available ELISA kits, it could also function as a potent marker of disease assessment. Further studies should focus on evaluating the advantage of SAA over other parameters and to optimize its role in managing inflammatory diseases.

Competing interests

The authors declare that they have no competing interests.

Citation

Singh NK, Varshney AN, Meena RP. Serum amyloid A as a sensitive marker of disease activity in rheumatic diseases. *IJRCI*. 2014;2(S1):SR1.

Received: 10 January 2014, **Accepted:** 5 May 2014 **Published:** 26 June 2014

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