Annals of Clinical Case Reports

Case Report
Published: 15 Oct, 2021

Diagnostic Value of Serum Amyloid A (SAA) in Assessing the Occurrence of Acute Phase Reaction in Athletes

Konrad Witek[1], Justyna Zaborowska[2], Dariusz Turowski[1] and Sylwia Lewandowska-Pachecka[2]*

[1]*Department of Biochemistry, Institute of Sport-National Research Institute, Poland*

[2]*Division of Laboratory Medicine, Medical University of Warsaw, Poland*

Abstract

Introduction: The aim of this study was to assess the usefulness of Serum Amyloid A (SAA) measurement in determining the presence or intensity of acute phase reaction in athletes in relation to selected diagnostic indicators.

Methods: Thirty male athletes from different sports disciplines participated in this study. Venous blood samples were collected in the morning, on an empty stomach and transferred to the laboratory within 1 h of collection. Concentrations of SAA, CRP hs, creatine kinase were measured in serum. Hematological indicators were measured in whole blood.

Results: In the study group there were found statistically significant positive relationship between SAA protein concentrations with CRP hs levels. Correlations between SAA concentration and some of the hematological indicators were also observed. SAA positively correlated with the percentage and count of neutrophils and monocytes, whereas the relationship with the percentage and count of lymphocytes was negative. Among the subjects there were no statistically significant correlations between serum Amyloid A concentration and CK activity. There were also observed lack of correlation between SAA concentration and White Blood Cells Count (WBC), Hematocrit (HCT) and the percentage and count of Eosinophils (EOS), Basophils (BASO) and Immature Granulocytes (IG).

Conclusion: The observed changes in the concentration of SAA does not permit clear identification of its advantages over widely used CRP measurements in the assessment of acute phase response occurrence in athletes. However, it appears that SAA could be an earlier indicator of acute phase response, but that requires more research.

Keywords: Serum Amyloid A; CRP; Acute phase reaction; Inflammation; Physical exercise; Athletes

OPEN ACCESS

Correspondence:
Konrad Witek, Department of Biochemistry, Institute of Sport–National Research Institute, Trylogii 2/16 st., 01-982 Warsaw, Poland, Tel: +48-22-569-99-41; Fax: +48-22-835-09-77; E-mail: konrad.witek@insp.waw.pl

Received Date: *15 Sep 2021*
Accepted Date: *30 Sep 2021*
Published Date: *15 Oct 2021*

Citation:
Witek K, Zaborowska J, Turowski D, Lewandowska-Pachecka S. Diagnostic Value of Serum Amyloid A (SAA) in Assessing the Occurrence of Acute Phase Reaction in Athletes. Ann Clin Case Rep. 2021; 6: 2030.

ISSN: 2474-1655

Introduction

For optimal athletes preparation it is very important to monitor not only physical performance but also a health status. One of the most important sources of information about general health of athletes is blood tests. Moreover, this is a common method of nutrients deficiency detection, e.g. Vitamins or iron. Unfortunately, concentration of some important blood parameters could be significantly influenced by coexistence of acute phase response [1].

Physical exercise has been shown to be a stressor capable of promoting an acute breakdown of the immune system stable state and promoting chronic adaptations [2]. Continuous aerobic exercise practice is likely to induce an increase in leukocytes, cytokines, interleukins, and tumor necrosis factor alpha in blood serum [3]. Several immunological markers change after long periods of physical exertion allowing the "open window" phenomenon to occur for 3 h to 72 h which decreases immunity and provides a greater onset of airway infections [4,5]. Also repeated short anaerobic bouts of exercise can lead to an acute phase response and signs of inflammation that are detectable even 24 h after cessation of exercise [6]. Thus, information about acute phase occurrence is very useful in propriate athlete's blood tests results analysis.

Serum Amyloid A (SAA) plays essential role in acute phase reaction and is used in clinical laboratories as an indicator of inflammation [7]. Although both SAA and C-Reactive Protein (CRP) are acute-phase proteins, the detection of SAA is more conclusive than the detection of CRP in

such cases like viral infections or severe acute pancreatitis [8]. The elevated SAA is closely related to inflammation-mediated diseases, such as liver diseases, autoimmune diseases, metabolism-related diseases, amyloidosis, and tumors [9-12]. Besides, in acute-phase reactions such as acute inflammation and trauma, the concentration of SAA in the blood can be rapidly increased within 5 h to 6 h under the stimulation of IL-1, IL-6, and TNF-α [13,14]. Therefore, SAA has important clinical value in the diagnoses, progression, and prognoses of diseases associated with inflammation. Due to the specific dynamics of changes in the concentration of SAA protein, slightly different from the currently widely used CRP [15], it may be an additional source of information about the health of players, and so far it is not widely used in sports practice.

The aim of this study was to assess the usefulness of Serum Amyloid A (SAA) measurement in determining the presence or intensity of acute phase reaction in athletes in relation to selected diagnostic indicators.

Case Presentation

Methods

Thirty male athletes from different sports disciplines participated in this study (anthropometric data in Table 1). Testing was conducted in a laboratory at the Department of Biochemistry, Institute of Sport-National Research Institute in Warsaw, Poland, having accreditation of the Polish Centre for Accreditation (no. AB946). This study was approved by the Ethical Research Committee at the Institute of Sport-NRI in Warsaw.

Participants were arrived at laboratory between 7:00 am and 9:00 am. Venous blood samples were collected after arrival on an empty stomach and transferred to the laboratory within 1 h of collection. All participants underwent medical examination, which did not detect any obvious signs of disease and they did not report any health problems.

Hematological parameters were measured in whole blood collected into tubes containing K3-EDTA. Blood samples for biochemical measurements were collected into tubes containing coagulation accelerator, and serum separator. All tubes were the Vacuette vacuum system tubes, Greiner bio-One (Austria). In order to obtain the serum for testing, blood samples were left to clot for 30 min and centrifuged at 2000 g for 10 min. The measurements of SAA concentration were made using a double-antibody sandwich Enzyme-Linked Immunosorbent (ELISA) Assay (Sunred Biological Technology, China). The measurement of Creatine Kinase (CK) and high sensitive C-Reactive Protein (CRP hs) were made with a spectrophotometric and turbidimetric method (respectively) using a biochemical analyzer Cobas Integra 400 Roche (Switzerland), using original manufacturer reagent kits. White blood cell, neutrophil, lymphocyte, monocyte, eosinophil, basophile and immature granulocyte count and haematocrit concentration measurements were made using a hematology analyzer Sysmex XN 1000 (Japan).

All data are presented as mean ± SD. The Shapiro-Wilk test was employed to determinate data normality. Because most of the data did not follow a normal distribution, relationships between variables were determined using Spearman's correlation coefficient. The level of $p \leq 0.05$ was considered significant. Variables were analyzed with Statistica 13 software (TIBCO Software INC., USA).

Results

Table 2 shows the values of measured parameters, and Table 3 shows correlation between SAA and other measured parameters.

Most of the parameters tested were within the accepted reference ranges. A mean value of CRP hs was slightly below reference range with cut off 5.0 mg/L was observed. Serum Amyloid A, for which the mean value was 10.1 μg/mL, was slightly abnormal (cut off 10.0 μg/mL). Moreover, an increased activity of creatine kinase was noted, for which the mean value was 205.1 U/L (cut off 190 U/L).

There were significant positive correlations between SAA and CRP hs, also between SAA and neutrophil count and percentage (R=0.37; p=0.04 and R=0.48; p=0.008, respectively) and monocyte count and percentage (R=0.56; p=0.001 and R=0.46; p=0.01, respectively). In contrast, it occurs significant but negative correlation between SAA and lymphocyte count and percentage (R= -0.50; p=0.01 and R= -0.66; p=0.0001, respectively). Any significant correlations were not found among SAA and anthropometric data. Interestingly, a result shows no significant correlation between SAA and CK, enzyme widely used in sport as a marker of the impact of physical exercises.

Relation between SAA and CRP hs in individual participants shows some differences in reaction among this two acute phase proteins. In three cases, elevated concentration of CRP hs occurs simultaneously with concentration of SAA within reference range (Figure 1).

Discussion

In available literature there is a lack of studies analyzing changes

Table 2: Values of measured parameters (mean ± SD).

Parameter	Units	Results
Serum amyloid A (SAA)	μg/ml	10.1 ± 18.1
C-reactive protein high sensitive (CRP-hs)	mg/l	4.9 ± 5.4
Hematocrit (HCT)	%	44.9 ± 2.4
Creatine Kinase (CK)	U/l	205.1 ± 99.7
White blood cells count	10^3/μl	6.4 ± 1.9
Neutrophil count	10^3/μl	3.6 ± 1.6
Neutrophil Percentage	%	53.8 ± 11.7
Lymphocyte count	10^3/μl	2.0 ± 0.6
Lymphocyte Percentage	%	31.8 ± 11.1
Monocyte count	10^3/μl	0.7 ± 0.2
Monocyte percentage	%	10.4 ± 2.9
Eosinophil count	10^3/μl	0.2 ± 0.2
Eosinophil percentage	%	2.9 ± 2.2
Basophile count	10^3/μl	0.034 ± 0.013
Basophile percentage	%	0.6 ± 0.2
Immature Granulocytes	10^3/μl	0.041 ± 0.026
Immature Granulocytes Percentage	%	0.6 ± 0.3
Erythrocytes Sedimentation Radio (ESR)	mm/h	3.6 ± 3.1

Table 1: Anthropometric characteristic of the study group (mean ± SD).

Sex	n	Age [years]	Body mass [kg]	Body height [m]	BMI
Male	30	23.8 ± 6.0	79.3 ± 14.1	184.4 ± 8.0	23.2 ± 2.5

Table 3: Correlations (R) between SAA and measured parameters.

	Serum amyloid A (SAA)
C-Reactive Protein high sensitive (CRP-hs)	0.43*
Hematocrit (HCT)	0.13
Creatine kinase (CK)	0.01
White blood cells count	0.18
Neutrophil count	0.37*
Neutrophil percentage	0.48*
Lymphocyte count	-0.50*
Lymphocyte percentage	-0.66*
Monocyte count	0.56*
Monocyte percentage	0.46*
Eosinophil count	-0.09
Eosinophil percentage	-0.23
Basophile count	-0.07
Basophile percentage	-0.15
Immature Granulocytes	-0.04
Immature Granulocytes Percentage	-0.01
ESR	0.19
Age	0.12
BMI	0,34

*Statistically significant correlation (p<0.05)

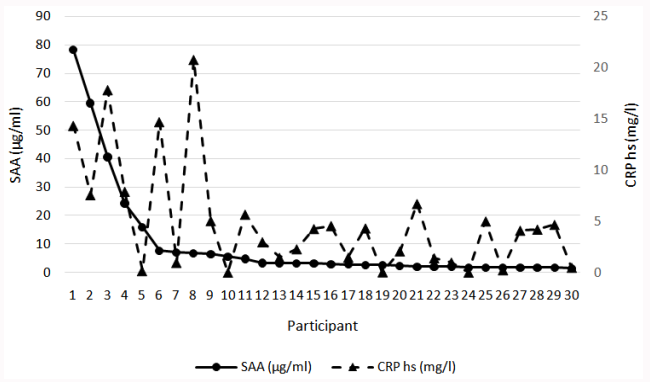

Figure 1: Relation between SAA and CRP hs in individual participants.

in the concentration of SAA protein in response to physical exercise in humans. A study conducted in Arabian horses showed a 10-fold increase in SAA protein concentration after participation in long-distance competitions, during medium-distance races the level of SAA increased about 2-fold [16]. Significant correlation was observed between the concentration of SAA protein and CRP, which is consistent with the results of studies examining the relationship between inflammatory markers and the clinical status of patients. In a study by Ahmed et al. [17] relating to the use of inflammatory markers as predictors in the assessment of a cardiovascular event, a positive correlation was found between these parameters. A similar correlation was observed in the study by Liu et al. [18] on the prognostic values of inflammatory markers in the assessment of the severity of the course of COVID-19.

This study focused on aspects where the indicated parameters could differ. Contrary to the CRP protein, in the case of the SAA protein, no correlation was found between its serum concentration and the OB value. The aforementioned lack of correlation may result from the greater adaptability of the organism to exercise in relation to the SAA protein than CRP. It is also possible that the level of SAA protein in the blood increases faster and faster returns to reference values. A clinical study by Oezcueruemez et al. [19] showed a peak increase in SAA protein at 24 h after the onset of pancreatitis symptoms. In the case of CRP protein its increase was not observed until 72 h [20]. This mechanism may be the cause of observed in our study differences in the concentrations of SAA and CRP individual participants.

According to the literature, intense exercise causes profound changes in the count and composition of white blood cells, which may continue long after training, is stopped, while the body regenerates. This period is characterized by an opposite effect in changes in the number of neutrophils and monocytes, and the number of lymphocytes. The training unit, which puts a lot of strain on the athlete's body, leads to a rapid, local neutrophilia and a subsequent increase in the number of neutrophils in the systemic circulation, which lasts up to 6 h after stopping exercise. Although the neutrophilia observed is related to that of bacterial infection, usually the neutrophil count returns to normal within 24 h of recovery. Exercise also causes changes in the amount of circulating monocytes in the blood; delayed monocytosis is observed within 1 h to 2 h of the onset of the initiating factor and usually returns to pre-workout values about 6 h after the end of activity. Lymphocyte counts decline after vigorous exercise and the observed lymphopenia often reaches levels equivalent to clinical lymphopenia ($<1.0 \times 10^3/\mu l$), but lymphocyte counts typically return to normal levels within the 4-6 hour recovery period [21-23].

Neutrophils and monocytes are part of the nonspecific immune response and are involved in the early defense response of the body, while lymphocytes are part of the late specific response. Based on the observed changes in the concentration of SAA protein, it can be concluded that serum Amyloid A is involved in the early stages of the body's defense reaction. Correlation between SAA and mention above subpopulation of leukocytes are stronger than in the case of CRP, and it seems, that SAA could be an earlier indicator of acute phase response.

Unexpectedly, there was no significant correlation between SAA and creatine kinase activity. Physical activity increases the level of CK in the blood, and its changes are inversely proportional to the level of training adaptation of players [24]. It is possible that a similar adaptation may also occur in SAA. Such a phenomenon was described in the case of horses [25]; however, there are no similar observations in the group of athletes.

Exercise also induces the production of Il-6, a potent pro-inflammatory cytokine, in response to metabolic changes and the bodies increased energy requirements during and after exercise [26]. Therefore, it cannot be ruled out that the lack of correlation between SAA and CK may indicate that SAA is produced in response to changes caused by metabolic processes, and not related to the muscle contractions themselves, as is the case with CK.

In conclusion, in the light of the obtained data, it cannot be unequivocally stated that SAA is a more sensitive or more specific indicator of the evaluation of the ongoing inflammation. It seems that SAA shows a dynamics of changes analogous to CRP hs. However, it appears that SAA could be an earlier indicator of acute phase response, but that requires more research. Our study has several weaknesses, i.e. Low group size, limited only to men or large variation in relation to the sport discipline. It provides the basis for further,

more detailed study on this topic.

Acknowledgment

The study was financially supported by Institute of Sport-National Research Institute, Grant number: 103.09.

References

1. Wang W, Knovich MA, Coffman LG, Torti FM, Torti SV. Serum ferritin: Past, present and future. Biochim Biophys Acta. 2010;1800(8):760-9.
2. Córdova A, Sureda A, Tur J, Pons A. Immune response to exercise in elite sportsmen during the competitive season. J Physiol Biochem. 2010;66(1):1-6.
3. Andersson H, Bohn S, Raastad T, Paulsen G, Blomhoff R, Kadi F. Differences in the inflammatory plasma cytokine response following two elite female soccer games separated by a 72-h recovery. Scand J Med Sci Sports. 2010;20(5):740-7.
4. Nieman DC, Pedersen BK. Exercise and immune function. Sports Med. 1999;27:73-80.
5. Gleeson M. Immune function in sport and exercise. J Appl Physiol (1985). 2007;103(2):693-9.
6. Meyer T, Gabriel HH, Ratz M, Muller HJ, Kindermann W. Anaerobic exercise induces moderate acute phase response. Med Sci Sports Exerc. 2001;33(4):549-55.
7. Barbierato M, Borri M, Facci L, Zusso M, Skaper SD, Giusti P. Expression and differential responsiveness of central nervous system glial cell populations to the acute phase protein serum amyloid A. Sci Rep. 2017;7:12158.
8. Zhang Y, Zhang J, Sheng H, Li H, Wang R. Acute phase reactant serum Amyloid A in inflammation and other diseases. Adv Clin Chem. 2019;90:25-80.
9. Dieter BP, Meek RL, Anderberg RJ, Cooney SK, Bergin JL, Zhang H, et al. Serum Amyloid A and Janus kinase 2 in a mouse model of diabetic kidney disease. PLoS One. 2019;14(2):e0211555.
10. Saulnier PJ, Dieter BP, Tanamas SK, McPherson SM, Wheelock KM, Knowler WC, et al. Association of serum Amyloid A with kidney outcomes and all-cause mortality in American Indians with type 2 diabetes. Am J Nephrol. 2017;46(4):276-84.
11. Todorov I, Gospodinova M, Bocheva Y, Gergana P. Serum Amyloid A protein in the course of infectious mononucleosis. Ther Adv Infect Dis. 2019;6:2049936118811208.
12. Zhang XY, Zhang G, Jiang Y, Liu D, Li MZ, Zhong Q, et al. The prognostic value of serum C-reactive protein-bound serum Amyloid A in early-stage lung cancer. Chin J Cancer. 2015;34(8):335-49.
13. Eklund KK, Niemi K, Kovanen PT. Immune functions of serum Amyloid A. Crit Rev Immunol. 2012;32(4):335-48.
14. Suzuki H, Sugaya M, Nakajima R, Oka T, Takahashi N, Nakao M, et al. Serum Amyloid A levels in the blood of patients with atopic dermatitis and cutaneous T-cell lymphoma. J Dermatol. 2018;45(12):1440-3.
15. Witkowska-Piłaszewicz OD, Żmigrodzka M, Winnicka A, Miśkiewicz A, Strzelec K, Cywińska A. Serum Amyloid A in equine health and disease. Equine Vet J. 2019;51(3):293-8.
16. Cywińska A, Szarska E, Górecka R, Witkowski L, Hecold M, Bereznowski A, et al. Acute phase protein concentrations after limited distance and long distance endurance rides in horses. Res Vet Sci. 2012;93(3):1402-6.
17. Ahmed MS, Jadhav AB, Hassan A, Meng QH. Acute phase reactants as novel predictors of cardiovascular disease. ISRN Inflamm. 2012;2012:953461.
18. Liu SL, Wang SY, Sun YF, Jia QY, Yang CL, Cai PJ, et al. Expressions of SAA, CRP, and FERR in different severities of COVID-19. Eur Rev Med Pharmacol Sci. 2020;24(21):11386-94.
19. Oezcueruemez-Porsch M, Kunz D, Hardt PD, Fadgyas T, Kress O, Schulz HU, et al. Diagnostic relevance of interleukin pattern, acute-phase proteins, and procalcitonin in early phase of Post-ERCP pancreatitis. Dig Dis Sci. 1998;43(8):1763-9.
20. Messmann H, Vogt W, Falk W, Vogl D, Zirngibl H, Leser HG, et al. Interleukins and their antagonists but not TNF and its receptors are released in post-ERP pancreatitis. Eur J Gastroenterol Hepatol. 1998;10(7):611-7.
21. Bøyum A, Wiik P, Gustavsson E, Veiby OP, Reseland J, Haugen AH, et al. The effect of strenuous exercise, calorie deficiency and sleep deprivation on white blood cells, plasma immunoglobulins and cytokines. Scand J Immunol. 1996;43(2):228-35.
22. Walsh NP, Gleeson M, Shephard RJ, Gleeson M, Woods JA, Bishop NC, et al. Position statement. Part one: Immune function and exercise. Exerc Immunol Rev. 2011;17:6-63.
23. Pedersen BK, Rohde T, Ostrowski K. Recovery of the immune system after exercise. Acta Physiol Scand. 1998;162(3):325-32.
24. Brancaccio P, Maffulli N, Limongelli FM. Creatine kinase monitoring in sport medicine. Br Med Bull. 2007;81-82:209-30.
25. Cywinska A, Witkowski L, Szarska E, Schollenberger A, Winnicka A. Serum Amyloid A (SAA) concentration after training sessions in Arabian race and endurance horses. BMC Vet Res. 2013;9:91.
26. Gleeson M, Bishop NC, Stensel DJ, Lindley MR, Mastana SS, Nimmo MA. The anti-inflammatory effects of exercise: Mechanisms and implications for the prevention and treatment of disease. Nat Rev Immunol. 2011;11(9):607-15.