

# Correlation Between Level of Soluble fas and Degree of Sepsis Severity Based on Apache II Score

*by* Erwin Astha Triyono

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## CORRELATION BETWEEN LEVEL OF SOLUBLE FAS AND DEGREE OF SEPSIS SEVERITY BASED ON APACHE II SCORE

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### ABSTRACT

A widely used scoring system to assess the severity of sepsis is Acute Physiology, Age, and Chronic Health Evaluation (APACHE) II scoring system, however there are some disadvantages in using this. Other parameters are needed to predict severity and outcome of sepsis. Proinflammatory cytokines and Fas receptors are increased in sepsis and their concentration elevations are correlated with disease severity. An increase of soluble Fas level will follow increasing Fas receptors. This study aimed to prove any correlation between the level of soluble Fas and degree of sepsis severity based on APACHE II score. A cross-sectional observational study was conducted in January-June 2015 on 30 septic patients. APACHE II scores were calculated from the patients' physiological data, age, and chronic health problem status. Levels of soluble Fas were measured using the ELISA method (Human FAS/ CD95 (Factor-Related Apoptosis) ELISA Kit, Elabscience Biotechnology). Levels of soluble Fas ranged between 1,049-2,783 pg/mL ( $1,855.7 \pm 477.27$  pg/mL). APACHE II scores varied between 4-29 ( $17.2 \pm 5.82$ ). Significant positive correlations between levels of soluble Fas and APACHE II score ( $r=0.347$ ,  $p=0.03$ ) were found. A prediction model of soluble Fas levels based on APACHE II score was made. Linear regression analysis produced a prediction model of soluble Fas levels based on APACHE II score, in which soluble Fas level =  $1,365.8 + 28.485 \times$  APACHE II score.

**Key words:** Sepsis, soluble Fas, APACHE II score

### INTRODUCTION

Sepsis is a systemic inflammatory response caused by microorganism infection. It is an essential clinical matter with high mortality and economic burden. The incidence of sepsis is still increasing although medical and technological improvements have been made. The incidence of severe sepsis in several developed countries ranged between 50 to 100 cases in every 100,000 persons in the population, while in developing countries, the incidences of sepsis, severe sepsis and septic shock are not known. A multi-cohort study involving 16 Asian countries by The Management of Severe Sepsis in Asia's Intensive Care units (MOSAIC) reported that death from sepsis by any cause was 44%. Sepsis is the 11<sup>th</sup> highest cause of death in the United States of America and responsible for 7% of all death in children.<sup>1-3</sup>

Sepsis in critically ill patients is correlated with a bad prognosis. Prognosis of the critically ill patients has been determined by assessing the degree of physical disorders of the patients' body for several decades.<sup>4</sup> A scoring system to determine patient's prognosis may help clinicians to compare their patients with the population mentioned in a study to decide whether an intervention can be implemented to the patient and whether it is beneficial for the patient.<sup>5</sup>

One of the scoring systems that is widely used to assess the degree of disease severity in sepsis, in group patients according to their mortality risk and also to determine their prognosis, is Acute Physiology, Age, and Chronic Health Evaluation (APACHE) II scoring system. The advantages in using APACHE II scoring system are its practical use, only simple data are needed to determine the score, definitions of every variable are clear and it has better sensitivity and reproducibility compared to APACHE III score.<sup>6,7</sup> Several disadvantages in using APACHE II scoring system are that its prediction of mortality risk is based on old data from the year 1979 to 1982, this scoring system is not designed to predict patient outcome individually and it is not designed for a particular disease. This scoring system weighs patients age too high so an old patient with a subtle disease will get a high APACHE II score. APACHE II scoring system also scores high for chronic health problems, so a young patient with severe sepsis but no chronic health problem will get a relatively low APACHE II score although the death risk from sepsis is high.<sup>8-10</sup>

Several clinical and experimental studies in sepsis that have been done previously aimed to find one sensitive parameter that can predict severity, and outcome of sepsis. One crucial thing to clinicians is this parameter can be determined when the diagnosis of sepsis is established, so they can decide the most appropriate

treatment method for their patients. Several studies about sepsis pathophysiology in animals have shown that apoptosis is an important cell death mechanism in sepsis and endotoxemia. The increasing apoptosis in the hematopoietic system and lymphoid tissue causes a decrease of mature T and B lymphocyte number causing immunosuppression during sepsis. Fas receptor (CD95/APO-1) is the central molecule involved in apoptosis in sepsis.<sup>11,12</sup>

Sepsis causes the level of proinflammatory cytokines and Fas receptor (Fas rec) to increase and the level of elevation is correlated with disease severity. Increasing level of soluble Fas will follow increasing level of Fas receptor.<sup>13,14</sup> Fleck *et al.* reported that the level of circulating Fas molecule (soluble Fas) risen significantly in patients with sepsis and septic shock, but it was still difficult to determine whether this level of soluble Fas correlated with severity and outcome of septic patients.<sup>11</sup> There were conflicting hypotheses about the role of soluble Fas in apoptosis, it was suspected that soluble Fas inhibits apoptosis by binding to Fas ligand and then inactivate it, but another study revealed that soluble Fas was released to exert proapoptogen effect.<sup>15,16</sup> This study aimed to find the correlation between the level of soluble Fas and degree of sepsis severity based on APACHE II score.

**METHODS**

The study was conducted in January-June 2015 using a cross-sectional observational design and samples were taken consecutively. Subjects consisted of non-traumatic sepsis patients in the Intermediate Care Ward and in the Internal Medicine Wards, Department of Internal Medicine, Dr. Soetomo Hospital Surabaya. The treating physicians determined diagnosis of sepsis based on sepsis criteria from the 1992 American College of Chest Physicians/Society of Critical Care Medicine Conference Consensus.

The treating physicians performed APACHE II score assessments within 24 hours after the diagnosis of sepsis was established, using an APACHE II score assessment sheet. The assessment of the APACHE II score for each study subject was done by collecting data of physical examination results, laboratory examination results, age, and history of disease from the subject's medical record.

Statistical analysis was performed using SPSS ver 16.0. Correlation between the levels of soluble Fas and degree of sepsis severity was analyzed using Pearson correlation test. If there were any positive correlation, linear regression analysis would be done to make a prediction model of soluble Fas levels based on APACHE II score. P-value <0.05 was considered as statistically significant, with a 95% confidence interval.

**RESULTS AND DISCUSSION**

The study subjects, consisted of 30 sepsis patients, 17 males (56.67%) and 13 females (43.33%) (Table 1). The reason for this gender tendency might be associated with a hormonal difference between sexes that contribute to different inflammation response and sepsis occurrence.<sup>17</sup> Several clinical and experimental studies have reported the significant influence of sex hormone to cellular and humoral immune response. Male hormone (androgen) appeared to have immunodepressive characteristics, while female hormone (estrogen) seemed to increase humoral and cellular immune response. An in-vitro study showed that estrogen had a stimulating effect on human mononuclear cell response (peripheral blood mononuclear cells) against lipopolysaccharide. This early immune response could contribute to increased resistance against sepsis in females.<sup>18</sup>

**Table 1.** Study subjects characteristics

Sample characteristics	Value
Gender, n (%)	
Male	17 persons (56.67%)
Female	13 persons (43.33%)
Age (years-old), mean ± SD	56.9 ± 16.18
Subjects with chronic health problems*, n (%)	22 persons (73.33%)
Diabetes mellitus	13 persons (43.33%)
Diabetes mellitus + diabetic nephropathy	4 persons (13.33%)
Chronic kidney disease	3 persons (10%)
Liver cirrhosis	2 persons (6.67%)
Subjects without chronic health problems*, n (%)	8 persons (26.67%)
Soluble Fas level, mean ± SD	1,855.7 ± 477.27 pg/ mL
APACHE II score, mean ± SD	17.2 ± 5.82

\*Chronic health problems: health problems that fit the criteria in the APACHE II score assessment sheet

Soluble Fas level measurement and APACHE II score assessment were performed in all subjects. Soluble Fas level measurements were performed in the Clinical Pathology Laboratory, Dr. Soetomo Hospital Surabaya. The soluble Fas level measurements were done within 24 hours after the diagnosis of sepsis was established, using Human FAS/CD95 (Factor-Related Apoptosis) ELISA Kit (Elabscience Biotechnology).

The majority of our study subjects were aged above 50 years old (mean age was 56.9 years old). Immune system dysregulation, malnutrition potential, increasing comorbidities, exposure to the resistant pathogen in a nursing home and increasing dependency to the invasive medical device might cause these older people to be more prone to infection and the complication that may accompany.<sup>19</sup>



Most subjects (22 subjects, 73.33%) had chronic health problems, which were liver cirrhosis, stage V chronic kidney disease, diabetes mellitus and diabetes mellitus with diabetic nephropathy. Some of the chronic health problems that had been known to increase the risk of sepsis occurrence were diabetes, chronic liver disease, HIV, and cancer.<sup>17</sup> Diabetic patients have defects in neutrophil function, such as decreasing adhesion, chemotaxis, phagocytosis, and intracellular killing function. Phagocytosis and chemotaxis function of monocytes are also decreased and the level of total immunoglobulin G (IgG) is lower in uncontrolled diabetes. Patients with a chronic liver disease have declining phagocytosis function and decreasing capability of producing reactive oxygen species from neutrophils. Neutrophils are also prone to be activated spontaneously and to lyse, that made toxic contents to be released and causing injury to surround tissues and organs. Sepsis risk in cancer patients is increased as a consequence of immunosuppression mechanism several from the disease itself and also caused by aggressive treatment, such as chemotherapy, radiotherapy, high dose glucocorticoids.<sup>19</sup>

All results of soluble Fas level measurements in this study were higher than usual, ranging from 1,049 to 2,783 pg/mL. De Freitas *et al.* found that the mean value of serum soluble Fas level from 6 healthy control subjects was 118 pg/mL (SD  $\pm$ 6 pg/mL).<sup>20</sup> Two subjects in this study who suffered from sepsis shock had quite high soluble Fas levels, which were 1,789 pg/mL and 2,137 pg/mL. This finding was in accordance with reports from Torre *et al.* who reported that circulating soluble Fas levels in sepsis patients were significantly higher than healthy controls.<sup>21</sup> De Freitas *et al.* also said similar results, levels of molecules that were associated with apoptosis (TNF- $\alpha$ / type 1-TNF receptor and Fas/ FasL) in the serum of patients with severe sepsis were higher than sepsis patients and healthy volunteers.<sup>20</sup>

APACHE II scores in this study ranged from 4 to 29. The highest APACHE II score was found in one subject with a diagnosis of stage V chronic kidney disease accompanied by health care-associated pneumonia and lung edema. APACHE II scores of two subjects suffering from sepsis shock accompanied by MODS were 18 and 14, lower than APACHE II scores of other subjects suffering sepsis only. Higher APACHE II score meant a more severe disease.<sup>22</sup> Discordance between degree of severity that was stated in the subject's diagnosis with the APACHE II score could be because APACHE II score system weighed patient's age too high, so a high score might be given to an old patient with a more subtle disease, on the contrary, a low score might be given to a young patient with more severe disease.<sup>9</sup> APACHE II score also weighed chronic health problems too high, so a young patient with severe sepsis but no chronic health problem would get a quite low APACHE II score although death risk because of sepsis was high.<sup>8</sup> Subjects with septic shock accompanied by MODS in our study did not have chronic health problems, so

APACHE II scores became low. Those were the disadvantages of using the APACHE II score to assess disease severity.

Pearson's correlation test result of our study showed a significant positive correlation between soluble Fas level and APACHE II score ( $p= 0.03$ ) with a correlation coefficient ( $r= 0.347$ ) (Figure 1), but the correlation was weak (correlation coefficient 0.3-0.5= weak correlation).<sup>23</sup> Previous studies from Milic *et al.*, revealed that soluble Fas concentrations positively correlated with APACHE II scores ( $r=0.6046$ ,  $p<0.001$ ).<sup>12</sup>

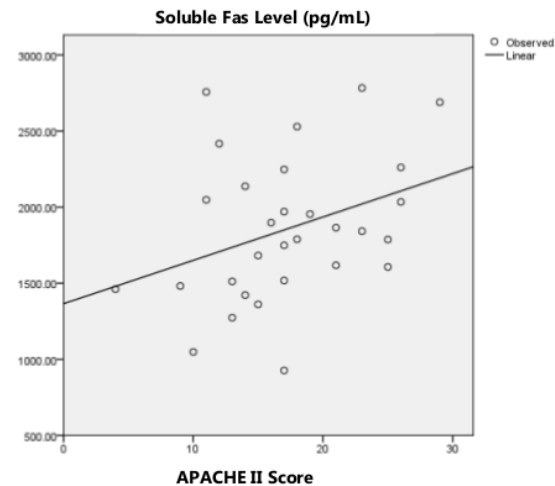


Figure 1. Graph of correlation between soluble Fas level, and APACHE II score

The weak correlation in our study was attributable to several possibilities, such as no uniformity in our study subjects' length of illness before soluble Fas levels were measured, or by factors of flaws in APACHE II score assessments. Soluble Fas levels in this study were indeed measured on the first-day sepsis was diagnosed by a physician, but durations of illness before the subjects came to the Dr. Soetomo Hospital's emergency room were varied. Some subjects were brought to the hospital after being ill for three days, some after five days-ill or more. Marsik *et al.*, studied Fas and Fas ligand expression regulation in human leukocytes with systemic inflammation. He found that soluble Fas level was not very increased within eight hours after endotoxin infusion, but increased significantly after 24 hours.<sup>14</sup> Subjects who had been suffering sepsis for several days will have a higher soluble Fas level on the day of arrival in the emergency room, compared to subjects who just suffered sepsis, with the same degree of severity. The disadvantage of the APACHE II score, that could cause weak correlation in this study, was that the APACHE II score weighed chronic health problems and patient's age too high.<sup>8,9</sup> This disadvantage caused APACHE II score to be less accurate in assessing a subject's severity degree, so the correlation became weak.

Linear regression analysis was performed to make a prediction model of soluble Fas level based on APACHE II score, which was: Soluble Fas level =  $1,365.8 + 28.485 \times \text{APACHE II score}$ . This prediction model allowed us to predict soluble Fas level of a sepsis patient based on APACHE II score of the patient and vice versa. Correlation coefficient (*r*) was 0.347, so determination coefficient value was 0.1204; thus the prediction accuracy of the prediction model obtained in this study was 12%. Correlation value in this study was weak, this caused the prediction model to be less accurate in predicting APACHE II score from soluble Fas level of septic patients and vice versa.

This study had several limitations, first was the variability of duration of illness before the subjects came to the Dr. Soetomo Hospital emergency room. Another limitation was the instrument we used in this study to assess sepsis severity degree was the APACHE II score. The APACHE II score had some disadvantages that could cause sepsis severity assessment to be not accurate. APACHE II score assessment also was done by the treating physician, so subjectivity element in the implementation of assessing APACHE II score could not be excluded.

### CONCLUSION AND SUGGESTION

There was a weak positive correlation between soluble Fas level and degree of sepsis severity based on APACHE II score. A further study with a broader population and specimen collected from sepsis patients with varying degrees of severity needs to be done in future. Length of illness of the patient before arriving at the hospital needs to be uniformed, so the soluble Fas levels might not be significantly different for sepsis patients with the same degree of severity. An advanced study using other parameters which can be measured objectively (e.g. procalcitonin) to measure the degree of sepsis severity needs to be done, in order to understand more about the role of soluble Fas in sepsis management.

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