Interleukin-10 serum level in acute coronary syndrome patients

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Abstrak

Tujuan *Membandingkan kadar IL-10 pada pasien sindrom koroner akut (SKA) dan pasien dengan penyakit jantung koroner (PJK).*

Metode *Subyek penelitian adalah pasien SKA yang dirawat di ruang rawat jantung intensif RSCM/FKUI, RS Persahabatan, RS MMC, dan RS Medistra Jakarta antara bulan Mei 2005 sampai Mei 2006. Pasien PJK rawat jalan diambil sebagai pembanding. Kadar seru interleukin 10 (IL-10) diukur pada kedua kelompok dengan metode radioimunoassay. Perbandingan kedua kelompok dilakukan dengan uji t-test tidak berpasangan. Untuk mengetahui apakah kadar IL-10 dapat digunakan sebagai prediksi SKA, maka dilakukan juga perhitungan sensitivitas dan spesifisitas IL-10.*

Hasil *Telah dianalisa data dari 146 penderita (84 SKA dan 62 PJK). Kadar IL-10 pada penderita SKA (7.37 pg/mL ± 7.81, CI 95% 5.68-9.07) lebih tinggi dibanding dengan penderita PJK (1.59 pg/mL ± 1.55, CI 95% 1.2-1.98). Cut-off point optimum untuk kadar IL-10 adalah >1.95 pg/mL, dengan sensitivitas 79.76 % dan spesifisitas 77.42 %.*

Kesimpulan *Kadar IL-10 pada kelompok SKA lebih tinggi secara bermakna dibanding kelompok PJK. Kadar IL-10 cukup baik digunakan sebagai prediksi SKA, walaupun tidak sebaik CRP.* ***(Med J Indones 2009;18:167-71)***

Abstract

Aim To compare plasma IL-10 concentrations in patients with Acute Coronary Syndrome (ACS) with those in Coronary Artery Disease (CAD).

Methods ACS patients hospitalized in intensive coronary care unit (ICCU) of Cipto Mangunkusumo Hospital/Faculty of Medicine University of Indonesia (CMH/FMUI), Persahabatan Hospital, MMC Hospital, and Medistra Hospital, Jakarta, between May 2005 and May 2006, were included in this study. The ambulatory CAD patients were taken as comparator. The serum IL-10 level was measured by immunoassay method, and compared by using Independent Student's t-test. To investigate whether IL-10 serum level could predict ACS, the sensitivity and specificity of this parameter towards SKA in various IL-10 serum levels were calculated as well.

Results In this observational study, as many as 146 subjects were analyzed, consisting of 84 ACS patients, and 62 coronary artery disease (CAD). The IL-10 level was higher in the group of ACS patients (7.37 pg/mL ± 7.81, CI 95% 5.68-9.07) than that in CAD patients (1.59 pg/mL ± 1.55, CI 95% 1.2-1.98). The optimal cut-off point for serum IL-10 level is >1.95 pg/mL, with 79.76 % sensitivity and 77.42 % specificity.

Conclusion The IL-10 level was higher in the ACS patients compared to that in CAD patients. Serum IL-10 measurement is a quite superior method to distinguish acute and stable condition, eventhough it is not as good as hsCRP for the same purpose. ***(Med J Indones 2009;18:167-71)***

Key words: Interleukin-10, acute coronary syndrome

Previous studies have shown that in the acute coronary syndrome (ACS), proinflammatory cytokines such as tumor necrosis factor-α (TNF-α), interleukin-1 (IL-1), and interleukin-6 (IL-6) are released from macrophages and T lymphocytes. These cytokines then stimulate the acute phase reactant, C- reactive protein (CRP) in the liver.[1]

Accumulating evidence showed a role of the immunoregulatory, antiinflammatory cytokine interleukin-10 (IL-10) in the aetiology of ACS.[2,3] Interleukin-10 (IL-10), a strong antiinflammatory cytokine, is secreted by Th2 subtype lymphocyte and most macrophages. It is known to inhibit many cellular processes and might have important roles in plaque progression, rupture, or thrombosis, including activation of NF-κB,[4] metaloproteinase production,[5] tissue factor,[6] cyclo-oxygenase expression,[7] and cell death.[8]

In patients with ACS, low admission levels of IL-10 have been associated with an increased risk of cardiovascular events and high IL-10 levels with a decreased risk.[9-11] However, recent study showed conflicting results.[1]

This study aimed to compare plasma IL-10 concentrations in patients with ACS with those in CAD subjects.

METHODS

This present study investigated, in a cross-sectional format, whether the bodily acute and chronic proinflammatory response as shown in the alteration of serum IL-10 level in ACS patients is different from those with coronary heart disease (CHD).

Subject

All ACS patients hospitalized in intensive coronary care unit (ICCU) in Cipto Mangunkusumo Hospital / Faculty of Medicine University of Indonesia (CMH/ FMUI), Persahabatan Hospital, MMC Hospital, Medistra Hospital were recruited for the study by consecutive sampling. Age and gender matched CHD patients visited the Cardiology Outpatient Clinic Department of Internal Medicine CMH/FMUI and Integrated Cardiac Service Outpatient Clinic CMH were recruited as control by consecutive sampling method as well. All patients in both groups fulfilling the inclusion criteria and without exclusion criteria, who gave informed consent after receiving explanation from the investigators, were enrolled as study subjects. The study was carried out between May 2005 and May 2006. All procedures in this study had been approved by the FMUI Research Ethics Committee.

Inclusion and Exclusion Criteria

ACS patients admitted to study centers mentioned above, fulfilling the ACS criteria that occurs not earlier than 72 hours from recruitment time were included in this study. CHD patients visiting study centers mentioned above and willing to participate were taken as comparator group.

The following patients were excluded from the study: currently in acute or chronic infection, patients with inflammatory response disorders such as autoimmune disease, connective tissue disease, neoplasm, trauma or surgery within 1 month prior to recruitment, currently receiving corticosteroid, NSAID, or immunosuppressive therapy, statin therapy in CHD group. In ACS group, statin is only given after collection of blood sample for measurements of inflammatory response (for observational study). Patients receiving thiazolidindione therapy, patients with chronic kidney or liver disease were also excluded.

Observed Parameters / Variables

History taking includes age, gender, education level, ethnic groups, history of previous chest pain and myocardial infarction, smoking habit, medication history, history of hypertension, history of DM, history of dyslipidemia, family history of heart disease. During physical examinations, blood pressure, body height, body weight, and waist circumference were measured and heart physical examination was performed.

Laboratory investigation includes: routine blood analysis (hemoglobin, hematocrite, leukocyte, thrombocyte); CK, CKMB, troponin T, fasting and postprandial serum glucose level, glyco Hb, total cholesterol level, direct LDL cholesterol level, HDL and triglyceride cholesterol level, ureum, creatinine, SGOT, SGPT. Inflammatory response: serum IL-10 level.

Definitions

Acute coronary syndrome (ACS)[14,15] is defined as a spectrum of heart emergency consists of: acute myocardial infarction with ST elevation (ST-elevated myocardial infarction/ STEMI), acute myocardial infarction without ST elevation (non ST-elevated myocardial infarction / NSTEMI), unstable angina pectoris (UAP).

Unstable angina pectoris[16] is identified if fulfilling one of the following criteria: 1. Angina at rest and lasts for a long time, usually more than 20 minutes. 2. New onset angina classified as IIIrd class or above in the Canadian Cardiovascular Society (CCS) classification. 3. Acceleration of new angina with signs of increased severity of angina at least 1 CCS class to at least IIIrd class of CCS.

Acute myocardial infarction (AMI)[17] is diagnosed if there are characteristic elevation and gradual decline of troponin or early elevation and decline of biochemical markers of myocardial necrosis with at least one of the following symptoms: a. Ischemic symptoms, b. Occurrence of pathologic Q wave in the ECG. c. Changes in ECG waves indicating ischemia (elevation or depression of ST segment). d. Coronary artery intervention (e.g. coronary angioplasty).

Coronary artery disease (CAD) is a coronary heart disease in patients not currently having acute coronary syndrome

based on anamnesis and ECG, proven history of CHD based on previous coronary angiography examination or having a history of acute myocardial infarction/ACS at least 6 months prior to the examination.

Body mass index (BMI) was measured in kg/m2, and waist circumference was measured using SECA 200 measuring tape. Inflammatory Response was detected by measuring serum interleukin-10 level (in pg/mL) using enzyme immunoassay method (QuantikineR kit. R&D System, Inc., Minneapolis, MN, USA). Normal value is from not detectable (ND)–5.16 pg/mL. Intra assay variability was 6.6 –8.5% and inter assay variability was 8.1–15.6%.

Blood sampling

For routine blood analysis and blood chemistry analysis, peripheral venous blood was collected after a 10-12 hours fasting. Two hours later, the second blood sample was taken for postprandial blood glucose. Bloods were collected using standard method and sent to the Laboratory. Peripheral venous blood samples for serum IL-10 level measurement was collected using standard method and sent to the Laboratory.

Data Processing and Analysis Technique

All data obtained as continuous data. The data were then coded, tabulated, calculated and analysed using STATA. Normally distributed variables were analysed using parametric tests, and variables with abnormal distribution will be transformed into normal data range by logarithmic transformation, and then analysed with parametric test. The significant value of $p < 0.05$ is accepted as significant.

To compare the inflammatory response between ACS and CHD patient, the student-t test for independent samples was performed.

RESULTS

In the observational study from 1 May 2005 to 5 May 2006, as many as 62 CHD patients and 84 ACS that met the study criteria were enrolled. Demographic analysis showed that there was no difference in ages among the two groups.

The risk factors systolic and diastolic blood pressures, waist circumference and body mass index (BMI) did not differ significantly. Laboratory parameters such as fasting blood glucose and Glyco Hb level, total cholesterol, LDL cholesterol, HDL cholesterol and triglyceride level were not significantly diffeterent in the two groups of patients.

Serum IL-10 level

The continuous data from IL-10 serum level was not normally distributed, we normalized the data using natural logarithm transformation as required for performing parametric statistical analysis. The serum IL-10 level is higher in ACS patients (Mean$\pm$SD) 7.37 $\pm$ 7.81 pg/mL; (95% CI: 5.68 - 9.07) compared with those in CHD patients (1.59 $\pm$ 1.55 pg/mL; 95% CI: 1.20 - 1.98).

To investigate whether IL-10 serum level could predict ACS, the sensitivity and specificity of this parameter towards SKA in various IL-10 serum levels were calculated. The optimal cut-off point with the highest sensitivity and specificity was IL-10 serum level > 1.95 pg/mL. The area under the receiver operating characteristic (ROC) curve for the sensitivity and specificity values for IL-10 serum level at various cut points was 0.88 as seen in Figure 1.

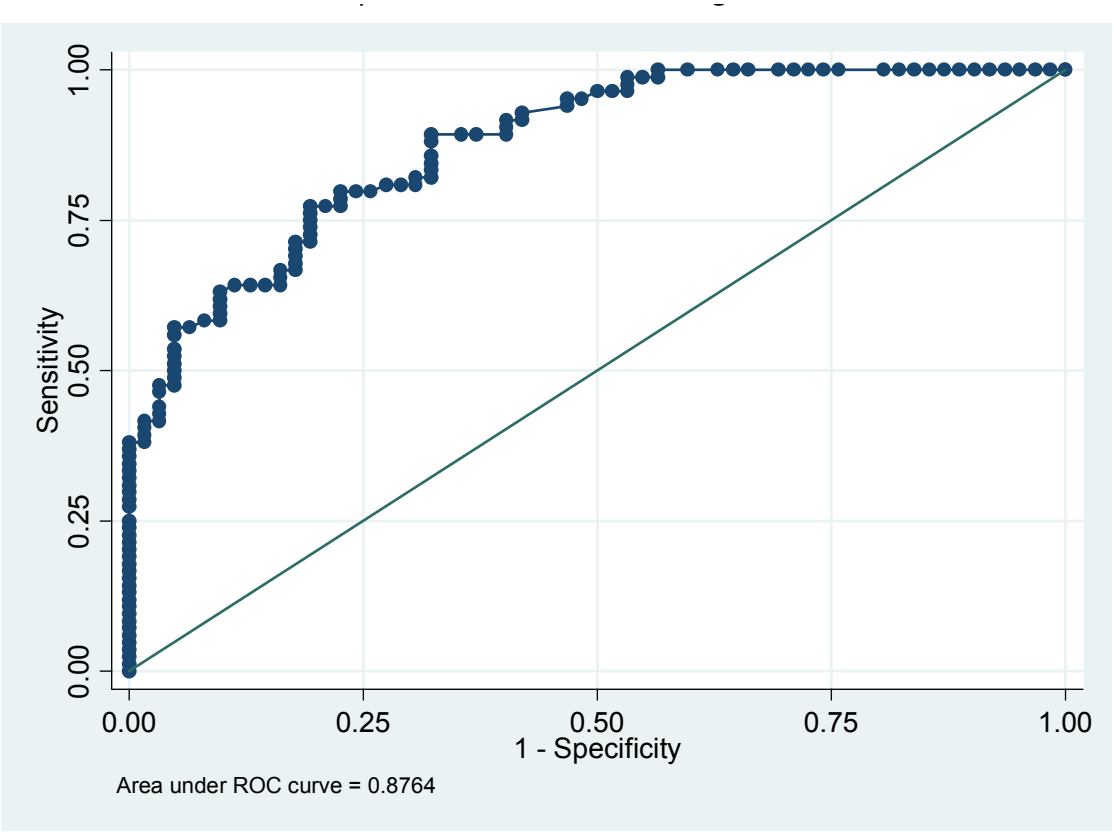

Figure 1. ROC of IL-10 in acute condition (ACS) comparee with non acute condition (CHD)

DISCUSSION

IL-10 serum level in ACS group

In this study, it was evident that the level of serum IL-10 in ACS patients was higher than that in CHD patients. The anti-inflammatory cytokine, IL-10 was abundantly released in the acute condition such as in ACS as a response to IL-6, compared with relatively stable CHD. This increase is supposedly aimed to maintain the balance of anti-inflammatory and pro-inflammatory factors in the body. This argument is supported by our finding that there was a strong, positive correlation between the level of serum IL-6 and IL-10 in ACS group (r=0.69; p=0.00, data not shown). As expected, there was only weak correlation between the level of serum IL-6 and IL-10 in CHD group (r=0.31; p=0.02).

In acute condition the body respons to maintain the balance between proinflammatory cytokine (IL-6) and antiinflamatory cytokine (IL-10), so any increase of IL-6 level in acute condition will be followed by an increase of IL-10.

In CAPTURE (c7E3 Antiplatelet Therapy in Unstable Refractory Angina) study, patients with increased IL-10 level have lower risk for death and for non-fatal myocardial infarction. Patients with increased both the hsCRP and IL-10 level showed lower risk compared to patients with increased hsCRP level alone without any increase in IL-10 serum level, suggesting that IL-10 might have a protective effect against the proinflammatory mediators in ACS.1

Studies on IL-10 level in other countries showed various results. A number of studies showed that in acute condition, there was a higher IL-10 serum level compared with that in stable condition. However, other studies reported lower IL-10 level in unstable angina pectoris.9,17 Smith et al.,9 firstly reported the lower IL-10 serum level in unstable angina pectoris compared with that in the stable angina pectoris. Mälarstig et al.,12 reported that patients with ACS have higher IL-10 plasma concentrations than healthy controls. In contrast to previous studies, a high concentration of IL-10 on admission was associated with a crude risk increase of death and MI.

To predict the occurance of ACS, the optimal cut-off point for serum IL-10 level is >1.95 pg/mL, with 79.76 % sensitivity and 77.42 % specificity. The sensitivity and specificity of various cut-off points for IL-10 level is also shown in Figure 1. Looking at the ROC curve, we are convinced that serum IL-10 measurement is a quite superior method to distinguish acute and stable condition, eventhough it is not as good as hsCRP for the same purpose.

It is concluded that the serum IL-10 level is higher in ACS patient group compared to CHD group.

REFERENCES

1. Hansson GK. Inflammation, atherosclerosis, and coronary artery disease. N Engl J Med 2005;352:1685–95.
2. Li JJ, Guo YL, Yang YJ. Enhancing anti-inflammatory cytokine IL-10 may be beneficial for acute coronary syndrome. Med Hypotheses 2005;65:103–6.
3. Tedgui A, Mallat Z. Anti-inflammatory mechanisms in the vascular wall. CircRes 2001;88:877–87.
4. Lentsch AB, Shanley TP, Sarma V, Ward PA. In vivo suppression of NF-kB and preservation of IkBα by interleukin-10 and interleukin-13. J Clin Invest 1997;100:2443-8.
5. Tziakas DN, Chalikias GK, Hatzinikolaou HI, Parissis JT, Papadopoulos ED, Trypsianis GA. Anti-inflammatory cytokine profile in acute coronary syndromes: behavior of interleukin-10 in association with serum metalloproteinases and proinflammatory cytokines. Int J Cardiol 2003;92:169–75.
6. Lindmark E, Tenno T, Chen J, Siegbahn A. IL-10 inhibits LPS-induced human monocyte tissue factor expression in whole blood. Br J Haematol 1998; 102:597-604.
7. Mertz PM, Dewitt DL, Stetlerstevenson WG, Wahl L M. Interleukin 10 suppression of monocyte prostaglandin H synthase-2: mechanism of prostaglandin- dependent matrix metalloproteinase production. J Biol Chem 1994;269:21322-9.
8. Cohen SB, Crawley JB, Kahan MC, Feldmann M, Foxwell BM. Interleukin-10 rescues T cells from apoptotic cell death: association with an upregulation of Bcl-2. Immunology 1997;92:1–5.
9. Smith DA, Irving SD, Sheldon J, Kaski JC. Serum levels of the antiinflammatory cytokine interleukin-10 are decreased in patients with unstable angina. Circulation 2001;104:746–9.
10. Anguera I, Miranda-Guardiola F, Bosch X, Filella X, Sitges M, Marin JL, et al. Elevation of serum levels of the anti-inflammatory cytokine interleukin-10 and decreased risk of coronary events in patients with unstable angina. Am Heart J 2002;144:811–7.
11. Heeschen C, Dimmeler S, Hamm CW, Fichtlscherer S, Boersma E, Simoon ML. Serum level of the anti-inflammatory cytokine interleukin-10 is an important prognostic determinant in patients with acute coronary syndromes. Circulation 2003;107:2109–14.
12. Mälarstig A, Eriksson P, Hamsten A, Lindahl B, Wallentin L, Siegbahn A. Raised interleukin-10 is an indicator of poor outcome and enhanced systemic inflammation in patients with acute coronary syndrome. Heart 2008;94;724-9
13. Antman EM, Anbe DT, Armstrong PW, Bates ER, Green LA, Hand M et al. ACC/AHA guidelines for the management of patients with ST-elevation myocardial infarction a report of the American College of Cardiology/American Heart Association Task Force on Practice Guidelines (Committee to revise the 1999 guidelines for the management of patients with acute myocardial infarction). Circulation 2004;110:588-636.
14. Braunwald E, Antman EM, Beasley JW, Califf RM, Cheitlin MD, Hochman JS. ACC/AHA guideline update for the management of patients with unstable angina and non-ST-segment elevation myocardial infarction–2002: Summary article. Circulation 2002;106:1893–900.
15. Braunwald E. Unstable angina : a classification. Circulation 1989;80:410-4.
16. Alpert JS, Thugesen K, Antman E. Myocardial infarction redefined-a consensus document of the joint European Society of Cardiology/American College of Cardiology committee for the redefinition of myocardial infarction. J Am Coll Cardiol 2000;36:959-69.
17. Yamashita H, Shimada K, Seki E, Mokuno H, Daida H. Concentrations of interleukins, interferon and C-reactive protein in stable and unstable angina pectoris. Am J Cardiol 2003;91:133-6.