GW25-e4328
The Effect of MicroRNA-21 on the Differentiations and Functions CD4+T Lymphocytes in Patients with Acute Coronary Syndrome
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Objectives: To investigate the effect of abnormal microRNA-21 expression on the differentiations and functions CD4+ lymphocyte in patients with acute coronary syndromes.

Methods: Twenty patients with ACS were enrolled in the study. Blood samples were taken from peripheral vein. The CD4+ T lymphocyte were isolated from mononuclear cells prepared with Ficoll-Hypaque density-gradients centrifugation from human peripheral blood by magnetic cell sorting system. The CD4+ T cells were seeded in culture plates of 6 wells. Each well contained 2ml RPMI-1640 medium without 10% fetal bovine serum. There are four group CD4+ T lymphocytes in the experiment: control groups, microRNA-21 groups, microRNA-21 inhibitor groups, and FAM-siRNA groups. After stimulated with phytohemagglutinin, the CD4+ T lymphocytes and culture supernatant were collected for the following experiments.

- The frequencies of Th1 and Th2 cells were measured by flow cytometry analysis (FACS).
- The total RNA and protein were extracted from CD4+ T lymphocytes using TRIzol and cell lysis buffer for western blotting, respectively.
- The level of IFN-γ, Th1, GATA-3 mRNA expression were measured by qRT-PCR.
- The level of IFN-γ, Th2, GATA-3 protein expression were examined using western blotting.
- The production of IFN-γ and IL-4 in culture supernatants of CD4+ T lymphocytes were detected by enzyme-linked immunosorbent assay (ELISA).

Pearson correlation analysis was conducted to examine the association between IFN-γ/Th2 and IFN-γ/IL-4.

Results: The FACS showed that microRNA-21 could promote CD4+ T lymphocytes to Th1 cells and the Th1/Th2 frequencies were also significantly increased in microRNA-21 group compared with the control group and microRNA-21 inhibitor group [47.2±8.2% vs 39.29% (47.2±8.2%)] %, P < 0.01; 63.2±8.6% vs 59.2±10.1 %, P = 0.024, respectively]. There was no significant difference between the three groups in the frequencies of Th2 cells [(P = 0.28, P = 0.798)]. In comparison with the control group, there was significantly increased level of Th1/Th2 in microRNA-21 group (P < 0.001). The level of IFN-γ mRNA expression in microRNA-21 inhibitor group was significantly lower than that in microRNA-21 group (P < 0.01). No significant differences were found between the three groups in the level of IFN-γ and GATA-3 mRNA expression (F = 1.055, P = 0.362; F = 1.601, P = 0.220, respectively). The level of IFN-γ/Th2 ratio protein expression in microRNA-21 group was significantly higher than that in microRNA-21 group and microRNA-21 inhibitor group (all P < 0.01). There was no significant difference between the three groups in the level of GATA-3 protein expression (F = 0.998, P = 0.097). The culture supernatant concentration of IL-4 was significantly increased in Th1 and Th2 (after 24-36 hours, after 1 week and after 4 weeks). Blood was centrifuged and plasma was separated. Monocytes were also isolated for further analysis. In plasma and monocytes, NLRP3, Cathepsin B, Interleukin-18 (IL-18), Pro-Interleukin-18 (Pro-IL-18), Interleukin-1β (IL-1β), Pro-IL-1β (Pro-IL-1β), oxidized low density lipoprotein (ox-LDL) expressions were appropriately evaluated with real time PCR.

Conclusions: Circulating monocytes in acute coronary syndrome (ACS) had a higher expression of NLRP3, Cathepsin B, and their downstream cytokines. Moreover, we demonstrated that in ACS patients there was good correlation between NLRP3 and Cathepsin-B. We also showed that high dose of statin had a more pronounced and desirable effect on the dynamic changes of NLRP3, Cathepsin B, their downstream mediators. These findings add new insights to the pathogenesis and management of ACS with NLRP3 as the potential target.

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Association of the neutrophil-lymphocyte ratio (NLR) with outcomes in patients admitted for an acute coronary syndrome
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Objectives: Patients with documented acute coronary syndromes or ACS exhibit a wide spectrum of early risk of death, ranging from 1 to 10%. An elevated leukocyte count has been identified as an independent predictor of an increased risk for long-term mortality and myocardial infarction. An elevated neutrophil count predicts a worse outcome in ACS. In contrast, a low lymphocyte count is related to high risks of adverse outcomes and mechanical complications, low ejection fraction, high degree of myocardial necrosis and mortality in patients with ACS. The neutrophil-lymphocyte ratio (NLR), therefore, integrates for two WBC subtypes with opposite actions in terms of vascular inflammation. Among patients diagnosed with ACS in the Philippine General Hospital, we aim to determine if an elevated NLR taken within 24 hours of admission is associated with higher rates of cardiovascular morbidity and mortality. Methods: A prospective cohort of adult patients admitted with a diagnosis of ACS (unstable angina, NSTEMI, STEMI) was conducted. The participants were stratified into two groups: low to intermediate NLR (NLR < 6.5) and high NLR (NLR > 6.5). The primary outcome was in-hospital mortality. Secondary outcomes include development of worsening of congestive heart failure (CHF), and the development of cardiogenic shock, re-infarction, dialysis-requiring renal failure, high-risk pneumonia, and arrhythmias.

Results: 117 patients with a mean age of 60 +/- 13 were included. Majority had 1-4 traditional risk factors for ACS. Diagnosis on admission was unstable angina (28%), NSTEMI (40%), and STEMI (37%). Analysis of data showed that the odds of in-hospital deaths among those with a high NLR is 5.71 times higher compared to those with a low-intermediate NLR (OR = 5.71 [1.53-21.23, p = 0.009]). Using linear regression, the NLR of patients who were non-survivors was computed at 9.91, while the NLR of patients who died of AC was 5.47. A high NLR was also predictive of the development or worsening of CHF (OR 4.75 [1.47 – 15.3, p = 0.009], shock (OR 5.0.0 [1.97 – 12.67, JACC Vol 64/16/Suppl C | October 16–19, 2014 | GW-ICC Abstracts/Cardiovascular Disease Clinical Research C135