ThGM cells as a prognostic and predictive marker for coronary heart disease

Dachao Mou (✉ moudachao@wchscu.cn)

Sichuan University West China Hospital   https://orcid.org/0000-0001-5914-4138

shasha wu

Sichuan University West China Hospital

Ling Jiao

Sichuan University West China Hospital

Yi Zhou

Sichuan University West China Hospital

xiufeng Bai

Sichuan University West China Hospital

Abstract

Background

Coronary heart disease (CHD) is causing by the aberrant aggregation of immune cells in plaque. This study aimed to identify abnormal T cell subtypes and inflammatory factors in CHD patients.

Methods

In total, 141 patients with CHD and 46 healthy controls at the West China hospital (Chengdu, China) between June 2020 and August 2020 were enrolled in this study. Patients were divided into five groups: Healthy control, SAP, UAP, AMI, and SCHD, based on the coronary angiography score and the clinical manifestations. Patients with any one of the following diseases were excluded: tumor, severe infectious disease, and severe inflammation. T cell subsets were analyzed using flow cytometry. Plasma concentration of cytokines were analyzed by Luminex.

Results

Flow cytometric analysis revealed that the number of ThGM cells was higher in CHD patients. The proportion of Th17 and Th1 cells were also increased in CHD patients. levels of IL-4, IL-5, IL-6, and IL-10 were significantly higher in CHD patients (P < 0.05). However, levels of GM-CSF were slightly lower in CHD patients. The multivariate analysis included 187 participants. Of these, 46 did not develop heart failure, 17 developed SCHD, 20 developed SAP, 69 developed UAP, and 35 developed AMI. In multivariate analysis, BMI, Hyperlipaemia, Hypertension, Total cholesterol, HDL, and LDL were not associated with CHD or GM-CSF expressing cell number. Using ANOVA for CHD clinical score studies. Using the student's t test to assess between groups.

Conclusions

Our results suggested that ThGM can be considered as a diagnostic marker of CDH.

Background

Coronary heart disease (CHD) is a kind of chronic inflammatory disorder involving a variety of immune cells and inflammatory mediators[1–3]. The pathogenesis of CHD is complex and heterogeneous, and its etiology remains unclear. Detection of early-stage CHD is important for maximizing the benefits of medical intervention. $CD4^+T$ cells, macrophages, neutrophil, High-sensitivity C-reactive protein, IL-6, and GM-CSF have been reported to play roles in the occurrence and development of CHD[4–7]. Atherosclerosis is a prerequisite for CHD. The main components of atherosclerotic plaques are immune cells, including T lymphoid cells, macrophages, and mast cells are from blood. Among them, T lymphoid

cells play an important role. T helper (Th)1 and T helper (Th)17 cells presented an activated pro-inflammatory state in the local atherosclerotic lesions[8]. Th1 cells predominantly produce interferon-gamma (IFN-γ), a strong pro-inflammatory cytokine, and have been observed in atherosclerotic plaques both in humans and animals[9]. Th17 cells play critical roles in the development of atherosclerotic lesions by producing IL-17 and, to a lesser extent, tumor necrosis factor (TNF)-α and Interleukin-6 (IL-6) [10]. Recently, the role of Granulocyte macrophage colony-stimulating factor (GM-CSF) producing Th cells (ThGM) cells in the initial and progression of some autoimmune diseases have been widely accepted, although its mechanism remains unclear. GM-CSF expression in ThGM was promoted by the IL-7-STAT5 pathway. GM-CSF are pro-inflammatory cytokines that participate in the pathogenesis of many kinds of autoimmune diseases, such as experimental autoimmune encephalomyelitis (EAE)[11], multiple sclerosis (MS)[12], and Myasthenia gravis (MG)[13]. The frequency of GM-CSF-producing T cells were reduced in blood but enriched in the inflamed thymi of MG patients. Besides, the frequencies of thymic GM-CSF-producing T cells were correlated with the clinical disease severity of MG patients[13]. The effector molecules, including IL6, GM-CSF, and IL-17, released by these immune cells accelerate the enlarged and rupture of plaques[14, 15]. The activation of inflammatory factors in the plaque is manifested as a marker of the acute coronary syndrome. The rupture of the plaque releases phospholipids, tissue factor, and platelet adhesion factor to the blood, which accelerate the formation of thrombus and lead to myocardial infarction[16–18]. Plaque rupture can be detected in 60 ~ 70% of patients with acute myocardial infarction[19, 20]. Current research confirmed that CHD is a systemic disease caused by abnormal immune cell function, dysregulated inflammatory factors, and the dysfunctional metabolic system[21, 22]. This study evaluated the role of T cells (Th1, Th17, and ThGM) and cytokines (GM-CSF, IFN-γ, IL-10, IL-17A, IL-1β, IL-2, IL-4, IL-23, IL-5, IL-6, IL-7, and TNF-α) in the development of CHD. We found a strong correlation between ThGM and CHD, this result may provide a new target in CHD drug development.

Methods

Patients

This study was approved by the Ethics Committee of West China Hospital (2019-332), and all patients signed informed consent. The study protocols were conducted in accordance with guidelines of WMA Declaration of Helsinki and CIOMS International Ethical Guidelines Biomedical Research Involving Human Subjects. All experimental protocols were approved by the the Ethics Committee of West China Hospital (2019-332). All samples were obtained from West China Hospital. Six scoring systems were included in the final analysis: gencini, syntax1, PCI syntax2 score, PCI 4-year mortality, CABG syntax2 score, CABG 4-year mortality. Peripheral blood samples were collected from 141 CHD patients (29 women and 112 men, aged 62.2 ± 11.9 years) and 46 healthy controls (8 women and 38 men, aged 61.24 ± 10.12 years) between June 2020 and August 2020. Patients were divided into 5 groups according to the coronary angiography score and the clinical manifestations: (1) Coronary atherosclerosis negative control group (n=46) including patients whose coronary artery examination showed no obvious stenosis; (2) Asymptomatic coronary heart disease group (N=17) including patients with coronary artery stenosis,

but no clinical symptoms; (3) Stable angina pectoris group (n=20) including patients with coronary artery stenosis, clinical manifestations of patients with stable angina pectoris; (4) Unstable angina pectoris group (n= 69) including patients with coronary artery stenosis, clinical manifestations of unstable angina; (5) Acute myocardial infarction group (n=35) including patients with acute myocardial infarction. Patients with any one of the following diseases were excluded: tumor, severe infectious disease, and severe inflammation.

Preparation of peripheral blood mononuclear cells (PBMC) and plasma

Blood samples were collected from patients with coronary heart disease and healthy controls. 10 mL of peripheral blood were collected from each patient and put in an EDTA anticoagulation vacuum tube (BD, 9036857). 2ml of whole blood were transferred into 15ml centrifuge tube, centrifuge at 400×g for 5min. The plasma was stored at -80°C. The remaining whole blood was transferred to a centrifuge tube (Greiner bio-one, 163288) containing 3mL ficoll solution (Solarbio, P8610), and centrifuged at 800×g for 15 minutes at room temperature (minimum acceleration and deceleration). After centrifugation, the middle buffy coat layer was collected and washed for twice with PBS to obtain PBMC.

Flow cytometry for detecting Th cells

T cell subtypes in PBMCs were quantified by flow cytometry (BD, FACSAria SORP). CD4$^+$IFN-γ$^+$T cells were considered to be Th1 cells. CD4$^+$ IFN-γ$^-$GM-CSF$^+$T cells were considered to be ThGM cells. CD4$^+$IL-17A$^+$ T cells were considered to be Th17 cells. 1×10^6 cells were transferred to a 1.5ml centrifuge tube containing 100ul FACS buffer (PBS solution containing 1% FBS). Anti-human CD4-PerCP-Cy5.5 flow cytometry antibody (BD, 552838) was added and incubated for 20min in the dark at room temperature. After washing with FACS buffer, 200ul of IC Fixation Buffer (invitrogen, 2235872) and Permeabilization Buffer (invitrogen, 1961659) were added into the tube, followed by incubating for 20 minutes at room temperature in the dark. After that, cells were resuspend using 100ul FACS buffer, add 5ul each of IFN-y-APC (BD, 554702), GM-CSF-PE (BD, 554507) and IL17A-BV421 (BD, 562933) into the tube and incubate for 30 minutes at room temperature. Then, resuspending cells with 300ul FACS buffer and detected using flow cytometry. Data were analyzed using Aria 2 software (BD, Inc.).

Luminex assay

levels of GM-CSF, IFN-γ, IL-10, IL-17A, IL-1β, IL-2, IL-4, IL- 23, IL-5, IL-6, IL-7, and TNF-α in plasma was detected using Luminex method. High throughput analysis was performed using the Milliplex Map Kit (Merck, HSTCMAG-28SK) according to the manufacturer's instructions. The operation method was shown in the instructions. Simply, setting up blank control wells (2), standard wells (14), QC wells (4) and sample wells (76) in the 96-well plate. Add 50ul of Serum Matrix to the blank well, 50ul of standards with different dilution concentrations into the standard wells, 50ul of quality control products into the QC wells, and 25ul of plasma and 25uL of Assay buffer into the sample wells. Add human monoclonal antibodies coupled with magnetic beads to 96 wells, 25uL per well and incubate with shaking for 16-18h at 4°C. Washing 3 times with wash buffer, and then add 50uL Detection Antibody to each well. After

incubating for 1 h at room temperature, add 50uL Streptavidin-Phycoerythrin (R-PE) into each well. Then, incubating for 30min at room temperature, wash 3 times in wash buffer, adding 150uL of Sheath Fluid and run plate on LuminexR 200^{TM} with Xponent software. Finally, calculating the amounts of cytokines based on the fluorescence intensity.

Statistical analysis

Using ANOVA for CHD clinical score studies. Using the student's t test to assess between groups. The correlation was analyzed by Pearson correlation analysis. All statistical analyses were performed using the Statistical Package for the Social Sciences version 20.0 (SPSS, USA). $P<0.05$ was considered significant.

Results

Clinical laboratory data of CHD patients

As shown in *Table 1*, this study enrolled 187 patients in west china hospital, all patients were detected by coronary angiography and were evaluated with Gencini and SYNTAX scores by two highly trained physicians. Based on the clinical conditions, patients were divided into healthy control group (Control) (n=46), asymptomatic coronary heart disease group (SCHD) (n=17), stable angina group (SAP) (n=20), unstable angina group (UAP) (n=69) and acute myocardial Infarct group (AMI) (n=35). The proportion of male patients is no different in Control, SAP, UAP, and AMI, except for SCHD (Table 1). Patients with autoimmune disease, tumor, severe infection, and infectious diseases were excluded.

Frequency of ThGM and other GM-CSF$^+$ T cell in PBMCs of CHD patients

ThGM is a special subtype of T cells. The IL-7-STAT5 axis promotes the generation of GM-CSF/IL-3-producing Th cells, which were designated as ThGM[11]. The proportion of ThGM is positively correlated with the severity of inflammation. To clarify the relationship between ThGM and CHD, we detected the T cell subsets using flow cytometry. ThGM is represented by CD4$^+$IFN-γ$^-$GM-CSF$^+$T cells (Fig. 1a). Interestingly, we found the proportion of GM-CSF positive CD4$^+$T cells was higher in CHD patients ($P<0.001$) (Fig. 1b). Besides, the number of ThGM was very high in SCHD (P⏐0.001), SAP (P=0.002), UAP (P⏐0.001), and AMI ($P<0.001$) patients (Fig. 1c).

The frequency of Th1 and Th17 cells are increased in PBMCs of CHD patients

Th1 and Th17 are two important T cell subsets. We found the number of the Th1 cells was slightly higher in SCHD (P=0.0027), SAP (P=0.0147), and UAP (P=0.0021) patients, but not in AMI (P<0.0558) (Fig. 2a). Furthermore, we found an increased ratio of Th17 cells in all disease stages ($P<0.001$) (Fig. 2b).

Cytokine levels were increased in plasma of CHD patients

Cytokines secreted by T cells play an important role in CHD development. To test the relationship between cytokine level and CHD severity, we detected GM-CSF, IFN-γ, IL-10, IL-17A, IL-1β, IL-2, IL-4, IL-23, IL-5, IL-6, IL-7 and TNF-α in patient blood using Luminex. Concentrations of IL-4, IL-5, IL-6, and IL-10 were higher in CHD patients. Levels of IL-4 and IL-5 increased significantly in SAP and UAP patients ($P<0.05$) (Fig. 3a,b). It is worth noting that levels of IL6 increased in concomitant with the severity of the disease ($P<0.05$) (Fig. 3c). levels of IL-10 gained significantly in patients with serious CHD patients ($P<0.0001$) (Fig. 3d). However, levels of GM-CSF, IFN-γ, IL-17A, IL-2, TNF-α were slightly lower in CHD patients ($P<0.05$) (Fig. 4a-e). Levels of IL-1β, IL-7, and IL-23 were normal in CHD patients (Fig. 4f-h).

Discussion

Number of ThGM cells are increased in CHD patients, but the plasmatic GM-CSF is not elevated. These data indicated that ThGM cells may be in an inactivated state in CHD patients. The proliferation of ThGM cells in vitro could be activated by anti-CD3 and anti-CD28. The in vivo mechanisms under ThGM proliferation and differentiation are more complex. The IL-7-STAT5 pathway activates the transcription of GM-CSF in ThGM cells[11]. Under specific circumstances, ThGM cells secretes GM-CSF to the plasma. This process must be precisely regulated to maintain the level of GM-CSF in plasma. The GM-CSF in plasma may in turn inhibit the excessive proliferation of ThGM cells to maintain homeostasis in the cell number. However, this feedback was destroyed in CHD patients, which leads to the paradox that the number of cells increases but the content of cytokines decreases. Further studies are needed to test this hypothesis.

Proportion of CD3$^+$IFN$^+$ cells is elevated in SAP and UAP patients[23]. Furthermore, the proportion of Th1 cells increased abnormally in the peripheral blood of CHD patients[24]. We found that the proportion of Th1 cells (CD4 IFN-γ$^+$ T cells) was significantly higher in CHD patients. It is worth noting that in AMI patients, the proportion of Th1 cells is normal. These results indicated that Th1 cells only take part in the early steps of atheromatous plaque formation.

IL-6 is an important inflammatory factor and promotes the development of CHD[14]. Levels of IL-6 were higher in CHD patients[25–27]. IL-6 promotes cell proliferation through JAK-STAT pathway[28]. The high levels of Th2 related cytokines, such as IL-4, IL-5, and IL-10, suggest that in addition to T cell-mediated inflammation, B cells may also participate in immune regulation in CHD patients. Besides, the proportion of Th17 cells (CD4$^+$IL-17A$^+$T cells) was significantly higher in the peripheral blood of CHD patients. Th17 cells are involved in the formation of atherosclerosis[29]. We found an increased Th17 cell level in SA and ACS patients. IL-17A binds to IL-17 receptor (IL-17R), activates NF-kB and JNK pathway by IL-17R-Act1-TRAF6 signaling axis. Although Th17 cells were increased in CHD patients, the plasmatic IL-17A remain unchanged. Unexpectedly, levels of IL-17A were decreased in SCHD, SAP, AMI patients. These results indicated that those Th17 cells, like ThGM cells, were in an inactivated state in CHD patients. The role of these silencing Th17 or ThGM cells needs to be discovered by further studies. Besides, unlike Th1, we found in this study that ThGM was in an inactivated state in the peripheral blood of CHD patients.

Strengths and limitations

In this study, we analyzed the blood sample from CDH patients using FACS and found the correlation between ThGM cell number and the severity of CHD. There are several limitations of our study. First, lack of further confirmation of the result by animal model study. Second, number of cytokines should be expanded to cover more potential key factors.

Conclusion

ThGM was a newly found Th subset, there was a strong connection between ThGM cell number and CHD seriousness. These results can provide new markers for CHD inspection and evaluation.

Declarations

Ethics approval and consent to participate

Prior to participation, the subjects were fully informed on the research procedure and each provided written informed consent. The study protocol was conducted in accordance with guidelines of WMA Declaration of Helsinki and CIOMS International Ethical Guidelines Biomedical Research Involving Human Subjects. All experimental protocols were approved by the the Ethics Committee of West China Hospital (2019-332).

Consent for publication

Not applicable.

Availability of data and materials

Raw data could be found in Supplementally Table1. All datasets used and/or analysed during the current study are available from the corresponding author on reasonable request.

Competing interests

The authors declare no conflict of interest that might have influenced the (presentation of the) work described in this manuscript.

Funding

Contributions

Author information

Affiliations

Laboratory of Human Diseases and Immunotherapies and National Clinical Research Center for Geriatrics, West China Hospital, Sichuan University, Chengdu, China

Dachao Mou, Shasha Wu, Ling Jiao, Yi Zhou & Xiufeng Bai

Clinical Institute of Inflammation and Immunology (CIII), Frontiers Science Center for Disease-related Molecular Network, West China Hospital, Sichuan University, Chengdu, China

Xiufeng Bai

Corresponding author

Correspondence to Xiufeng Bai

References

1. Bäck M, Yurdagul A, Jr., Tabas I, Öörni K, Kovanen PT. Inflammation and its resolution in atherosclerosis: mediators and therapeutic opportunities. Nature reviews Cardiology. 2019;16:389-406. https://doi.org/10.1038/s41569-019-0169-2

2. Jaiswal S, Libby P. Clonal haematopoiesis: connecting ageing and inflammation in cardiovascular disease. Nature reviews Cardiology. 2020;17:137-44. https://doi.org/10.1038/s41569-019-0247-5

3. Vergallo R, Crea F. Atherosclerotic Plaque Healing. The New England journal of medicine. 2020;383:846-57. https://doi.org/10.1056/NEJMra2000317

4. Huynh K. Distinct immune microenvironments in atherosclerotic plaques. Nat Rev Cardiol. 2020;17:7. https://doi.org/10.1038/s41569-019-0304-0

5. Swerdlow DI, Holmes MV, Kuchenbaecker KB, Engmann JE, Shah T, Sofat R, et al. The interleukin-6 receptor as a target for prevention of coronary heart disease: a mendelian randomisation analysis. Lancet (London, England). 2012;379:1214-24. https://doi.org/10.1016/s0140-6736(12)60110-x

6. Gopal K, Nagarajan P, Jedy J, Raj AT, Gnanaselvi SK, Jahan P, et al. β-Carotene Attenuates Angiotensin II-Induced Aortic Aneurysm by Alleviating Macrophage Recruitment in Apoe(-/-) Mice.

PLoS One. 2013;8:e67098. https://doi.org/10.1371/journal.pone.0067098

7. Gopal K, Nagarajan P, Shankar EM, Kamarul T, Kumar JM. High-fat diet- and angiotensin II-induced aneurysm concurrently elicits splenic hypertrophy. Eur J Clin Invest. 2014;44:1169-76. https://doi.org/https://doi.org/10.1111/eci.12351

8. Fernandez DM, Rahman AH, Fernandez NF, Chudnovskiy A, Amir ED, Amadori L, et al. Single-cell immune landscape of human atherosclerotic plaques. Nat Med. 2019;25:1576-88. https://doi.org/10.1038/s41591-019-0590-4

9. Butcher MJ, Filipowicz AR, Waseem TC, McGary CM, Crow KJ, Magilnick N, et al. Atherosclerosis-Driven Treg Plasticity Results in Formation of a Dysfunctional Subset of Plastic IFNγ+ Th1/Tregs. Circ Res. 2016;119:1190-203. https://doi.org/10.1161/circresaha.116.309764

10. Taleb S, Tedgui A, Mallat Z. IL-17 and Th17 cells in atherosclerosis: subtle and contextual roles. Arterioscler Thromb Vasc Biol. 2015;35:258-64. https://doi.org/10.1161/atvbaha.114.303567

11. Sheng W, Yang F, Zhou Y, Yang H, Low PY, Kemeny DM, et al. STAT5 programs a distinct subset of GM-CSF-producing T helper cells that is essential for autoimmune neuroinflammation. Cell research. 2014;24:1387-402. https://doi.org/10.1038/cr.2014.154

12. Aram J, Frakich N, Morandi E, Alrouji M, Samaraweera A, Onion D, et al. Increased IL-2 and Reduced TGF-β Upon T-Cell Stimulation are Associated with GM-CSF Upregulation in Multiple Immune Cell Types in Multiple Sclerosis. Biomedicines. 2020;8. https://doi.org/10.3390/biomedicines8070226

13. Ingelfinger F, Krishnarajah S, Kramer M, Utz SG, Galli E, Lutz M, et al. Single-cell profiling of myasthenia gravis identifies a pathogenic T cell signature. Acta Neuropathol. 2021. https://doi.org/10.1007/s00401-021-02299-y

14. Held C, White HD, Stewart RAH, Budaj A, Cannon CP, Hochman JS, et al. Inflammatory Biomarkers Interleukin-6 and C-Reactive Protein and Outcomes in Stable Coronary Heart Disease: Experiences From the STABILITY (Stabilization of Atherosclerotic Plaque by Initiation of Darapladib Therapy) Trial. Journal of the American Heart Association. 2017;6. https://doi.org/10.1161/jaha.116.005077

15. Frostegård J, Zhang Y, Sun J, Yan K, Liu A. Oxidized Low-Density Lipoprotein (OxLDL)-Treated Dendritic Cells Promote Activation of T Cells in Human Atherosclerotic Plaque and Blood, Which Is Repressed by Statins: microRNA let-7c Is Integral to the Effect. Journal of the American Heart Association. 2016;5. https://doi.org/10.1161/jaha.116.003976

16. Rashid I, Maghzal GJ, Chen YC, Cheng D, Talib J, Newington D, et al. Myeloperoxidase is a potential molecular imaging and therapeutic target for the identification and stabilization of high-risk atherosclerotic plaque. European heart journal. 2018;39:3301-10. https://doi.org/10.1093/eurheartj/ehy419

17. Jamasbi J, Megens RT, Bianchini M, Münch G, Ungerer M, Faussner A, et al. Differential Inhibition of Human Atherosclerotic Plaque-Induced Platelet Activation by Dimeric GPVI-Fc and Anti-GPVI Antibodies: Functional and Imaging Studies. Journal of the American College of Cardiology. 2015;65:2404-15. https://doi.org/10.1016/j.jacc.2015.03.573

18. Nilsson J. Atherosclerotic plaque vulnerability in the statin era. European heart journal. 2017;38:1638-44. https://doi.org/10.1093/eurheartj/ehx143

19. Khalifa AKM, Kubo T, Ino Y, Terada K, Emori H, Higashioka D, et al. Optical Coherence Tomography Comparison of Percutaneous Coronary Intervention Among Plaque Rupture, Erosion, and Calcified Nodule in Acute Myocardial Infarction. Circulation journal : official journal of the Japanese Circulation Society. 2020;84:911-6. https://doi.org/10.1253/circj.CJ-20-0014

20. Teraguchi I, Imanishi T, Ozaki Y, Tanimoto T, Orii M, Shiono Y, et al. Impact of glucose fluctuation and monocyte subsets on coronary plaque rupture. Nutrition, metabolism, and cardiovascular diseases : NMCD. 2014;24:309-14. https://doi.org/10.1016/j.numecd.2013.08.010

21. Dregan A, Charlton J, Chowienczyk P, Gulliford MC. Chronic inflammatory disorders and risk of type 2 diabetes mellitus, coronary heart disease, and stroke: a population-based cohort study. Circulation. 2014;130:837-44. https://doi.org/10.1161/circulationaha.114.009990

22. Li H, Sun K, Zhao R, Hu J, Hao Z, Wang F, et al. Inflammatory biomarkers of coronary heart disease. Frontiers in bioscience (Scholar edition). 2018;10:185-96. https://doi.org/10.2741/s508

23. Fernandes JL, Mamoni RL, Orford JL, Garcia C, Selwyn AP, Coelho OR, et al. Increased Th1 activity in patients with coronary artery disease. Cytokine. 2004;26:131-7. https://doi.org/10.1016/j.cyto.2004.01.007

24. Ding R, Gao W, He Z, Wang H, Conrad CD, Dinney CM, et al. Overrepresentation of Th1- and Th17-like Follicular Helper T Cells in Coronary Artery Disease. Journal of cardiovascular translational research. 2015;8:503-5. https://doi.org/10.1007/s12265-015-9662-0

25. Martínez GJ, Robertson S, Barraclough J, Xia Q, Mallat Z, Bursill C, et al. Colchicine Acutely Suppresses Local Cardiac Production of Inflammatory Cytokines in Patients With an Acute Coronary Syndrome. Journal of the American Heart Association. 2015;4:e002128. https://doi.org/10.1161/jaha.115.002128

26. Kaptoge S, Seshasai SR, Gao P, Freitag DF, Butterworth AS, Borglykke A, et al. Inflammatory cytokines and risk of coronary heart disease: new prospective study and updated meta-analysis. European heart journal. 2014;35:578-89. https://doi.org/10.1093/eurheartj/eht367

27. Goldhammer E, Tanchilevitch A, Maor I, Beniamini Y, Rosenschein U, Sagiv M. Exercise training modulates cytokines activity in coronary heart disease patients. International journal of cardiology. 2005;100:93-9. https://doi.org/10.1016/j.ijcard.2004.08.073

28. Lim CP, Fu XY. Multiple roles of STAT3 in cardiovascular inflammatory responses. Progress in molecular biology and translational science. 2012;106:63-73. https://doi.org/10.1016/b978-0-12-396456-4.00010-9

29. Wang Z, Lee J, Zhang Y, Wang H, Liu X, Shang F, et al. Increased Th17 cells in coronary artery disease are associated with neutrophilic inflammation. Scandinavian cardiovascular journal : SCJ. 2011;45:54-61. https://doi.org/10.3109/14017431.2010.491123

Tables

Table 1. Clinical characteristics of the CHD patients and healthy controls.

Characteristics	Control (n=46)	SCHD (n=17)	SAP (n=20)	UAP (n=69)	AMI (n=35)
Clinical characteristics					
Male, n (%)	38 (82.6)	9 (52.94)*	18 (90)	55 (79.71)	30(85.71)
Age (years)	61.24±10.12	68.63±9.15	60.65±11.39	62.27±11.86	60.71±12.82
BMI	23.82±4.37	22.62±14.61	24.61±2.33	24.32±3.35	24.3±3.38
Hypertension, n (%)	27 (58.7)	8 (47.06)	13 (65)	38 (55.07)	12 (34.29)
Diabetes, n (%)	4 (8.7)	4 (23.52)	4 (20)	19 (27.54)	9 (25.71)
Hyperlipaemia, n (%)	5 (10.9)	1 (5.89)	1(5)	4 (5.8)	6(17.14)
Laboratory characteristics					
Hematocrit (L/mL)	40.33±6.12	35.86±5.84	43.8±3.37	40.1±4.54	40.54±4.97
CK-MB (ng/mL)	4.95±10.46	2.87±2.57	2.01±1.21	1.65±0.96	45.97±81.85*
Total cholesterol (mmol/L)	3.78±1.22	3.56±0.81	3.38±1	4.42±0.93	4.15±1.29
HDL cholesterol (mmol/L)	1.34±0.45	1.48±0.36	1.509±0.61	1.32±1.07	1.2±0.38
LDL cholesterol (mmol/L)	1.89±1.06	1.86±0.67	5.2±15.5*	1.81±0.73	2.51±1.22
Coronary angiographic score					
gencini score	0	8.06±10.92*	44.9±32.34*	75.12±49.72*	72.03±47.73*
syntax1 score	0	1.58±4.23*	14.8±9.57*	22.14±12.35*	18.1±12.15*

The values are expressed as proportions or the mean ± standard deviations. CHD, coronary heart disease; SCHD, asymptomatic coronary heart disease; SAP, stable angina pectoris; UAP, unstable angina pectoris; AMI, acute myocardial infarction. BMI, body mass index; HDL, high-density lipoprotein; LDL, low-density lipoprotein. *P<0.05, all kinds of CHD vs control group.
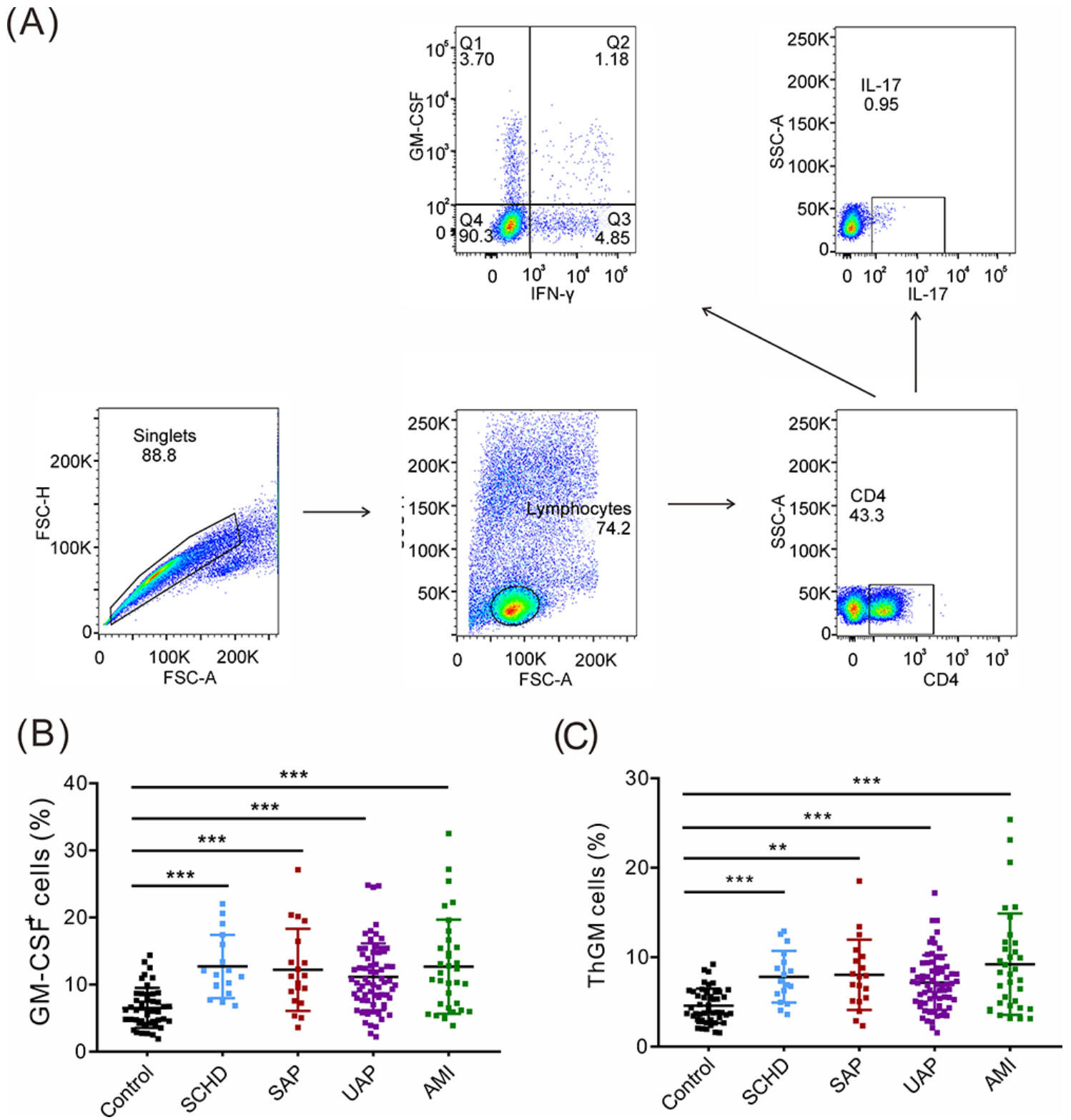
Figures

(A)

(B)

(C)

Figure 1
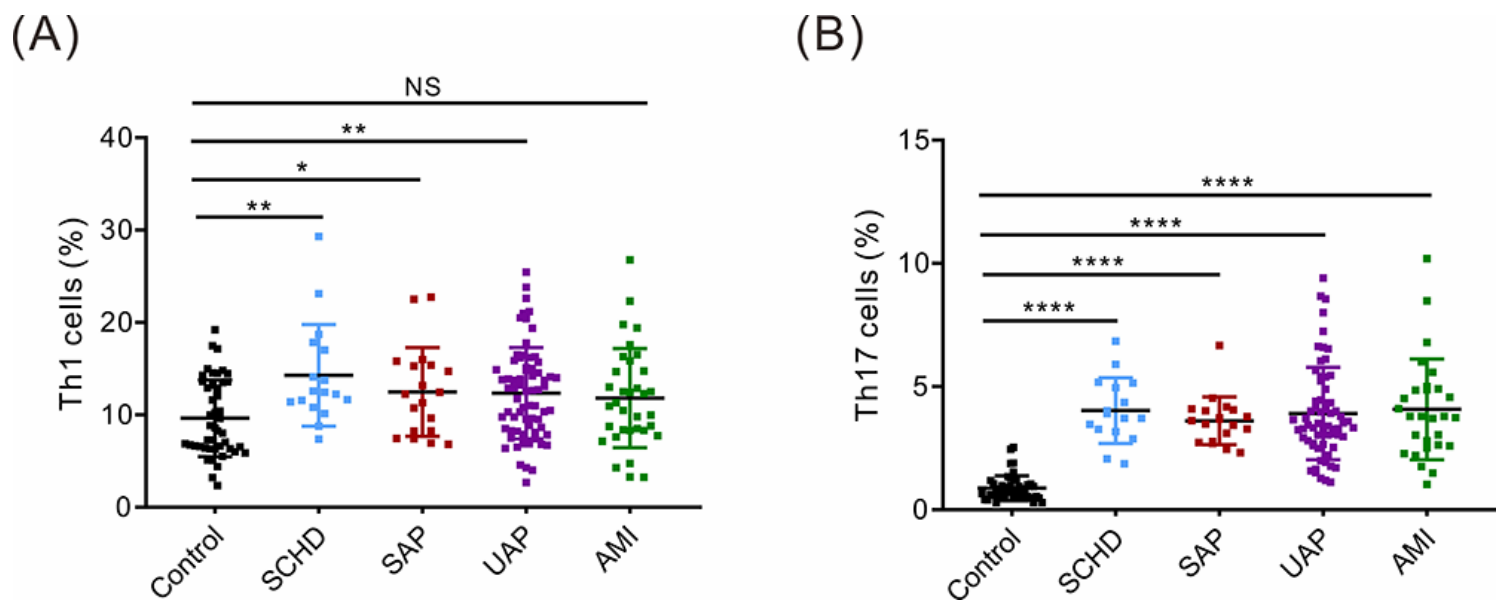
Legend not included with this version

Figure 2
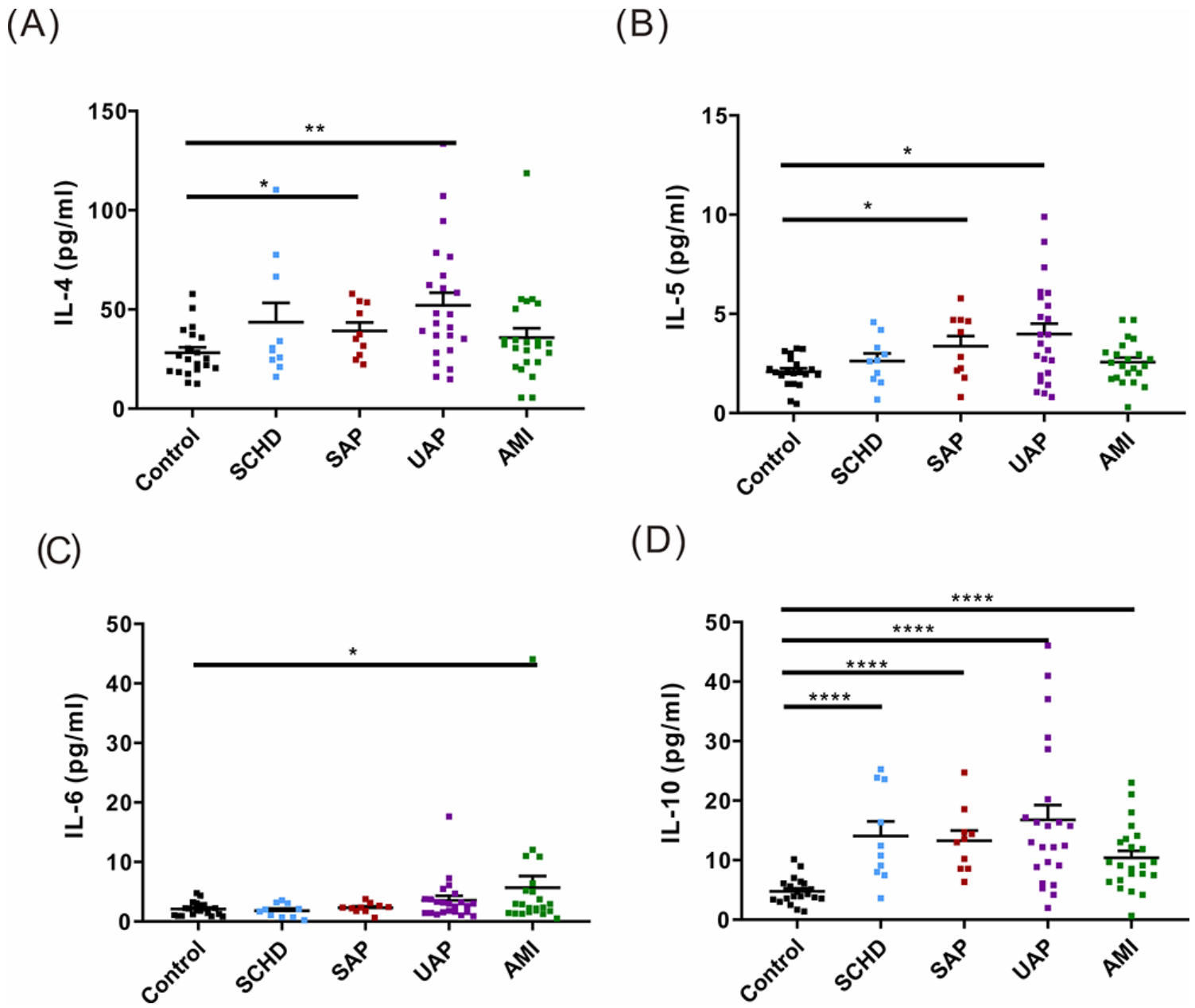
Legend not included with this version

Figure 3
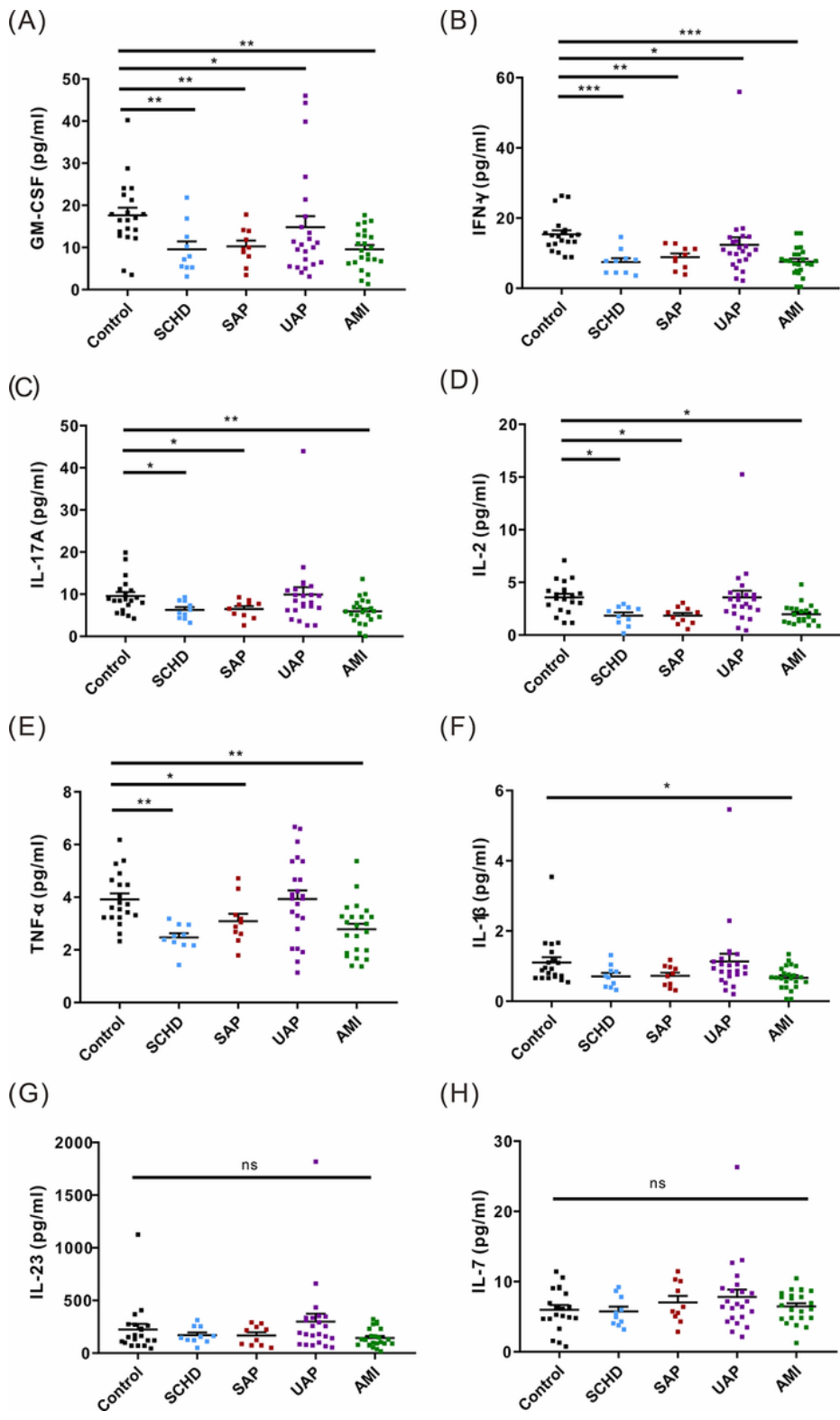
Legend not included with this version

Figure 4

Legend not included with this version

Supplementary Files

This is a list of supplementary files associated with this preprint. Click to download.

- SupplementallyTable1.xlsx