Serum apolipoprotein A-1 is related to inflammatory factors in the acute phase of gout

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Research

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Abstract

BACKGROUND

To observe the correlation between serum apolipoprotein A-1 (apoA-1) and inflammatory factors in patients with gouty arthritis.

METHODS

From February to September 2020, 97 patients with gout (gout group) and 70 healthy controls (control group) were selected as the study subjects. All patients were admitted to the outpatient department of Beijing Luhe Hospital affiliated to Capital Medical University. Of 97 patients in the gout group, 7 patients had gout in the acute phase. Serum concentrations of apoA-1, NLRP3 inflammasome (NACHT-LRR-PYD protein 3, NLRP3), interleukin-1 (IL-1) and interleukin-9 (IL-9) were detected. The correlation of serum apoA-1 concentration, gout-related inflammatory factors (NLRP3, IL-1 beta, IL-9) and other clinical and laboratory indicators was analyzed.

RESULTS

The serum apoA-1 concentration in the gout group was lower than that in the control group (P<0.05). With the increase of serum uric acid level, serum apoA-1 concentration decreased (R^2=0.3160, P<0.05). Multiple linear analyses were performed to increase blood glucose, blood lipid, liver and kidney, etc., and the correlation remained (R=3.36, P<0.05). With the increase of serum IL-1 beta concentration, serum apoA-1 concentration decreased (R^2=0.3993, P<0.05). Multiple linear analyses were performed to increase blood glucose, blood lipid, liver and kidney, etc., and the correlation remained (R=2.95, P<0.05).

CONCLUSIONS

In the acute stage of gout, as the serum uric acid level increases, the serum IL-1β concentration increases and the apoA-1 concentration gradually decreases, which may indicate that apoA-1 participates in the inflammatory response of gout to a certain extent.

Background

Gouty arthritis, purine metabolism, and (or) uricosuric disorder caused by a heterogeneous group of disorders. Hyperuricemia (400umol/L at 37 degrees Celsius, or 6.8mg/dl) is the main biochemical basis of gout, but not all patients with hyperuricemia eventually develop gout, and blood uric acid levels are not absolutely related to gout[1]. In recent years, the factors that promote the development of gout in patients with hyperuricemia have become a research hotspot[2]. As we all know, gout patients are often accompanied by hyperlipidemia, apoA-1 is mainly synthesized by liver cells and intestinal cells, and is also the main component of high-density lipoprotein (HDL). Research in the cardiovascular field confirms that HDL has a pleiotropic anti-atherosclerotic effect on the one hand, and the second main feature is its anti-inflammatory properties[3]. HDL may confer anti-atherosclerosis and anti-inflammatory properties of apoA-1. However, there are few reports on the anti-inflammatory properties of apoA-1 in gout. In this study, we tested the levels of apoA-1, gout-related inflammatory factors (NLRP3, IL-1β, IL-9), and apoA-1 in patients with gout correlation with inflammatory factors. In order to better understand the relationship between apoA-1 and the occurrence and development of gout.

Methods

All patients in this study were admitted to the Department of Rheumatology and Immunology, Beijing Luhe Hospital, Capital Medical University (Beijing, China) from February to September 2020. Participants in this study were divided into two groups, including 97 patients with gout, and 70 healthy controls who were selected to match the BMI of the gout group. To exclude the effects of varying estrogen levels, all patients were male. All patients with gout met the diagnostic criteria for gout arthritis jointly issued by the American College of Rheumatology/European League Against Rheumatism in 2015. Subjects in the gout group were all in the acute stage of gout (within 7 days after the onset of arthritis). No gout patients required a low-fat/low-purine diet or had taken drugs that affect uric acid metabolism within 2 weeks before enrollment. The exclusion criteria were subjects with hyperlipidemia, diabetes, coronary atherosclerotic heart disease, hypertension, abnormal liver function, renal insufficiency, thyroid disease, other chronic diseases, and infections such as bacteria, fungi, and viruses. A visit was scheduled within 7 days in fasting conditions for blood sampling (approximately 4 mL), which was followed by a visit and an interview by the same investigator to collect arthritis risk and health information. All subjects were tested for serum NLRP3, IL-1β, IL-9 and biochemical indicators. Additionally, all subjects were tested by ELISA for APOA1.

Biochemical measurements. Blood was collected in specific tubes for routine biochemistry (blood glucose, lipids, liver function tests, creatinine) and in one 10mL EDTA tubes (4 mL) that were immediately centrifuged to collect the plasma that was stored in 1mL aliquots at -80°C for the subsequent analysis. All the assays were performed in a double-blinded manner.

ELISA for NLRP3, IL-1β, IL-9, APOA1. Plasma level of NLRP3, IL-1β, IL-9, APOA1 was quantified by ELISA, according to the instruction (Shanghai Enzyme-linked Biotechnology Co., Ltd., Shanghai, China).

Data analysis. Statistical analysis of the experimental results using JMP Pro14.0 statistical software. Quantitative data is expressed as the mean value ± standard deviation. The t-test was used for statistical comparison between the two groups. ANOVA test and rank sum test were used for comparison between multiple groups. The correlation analysis uses Spearman correlation analysis. P value <0.05 was considered statistically significant. R programming language, version 3.5.1 matching software was used to match the research subjects of gout group and control group.

Results
Basic situation. There was no difference in gender and BMI among all subjects, and the gout group was younger. The blood pressure, blood creatinine, blood glucose and blood lipid of all the subjects were normal. Blood pressure, blood creatinine, and blood glucose increased in the gout group. High-density lipoprotein cholesterol decreased in gout group; blood uric acid increased in gout group:

The subjects in the two groups (gout group and control group) were all male, and there was no statistically significant difference between the sexes (P > 0.05). There was no significant difference in body mass index (BMI) between the two groups of subjects (P > 0.05). There was no significant difference in age between the gout group (remission period) and the control group (P > 0.05).

The blood pressure, blood creatinine, blood sugar and blood lipids of the two groups of subjects were all within the normal range. Among them, the blood pressure, blood creatinine, and blood glucose levels of the gout group were higher than those of the control group, and the difference was statistically significant (P value < 0.05). The high-density lipoprotein cholesterol in the gout group (acute phase and remission phase) was lower than that in the control group, the difference was statistically significant (P value < 0.05). The blood uric acid in the gout group was higher than that in the control group, the difference was statistically significant (P < 0.05). (Table.1)

| Table 1  | Clinical characteristics of the hospital-based study population |
|----------|---------------------------------------------------------------|
| n        | age   | BMI  | SBP | DBP | Cr  | GLU  | UA  | apoA-1 | NLRP3 | IL-1β | IL-9 | TG | TC |
|          | years | kg/m² | mmHg | mmHg | umol/L | mmol/L | ug/ml | pg/ml | pg/ml | pg/ml | mmol/L | mmol/L |
| threshold values | 57–97 | 3.9–6.1 | 208–428 |
| Gout     | 97    | 33.71±10.69 | 25.94±3.39 | 120.36±8.17 | 80.06±7.10 | 86.16±10.83 | 5.32±0.47 | 485.79±96.41 | 11.75±4.72 | 116.86±81.25 | 91.53±49.19 | 34.46±14.57 | 1.19±0.35 | 4.48±0.64 |
| Control  | 70    | 37.10±8.03 | 25.10±3.26 | 113.17±15.36 | 72.97±9.06 | 82.22±8.06 | 4.97±0.42 | 368.37±34.79 | 20.54±11.10 | 97.50±19.34 | 41.46±6.53 | 23.64±4.49 | 1.08±0.36 | 4.52±0.56 |
| P        | 0.0621 | 0.2165 | <0.0001 | 0.0001 | 0.0353 | 0.0002 | <0.0001 | <0.0001 | 0.0258 | <0.0001 | <0.0001 | 0.1168 | 0.7081 |

P in the table refers to the P values of the paired t-test in the healthy group and the gout group. BMI: body mass index, SBP: systolic blood pressure, DBP: diastolic blood pressure, Cr: creatinine, GLU: blood glucose, UA: uric acid, IL-1β: interleukin-1β, IL-9: interleukin-9, TG: triglycerides, TC: total cholesterol, HDL-C: high-density lipoprotein cholesterol, LDL-C: low-density lipoprotein cholesterol.

**apoA-1 and inflammation index level**

The plasma apoA-1 decreased in the gout group, and the plasma IL-1β, IL-9, NLRP3 increased

the plasma apoA-1 in the gout group was lower than the control group, the difference was statistically significant (P < 0.05). The plasma IL-1β, IL-9 and NLRP3 in the gout group (acute phase and remission phase) were higher than the control group, the difference was statistically significant (P value < 0.05). (Table.1)

**Correlation analysis**

With the increase of blood uric acid, the concentration of apoA-1 decreased, R²=0.3160, P < 0.05. As IL-1β increased, the apoA-1 concentration decreased, R²=0.3993, P < 0.05. (Fig. 1) Multivariate linear analysis of plasma apoA-1 levels with age, BMI, blood pressure, blood creatinine, blood glucose, blood lipids, blood uric acid, inflammation indicators (NLRP3, IL-1β, IL-9) to confirm the relationship between plasma apoA-1 and blood uric acid, the correlation holds, β=0.03, OR=3.36, P < 0.05; the correlation between apoA-1 and IL-1β holds, β=0.065, OR=2.95, P < 0.05. (Table.2)
The study was approved by the Ethics Committee of Beijing Luhe hospital, Capital Medical University (2020-LHKY-019-03).


declarations

Discussion

With the improvement of living standards, the prevalence of gout has risen sharply in the past two decades, becoming an increasingly serious public health problem[5]. Repeated attacks of acute gout affect people's health-related quality of life[6]. Chronic gout cause joint damage and dysfunction [7]. As a result, hospitalizations caused by gout continue to increase, further increasing the social and economic impact of the disease. Hyperuricemia is the biochemical basis for the occurrence of gout. Abnormal blood uric acid metabolism and blood lipid metabolism are inextricably linked[8,9]. Therefore, apoA-1 is selected as the core of the research. To explore the correlation with blood uric acid level and inflammation-related indexes of gout.

Apolipoprotein A-I (apoA-1), mainly synthesized by hepatocytes and intestinal cells, is the main component of high density lipoprotein (HDL) [2]. HDL has a versatile anti-atherosclerotic effect on the one hand, the second main feature is its anti-inflammatory properties [3,10]. HDL may be given an anti-atherosclerotic and anti-inflammatory properties of apoA-1. The anti-inflammatory properties of HDL in experimental models in vitro and in vivo are consistent [11-13]. In the study of rheumatoid arthritis, the plasma apoA-1 and HDL levels of newly diagnosed rheumatoid arthritis patients were lower than those of normal people. In contrast, apoA-1 levels in the synovial fluid of patients with rheumatoid arthritis are elevated. Although the level of apoA-1 in joint fluid is only less than 1/10 of the plasma level, it indicates that apoA-1 infiltrates in the inflammatory joints of rheumatoid arthritis [14]. In immune-related inflammation, immune inflammatory cells infiltrate the target tissue, resulting in tissue damage. In chronic inflammation, T lymphocytes are activated, thereby activating monocytes, and monocytes produce a large number of inflammatory factors. These inflammatory factors include Tumor Necrosis Factor-α (TNF-α) and IL-1β, which in turn triggers a series cascade reaction. The imbalance of inflammatory factors and their inhibitors is one of the characteristics of chronic inflammation. Studies have confirmed that apoA-1 inhibits the activation of monocytes by T lymphocytes. Thereby inhibiting the production of TNF-α and IL-1β, apoA-1 interferes with the interaction of T lymphocytes and monocytes[15,16].

Gouty arthritis has the activation of NLRP3 inflammatory body, and the subsequent maturation and secretion of IL-1β[17,18]. IL-9 is produced by various immune cells, such as helper T cells, mast cells, eosinophils, and type 2 congenital lymphocytes, and depends on them to perform various functions. A large number of studies have confirmed the various in vivo functions of IL-9, including the promotion of allergic and immune diseases. This study found that plasma NLRP3, IL-1β, and IL-9 in the gout group were all elevated, which was consistent with previous studies[19,20]. This study confirmed that with the increase of blood uric acid level, IL-1β concentration increased, plasma apoA-1 concentration decreased. It suggests that apoA-1 may show anti-inflammatory activity by affecting inflammatory factors. The mechanism of action of apoA-1 has not been fully elucidated, and further research is to be done.

Declarations

Ethics approval and consent to participate

The study was approved by the Ethics Committee of Beijing Luhe hospital, Capital Medical University (2020-LHKY-019-03).

Consent for publication

The value of each factor was analyzed using the general linear model for unbalanced data with Bonferroni’s multiple comparison tests. IL-1β, Interleukin-1β, IL-9: Interleukin-9, BMI: Body mass index, SBP: Systolic blood pressure, DBP: Diastolic blood pressure, Cr: Creatinine, Glu: Blood glucose, UA: Uric acid, TG: Triglycerides, TC: Total cholesterol, LDL-C: Low-density lipoprotein cholesterol, HDL-C: High-density lipoprotein cholesterol.

Table 2
Multivariate correlation analysis of basic data of two groups of patients

| Variable         | β     | Standard error | OR    | 95% Confidence interval | P    |
|------------------|-------|----------------|-------|-------------------------|------|
| IL-1β(pg/ml)     | -0.065| 0.02           | -2.95 | -0.10~0.02              | 0.0040|
| NLRP3(pg/ml)     | 0.01  | 0.02           | 0.56  | -0.02~0.04              | 0.5737|
| IL-9(pg/ml)      | 0.13  | 0.11           | 1.18  | -0.08~0.35              | 0.2426|
| age(years)       | 0.15  | 0.08           | 1.72  | -0.02~0.33              | 0.0887|
| BMI(kg/m²)       | 0.12  | 0.30           | 0.4   | -0.48~0.73              | 0.6908|
| SBP(mmHg)        | -0.12 | 0.10           | -1.15 | -0.33~0.08              | 0.2525|
| DBP(mmHg)        | -0.12 | 0.14           | -0.89 | -0.40~0.15              | 0.3746|
| Cr(umol/L)       | 0.06  | 0.08           | 0.72  | -0.11~0.24              | 0.4707|
| Glu(mmol/L)      | -3.08 | 1.87           | -1.65 | -6.8~0.63               | 0.1029|
| UA(umol/L)       | -0.03 | 0.01           | -3.36 | -0.05~0.01              | 0.0011|
| TG(mmol/L)       | 3.85  | 3.13           | 1.23  | -2.36~10.08             | 0.2218|
| TC(mmol/L)       | 3.10  | 5.48           | 0.57  | -7.77~13.98             | 0.5720|
| LDL-C(mmol/L)    | -3.83 | 5.51           | -0.7  | -14.77~7.09             | 0.4876|
| HDL-C(mmol/L)    | 1.07  | 7.46           | 0.14  | -13.74~15.88            | 0.8861|

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All authors provide consent for publication of this paper.

**Availability of data and materials**

All data generated and analyzed in this study are included in this published article. The datasets are available from the corresponding author on reasonable request.

**Competing interests**

All authors of this paper have no competing interests to disclose.

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**Authors' contributions**

Yuan Wang contributed to the study conception and design. Material preparation and data collection were performed by all authors. The data analysis and first draft of the manuscript was written by Yuan Wang, and all authors commented on previous versions of the manuscript. All authors read and approved the manuscript.

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Figures

Figure 1

apoA-I is associated with blood uric acid and IL-1β