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Original Article

The relationship between serum levels of cholesterol and disease activity in Sjogren's syndrome: a retrospective study

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Abstract: Lipid metabolism is dysfunctional in many non-metabolic diseases. In past studies, it has been proved that lipid levels are related to disease activities in some autoimmune diseases. However, there are few studies on lipid metabolism in Sjogren's syndrome (SS). Therefore, in this present case-control study, we compared the cholesterol levels of 93 SS patients and 73 healthy controls using an independent t-test. The results showed that the levels of total cholesterol (TC), high-density lipoprotein cholesterol (HDL-C), and low-density lipoprotein cholesterol (LDL-C) in SS patients were all lower. The clinical data of these patients were converged and analyzed with the patients' serum cholesterol levels using a Pearson correlation. The serum levels of TC, HDL-C, and LDL-C were all correlated with albumin, globulin, IgG, and the EULAR Sjogren's syndrome disease activity index (ESSDAI). Furthermore, patients with anti-SSA antibodies, as well as those with cytopenia, had lower levels of TC and LDL-C, but not HDL-C. In contrast, patients with anti-SSB antibodies had lower levels of TC, HDL-C, and LDL-C. In conclusion, cholesterol metabolism disorders exist in SS patients and are associated with autoantibody production and the ESSDAI, which indicates that serum cholesterol levels might be an effective indicator for assessing disease activity in SS.

Keywords: Total cholesterol, low-density lipoprotein cholesterol, high-density lipoprotein cholesterol, autoantibody, Sjogren's syndrome

Introduction

Lipid metabolism has been shown to be dysfunctional in many non-metabolic diseases, including some autoimmune diseases. For example, some studies have indicated higher serum levels of total cholesterol (TC), low-density lipoprotein cholesterol (LDL-C), triglyceride (TG) [1] and lower levels of high-density lipoprotein cholesterol (HDL-C) in rheumatoid arthritis (RA) [2], which may be related to a high risk of cardiovascular diseases [3]. Some studies verified the association between cardiovascular diseases and impaired serum cholesterol efflux capacity (CEC) [4]. Systemic lupus erythematosus (SLE) patients have been shown to have lower levels of HDL-C and abnormal CEC [5].

Sjogren's syndrome (SS) is a common autoimmune disease characterized by ocular and oral dryness. A large amount of inflammatory cell infiltration and autoantibody production have been confirmed in the salivary glands of SS patients and mice. In addition, lipid deposition in the salivary glands of NOD mice was found to be related to lymphocyte infiltration [6], to which impaired lipid efflux might contribute. However, few studies have reported on serum lipid metabolism in SS patients or mice. The purpose of our study is to prove the metabolism dysfunction of serum cholesterol and its correlation with disease activity.

Patients and methods

The international classification criteria (2016) for primary Sjogren's syndrome was used as our inclusion criteria. The exclusion criteria included infection, tumor, hypertension, diabetes, cardiovascular diseases, and other autoimmune diseases. None of the participants took glucocorticoids or lipid-lowering drugs within 1 month prior to this study.

Table 1. A comparison of the cholesterol levels between patients with Sjogren's syndrome and the healthy controls

	TC (mmol/l)	HDL-C (mmol/l)	LDL-C (mmol/l)
SS patients (n=93)	4.33±0.94	1.15±0.29	2.17±0.65
Healthy controls (n=73)	4.67±0.74	1.40±0.31	2.41±0.62
t value	-2.534	-5.467	-2.402
p value	0.012	<0.001	0.017

We collected the clinical data of 93 SS patients and 73 healthy controls from January 2013 to June 2018 in the Third Affiliated Hospital of Soochow University. The collected data included serum cholesterol levels, age, gender, course of the disease, the erythrocyte sedimentation rate (ESR), C-reactive protein (CRP), blood routine, liver function, kidney function, immunoglobulin (Ig), complements, autoantibody (ANA), extractable nuclear antibody (ENA), and the rheumatoid factor (RF). The disease activity of the SS patients was evaluated using the EULAR Sjogren's syndrome disease activity index (ESSDAI).

The cholesterol levels between the SS patients and the healthy controls were compared using an independent t-test. One-way ANOVA was used to compare cholesterol levels among the healthy controls and the SS patients with or without anti-SSA or anti-SSB antibodies or system involvement. The correlations between cholesterol levels and other clinical data were analyzed, and then a multiple linear regression was tested among these data. All the statistical analyses were conducted using SPSS 19.0.

Results

SS patients have lower serum levels of TC, HDL-C, and LDL-C

Ages, BMI and levels of TC, HDL-C and LDL-C in both groups were normally distributed. The mean (S.D.) age of SS patients and healthy controls was 55.42 (12.44) years and 52.47 (8.57) years, respectively (P>0.05). There was no difference in BMI between SS (22.74±3.17) and the healthy controls (23.53±2.93) either. Two patients were males and the others were females, the same as in the healthy control group.

The serum levels of TC were lower in the SS patients compared with the healthy controls (4.33±0.94 mmol/l *vs.* 4.67±0.74 mmol/l, P<0.05). The mean (S.D.) level of HDL-C was 1.15 (0.29) mmol/l and 1.40 (0.31) mmol/l in the SS patients and healthy controls, respectively (P<0.05). The mean (S.D.) level of LDL-C was 2.17 (0.65) mmol/l and 2.41 (0.62) mmol/l in the SS patients and healthy controls, respectively (P<0.05) (**Table 1**).

Serum TC, HDL-C and LDL-C levels are correlated with Ig and ESSDAI

Table 2 shows the correlations between the serum TC, HDL-C, LDL-C levels and the other clinical data in the SS patients. All TC, HDL-C, and LDL-C levels were correlated with globulin, albumin, IgG and ESSDAI. Furthermore, the TC and LDL-C levels were positively correlated with age, disease course, lymphocyte count, platelet count, creatinine, blood urea nitrogen (BUN), and their complement levels.

Since the multiple linear regression was tested among the cholesterol levels and the correlated data, TC correlated with globulin, IgG, and IgA. LDL-C correlated with the lymphocyte count and C4 (**Table 3**).

Patients with anti-SSA/SSB antibodies have lower serum levels of cholesterol

As the TC, HDL-C, and LDL-C levels were all negatively correlated with globulin and IgG, we speculated that the cholesterol levels were relevant to ANA. According to the ENA results, the patients were divided into two groups. Their cholesterol levels were compared with the healthy controls using a one-way ANOVA.

Compared with the other two groups, the patients with anti-SSA antibodies had lower levels of TC, but the anti-SSB positive group showed lower levels of TC, HDL-C, and LDL-C (**Table 4**).

Patients with cytopenia have lower levels of cholesterol

Since the serum TC, HDL-C, and LDL-C levels were negatively correlated with ESSDAI, we suspected that the serum cholesterol levels correlated with disease activity. According to the details of the ESSDAI, the patients were divided into positive and negative groups based on their systemic symptoms, such as pulmo-

Table 2. The correlation between TC, HDL-C, LDL-C levels and other clinical data in SS patients

	TC		HDL-C		LDL-C	
	R value	*p* value	R value	*p* value	R value	*p* value
Age (yr)	0.282	0.006	0.063	0.548	0.278	0.007
Course (yr)	0.251	0.015	0.130	0.214	0.255	0.014
WBC ($\times 10^9$)	0.145	0.164	0.065	0.536	0.122	0.243
Neutrophils ($\times 10^9$)	0.012	0.912	0.001	0.989	-0.012	0.912
Lymphocytes ($\times 10^9$)	0.414	<0.001	0.167	0.110	0.416	<0.001
Hb (g/l)	0.186	0.074	0.215	0.039	0.140	0.181
PLT ($\times 10^9$)	0.209	0.044	0.154	0.142	0.263	0.011
ESR (mm/h)	-0.099	0.346	-0.136	0.193	-0.070	0.503
CRP (mg/l)	-0.038	0.716	-0.145	0.166	0.049	0.638
ALT (u/l)	-0.137	0.191	-0.049	0.643	-0.131	0.211
AST (u/l)	-0.090	0.389	0.117	0.265	-0.099	0.346
γGT (u/l)	0.003	0.977	-0.116	0.267	0.019	0.860
ALP (u/l)	0.075	0.478	-0.129	0.216	-0.031	0.771
LDH (u/l)	0.139	0.185	0.184	0.077	0.107	0.306
TBil (mmol/l)	-0.043	0.686	-0.015	0.883	-0.19	0.853
DBil (mmol/l)	-0.209	0.044	-0.073	0.485	-0.154	0.141
IBil (mmol/l)	0.075	0.467	0.025	0.809	0.072	0.491
Globulin (g/l)	-0.287	0.005	-0.204	0.049	-0.289	0.005
Albumin (g/l)	0.292	0.005	0.217	0.037	0.218	0.036
SCr (umol/l)	0.335	0.001	0.055	0.599	0.297	0.004
BUN (mmol/l)	0.345	0.001	0.113	0.280	0.320	0.002
BUA (mmol/l)	0.091	0.386	-0.100	0.340	0.159	0.127
GLU (mmol/l)	0.165	0.113	-0.122	0.244	0.168	0.108
IgA (g/l)	-0.251	0.015	-0.040	0.706	-0.239	0.021
IgG (g/l)	-0.369	<0.001	-0.231	0.026	-0.329	0.001
IgM (g/l)	0.008	0.937	-0.072	0.495	-0.001	0.990
C3 (g/l)	0.224	0.031	0.011	0.914	0.265	0.010
C4 (g/l)	0.284	0.006	-0.020	0.848	0.361	<0.001
RF	-0.270	0.018	-0.159	0.171	-0.265	0.021
ANA	-0.194	0.063	-0.048	0.649	-0.120	0.250
ESSDAI	-0.271	0.009	-0.248	0.017	-0.212	0.042

nary diseases, skin lesions, cytopenia, and so on. Patients with cytopenia had lower levels of TC, HDL-C, and LDL-C compared with the healthy controls and lower levels of TC compared with the cytopenia-negative group (**Table 5**).

Discussion

Dyslipidemia is widespread in the general population, in which hyperlipidemia is more commonly detected. Abnormal lipid metabolism is closely related to coronary heart disease [7]. A high incidence of dyslipidemia has been observed in patients with autoimmune diseases and partially correlates with disease activity. It has been confirmed that SLE patients with hyperlipidemia are more susceptible to lupus encephalopathy, nephritis, and femoral head necrosis [8]. Though they have lower levels of serum cholesterol, a higher risk of cardiovascular accident has been proved in RA patients [9]. Furthermore, the HDL-C level is negatively correlated with the disease activity of RA [10].

As a common autoimmune disease with multisystem involvement, SS is characterized by lymphocyte infiltration and ANA production. Metabolic syndrome has been reported in SS [11], but the risk of acute myocardial infarction is similar between SS patients and the healthy controls [12]. In the present study, lower serum levels of TC, HDL-C, and LDL-C were detected in SS patients. To determine the link between cholesterol levels and the disease condition of SS, we further explored the correlation between cholesterol levels and other clinical data.

Through the correlation analysis, we found that the serum levels of cholesterol all correlated with globulin and IgG. Patients with anti-SSA/SSB antibodies had lower serum levels of cholesterol. Therefore, we suspected that the lower cholesterol levels were related to ANA production. Meritxell et al. have confirmed that feeding mice a high-cholesterol diet (HCD) results in an up-regulated marginal zone B (MZB) cell surface expression of PDL1 and increases the interaction between MZB cells and pre-Tfh (T follicular helper) cells, leading to PDL1-mediated suppression of Tfh cell motility, the alteration of Tfh cell differentiation, reduced Tfh abundance, and the suppression of the proath-

Table 3. A multiple linear regression among cholesterol levels and the correlated data

	Independent variables	β value	t value	*p* value	95% CI
TC	globulin	0.625	2.323	0.024	0.010~0.131
	IgG	-0.681	-2.770	0.007	-0.178~-0.029
	IgA	-0.454	-2.340	0.023	-0.288~-0.023
LDL-C	lymphocyte count	0.384	2.905	0.005	0.129~0.697
	C4	0.275	2.121	0.038	0.147~5.002

Table 4. A comparison of the cholesterol levels among anti-SSA/SSB positive, negative and the healthy control groups

anti-SSA	TC (mmol/l)	HDL-C (mmol/l)	LDL-C (mmol/l)
Positive group (n=77)	4.23±0.90	1.13±0.30	2.12±0.64
Negative group (n=16)	4.81±0.98	1.23±0.23	2.42±0.66
Healthy controls (n=73)	4.67±0.74	1.40±0.31	2.41±0.62
F value	6.443	15.610	4.324
p value	0.002	<0.001	0.015
anti-SSB	TC (mmol/l)	HDL-C (mmol/l)	LDL-C (mmol/l)
Positive group (n=30)	4.00±0.70	1.05±0.28	1.98±0.56
Negative group (n=63)	4.49±1.00	1.20±0.29	2.27±0.67
Healthy controls (n=73)	4.67±0.74	1.40±0.31	2.41±0.62
F value	6.682	18.104	5.146
p value	0.002	<0.001	0.007

Table 5. A comparison of cholesterol levels among patients with or without cytopenia and the healthy controls

Cytopenia	TC (mmol/l)	HDL-C (mmol/l)	LDL-C (mmol/l)
Positive group (n=49)	4.12±0.95	1.10±0.29	2.05 ±0.67
Negative group (n=44)	4.56±0.88	1.20±0.29	2.31±0.61
Healthy controls (n=73)	4.67±0.74	1.40±0.31	2.41±0.62
F value	6.431	16.552	4.818
p value	0.002	<0.001	0.009

erogenic Tfh response [13]. On the contrary, we speculated that SS patients with lower cholesterol levels had more Tfh differentiation, leading to promoted B cell differentiation and ANA production. The receptor of HDL-C participates in cholesterol efflux via ABCA1/ABCG1, which is an important pathway of cholesterol metabolism. Impaired CEC has been detected in patients with autoimmune diseases [14]. Furthermore, Abca1/g1-deficient mice develop enlarged lymph nodes and glomerulonephritis suggestive of SLE. This lupus-like phenotype was recapitulated in mice with knockouts of Abca1/g1 in dendritic cells (DCs). DC-Abca1/g1 deficiency enhances T cell activation and the polarization of Th1 and Th17 cells [15], which participate in humoral immunity. Therefore, we supposed that the cholesterol efflux was impaired in SS patients, and abnormal cholesterol metabolism in DCs might promote T cell activation and polarization. Once the ANA of ABCA1 is detected [16], other ANAs that affect cholesterol metabolism might be produced in SS patients.

In our present study, serum TC and LDL-C levels were lower in patients with cytopenia, especially in those with lymphopenia and thrombocytopenia. Menno et al. have proved that SR-BI$^{-/-}$ mice suffer from thrombocytopenia [17]. When SR-BI, a marker on platelets and a receptor of HDL, is knocked out, abnormal cholesterol metabolism and thrombocytopenia appear. It remains unclear whether lymphocytes or platelets express a receptor of LDL. Cholesterol sulfate is present on a variety of cells and in human LDL. Platelets adhere to cholesterol sulfate in a concentration-dependent and saturable manner [18]. In patients with thrombocytopenia, the platelet adhesion ability is decreased, which may be attributed to lower levels of LDL-C. Amy et al. have confirmed that B lymphocytes can secrete antibodies against oxidized LDL [19]. Therefore, patients with lymphopenia have lower levels of LDL-C.

Furthermore, we found that LDL-C was positively correlated with SCr, BUN and their complement levels. Abnormal lipid metabolism exists in patients with chronic kidney diseases, which may be related to lipoprotein leakage. Therefore, cholesterol levels might be associated with kidney function. It has been observed that cholesterol crystals can induce complement-dependent inflammasome activation and cytokine release [20], which is consistent with our result of a positive relationship between LDL-C and the complement levels. Furthermore, the

complement system participates in the pathophysiology of atherosclerosis through TLRs [21].

Our retrospective study showed decreased serum cholesterol levels and their relationship with other clinical data in SS. However, the reason for such a reduction and its effect on the disease are still unknown. In a future study, we will further explore the mechanism of abnormal cholesterol metabolism and its impact on the immune system in SS.

In conclusion, the abnormality of cholesterol metabolism in SS patients correlates with autoantibody production and disease activity. It is expected to be an effective indicator for assessing disease activity in SS. We speculate that this abnormality might participate in humoral immunity in SS. Further studies are needed to determine the relationship between cholesterol metabolism and immune reaction in SS.

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Disclosure of conflict of interest

None.

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