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Original research article

Expression of several long noncoding RNAs in peripheral blood mononuclear cells of patients with systemic lupus erythematosus

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ABSTRACT

Purpose: Accumulating evidence has linked long noncoding RNAs (lncRNAs) to autoimmune and inflammatory disorders. This study aimed to detect the expression levels of five lncRNAs (lnc0640, lnc3643, lnc5150, lnc7514 and lncagf) in peripheral blood mononuclear cells (PBMCs) of patients with systemic lupus erythematosus (SLE), as well as their correlation with clinical and laboratory features.

Materials/methods: We recruited 76 patients with SLE and 71 normal controls into the present study, and obtained PBMCs from the blood samples of all study subjects. Expression levels of lncRNAs were determined by quantitative real-time reverse transcription polymerase chain reaction and their associations with clinical and laboratory characteristics were analyzed.

Results: Lnc5150 expression levels were statistically significantly decreased ($Z = -6.016$, $P < 0.001$) compared with normal controls. Lnc3643 levels were also statistically significantly decreased in SLE patients with proteinuria compared with those without ($Z = -2.934$, $P = 0.003$), and the lnc7514 levels were statistically significantly lower in anti-dsDNA(+) patients compared with anti-dsDNA(-) patients. The expression levels of lnc3643 were correlated with C-reactive protein and erythrocyte sedimentation rate (ESR), lnc7514 was correlated with disease activity and ESR (all $P < 0.01$).

Conclusions: The aberrant lncRNA expression levels and their associations with laboratory features in SLE suggest their important role in SLE pathogenesis.

1. Introduction

Systemic lupus erythematosus (SLE) is a severe, prototypic multisystem autoimmune disease [1,2]. The immune system attacks the healthy tissues and cells because the body loses the immunological tolerance to self-antigens in autoimmunity [3]. Therefore, SLE has a characteristics of a multitude of autoantibody production, immune complex deposition and systemic inflammation [4]. This inflammatory reaction can be widespread and affects multiple tissues and organs of the body such as skin, kidneys, musculoskeletal system and nervous system [5]. The etiology and pathogenesis of SLE are complex and not fully elucidated, it is generally accepted that both genetic and environmental factors are implicated in the development and progression of this disease [6]. The emergence of genomics got us a brand new perspective to explore these complex mechanisms and shed light on a new epoch for research of diseases [7]. For a long time, the researchers neglected the importance of long noncoding RNAs (lncRNAs). Fortunately, many studies showed that lncRNAs have been identified to be tightly linked to diverse human diseases including SLE [8,9].

LncRNAs include more than 200 nucleotides in length but without evident protein coding potential on the function. LncRNAs on average have a lower expression level than protein-coding mRNAs, frequently reside in the nucleus and have poorer interspecies sequence conservation [10]. LncRNAs, on the basis of their genomic proximity to protein-coding genes, are classified into sense, antisense, bidirectional, intronic and intergenic [11]. The functions of lncRNAs are far from understood

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now, but a number of researches have reported that some of them are implicated in diverse gene-regulatory mechanisms including gene transcription, RNA splicing, chromatin remodeling and protein transport [11,12].

Emerging studies have revealed association between lncRNAs and autoimmune diseases. Following involvement of toll-like receptors (TLRs), lncRNAs form functional lncRNA–protein complexes that remove repressors of transcription or recruit activators, giving rise to rapid expression of inflammatory mediators [13]. IIott et al. [14] inferred that lncRNAs are involved in the regulation of human innate immunity, and 216 lncRNAs and 35 regions of bidirectional transcriptions (RBTs) expressed differentially in response to lipopolysaccharide (LPS), therefore lncRNA may be essential to monocyte activation. Wu et al. [15] discussed that lncRNA may be involved in innate and adaptive immune responses, and development of immune cell in autoimmune diseases. Recently, Wu et al. [8] found that the linc0949 and linc0597 expression levels decreased dramatically in peripheral blood mononuclear cells (PBMCs) of SLE patients, and their results also validated that lncRNAs were certainly involved in the regulation of innate immunity. Aune et al. [16] showed that the loci of transcribing novel lncRNAs were not randomly distributed across the genome, but co-located with leukocyte transcriptional enhancers, and adjacent genetic mutations related to the risk of autoimmune disease risk.

Based on these recent findings, we hypothesized that lncRNAs may exert a vital role in SLE pathogenesis and serve as potential disease markers. Our recent study [17] showed that the plasma levels of 5 lncRNAs (ENST00000450640: lnc0640; ENST00000583643: lnc3643; ENST00000425150: lnc5150; ENST00000507514: lnc7514; uc001agf.1: lncagf) were statistically significantly different between SLE and healthy controls. In the present study, in order to further determine the relationship between lncRNAs and SLE, we determined the expression levels of these 5 lncRNAs in PBMCs from patients with SLE, and investigated their relationship with disease manifestations and laboratory indexes.

2. Materials and methods

2.1. Subjects

We enrolled 76 SLE patients (69 females, 7 males) and 71 healthy controls (62 females, 9 males) in this study. All the SLE patients in our study were consecutively selected from Anhui Provincial Hospital and the First Affiliated Hospital of Anhui Medical University (China) between February 2016 and May 2016. The 76 SLE patients met the American College of Rheumatology (ACR) diagnostic criteria for the classification of SLE as revised in 1997 [18]. The SLE patients with renal involvement were defined based on any one of the following criteria: (1) persistent proteinuria (≥0.5 g per day); (2) hematuria or the presence of active cellular casts by microscopic examination of urinary sediment; (3) biopsy evidence of lupus nephritis (LN) [19]. The disease severity was assessed by SLE disease activity index 2000 (SLEDAI-2 K) [20]. We obtained healthy controls samples from the Health Examination Center of the Second Affiliated Hospital of Anhui Medical University (China). The healthy controls were defined as healthy people who have no history of any autoimmune and inflammatory diseases, or cancer, and they did not meet any criterion of the ACR diagnostic criteria for the SLE classification as revised in 1997. In the last month before inclusion in the study, the healthy controls were healthy and did not receive any hormones or immunosuppressants and there was no history of autoimmune diseases among the direct relatives of the healthy controls. The demographic, clinical and laboratory data were also obtained.

The mean age of the 76 SLE patients was 37 years (range from 26 to 46), while the mean age of the 71 healthy controls was 40 years (range from 26 to 51). In Table 1 we summarized the basic features and clinical information on the study subjects. The main clinical manifestations were lupus nephritis (42.1%), arthritis (27.6%), rash (46.1%), alopecia (34.2%), and the major laboratory index changes included anti-SSA (72.4%), low complement (64.5%), proteinuria (46.1%), and anti-dsDNA (43.4%).

Table 1
Characteristics of SLE patients and healthy controls.

Characteristics	SLE patients	Healthy controls	*Z*	*P*
Demographic characteristics	76	71	–	–
Age (years), M(P_{25},P_{75})	37(26,46)	40(26,51)	−1.734	0.083
Sex (female/male)	69/7	62/9	−11.412	< 0.001
Disease duration (years), M(P_{25},P_{75})	4.74(0.97,8.69)	–	–	–
SLEDAI-2 K, M(P_{25},P_{75})	12(7,19)	–	–	–
Clinical manifestations, n(%)				
Lupus nephritis	32(42.1)	–	–	–
Arthritis	21(27.6)	–	–	–
Myositis	5(6.6)	–	–	–
Rash	35(46.1)	–	–	–
Alopecia	26(34.2)	–	–	–
Oral ulcer	13(17.1)	–	–	–
Pleuritis	5(6.6)	–	–	–
Fever	25(32.9)	–	–	–
Vision disorder	7(9.2)	–	–	–
Neurological disorder	8(10.5)	–	–	–
Laboratory measurements, n(%)				
Anti-dsDNA	33(43.4)	–	–	–
Anti-Sm	31(40.8)	–	–	–
Anti-SSA	55(72.4)	–	–	–
Anti-SSB	6(7.9)	–	–	–
Anti-RNP	27(35.5)	–	–	–
Anti-Ribosomal P	23(30.3)	–	–	–
Leukopenia	13(17.1)	–	–	–
Thrombocytopenia	27(35.5)	–	–	–
Hematuria	30(39.5)	–	–	–
Proteinuria	35(46.1)	–	–	–
Low complement	49(64.5)	–	–	–
Medical therapy, n(%)				
Prednisone (≥30 mg/day)	26(34.2)	–	–	–
Immunosuppressants	27(35.5)	–	–	–
HCQ	62(81.6)	–	–	–

Abbreviations: M(P_{25},P_{75}) - median (interquartile range); SLEDAI-2K - Systemic Lupus Erythematosus Disease Activity Index 2000; dsDNA - double-stranded DNA; Sm - Smith; SSA - Sjögren's syndrome-related antigen A; SSB - Sjögren's syndrome-related antigen B; RNP - Ribonucleoprotein; Immunosuppressants - azathioprine, cyclophosphamide, cyclosporine, tacrolimus, leflunomide, mycophenolate mofetil and methotrexate; HCQ - Hydroxychloroquine.

2.2. Ethical issues

The protocol for this study was consistent with the provisions of the World Medical Association Declaration of Helsinki 1964 with later amendments, and the study was approved by the Ethics Committee of Anhui Medical University (No.: 20150115). All subjects have given informed written consent before inclusion in the study.

2.3. Clinical and laboratory features

Clinical data of patients with SLE including arthritis, myositis, rash, alopecia, oral ulcer, pleuritis, vision disorder, fever and neurological disorder were surveyed. Laboratory parameters were recorded as well, including thrombocytopenia ($< 100 \times 10^9$/L), leukopenia ($< 3.0 \times 10^9$/L), hematuria (> 5 RBC/HP), pyuria (> 5 WBC/HP) and proteinuria (> 0.5 g/24 h). Meanwhile, elevated C-reactive protein (CRP), increased erythrocyte sedimentation rate (ESR) (> 20 mm/h),

Table 2
Comparison the lncRNAs expression level between different subgroups.

Group	Number	Lnc0640	Lnc3643	Lnc5150	Lnc7514	Lncagf
Healthy controls	71	0.32(0.22,0.65)	0.78(0.56,1.11)	0.90(0.60,1.10)	0.64(0.49,1.25)	0.27(0.20,0.45)
SLE	76	0.53(0.25,1.47)	0.85(0.65,1.34)	0.37(0.24,0.71)[a]	0.59(0.38,1.09)	0.31(0.17,0.48)
P		0.026	0.164	< 0.001	0.162	0.897
SLE patients						
SLE with nephritis	32	0.53(0.18,1.89)	0.76(0.55,0.97)	0.29(0.17,0.66)	0.50(0.33,0.98)	0.25(0.18,0.55)
SLE without nephritis	44	0.53(0.27,1.20)	0.94(0.72,1.44)	0.42(0.29,0.75)	0.62(0.46,1.27)	0.33(0.16,0.45)
P		0.801	0.015	0.053	0.092	0.771
SLE patients						
Female	69	0.55(0.26,1.83)	0.87(0.65,1.38)	0.37(0.21,0.70)	0.61(0.38,1.09)	0.30(0.17,0.49)
Male	7	0.42(0.13,0.90)	0.75(0.55,0.88)	0.57(0.35,1.08)	0.57(0.35,0.73)	0.32(0.24,0.41)
P		0.355	0.159	0.199	0.572	0.716

All the expression levels are displayed as median value (interquartile range).
Abbreviations: SLE - systemic lupus erythematosus; LN - lupus nephritis.
[a] Patients with SLE had statistically significantly lower lnc5150 levels than healthy controls ($Z = -6.016$, $P < 0.001$).

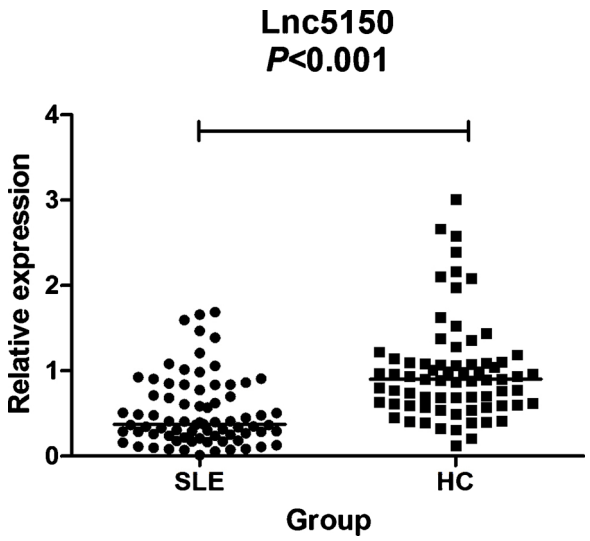

Fig. 1. Comparison of expression of lnc5150 between different groups.
Each symbol represents an individual subjects; horizontal lines indicate median values. The expression levels of the lncRNAs in 76 SLE patients, 71 healthy controls were analyzed by qRT-PCR and normalized by β-actin. SLE: Systemic Lupus Erythematosus; HC: healthy control.

anti-dsDNA, anti-Sm, anti-nuclear antibody (ANA), anti-SSA, anti-SSB, anti-RNP, and anti-ribosomal P; C3/C4 (by immunoturbidimetry) were also collected. In addition, medications used by the study subjects were reviewed.

2.4. PBMC and total RNA

Samples of peripheral blood (5 ml) from each study subject were collected. PBMCs were purified from peripheral blood samples by using Ficoll-Hypaque density gradient centrifugation. Total RNA was extracted from PBMCs using TRIzol reagent (Invitrogen, Carlsbad, CA, USA). RNAs concentration and purity were determined using a NanoDrop™ 2000 spectrophotometer (Thermo Scientific, USA).

2.5. Quantitative reverse transcription polymerase chain reaction (qRT-PCR)

First, residual DNA in the total RNA was removed and then total RNAs were reverse-transcribed into cDNA using the PrimeScript™ RT reagent Kit (Takara Bio Inc, Japan). We dissolved the cDNA samples with DNA-free water and prepared the qRT-PCR reaction system following the manufacturer's instructions. qPCR was performed with SYBR Green (SYBR® Premix Ex Taq™ II, Takara Bio Inc, Japan) and ABI ViiA™ 7 Real-Time PCR System (Applied Biosystems, Foster City, CA, USA) was used to collect the data. Thermal cycle conditions included 1 cycle at 95 °C for 1 min, and 42 cycles at 95 °C for 10 s, followed by 60 °C for 30 s and 72 °C for 1 min. Housekeeping gene β-actin was set as internal reference gene. The primer sequences used for qPCR are shown in Supplement Table S1. Comparative quantification data of lncRNAs expressions were analyzed using the 2-△△Ct method [21].

2.6. Statistical analysis

Statistical analysis was conducted using SPSS 20.0 software (IBM Corp., Armonk, NY, USA). Nonparametric distribution data was expressed as median value and interquartile range (IQR). The gene expression levels were compared between the two groups by nonparametric Mann-Whitney U test. The correlation analyses were performed by Spearman's rank correlation test. Figures were made by GraphPad Prism version 5.0 (GraphPad Software, La Jolla, CA, USA). Receiver operating characteristic (ROC) curves were constructed by MedCalc Statistical Software version 15.2 (MedCalc Software bvba, Ostend, Belgium). The area under curve (AUC) was applied to determine the specificity and sensitivity of lncRNAs which may serve as disease markers. Bonferroni correction was used for multiple testing and a significance threshold of 0.01(0.05/5) was applied in the analysis of the five lncRNAs.

3. Results

3.1. Expression level of lncRNAs in SLE patients

The expression levels of the five lncRNAs (lnc0640, lnc3643, lnc5150, lnc7514 and lncagf) in the PBMCs of 76 SLE patients and 71 healthy controls were detected by qRT-PCR. As shown in Table 2 and Fig. 1, SLE patients had statistically significantly lower lnc5150 levels than the healthy controls ($Z = -6.016$, $P < 0.001$). ROC analysis documented clear separation of SLE patients and controls (AUC 95% *CI*: 0.788(0.713-0.851), $P < 0.001$) (Fig. 2). No statistically significant changes regarding other lncRNAs were observed in SLE (all $P > 0.01$) (Table 2).

When we divided the SLE patients into lupus nephritis (LN) and no nephritis, the expression levels of those five lncRNAs showed no

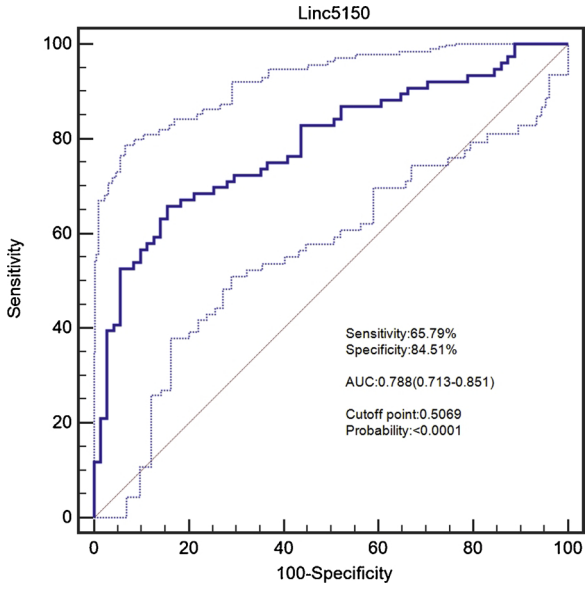

Fig. 2. Receiver operating characteristic (ROC) curve analysis of lnc5150. SLE patients *vs* healthy controls; Area under the curve was 0.788(0.713-0.851), $P < 0.001$. Employing a cut-off of 0.56069 yielded a sensitivity of 65.79% and specificity of 84.51%.

statistically significant difference between other groups ($P > 0.01$). In the SLE group, the expression of five lncRNAs showed no statistically significant difference between female and male (all $P > 0.01$) (Table 2).

3.2. Association of lncRNAs levels with clinical and laboratory characteristics

We assessed the correlations of lncRNAs levels with clinical features or laboratory indexes of SLE patients, however, no statistically significant association between the five lncRNAs expression and any clinical manifestations were observed (all $P > 0.01$) (Table S2).

In laboratory characteristics, as shown in Table 3, Table S3 and Fig. 3, lnc3643 expression was statistically significantly lower in SLE patients with proteinuria than those without ($Z = -2.934$, $P = 0.003$). Moreover, as shown in Fig. 4, the lnc7514 expression in SLE patients with positive anti-double stranded DNA (dsDNA) was decreased compared with those without ($Z = -2.699$, $P = 0.007$). Correlation analysis demonstrated that CRP was correlated with the expression levels of lnc3643, SLEDAI-2 K was correlated with the expression levels of lnc7514, and ESR was negatively correlated with the expression levels of the three lncRNAs (all $P < 0.01$) except the lnc0640 and lncagf. However, complements C3 and C4 as well as the disease duration did not have statistically significant correlations with the lncRNAs expression levels (Table 4).

3.3. Effect of treatment on expression levels of lncRNAs

There was no statistically significant difference in the expression levels of the 5 lncRNAs between patients receiving medium or high-dose prednisone (> 30 mg/d) and patients receiving low-dose prednisone. Additionally, no statistically significant difference was found in the expression levels of the 5 lncRNAs between SLE patients being treated with immunosuppressants and those without immunosuppressants ($Z = -2.350$, $P = 0.019$) (Table 5).

4. Discussion

LncRNAs biochemically resemble mRNAs presented by Jacob and Monod, yet do not serve as the template for protein synthesis [22]. However, accumulating evidence has revealed that lncRNAs may play a vital role in maintaining cellular and tissue homeostasis [23,24]. LncRNAs, which comprise the bulk of human noncoding transcriptome, can be cytoplasmic or nuclear. Currently, lncRNAs have been extensively studied in the field of genomic imprinting and cell differentiation, and considered as the key regulators of various biological processes including autoimmunity [15]. Although the molecular mechanisms of lncRNAs are poorly understood, increasing evidence suggests that lncRNAs could respond to TLRs stimulus, such as lincRNA-Cox2, THRIL and NEAT1 [15]. Wu et al. [8] explored the expression levels of linc0949, linc0597, linc1992 and linc3995 in SLE patients and the results revealed that the expression level of linc0949 was decreased in SLE and that it statistically significantly increased after treatment. It has also been reported that plasma levels of Gas5, lnc-DC and linc0597 were statistically significantly different between healthy controls and SLE patients [25]. LncRNAs may break a new ground for diagnostic and therapeutic targets in SLE by recognition of their roles.

In the current study, we detected the expression levels of five lncRNAs (lnc0640, lnc3643, lnc5150, lnc7514, lncagf) in PBMCs and explored their correlation with clinical and laboratory characteristics of SLE patients. The results demonstrated lower expression of lnc5150 in SLE patients compared with healthy controls. Lnc5150 might disclose a diagnostic value for SLE with AUC as high as 0.788. Moreover, lnc5150 expression levels were statistically significantly correlated with elevated ESR in patients. In addition, the expression levels of lnc3643 were associated with proteinuria, ESR and CRP in SLE patients. As to lnc7514, the expression level was reduced in SLE patients with positive dsDNA compared with negative dsDNA, and correlated with the ESR and SLEDAI-2 K, which demonstrated that lower levels of lnc7514 may thus be beneficial to identify SLE patients with severe disease.

It is well demonstrated that lncRNAs could regulate different kinds

Table 3
Associations between lncRNAs expression level with laboratory parameters of SLE patients.

Group	Number	lnc3643	lnc5150	lnc7514	lncagf
Anti-dsDNA					
+	33	0.86(0.61,1.29)	0.40(0.25,0.82)	0.48(0.32,0.75)[a]	0.29(0.17,0.50)
–	43	0.84(0.66,1.39)	0.36(0.18,0.62)	0.73(0.53,1.09)	0.35(0.17,0.47)
P		0.996	0.393	0.007	0.757
Proteinuria					
+	35	0.72(0.55,0.97)[b]	0.34(0.17,0.49)	0.57(0.33,0.76)	0.27(0.18,0.47)
–	41	0.98(0.78,1.47)	0.48(0.29,0.81)	0.73(0.44,1.30)	0.37(0.17,0.49)
P		0.003	0.060	0.064	0.337

All the expression levels are displayed as median value (interquartile range).
Abbreviations: dsDNA - double-stranded DNA.
[a] lnc7514 expression in SLE patients with positive anti-double stranded DNA (dsDNA) was decreased compared with those without ($Z = -2.699$, $P = 0.007$).
[b] lnc3643 expression was statistically significantly lower in SLE patients with proteinuria than those without ($Z = -2.934$, $P = 0.003$).

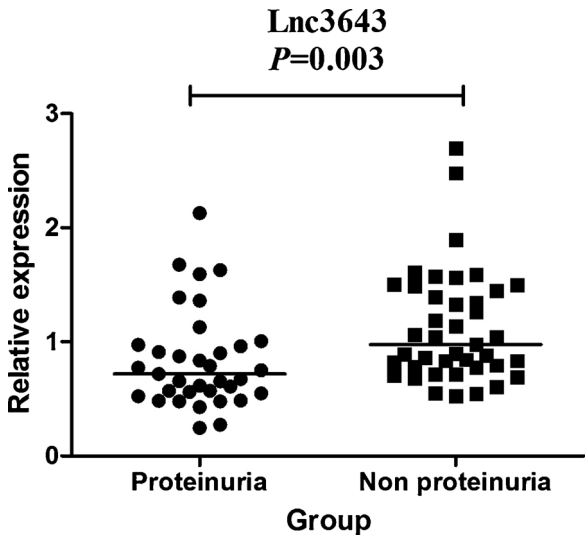

Fig. 3. Comparison of expression of lnc3643 between different groups.
Each symbol represents an individual subjects; horizontal lines indicate median values.
The expression levels of lnc3643 in SLE with proteinuria compared with those without.

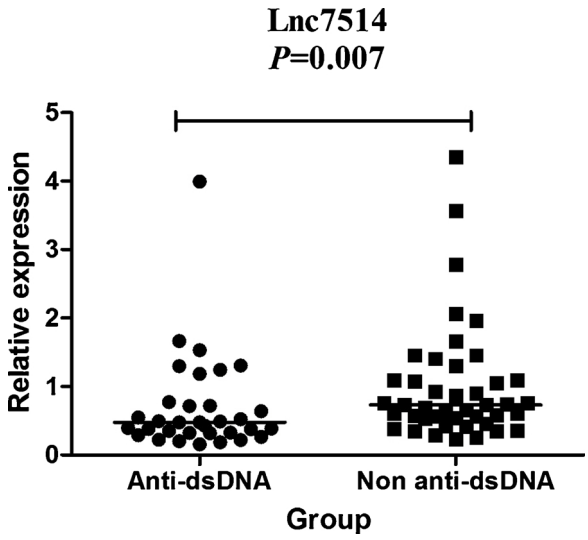

Fig. 4. Comparison of expression of lnc7514 between different groups.
Each symbol represents an individual subjects; horizontal lines indicate median values.
The expression levels of lnc7514 in SLE with anti-dsDNA(+) compared with anti-dsDNA(-).

of cell and tissue development, such as stem cells, erythrocytes, adipocytes [26–29]. In addition, activation, differentiation, and imbalance expression of T cells, B cells, macrophages, and NK cells are directly linked with lncRNAs, which also play a vital role in the autoimmune disease [11,30]. TLRs play a key role in the pathogenesis of SLE [31,32], coincidentally, TLR signaling induces the expression of lncRNAs which was involved in immune regulation [33–35]. Thereby, lncRNA may participate in the pathogenesis and development of SLE.

Li et al. [33] found that the lncRNA THRIL (lincRNA1992) bound specifically to heterogenous nuclear ribonucleoprotein L (hnRNPL) and formed a functional THRIL–hnRNPL complex, which could facilitate the transcription of the tumor necrosis factor-α (TNF-α) gene by binding to its promoter. THRIL was required for expression of many immune-response genes, and the increasing evidence demonstrated that THRIL plays a key role in the innate immune response in humans. Additionally, Rapicavoli et al. [36] found that the production of "Lethe" increases when TNF-α activates Nuclear factor κB (NF-κB), and Lethe could also bind to NF-κB and prevent NF-κB from interacting with DNA, thereby downregulating the production of various inflammatory proteins. Therefore, Lethe also acts as a negative-feedback regulator of the NF-κB signaling pathway to restrain inflammation. It has been revealed that linc0949, linc0597, linc1992 and linc3995 also regulate the induction of TNF-α [33]. However, the pro-inflammatory cytokine TNF-α plays key roles in defensing against inflammatory and immune responses of SLE [37–39]. Zhang et al. [9] found that the expression of lncRNA NEAT1 were abnormally increased in SLE patients, which might attribute to the activation of p38 induced by LPS. However, the exact functions of the lncRNAs in SLE remain to be further explored.

We also evaluated the effect of drugs on the lncRNAs expression. The expression levels of the studied lncRNAs were not statistically significantly different between patients treated with medium to high doses of prednisone and patients receiving low doses of prednisone. In addition, no statistically significant effect of immunosuppressants on lncRNAs expression levels were found. This implied that medical treatment might not affect the expression levels of the studied lncRNAs.

4.1. Limitations of the study

Several limitations should be acknowledged in the present study. First, the sample size was relatively small. Therefore, the conclusion should be treated with caution and further studies with large sample size are still awaited to support the preliminary results. Second, we did not perform a functional study of these 5 lncRNAs, thus the underlying mechanism still needs further investigation.

5. Conclusion

In summary, our study reveals aberrant expression of lnc5150 in PBMCs of SLE patients. In addition, the expression levels of lnc3643 and lnc7514 are correlated with disease activity or CRP, further indicating

Table 4
Correlation of lncRNAs expression levels with several parameters of SLE patients.

Parameters	Number	lnc0640		lnc3643		lnc5150		lnc7514		lncagf	
		rs	*P*	*rs*	*P*	*rs*	*P*	*rs*	*P*	*rs*	*P*
C3	72	−0.120	0.315	0.079	0.512	−0.077	0.522	0.000	0.997	−0.055	0.647
C4	60	−0.063	0.634	−0.087	0.509	−0.142	0.278	0.055	0.677	−0.067	0.614
ESR	72	−0.176	0.139	−0.408	< 0.001	−0.310	0.008	−0.380	< 0.001	−0.271	0.022
CRP	66	−0.157	0.207	−0.442	< 0.001	−0.162	0.194	−0.262	0.033	−0.018	0.884
SLEDAI-2 K	76	−0.159	0.170	−0.242	0.035	−0.083	0.475	−0.308	0.007	−0.187	0.108
Disease duration	76	−0.127	0.273	0.114	0.329	0.054	0.642	0.000	0.998	−0.093	0.428
Age	76	0.126	0.277	0.046	0.692	−0.086	0.461	0.216	0.062	0.055	0.640

Abbreviations: C3 - complements 3; C4 - complements 4; ESR - erythrocyte sedimentation rate; CRP - C-reactive protein; SLEDAI-2K - Systemic Lupus Erythematosus Disease Activity Index 2000.

Table 5
Associations between lncRNAs expression levels and medical therapy of SLE.

Group	Number	Lnc0640	Lnc3643	Lnc5150	Lnc7514	Lncagf
Prednisone(mg/day)						
≥15	43	0.45(0.24,0.95)	0.88(0.72,1.26)	0.43(0.26,0.84)	0.61(0.42,1.09)	0.31(0.16,0.44)
< 15	33	0.62(0.29,1.97)	0.83(0.59,1.38)	0.35(0.20,0.59)	0.58(0.33,1.13)	0.29(0.18,0.55)
P		0.363	0.450	0.405	0.352	0.497
HCQ(mg/day)						
Yes	62	0.55(0.27,1.77)	0.89(0.64,1.39)	0.37(0.25,0.64)	0.61(0.38,1.08)	0.29(0.17,0.49)
No	14	0.46(0.16,1.09)	0.71(0.64,0.93)	0.53(0.17,0.93)	0.54(0.38,1.14)	0.44(0.17,0.48)
P		0.630	0.213	0.592	0.820	0.446
Immunosuppressants*						
Yes	27	0.69(0.27,1.97)	1.00(0.82,1.50)	0.34(0.24,0.59)	0.69(0.38,1.30)	0.31(0.17,0.49)
No	49	0.50(0.22,1.29)	0.76(0.61,1.05)	0.40(0.23,0.74)	0.57(0.37,0.91)	0.29(0.17,0.48)
P		0.394	0.019	0.464	0.275	0.956

All the expression levels were displayed as median value (interquartile range).
* Immunosuppressants (including: azathioprine, cyclophosphamide, cyclosporine, tacrolimus, leflunomide, mycophenolate mofetil and methotrexate).

the important role of lncRNAs in SLE pathogenesis. However, in addition to the 5 lncRNAs analysed in the present study, there may be other lncRNAs linked to SLE. Therefore, further studies are still needed to comprehensively explore the role of lncRNAs in SLE.

Appendix A. Supplementary data

Supplementary material related to this article can be found, in the online version, at doi: 10.1016/j.advms.2019.08.002.

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Declaration of Competing Interest

The authors declare no conflict of interests.

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