regulation of oxidative metabolism and potential roles in development of systemic lupus erythematosus (SLE).

Methods CD4 + T lymphocytes isolated from 97 SLE patients, 30 RA patients, 10 AS patients and 20 healthy donors were used in our in vitro experiments. Total RNA of CD4 + T lymphocytes isolated from 3SLE patients and 3 healthy donors were used to perform small RNA sequencing. Total RNA of CD4 + T lymphocytes transfected with synthetic tRF-3009 or negative control (a random sequence, single strand) were used to perform next generation sequencing. CD4 + T lymphocytes isolated from healthy donors were transfected with tRF-3009/negative control or tRF-3009 siRNA/si-NC in vitro, with/without IFN-alpha treatment, to analyze OCRROS and ATP concentration. Real-time qRT-PCR was performed to analyze expression of tRF-3009 and related gene expressions.

Results We have identified a series of tRFs expressed abnormally in SLE CD4 + T lymphocytes. Interestingly, almost all up regulated tRFs were from 3’end of mature trna while down regulated ones were from 3’end of trna precursors. We have found that expression of tRF-3009 was correlated with lupus nephritis (LN) and urine protein (PRO). The expression of tRF-3009 could be induced by IFN-alpha treatment in vitro. Knockdown of tRF-3009 by siRNA could rescue the metabolism change of CD4 + T lymphocytes induced by IFN-alpha. Transfection of tRF-3009 alone could up regulate oxidative phosphorylation of CD4 + T lymphocytes in vitro.

Conclusions This study identified a series of abnormally expressed tRFs existed in CD4 + T lymphocytes isolated from SLE patients. One of these small RNAs, tRF-3009 also enriched in kidney and correlated with occurrence of lupus nephritis. tRF-3009 anticipated in metabolism regulation, may play important roles in SLE pathogenesis. (Suppl 1):A1

Next, 13 variables with a p-value<0.20 were entered into a binary logistic model using backwards stepwise selection. Results 2028 individuals reported currently taking HCQ with 1581 (78.0%) individuals reporting taking the medication exactly as prescribed all of the time, 385 (19.0%) reporting some of the time, and 62 (3.1%) reporting sometimes or never taking HCQ as prescribed. People with lupus were twice as likely to adhere to HCQ if they reported having a rheumatologist as the primary doctor treating their lupus (p=0.046, OR=2.00) and more than twice as likely if they reported having a positive impression of HCQ (p=0.029, OR=2.14) or experienced improvement in treating their symptoms using HCQ (p=0.009, OR=2.49). Notably, African Americans and those indicating race as Other (mixed, American Indian/Alaskan Native, Pacific Islander or Middle Eastern) were 65% less likely to adhere to HCQ treatment (p=0.002, OR=0.35; p=0.021, OR=0.35, respectively).

Conclusions The study confirms the importance of experiential factors such as medication impression and patient-reported symptom improvement in HCQ treatment adherence. Additionally, results highlight the potential significance of having a specialist as the primary lupus doctor. This may be due to rheumatologists higher awareness of the value of HCQ for lupus patients compared to other providers. Further exploration is needed on cultural factors negatively influencing treatment adherence among certain racial groups, particularly African Americans and Other racial groups (mixed, American Indian/Alaskan Native, Pacific Islander or Middle Eastern). Treatment adherence is particularly important in lupus given the complex nature of the disease and fluctuation of disease activity, which may be exacerbated by poor adherence.

Funding Source(s): The National Key Research and Development Program of China(2016YFC0903900 and 2017YFC0909000)
Calculated cell numbers indicate elevated T cells in SLE patients and suppressed T cells in EA ANA+ healthy individuals. 20 cell surface marker expression is shown using dimensionality reduced t-SNE plots from PBMC data (110,000 cells) derived from 72 samples. (A) A density map is shown depicting the density of cells and are numbered according to phenotypic subset. (B) Density maps depicting European and African American ANA-, ANA+ and SLE patient PBMC t-SNE plots created using all 33 surface markers are plotted. All plots were derived from cumulative data from 12 individuals per group. (C) Cells numbers were calculated from cell subsets using frequencies and total cell counts. Cell numbers for T cells, CD4+ T cells, and CD8+ T cells are shown. *p<0.05, **p<0.01, ***p<0.001
and will never develop overt disease. Understanding differences in immune cell physiology between ANA+healthy individuals and individuals with clinical SLE remains a critical goal in the understanding of SLE pathogenesis across ethnicities.

Methods Blood specimens and information on disease activity were collected from European (EA) and African American (AA) individuals classified and matched in groups as ANA-healthy controls (n=24), ANA+healthy (n=24) or SLE patients (n=24). Single-cell analysis of cell surface markers was completed by mass cytometry on PBMCs and cellular heterogeneity was visualized using tSNE (figure 1A–B) and manual gating. Further, phospho-specific flow cytometry was used to measure basal levels of pERK, pPLCg2 and p38 and expression following CD3/CD28 (TCR) and anti-IgG and IgM (BCR) stimulation. Whole genome RNA-sequencing was performed on flow cytometry sorted T cells, B cells and monocytes from 35 matched ANA-, ANA+ and SLE patients followed by weighted correlation network analysis (WGCNA) and pathway enrichment analyses.

Results Both European and African American SLE patients were distinguished from healthy individuals by T cell proliferation (p=0.0002) (figure 1C), plasmacytoid dendritic cell activation (p=0.021) and elevated stem cell factor (p=0.0003). EA ANA+healthy individuals exhibited greater immune regulation with reduced T cell numbers (p=0.002) (figure 1C), decreased activation of dendritic cells (p=0.039) and transitional B cells (0.033), and elevated expression of the inhibitory receptor CD85j (p=0.042) on specific immune cell subsets compared to ANA- healthy subjects. Further, a module associated with hematopoesis, T cell activation and intrinsic apoptosis signaling pathways is expressed at a higher level in T cells of EA ANA+individuals. In contrast, AA ANA+healthy individuals had elevated plasma levels of IL-6 (p=0.018) and reduced inhibitory receptor expression (p=0.0089) compared to ANA- healthy controls. Gene expression modules associated with viral responses and type I IFN pathway activation were identified in AA SLE patient B cells, while similar expression modules were only found in the monocytes of European American SLE patients.

Conclusions These results highlight the importance of stem cell factor and T cell expansion in SLE pathogenesis, and suggest that mechanisms of SLE pathogenesis differ by ethnicity. ANA+European Americans may have more effective regulatory mechanisms in place to prevent transition to classified autoimmune disease.

Funding Source(s): This work was supported by the NIH under award numbers U54GM104938, U01AI101934, U19AI082714, and P30AR053483.