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ORIGINAL PAPER

# CMPK2 Promotes CD4$^+$ T Cell Activation and Apoptosis through Modulation of Mitochondrial Dysfunction in Systemic Lupus Erythematosus

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## Abstract

Systemic lupus erythematosus (SLE) is a classic autoimmune disease characterized by abnormal autoantibodies, immune complex deposition, and tissue inflammation. Despite extensive research, the exact etiology and progression of SLE remain elusive. Cytidine/uridine monophosphate kinase 2 (CMPK2), a mitochondrial nucleoside monophosphate kinase, has garnered attention for its potential involvement in the development of various diseases, including SLE, where it has been observed to be dysregulated in affected individuals. However, the specific involvement of CMPK2 in the pathogenesis of SLE remains unclear. This study aims to clarify the expression level of CMPK2 in SLE CD4$^+$ T cells and explore its impact on CD4$^+$ T cells. The expression levels of the CMPK2 gene and the corresponding CMPK2 protein in CD4$^+$ T cells of SLE patients were quantified using RT-qPCR and Western blot, respectively. Immunofluorescence and RT-qPCR were used to assess the mitochondrial function of SLE CD4$^+$ T cells. Flow cytometry was used to assess CD4$^+$ T cell activation and apoptosis levels. The impact of CMPK2 on CD4$^+$ T cells was investigated by gene transfection experiment. We found that CMPK2 was significantly upregulated in SLE CD4$^+$ T cells at both gene and protein levels. These cells demonstrated aberrant mitochondrial function, as evidenced by elevated mitochondrial reactive oxygen species (mtROS) levels, mitochondrial membrane potential, and mitochondrial DNA (mtDNA) copy number. Flow cytometry revealed a notable increase in both apoptosis and activation levels of CD4$^+$ T cells in SLE patients. Gene transfection experiments showed that suppressing CMPK2 led to a significant improvement in these conditions. These findings suggest that CMPK2 may be involved in the pathogenesis of SLE by regulating mitochondrial dysfunction in CD4$^+$ T cells and thus affecting CD4$^+$ T cell activation and apoptosis. Our study may provide a new target for the treatment of SLE.

**Keywords** Systemic lupus erythematosus · CD4$^+$ T cell · CMPK2 · Mitochondrial dysfunction · Apoptosis · CD4$^+$ T cell activation

## Introduction

Systemic lupus erythematosus (SLE) is an incurable chronic autoimmune disease characterized by autoantibody abnormalities, immune complex deposition, and tissue inflammation [1]. The pathogenesis of SLE is complex and not fully understood. Although there have been remarkable

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advances in the survival rates of SLE patients, achieving the remission remains a challenge [2]. Therefore, it is essential to further our understanding and research into the pathogenesis of SLE.

Recent research has highlighted that abnormalities in immune cells play a significant role in the pathological mechanisms of SLE [3–5]. Among the vital components of the adaptive immune system, T cells occupy a prominent position, with the ability to differentiate into two distinct subtypes: $CD4^+$ T cells and $CD8^+$ T cells. $CD4^+$ T cells, colloquially known as helper T cells, encompass various subgroups, including Th1, Th2, Th17, Treg, and Tfh cells. The activation of $CD4^+$ T cells is integral to their functionality, and investigations have revealed that in SLE patients, $CD4^+$ T cells are excessively activated. The excessive activation of $CD4^+$ T cells not only stimulates B cells to produce autoantibodies but also leads to the destruction of macrophages, which in turn elevates the levels of autoantigens in circulation and impairs the clearance of immune complexes. Consequently, these autoantibodies and autoantigens form immune complexes that accumulate in tissues and organs, aggravating tissue and organ damage, and thus accelerating the onset and progression of SLE [6]. Notably, the activation and proliferation of $CD4^+$ T cells are tightly dependent on the fine regulation of cellular metabolism, where resting cells maintain a low energy requirement, whereas activated cells demand a higher energy supply. Mitochondria, the energy-producing organelles within cells, play a pivotal role in regulating cellular activation and apoptosis [7–9]. Recently, numerous studies have highlighted abnormal mitochondrial function in multiple immune cell types in SLE patients, notably in B cells and neutrophils, which have been implicated in disease pathogenesis [10, 11]. Specifically, mitochondrial dysfunction in B cells has been linked to plasma cell differentiation and disease activity. In neutrophils, the modulation of aberrant mitochondrial function has been shown to decrease mitochondrial reactive oxygen species (mtROS) production and NET formation in mouse models. Similarly, in $CD4^+$ T cells, mitochondrial dysfunction is pronounced, evident by aberrant elevations in mitochondrial membrane potential and mtROS levels, which have been documented in SLE patients [12]. Studies have shown that mtROS potentiates T cell activation and apoptosis [13, 14], and it may contribute significantly to the progression of SLE. Therefore, targeting and alleviating mitochondrial dysfunction in $CD4^+$ T cells emerge as a promising therapeutic strategy for mitigating SLE progression and attenuating disease severity.

Cytidine/uridine monophosphate kinase 2 (CMPK2) is a 449-amino-acid protein that plays an important role in the immune system [15]. A study observed a substantial elevation in the activation level of IFN-α2b-treated $CD4^+$ T cells. Subsequent RNA sequencing and quantification revealed a significant upsurge in CMPK2 expression in IFN-α2b-treated $CD4^+$ T cells, in comparison to the untreated group [16], thereby implying a potential role for CMPK2 in facilitating $CD4^+$ T cell activation. Furthermore, a study investigating familial cerebral calcification identified mutations in the CMPK2 gene that were associated with mitochondrial dysfunction [17]. Additionally, a separate study revealed that inhibiting CMPK2 expression mitigated glucocorticoid-induced cellular senescence in the context of osteoporosis, and facilitated osteogenic differentiation through enhanced mitochondrial function [18]. These cumulative findings underscore the pivotal importance of CMPK2 in maintaining mitochondrial function.

In this study, we demonstrated that CMPK2 may promote $CD4^+$ T cell activation and apoptosis through its ability to regulate mitochondrial function, which in turn is involved in the pathogenesis of SLE. This study may provide a new therapeutic target for SLE.

## Patients and Methods

### Ethics Statement

The current study was approved by the Ethics Committee of The First Affiliated Hospital of USTC (No. 2023KY283), and adhered strictly to the principles outlined in the Declaration of Helsinki. Signed informed consent was obtained from all participants before sample collection.

### Study Subjects

A total of 84 SLE patients were recruited from the ward of The First Affiliated Hospital of USTC. Relevant clinical information regarding the patients was provided in the Supplementary Table 1. Additionally, 123 healthy controls (HCs) were recruited from the Physical Examination Center. All patients included in our study met the American College of Rheumatology's revised criteria for the classification of SLE.

### $CD4^+$ T Cell Isolation and Culture

Peripheral blood mononuclear cells (PBMCs) were isolated from fresh heparinized blood using Ficoll-Hypaque density gradient centrifugation (LTS1077; TBD Sciences, China). $CD4^+$ T cells were positively selected using anti-CD4 MACS beads (5220602189; Miltenyi Biotec, Germany) according to the manufacturer's instruction, and were subsequently cultured in RPMI 1640 complete medium (C3010-0500; Vivacell, China) supplemented with 10% fetal bovine serum (FBS) (2148384; BioGro, Israel), at 37 °C with 5% $CO_2$ for 48 h.

## siRNA Transfection

$CD4^+$ T cells from SLE patients were transfected with 50 nM of si-CMPK2, and si-NC (both from RIBOBIO, China) was used as a negative control, and then cultured in RPMI 1640 complete medium supplemented with 10% FBS. RNA, DNA, and proteins were extracted after 48 h of transfection in an incubator at 37 °C with 5% $CO_2$.

## Real-time quantitative PCR (RT-qPCR)

Total RNA was isolated from the cells using a HiPure Total RNA Mini Kit (R4111-02; Magen, China). Preparation of complementary DNA (cDNA) was done using a BioRT Master HiSensi cDNA First Strand Synthesis Kit (BSB40M1; BioFlux, China). Primers specific for human CMPK2 and GAPDH were obtained from Beijing Tsingke Biotech Co., Ltd,. 2×SYBR Green PCR Master Mix (A4004M; Mum, China) was used for RT-qPCR, which was run using a Roche LightCycler 480. Data were analyzed with the $2^{-\Delta\Delta CT}$ method to determine relative gene expression levels. The primer sequences used are listed in Table 1.

## Western Blot

PBMCs were isolated from fresh heparinized blood by Ficoll-Hypaque density gradient centrifugation. $CD4^+$ T cells were isolated by positive selection with anti-CD4 MACS beads according to the manufacturer's instruction. Proteins were resolved by sodium dodecyl sulfate-polyacrylamide gel electrophoresis and then transferred to PVDF membranes (IPVH00010; Merck, USA). Subsequently, the membranes were blocked using 5% skimmed milk for 2 h at room temperature. Then the membrane was incubated with primary antibodies diluted in Tris-buffered saline containing 0.1% Tween-20 (TBST) at 4 °C overnight with gentle agitation. Afterward, the membrane was washed with TBST three times, followed by incubation with an anti-rabbit peroxidase (HRP)- conjugated secondary antibody (1:10000, 511203; ZEN-BIOSCIENCE, China) for 1.5 h at room temperature. Images were visualized using an Omega Lum C imaging system (GE, USA), and the bands were quantified using Image J. The primary antibodies used were CMPK2 (1:4000, 25877-1-AP; Proteintech, China), GAPDH (1:5000, 380646; ZEN-BIOSCIENCE, China).

**Table 1** The primer sequence used for RT-qPCR

| Gene Name | Primer Sequences |
|---|---|
| CMPK2 | Forward: 5′-GTACCTCCTTTATTCCTGAAGCC-3′ |
| | Reverse: 3′-ATGGCAACAACCTGGAACTTT-5′ |
| hGAPDH | Forward: 5′-GTCTCCTCTGACTTCAACAGCG-3′ |
| | Reverse: 3′-ACCACCCTGTTGCTGTAGCCAA-5′ |

## Immunofluorescence Detection of mtROS

Mitochondrial ROS levels were assessed utilizing MitoSOX Red Mitochondrial Superoxide Indicator (40778ES50; Yeasen Biotech, China). Cells were cultured in 24-well plates containing poly-L-lysine coated slides. After pretreatment, the cell culture supernatant was gently aspirated, and the cells were rinsed three times with PBS. Subsequently, 250 µL of MitoSOX Red Mitochondrial Superoxide Indicator Probe Working Solution, diluted to a final concentration of 5 µM, was added and incubated for 10 min at 37 °C in the dark. Afterward, the cells were rinsed three times with pre-warmed HBSS buffer (BL561A; Biosharp, China), and DAPI (P0131; Beyotime Biotechnology, China) was added to observe the cells under a fluorescence microscope. The excitation/emission wavelengths used were 510/580 nm.

## Mitochondrial Membrane Potential Detection

Cellular mitochondrial membrane potential was assessed according to the protocol of Mitochondrial Membrane Potential Assay Kit (C2003; Beyotime Biotech, China). The staining solution was diluted at a 1:200 ratio of JC-1 to culture medium. For cells in 24-well plates, 250 µL of Staining Solution was added and the cells were incubated for 20 min. Subsequently, the cells were washed three times with JC-1 buffer solution. Then, the cells were imaged with a fluorescence microscope. The excitation/emission wavelength for JC-1 monomers is 488/525 nm. The excitation/emission wavelength for JC-1 aggregates is 560/595 nm.

## Detection of mtDNA Copy Number

Whole genomic DNA was extracted using QlAamp DNA Mini Kit (169020441; Qiagen, Germantown, MD). The purity and concentration of the DNA were assessed using the DeNovix DS-11 (DeNovix, USA). If the ratio of A260/280 fell between 1.8 and 2.0, the extracted DNA met the detectable standard. Then relative copy number of mitochondrial DNA (mtDNA) was quantified by RT-qPCR using nuclear DNA (nDNA) content as a standard. Human Mitochondrial DNA (mtDNA) Monitoring Primer Set (7246; Takara, China), TB Green® Premix Ex Taq™ II (RR820; Takara, China).

## Flow Cytometry

### $CD4^+$ T Cell Activation Level Detection

The cells were collected, washed twice with PBS, and resuspended in 100 µL of PBS. Subsequently, antibodies

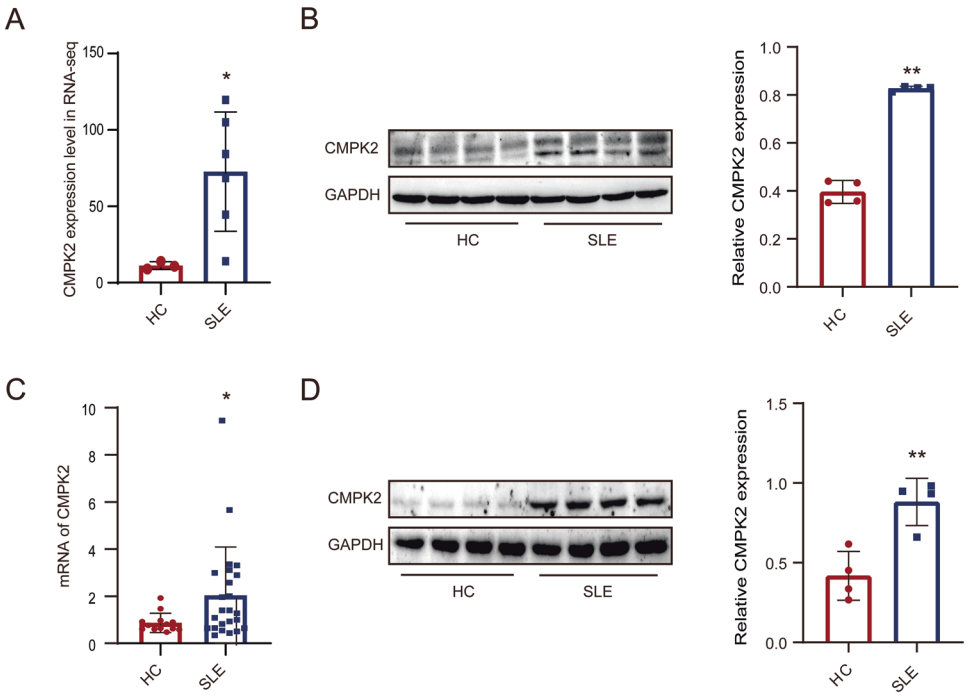


**Fig. 1** CMPK2 was highly expressed in SLE CD4$^+$ T cells. **A** The expression level of CMPK2 in PBMC was analyzed based on sequencing data (HC n = 3; SLE n = 6). **B** Western blot assessed CMPK2 protein expression in PBMCs from SLE patients and HCs (HC n = 8; SLE n = 4). **C** RT-qPCR evaluated CMPK2 gene expression in CD4$^+$ T cells from SLE patients and HCs (HC n = 13; SLE n = 24). **D** Western blot determined CMPK2 protein expression in CD4$^+$ T cells from SLE patients and HCs (HC n = 28; SLE n = 14). Data were shown as mean ± standard deviation. $^*P < 0.05$; $^{**}P < 0.01$

against CD45 (564047; BD Biosciences, USA), CD3 (563798; BD Biosciences, USA), CD4 (300545; Biolegend, USA), CD25 (E-AB-F1194E; Elabscience Biotechnology Co.,Ltd, China), and HLA-DR (E-AB-F1111C; Elabscience Biotechnology Co.,Ltd, China) were added sequentially. The cells were then incubated for 30 min at room temperature in the dark, followed by two washes with PBS. Finally, resuspended in 500 µL of PBS for analysis. A FACS-can flow cytometry (Beckman Coulter, USA) was adopted to analyze cell fluorescence.

### Apoptosis Rate Detection

Apoptosis rate was determined according to the kit instructions (C1062; Beyotime Biotech, China). Specifically, cells were collected and washed twice with cold PBS, followed by resuspension in 195 µL of Annexin-V binding buffer. Subsequently, 5 µL of Annexin V-FITC and then 10 µL of propidium iodide staining solution were added. The cells were then incubated at room temperature for 15 min in the dark. Cells were then analyzed and quantified using flow cytometry (BD Biosciences, USA). The flow data were analyzed using Flowjo software version 10.6.2 (FlowJo LLC, Ashland, OR).

### Enzyme-Linked Immunosorbent Assay

The cell supernatant was collected by centrifugation at 1000 g for 20 min. Human IL-2, TNF-α and IFN-γ ELISA kits (MM-0055H1; MM-0122H1; MM-0033H1; Jiangsu Meimian Industrial Co., Ltd, China) were used to detect cytokine levels as the manufacturer's instructions. Assay range of the ELSIA kits used was 10-320 pg/mL (sensitivity 1.0 pg/mL) for IL-2, 1.0−80 pg/mL (sensitivity 1.0 pg/mL) for TNF-α, and 5.0−800 pg/mL (sensitivity 1.0 pg/mL) for IFN-γ. The intraassay and interassay coefficients of variation were both <15%.

### Statistical analysis

All analyses were conducted using GraphPad Prism 8.0.2. The results were presented as the mean ± SD. The difference between the two groups was assessed utilizing the Student's *t* test. Statistical significance was defined as $P < 0.05$.

## Results

### CMPK2 was Significantly Upregulated in SLE CD4$^+$ T Cells

Upon analyzing the sequencing data of the subject group, we found that CMPK2 was highly expressed in PBMCs of SLE patients (Fig. 1A). The Western blot results were consistent with the sequencing findings (Fig. 1B). Given CD4$^+$ T cells are crucial in the pathogenesis of SLE and it has been demonstrated that CMPK2 gene levels were significantly elevated in SLE CD4$^+$ T cells [19], we further examined the expression of CMPK2 in SLE CD4$^+$ T cells. Our experimental results indicate that both the gene and protein levels of CMPK2 in SLE CD4$^+$ T cells were significantly higher than those in the HC group (Fig. 1C, D).

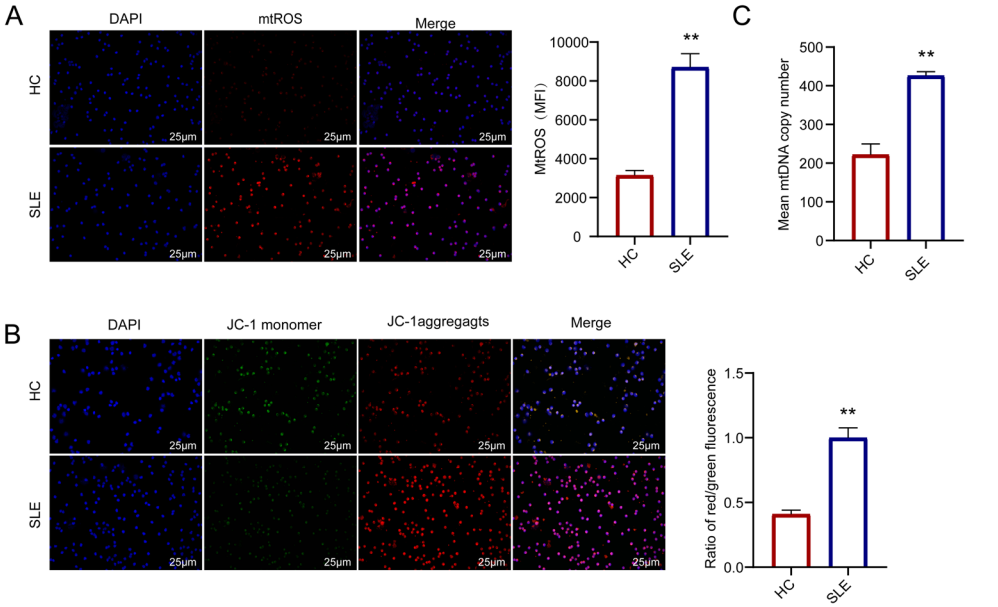


**Fig. 2** Mitochondrial dysfunction was observed in SLE $CD4^+$ T cells. **A** Immunofluorescence assessed mtROS levels in $CD4^+$ T cells from SLE patients and HCs (HC n = 14; SLE n = 6). **B** Mitochondrial membrane potential in $CD4^+$ T cells from both SLE patients and HCs was determined using immunofluorescence (HC n = 14; SLE n = 6). **C** The mtDNA copy number in $CD4^+$ T cells from SLE patients and HCs was assessed via RT-qPCR (HC n = 21; SLE n = 15). Data were shown as mean ± standard deviation. $^{**}P < 0.01$

## Mitochondrial Dysfunction was Observed in SLE $CD4^+$ T Cells

Mitochondrial dysfunction has a profound impact on cellular functions such as activation, apoptosis, and inflammatory responses in SLE. The presence of mitochondrial damage in SLE patients is evidenced by elevated mitochondrial membrane potential, increased mtROS levels [20]. Therefore, we examined indicators of mitochondrial damage in SLE $CD4^+$ T cells. The fluorescence intensity of mtROS in $CD4^+$ T cells of SLE patients was significantly elevated compared to the HC group (Fig. 2A). Furthermore, the mitochondrial membrane potential in SLE patients was notably higher than in the HC group (Fig. 2B). Additionally, the mtDNA copy number in $CD4^+$ T cells was significantly elevated in SLE patients (Fig. 2C).

## Activation and Apoptosis of $CD4^+$ T Cells were Increased in SLE Patients

SLE is characterized by overactivated $CD4^+$ T lymphocytes, a key pathological factor in the disease [21]. Our study observed markedly elevated levels of CD25 and HLA-DR expression in SLE $CD4^+$ T cells compared to HC group (Fig. 3A). Refer to Supplementary Fig. 1 for a detailed illustration. Furthermore, the concentrations of IL-2, TNF-α, and IFN-γ cytokines were notably higher in SLE patients than in HC group (Fig. 3B), indicating increased activation of $CD4^+$ T cells in SLE. Literature review indicates that activated $CD4^+$ T cells can induce apoptosis [22]. Our results consistently showed a significantly higher rate of $CD4^+$ T cell apoptosis in SLE patients compared to HC group (Fig. 3C).

## Inhibition of CMPK2 Ameliorates Mitochondrial Dysfunction in $CD4^+$ T cells

The above experiments have confirmed that CMPK2 was highly expressed in SLE $CD4^+$T cells, indicating its potential influence on $CD4^+$ T cell function or phenotype. Research has shown that CMPK2 has an impact on mitochondrial dysfunction [18]. To investigate its role in $CD4^+$ T cells, we utilized small interfering RNA to silence the CMPK2 gene in SLE $CD4^+$ T cells. The figure shows successful silencing of CMPK2 (Fig. 4A). Subsequently, we measured the mitochondrial function indicators in $CD4^+$ T cells. Immunofluorescence experiments displayed a significant decrease in mtROS fluorescence intensity in $CD4^+$ T cells of the si-CMPK2 group compared to the control group (Fig. 4B). Moreover, the mitochondrial membrane potential and mtDNA copy number in $CD4^+$ T cells were notably lower in the si-CMPK2 group (Fig. 4C, D).

## Inhibiting CMPK2 Suppresses $CD4^+$ T Cell Activation and Apoptosis

The aforementioned experiments have confirmed that inhibiting CMPK2 can alleviate mitochondrial dysfunction in SLE $CD4^+$ T cells. Studies have shown that mtROS is also essential for the activation of $CD4^+$ T cells [14]. Moreover, mitochondrial dysfunction may contribute to increased apoptosis in $CD4^+$ T cells. Subsequently, we examined the levels of activation and apoptosis in $CD4^+$ T cells following

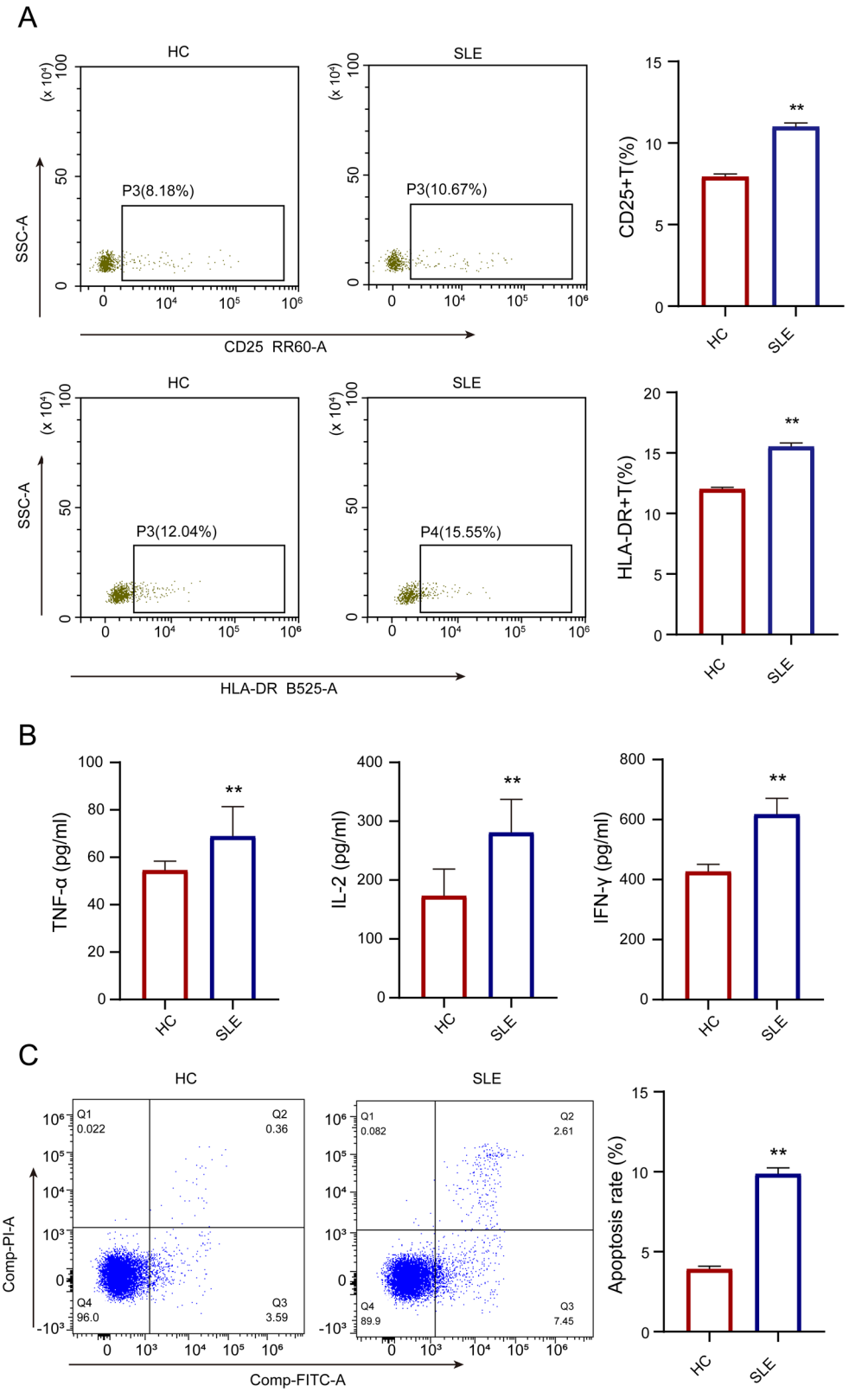


**Fig. 3** Activation and apoptosis of $CD4^+$ T cells were increased in SLE patients. **A** Flow cytometry was used to detect CD25 and HLA-DR expression in $CD4^+$ T cells from both SLE patients and HCs (HC n = 8; SLE n = 6). **B** ELISA was used to measure the levels of IL-2, TNF-α, and IFN-γ in the culture supernatants of $CD4^+$ T cells from both SLE patients and HCs (HC n = 30; SLE n = 24). **C** The level of $CD4^+$ T cell apoptosis in both SLE patients and HCs was determined by flow cytometry (HC n = 14; SLE n = 6). Data were shown as mean ± standard deviation; biological duplicates were performed. $^*P < 0.05$; $^{**}P < 0.01$

CMPK2 silencing. The results indicate that silencing CMPK2 reduces the expression levels of CD25 and HLA-DR, surface markers of $CD4^+$ T cells (Fig. 5A). Additionally, the levels of the cytokines IL-2, TNF-α, and IFN-γ were decreased (Fig. 5B). The apoptosis rate was also notably inhibited (Fig. 5C).

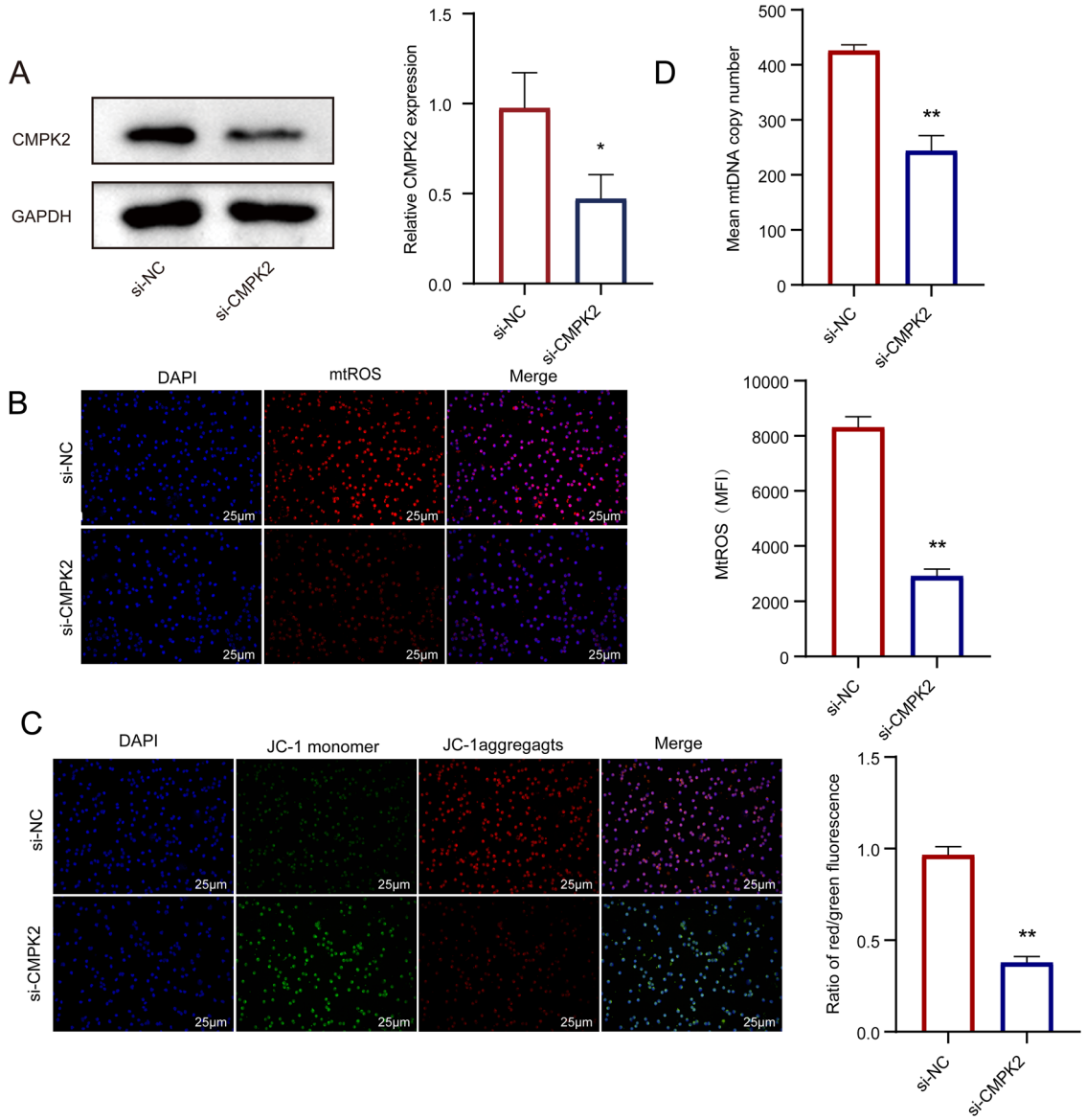


**Fig. 4** Inhibition of CMPK2 ameliorates mitochondrial dysfunction in $CD4^+$ T cells. **A** CMPK2 protein levels in SLE $CD4^+$ T cells transfected with si-CMPK2 or si-NC (n = 5). **B** MtROS levels in SLE $CD4^+$ T cells transfected with si-CMPK2 or si-NC (n = 6). **C** Mitochondrial membrane potential of $CD4^+$ T cells from SLE patients transfected with si-CMPK2 or si-NC (n = 6). **D** MtDNA copy number of SLE $CD4^+$ T cells transfected with si-CMPK2 or si-NC (n = 15). Data were shown as mean ± standard deviation; biological duplicates were performed. $^*P < 0.05$; $^{**}P < 0.01$

## Discussion

SLE is a complex autoimmune disease characterized by the breakdown of systemic immune tolerance and intense inflammatory reactions [23]. T cells, crucial in the inflammatory response, play a role in enhancing innate immune responses by producing a variety of soluble or membrane-bound mediators, exacerbating chronic inflammation and tissue damage in SLE patients [24]. $CD4^+$ T cells, a key subset among T lymphocytes, not only assist in triggering cellular and humoral immunity but also support B lymphocytes in antibody production and macrophage activation [25]. Upon activation, $CD4^+$ T cells release a myriad of cytokines, regulating the activity of T lymphocytes, B lymphocytes, monocytes/macrophages, and other cells, leading to the generation of numerous autoantibodies in the body and causing damage across multiple systems [26]. These findings indicate that regulating $CD4^+$ T cells could help mitigate the progression of SLE.

Mitochondria serve as vital intracellular energy producers, generating ATP to fuel cellular functions. Additionally, they play a role in regulating apoptosis, immune responses, and other biological processes [27]. Recent studies indicate that mitochondrial damage and metabolic

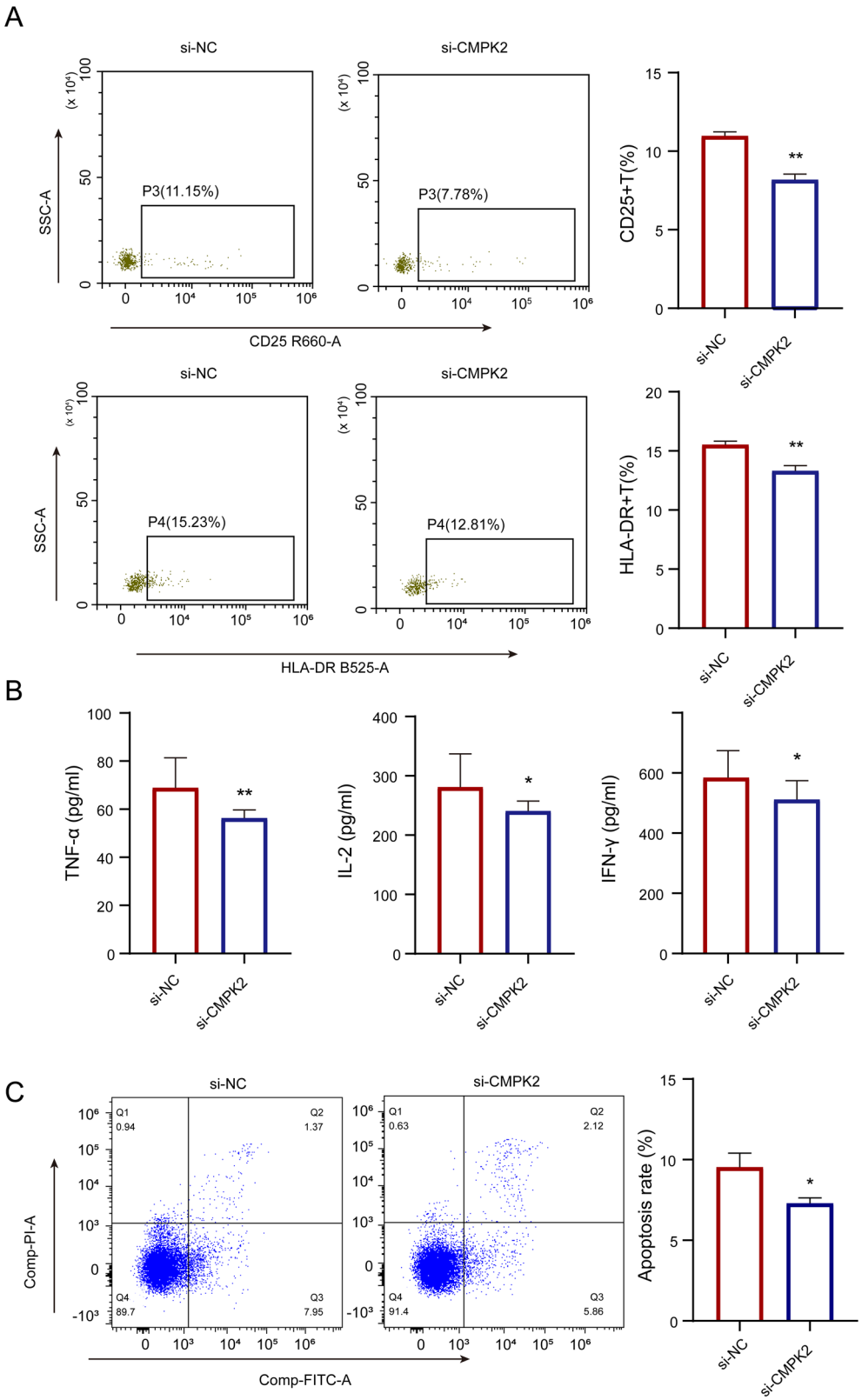


**Fig. 5** Inhibiting CMPK2 suppresses $CD4^+$ T cell activation and apoptosis. **A** CD25 and HLA-DR levels in SLE $CD4^+$ T cells transfected with si-CMPK2 or si-NC (n = 6). **B** IFN-γ, IL-2, and TNF-α levels in culture supernatants of SLE $CD4^+$ T cells transfected with si-CMPK2 or si-NC (n = 24). **C** Apoptosis levels in SLE $CD4^+$ T cells transfected with si-CMPK2 or si-NC (n = 8). Data were shown as mean ± standard deviation; biological duplicates were performed. $^*P < 0.05$; $^{**}P < 0.01$

dysfunction in T lymphocytes may significantly contribute to autoimmune diseases such as SLE [13, 28–34]. In a study involving 25 SLE patients and 25 healthy individuals, peripheral blood mononuclear cells were analyzed, revealing elevated levels of mitochondrial membrane potential in SLE patients, indicating mitochondrial damage [12]. Research by Lee et al. [35] showed that increased reactive oxygen species production due to mitochondrial damage could disrupt cytokine levels, potentially driving SLE progression. Our study confirmed mitochondrial dysfunction in $CD4^+$ T cells from SLE patients, as evidenced by heightened mitochondrial membrane potential, increased mtDNA copy number, and elevated mtROS levels.

CMPK2, a mitochondrial nucleoside monophosphate kinase, has been reported for its potential antiviral activity and crucial role in intrinsic immunity [16, 36, 37]. In this study, we observed a significant increase in CMPK2 expression in $CD4^+$ T cells of SLE patients. Inhibiting CMPK2 expression notably alleviated mitochondrial dysfunction in these cells. In line with this, CMPK2 has also been shown to be associated with mitochondrial function in a study of primary sjogren's syndrome [38]. Given that mitochondrial mtROS play a crucial role in T cell activation and are linked to apoptosis [13, 14], we further investigated the impact of inhibiting CMPK2 on $CD4^+$ T cell activation and apoptosis. Our findings revealed a notable suppression of activation and apoptosis in $CD4^+$ T cells following CMPK2 inhibition. These findings indicated that CMPK2 inhibition can effectively decrease the levels of $CD4^+$ T cell activation and apoptosis, potentially due to the amelioration of mitochondrial dysfunction by CMPK2.

## Conclusions

In summary, our findings underscore the significant upregulation of CMPK2 in $CD4^+$ T cells of SLE patients, which may have implications for SLE pathogenesis. Furthermore, we propose that CMPK2 may contribute to SLE pathogenesis by modulating mitochondrial dysfunction in $CD4^+$ T cells, thereby influencing their activation and apoptosis. To the best of our knowledge, this study is the first to investigate the involvement of CMPK2 in the pathogenesis of SLE. These findings not only enhance our understanding of SLE pathogenesis but also open avenues for the development of novel therapeutic strategies targeting CMPK2 and its downstream effects on mitochondrial function and T cell biology. Further studies are warranted to elucidate the precise mechanisms by which CMPK2 modulates mitochondrial function and T cell behavior in SLE, and to explore the potential of CMPK2 inhibition as a therapeutic strategy for this autoimmune disorder.

**Supplementary information** The online version contains supplementary material available at https://doi.org/10.1007/s12013-024-01443-1.

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**Author Contributions** Min Zhang and Xiao-Yi Jia conceptualized and designed this study and critically revised the article for important intel actual content; Ya-Nan Tan, analyzed the data and wrote the article; Yan-Ma, Li Jin and Nan-Xiang were responsible for clinical sample collection, Ya-Nan Tan, Ge-Ge Jiang, Xiang-wen Meng and Zhi-Yuan Lu were responsible for blood sample processing, WB, Immunofluorescence and RT-qPCR; Xiao-Ge Sun, Qian Wang, and Xue Wang performed the ELISA kits. All the authors approved the final version of the article to be submitted. The authors agreed to be accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved.

## Compliance with Ethical Standards

**Conflict of Interest** The authors declare no competing interests.

**Ethics Approval and Consent to Participate** The current study was approved by the Ethics Committee of The First Affiliated Hospital of USTC (No. 2023KY283), and all experimental protocols conformed to the Declaration of Helsinki. Signed informed consent was obtained from all participants before sample collection.

## Abbreviations

| | |
|---|---|
| SLE | systemic lupus erythematosus |
| CMPK2 | cytidine/uridine monophosphate kinase 2 |
| mtROS | mitochondrial reactive oxygen species |
| mtDNA | mitochondrial DNA |
| PBMC | peripheral blood mononuclear cell |
| HC | healthy control |
| nDNA | nuclear DNA |

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