

## Biomarkers for systemic lupus erythematosus: an update

Systemic lupus erythematosus (SLE) is an inflammatory autoimmune disease characterized by diverse autoantibodies and varied clinical features. There is much interest in biomarkers, but diagnostic and disease activity markers in actual clinical practice remain complement, autoantibodies including anti-dsDNA antibody, leukocyte count and urinalysis. This article reviews recently studied epigenetic, protein and metabolomic biomarkers. Although there are many studies about the SLE pathogenesis and biomarkers, only some biomarkers have been validated. This is most commonly because there are few prospective longitudinal and interventional studies that validate the biomarker utility. Recently elucidated candidate biomarkers need well-designed prospective studies and standardized assays for commercialization. Moreover, continuous research for SLE pathogenesis can promote biomarker development using new technologies, such as microarrays, proteomics and metabolomics techniques.

**Keywords:** autoantibody • biomarker • cytokine • epigenetic • metabolomics • systemic lupus erythematosus

Systemic lupus erythematosus (SLE) is a chronic inflammatory disease characterized by diverse autoantibodies and varied clinical manifestations. Although the pathogenesis of SLE is still unclear, multiple genetic, environmental and hormonal factors are thought to contribute and interact in the development of the disease [1]. Immune system dysregulation such as incomplete clearance of apoptotic materials, hyperactivation of immune cells, overproduction of antibodies and abundant immune complex deposits are prominent in SLE [2].

SLE patients can have diverse clinical manifestations, ranging from mild constitutional symptoms, such as myalgias and malaise, to life-threatening presentations, such as neuropsychiatric lupus and pulmonary hemorrhage; various laboratory abnormalities are seen, including leukopenia, thrombocytopenia, hypocomplementemia, elevated autoantibodies and proteinuria. SLE has complex laboratory findings, heterogeneous manifestations and an unpredictable course,

making the diagnosis and evaluation of disease activity difficult and remains a major challenge to physicians. The classification criteria published in 1982 and 2012 consist of several symptoms and laboratory findings, and are important tools for research studies [3,4]. However, these criteria provide no predictive information on the prognosis and course of the disease. Disease activity in SLE is also varied, ranging from mild to serious manifestations. Therefore, accurate assessment of disease activity is important for appropriate therapeutic decision-making and data presentation useful for new drug development. Several immunologic markers, including complement and antidouble stranded DNA (dsDNA) antibody, are used in laboratory monitoring of disease activity. However, these markers correlate better with certain clinical manifestations of the disease (especially nephritis) than the disease activity itself [5]. Disease activity can be assessed using several indices, such as SLE Disease

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Activity Index (SLEDAI), British Isles Lupus Assessment Group (BILAG) and Systemic Lupus Activity Measure (SLAM) [6]. These measurements reflect the diverse clinical manifestations and laboratory findings of SLE, and have performed effectively and reliably in studies with correlating with each other [7]. However, they are too complicated for use in routine clinical practice, and some are inadequate for assessing therapeutic efficacy in clinical trials.

A biomarker is an objective measurement, characterized as a genetic, biological, biochemical, molecular or imaging event, whose alterations correlate with disease pathogenesis or manifestations [8]. A valuable and reliable biomarker for a disease must be biologically and pathophysiologically relevant, and a measure of a product or cell that contributes to disease pathogenesis. It must be simple for use in clinical practice, and should change accurately and sensitively according to disease activity. Although many studies on pathogenesis and biomarkers in SLE have been published, no biomarker has been validated recently for actual clinical practice, primarily because there are few prospective longitudinal and interventional studies of their utility. Therefore, in this work, we found simple and critical serologic biomarkers, which are associated with susceptibility or pathogenesis in SLE during the past 10 years, and introduce candidate serologic biomarkers for diagnosis and evaluation of disease activity in SLE, divided into categories of epigenetic, protein (such as cytokines, autoantibodies and complement activation proteins) and metabolomic biomarkers.

### Epigenetic biomarkers

Epigenetic modification can influence gene expression through chromatin structural change that modulates the access to transcription factors and alters cellular function without modifying the genomic sequence [9]. The transcription factors must bind regions of DNA targets for modulation of gene expression. Therefore, an efficient mechanism to avoid the expression of a target gene is to disrupt the binding of transcription factor to DNA through DNA methylation [10]. In addition, the packing of DNA with histones in a structure known as nucleosome plays an important regulatory function in eukaryotic cells [9]. Histones can effect reversible modifications with alteration of chromatin structure that affects DNA accessibility to transcription factors [11]. MicroRNAs (miRNAs) are a class of small noncoding RNAs consisting of 21–23 base-pair structures that regulate post-transcriptional events, transcript degradation or translational suppression [12]. These epigenetic mechanisms are involved in various autoimmune diseases, including SLE, by regulating immunogenicity and autoantibody production [10].

### DNA methylation

Methylation of DNA is one of the commonly evaluated epigenetic modifications, and cytosine methylation in the regulatory regions of DNA results in the transcriptional inactivation of genes [10]. Therefore, hypomethylation is associated with the activation of transcription, and impaired DNA methylation has been found in T, B or NK cells of SLE patients [9,13,14]. A recent study performed a high-resolution analysis of the characteristics of plasma DNA abnormalities in patients with SLE using massively parallel genomic and methylomic sequencing [15]. They showed that plasma DNA aberrations in measured genomic representations, increased proportion of short DNA fragment and decreased methylation densities. Furthermore, the binding of anti-dsDNA antibody to plasma DNA is an important factor associated with these abnormalities including hypomethylation and DNA fragment size shortening in SLE patients.

### Histone modification

Post-translational modifications of histones, such as methylation and acetylation, result in remodeling of chromatin, which alters and modulates the accessibility of DNA to transcription factors [11]. Global hypoacetylation of histone H3 and H4 was observed in CD4<sup>+</sup> T cells of SLE patients, and the degree of H3 acetylation correlated negatively with increased disease activity [16]. Another study showed that histone H3 acetylation and dimethylated H3 lysine 4 levels in *TNFSF7* promoter were significantly higher in SLE CD4<sup>+</sup> T cells, and both factors correlated positively with disease activity in lupus patients [17]. These results suggested that aberrant histone modifications may contribute to the development of the disease. Studies reported that histone modifications have been observed in CD4<sup>+</sup> T cells, B cells or monocytes of lupus patients, but not in serum or plasma [16–18].

### miRNAs

Because miRNAs control immune cell differentiation and regulate innate and adaptive immune responses, dysregulated miRNA levels could play an important role in SLE pathogenesis. In addition, several studies already showed that miRNAs could represent biomarkers that help monitor disease activity. miRNA microarray techniques and miRNA sequencing with peripheral blood mononuclear cells, CD4<sup>+</sup> cells or renal biopsies revealed dysregulated miRNAs in SLE, including *miR-21*, *25*, *31*, *15a*, *126*, *146a*, *148a*, *155*, *182–96–183* cluster and *186* [19,20]. Most miRNAs originate from blood cells, including platelets and endothelial cells, and some data of aberrant miRNAs expression of circulating cells were shown in SLE [21].

Furthermore, recent data demonstrated the stability of cell-free circulating miRNAs [22–24]. In a study evaluating serum and urinary miRNAs, the serum levels of *miR-200a*, *miR-200b*, *miR-200c*, *miR-429*, *miR-205* and *miR-192* of lupus patients were lower than those of controls [24]. In addition, serum *miR-200a* inversely correlated with the SLEDAI, and serum *miR-200b* and *miR-192* correlated with platelet count. Another study reported upregulation of 19 miRNAs and downregulation of 32 miRNAs in a Chinese SLE population [22]. The study showed that *miR-126* was highly expressed only in the serum of SLE; however, four other miRNAs (*miR-21*, *miR-451*, *miR-223* and *miR-16*) were upregulated in SLE and rheumatoid arthritis (RA). A recent study showed increased expression of *miR-142-3p* and *miR-181a*, and decreased expression of *miR-106a*, *miR-17*, *miR-20a*, *miR-203* and *miR-92a* in SLE [23]. A 4-miRNA predictive model differentiated SLE from disease controls efficiently, except for those with vasculitis. The authors suggested that circulating miRNA patterns distinguish SLE from other immunoinflammatory diseases.

## Protein biomarkers

### Cytokines & cytokine receptors

Cytokines and their receptors play an important role in lupus pathogenesis, and their balance determines disease activity [25,26]. These cytokines are soluble factors for differentiation, maturation and activation of the various immune cells, and their imbalance would result in local inflammatory processes and tissue damage [27]. Serum levels of IL-6, IL-10, IL-17, type I interferon (IFN), soluble IL-2 receptor (sIL-2R) and soluble tumor necrosis factor receptor (sTNFR) suggest pivotal functions in maturation and activation of various inflammatory cells, and may be promising biomarkers of disease activity in SLE.

IL-6 is a multifunctional cytokine with various biological activities, such as regulation of immune responses, hematopoiesis and the acute phase response, and one of the first cytokines evaluated in lupus pathogenesis due to its close link with B lymphocytes [27,28]. Studies on serum IL-6 in SLE showed that some patients had elevated levels, and these levels correlated with disease activity and anti-dsDNA levels [29,30]. Our study showed that the serum IL-6 of SLE patients were higher than those of normal controls and were correlated with SLEDAI, erythrocyte sedimentation rate (ESR) and C-reactive protein (CRP) [30]. However, most SLE patients do not have elevated levels, but elevated serum IL-6 levels with increased CRP have been observed in SLE patients with infection or serositis. We confirmed that cytokine IL-6 and CRP

responses were related in SLE; the response in infection was higher than that of serositis and tissue injury, and nearly no response in other noninfectious lupus disease activity [31].

IL-10, which is produced by activated monocytes and T cells, is also a potent stimulator of B cell proliferation and immunoglobulin production, and its role in lupus pathogenesis is hypothesized [26]. IL-10 levels were elevated in human SLE, and these levels have shown a significant correlation with disease activities [30,32]. Our prospective study revealed serum IL-10 levels were higher in patients with SLE than normal controls [30]. In addition, serum IL-10 was correlated with SLEDAI, anti-dsDNA, C3, C4 and lymphopenia. These results suggest that IL-10 may be a possible biomarker of disease activity in SLE.

IL-17 is primarily released by activated CD4<sup>+</sup> T lymphocytes, named Th17 cells. IL-17 has a potent capacity to recruit monocytes and neutrophils, assist T-cell infiltration and upregulate adhesion molecule expression [33]. Furthermore, IL-17 assumes a critical role for the proliferation, survival and antibody secretion by B cells in human SLE [34]. Increased IL-17 levels were observed in lupus patients and correlated with some disease manifestations with conflicting results [35].

IFN is a cytokine that possesses the capacity to halt viral replication, and comprises both type I and type II IFN. Type I IFN is composed of IFN- $\alpha$ , IFN- $\beta$ , IFN- $\gamma$ , IFN- $\kappa$  and IFN- $\delta$ , and type II subgroup is represented by IFN- $\gamma$ . Type I IFNs have various immunomodulatory effects, such as the promotion of survival of activated T and B lymphocytes, induction of Fas mediated apoptosis and maturation of dendritic cells [26]. Mechanisms for IFN contribution to lupus pathogenesis have been proposed, and important clues that IFN may contribute to SLE came from several human clinical trials using IFN- $\alpha$  as a therapeutic agent for carcinoid tumor or viral hepatitis [25]. Elevated serum IFN- $\alpha$  levels were reported, and were correlated with disease activity and severity [36]. However, direct measurement of type I IFN levels in the serum of lupus patients is difficult due to the antibodies used to detect them or the effects of proteins associated with circulating IFN- $\alpha$ , such as soluble IFN receptors or anti-IFN antibodies [37]. Also, gene expression profiling revealed that lupus patients expressed IFN-inducible genes in peripheral blood cells, and their expression correlated with disease activities [38]. In addition to the IFN gene signature, elevated IFN-regulated chemokines are reported, and are associated with disease activity and clinical test results [32,39]. A longitudinal study was conducted in lupus patients for a year to validate the potential utility of IFN-regulated chemokines, such as CXCL10, CCL2 and CCL19, as biomarkers of dis-

ease activity [40]. These serum chemokine levels correlated with SLEDAI, and also predicted future disease flares. A recent study showed IFN- $\alpha$ , CXCL10 (IP-10) and sialic acid binding immunoglobulin-like lectin-1 have been significantly correlated with SLEDAI and BILAG [41].

sIL-2R is a complex formed by  $\alpha$ -,  $\beta$ - and  $\gamma$ - chains, and the  $\alpha$ -chain is highly sensitive to upregulation by activation. The presence of sIL-2R in the serum can be considered as a marker of T-lymphocyte activation *in vivo* because activated T cells release the soluble  $\alpha$ -chain of IL-2R [26]. Serum sIL-2R levels are elevated compared with normal controls and correlated linearly with disease activity [42].

TNF- $\alpha$  is a proinflammatory cytokine, and its biological activity is induced upon interaction with two receptors, TNF receptor type I (TNFR1) and TNFR2 [26]. TNF- $\alpha$  induces shedding of membrane TNFR in T lymphocytes and macrophages, and sTNFR serves as a physiologic regulator of TNF- $\alpha$  activity. Several studies revealed that the titers of circulating sTNFR1 and sTNFR2 were significantly increased in patients with SLE and correlated with disease activity [43,44]. Furthermore, a recent study showed that sTNFR1 and sTNFR2 were significantly higher in preflare lupus patients, and suggested that these mediators may be useful to identify patients at risk of disease flare [45].

Although some cytokines inconsistently showed positive results with limited disease manifestations, such as nephritis and neuropsychiatric lupus, these cytokines and soluble cytokine receptors may be promising candidate serologic biomarkers for lupus activity on a pathophysiologic basis.

### Autoantibodies

Antinuclear antibody (ANA), anti-dsDNA antibody and anti-Sm antibody are used in the classification criteria of SLE; however, they are not consistent and lack any definitive role in the diagnosis. ANA is insensitive and is not useful in evaluating disease activity; anti-dsDNA antibody is highly specific, but its sensitivity is only 57.3% [28,46]. Recently, many researchers have evaluated the use of other autoantibodies in diagnosis and monitoring in patients with SLE. Among those autoantibodies, antichromatin/antinucleosome antibodies and anti-C1q antibodies have been studied and shown to be promising markers for diagnosis or as new measures of renal involvement [47,48].

Chromatin is the complex molecule including DNA and histone found in the nucleus of eukaryotic cells, and nucleosomes are the fundamental unit of chromatin and a normal product of cell apoptosis. In SLE, massive amounts of circulating nucleosomes

are recognized by the immune system due to defective removal of apoptotic materials [49]. One study showed that the nucleosome is major autoantigen recognized by T and B lymphocytes in SLE [50]. Several investigations have revealed antichromatin antibodies to be a highly accurate diagnostic marker for SLE, with sensitivity between 45 and 100%, and specificity between 90 and 99% [47,51]. Antichromatin antibodies are positive in some cases with negative anti-dsDNA antibodies [52]. Furthermore, we observed that antichromatin antibody levels were elevated in incomplete lupus patients (satisfying only two or three criteria for SLE) compared with normal controls [53]. Some studies have shown that antichromatin antibodies may serve as a reliable biomarker in the diagnosis of lupus nephritis [54,55]. One prospective study showed that antichromatin antibody correlated with anti-dsDNA antibody, serum albumin and urine protein-creatinine ratio, and its level was significantly reduced in patients with clinical remission [54]. Furthermore, one study evaluated autoantibody profiles in SLE patients with nephritis and lupus nephritis patients received kidney transplantation, and showed that nucleosome autoantibodies were more frequent and much higher in lupus nephritis patients requiring transplantation than SLE patients with nephritis [56]. However, our study did not show any correlation between antichromatin levels and lupus nephritis [53]. A recent meta-analysis of antichromatin antibody as a diagnostic marker for lupus revealed no correlation with kidney involvement [57].

C1q has a critical role in the clearance of immune complexes and apoptotic bodies. C1q deficiency is one of the strongest risk factor for SLE. Recently, anti-C1q antibodies have been associated with hypocomplementemic urticarial vasculitis and SLE [58]. The prevalence of anti-C1q antibody in patients with SLE ranges from 30 to 60%, and its presence is correlated with disease activity and severity of lupus nephritis [48,59]. An increase in anti-C1q antibodies was suggested to predict renal flares, and the absence of anti-C1q antibodies was reported to exclude a diagnosis of lupus nephritis [59,60]. A recent meta-analysis of 25 studies to evaluate diagnostic accuracy and correlation of disease activity of anti-C1q antibodies in lupus nephritis confirmed that it has relatively good sensitivity and specificity in the diagnosis of lupus nephritis and is a valuable adjunct for assessing renal activity [61]. These results suggest that anti-C1q antibody may provide a useful means to monitor renal involvement or predict renal flares.

### B-lymphocyte stimulator

B-lymphocyte stimulator (BLyS) is a member of the TNF ligand superfamily, and is also known as B-cell activating factor (BAFF). It is expressed as a trans-

membrane protein on monocytes, macrophages and dendritic cells [62]. After release from the cell surface, a soluble form of BLYS is activated and stimulates B-cell proliferation and immunoglobulin production. One longitudinal observational study showed that lupus patients exhibited variable serum BLYS levels, with half of the patients having persistently or intermittently elevated levels over the follow-up period [63]. However, changes of serum BLYS levels did not correlate with changes of disease activity or specific organ involvement. A subsequent longitudinal prospective study evaluated 254 SLE patients over a 2-year period and found that plasma BLYS levels were correlated with anti-dsDNA antibody levels and SELENA-SLEDAI [64]. In addition, multivariate analyses revealed that an increase in the SELENA-SLEDAI score at a follow-up visit was significantly correlated with high BLYS levels at previous visits. These results suggest a delayed causative relationship between BLYS levels and lupus disease activity. Recently, two studies showed that BLYS levels were correlated with disease activity in white and Chinese SLE patients, but not in African American patients [65,66]. They also demonstrated a significant correlation between BLYS levels and IFN- $\alpha$  levels.

### Complement activation products

Serum complement levels were included as immunologic criteria in recently published classification criteria for SLE, and have been used to monitor disease activity in lupus patients [3]. Consumption of complements is represented by decreased levels of C3 and C4, lowered total hemolytic activity or the presence of complement activation products [67]. However, because low complement levels are less than ideal for assessing disease activity, many studies have investigated the potential of a number of complement activation products, such as C3a, C5a, C3d and C4d, as disease activity mark-

ers [68–70]. One study showed that assessment of serum complement activation products (C4d, Bb, C5b-9) was better than conventional C3 and C4 in distinguishing patients with varying degrees of lupus disease activity [69]. They concluded that C4d and Bb were sensitive biomarkers of moderate-to-severe lupus disease activity and might be most helpful in situations where conventional C3 and C4 levels remained normal despite evidence of clinical disease activity. In addition, the terminal complex C5b-9 was reported to have a strong correlation with lupus disease activity [68]. However, complement activation occurs in other comorbidities such as infection, and complement activation products have low specificity, providing only a rough estimate of disease activity in SLE [70]. Recently, many studies demonstrated that cell-bound complement activation products, such as E-C4d and B-C4d, are potential markers for lupus diagnosis or evaluation of disease activity [71,72]. These cell-bound complement activation products are determined using flow cytometry.

### TAM receptor & ligands

Impaired clearance of apoptotic cells was detected in the germinal centers of lymph nodes of lupus patients, and has been theorized to have a pivotal role in the pathogenesis of SLE [2]. The Tyro3, Axl and Mer (TAM) kinases are major regulators of innate immunity and phagocytosis of apoptotic cells. The 2 ligands, Gas6 and protein S, interact with TAM, and regulate cell survival and proliferation, cell adhesion and migration and inflammatory cytokine release [73].

A study measuring plasma soluble Mer (sMer) and Tyro3 (sTyro3) levels showed that SLE patients had increased levels of sMer and sTyro3, and a strong correlation was found between sMer and SLEDAI [74]. In a study measuring sMer and soluble Axl (sAxl), both were associated with antichromatin and antiphos-

**Table 1. Summary of candidate serologic markers in systemic lupus erythematosus.**

Category	Marker
<b>Epigenetic biomarkers</b>	
miRNAs	<i>miR-200a, miR-200b, miR-200c, miR-429, miR-205, miR-192, miR-142-3p, miR-181a, miR-106a, miR-17, miR-20a, miR-203 and miR-92a</i>
<b>Protein biomarkers</b>	
Cytokine	IL-6, IL-10, IL-17, IFN and IFN gene signature and sIL-2R
Autoantibody	Antinucleosome antibody and anti-C1q antibody
<b>B-lymphocyte stimulator</b>	
Complement activation products	C3a, C5a, C3d and C4d
TAM receptor and ligands	Soluble Tyro3, soluble Mer, soluble Axl and Gas6
<b>Metabolomic biomarkers</b>	Energy substrate, low-density lipoproteins, lactate, markers of oxidative stress and lipid profiles

This table is not meant to be exclusive. Some of the less-studied markers are not listed.

pholipid antibodies, and with hematologic and renal involvement [75]. However, strong correlations with SLEDAI, complement reduction and anti-dsDNA antibody titer were found for only sMer, not for sAxl.

An initial study evaluating plasma Gas6 in 107 SLE patients showed that levels were elevated in patients with neurologic or hematologic involvement, but were not correlated with disease activity markers [76]. However, two recent studies showed that Gas6 levels were elevated in lupus patients and correlated with disease activity markers, including SLEDAI, ESR and complement levels [77,78]. In addition, our receiver-operating characteristics analysis for identifying patients with active lupus showed that the area under the curve for Gas6 was higher than that of anti-dsDNA antibody, C3, C4 and ESR [77]. Several studies have suggested that the levels of free protein S may be lower in lupus patients; we showed that free protein S levels were

decreased in SLE patients with serositis, hematologic, immunologic and neurologic disorders, and correlated with C3 and C4 [76]. A recent study evaluated plasma levels of components of the TAM system in lupus patients, and showed that plasma Gas6 and all 3 soluble receptor levels were higher in lupus, and that free protein S was lower [79]. Those parameters correlated with SLEDAI scores, and Gas6 was higher in the most severe cases, while free and total protein S were lower.

These studies support the possibility that TAM kinases and their two ligands could be promising biomarkers. For further elucidation of the role of TAM and their ligands in the pathogenesis of SLE, future research efforts, including a human genetic study, are needed.

### Metabolomic biomarkers

Metabolomics is the quantitative measurement of the concentration of low molecular weight compounds

#### Executive summary

##### Definition of a biomarker & candidate serologic markers in systemic lupus erythematosus

- A biomarker is an objective measurement, characterized as a genetic, biological, biochemical, molecular or imaging event, whose alterations correlate with disease pathogenesis or manifestations.
- Candidate serologic biomarkers for diagnosis and evaluation of disease activity in systemic lupus erythematosus (SLE) are divided into categories of epigenetic, protein and metabolomic biomarkers.

##### Epigenetic biomarkers

- Most studies reported that DNA methylation and histone modifications have been observed in CD4<sup>+</sup> T cells, B cells or monocytes of lupus patients, but not in serum or plasma.
- The serum levels of *miR-200a*, *miR200b*, *miR200c*, *miR429*, *miR-205* and *miR-192* of lupus patients were lower than those of controls.
- Recent study showed increased expression of *miR-142-3p* and *miR-181a*, and decreased expression of *miR-106a*, *miR-17*, *miR-20a*, *miR-203* and *miR-92a* in SLE.

##### Protein biomarkers

- Cytokines IL-6, IL-10, IL-17, IFN and IFN gene signature, and sIL-2R, may be promising candidate serologic biomarkers for lupus activity on a pathophysiologic basis.
- Antichromatin/antinucleosome antibodies and anti-C1q antibodies have been shown to be promising markers for diagnosis or as new measures of renal involvement.
- Several studies showed that BlyS levels were correlated with disease activity and IFN- $\alpha$  levels.
- Soluble Mer, Tyro3 or Gas6 levels were correlated with disease activity markers of SLE, such as SLE Disease Activity Index.

##### Metabolomic biomarkers

- The metabolomic study utilizing nuclear magnetic resonance spectroscopy found reductions in energy substrates, increased low-density lipoproteins and increased lactate.
- Another metabolomic study with liquid chromatography/MS and gas chromatography/MS platforms exhibited reduction of energy substrates, elevation of markers of oxidative stress and inflammation markers and altered lipid profiles.
- Because the potential of metabolomics is not yet fully realized, additional prospective studies for its role as a biomarker are needed in SLE.

##### Conclusion

- Although many studies on biomarkers in SLE have been published, there is no new biomarker validated recently for clinical practice, because of the extreme heterogeneity of SLE and few prospective longitudinal and interventional studies.
- Several potential serologic biomarkers for diagnosis and assessment of disease activity are miRNA, IFN- $\alpha$ , IFN-regulated chemokines, sIL-2R, antichromatin antibody, TAM receptor and ligands and anti-C1q antibody.
- The role of epigenetic and metabolomic biomarkers in SLE must be confirmed with prospective large scale studies.

present in biological fluids [80]. Metabolites are non-peptide molecules representing the result of specific cellular processes. The change of metabolite concentration indicates the spectrum of biochemical effects induced by a disease or its treatment. Metabolomics has become feasible with the use of new technologies, such as mass spectrometry (MS) and high-resolution proton nuclear magnetic resonance. The first metabolomic study utilizing nuclear magnetic resonance spectroscopy was performed on SLE serum, and found reductions in energy substrates, increased low-density lipoproteins and increased lactate [81]. Another metabolomic study in SLE was performed with liquid chromatography/MS and gas chromatography/MS platforms. Serum from lupus patients exhibited reduction of energy substrates, elevation of markers of oxidative stress and inflammation markers, and altered lipid profiles [82]. Because the potential of metabolomics is not yet fully realized, additional prospective studies for its role as a biomarker are needed in SLE.

## Conclusion

Although many studies on pathogenesis and biomarkers in SLE have been published, there is no new biomarker validated recently for clinical practice. One reason for this is the extreme heterogeneity of SLE. The second is that there are few prospective longitudinal and interventional studies validating the utility of the biomarkers. The third is that much research on SLE biomarkers does not progress to studies on their practical application.

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## Future perspective

Recently identified candidate biomarkers need well-designed prospective studies with collaborating investigators and development of standardized assays for commercialization. Moreover, ongoing research on the pathogenesis of SLE can assist in biomarker development through use of new and powerful technologies, such as protein and antibody microarrays, proteomics and metabolomics. A recently emerging spectrum of 'omics' could be used to identify better biomarkers for diagnosis and assessment of disease activity in SLE.

## Financial & competing interests disclosure

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